RLX0120

CLINICAL STUDY PROTOCOL

Title: Multicenter, adaptive, randomized, placebo-controlled, double blind, parallel-group Phase 2/3 trial, to study efficacy and safety of two doses of raloxifene in adult paucisymptomatic COVID-19 patients.

Study Number:	RLX0120
EudraCT Number/IND:	2020-003936-25
Investigational Product:	Raloxifene
Phase of the study:	II/III, adaptive trial
Protocol Version - Date:	**Version No. 3.0 – FINAL 22/DEC/2020**

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Table of Contents

List of Abbreviations and Definitions of Terms

ADR	Adverse Drug Reaction
AE	Adverse Event
AESI	Adverse Event of Special Interest
ALCOAC	Attributable-Legible-Contemporaneous-Original-Accurate-Complete
ARDS	Acute RespiratoryDistress Syndrome
AUC	Area Under the Curve
BP	Blood Pressure
BT	Body Temperature
CFR	Code of Federal Regulations
CM	Concomitant Medications
CP	Conditional Power
CPMP	Committee for Proprietary Medicinal Products
CRA	Clinical Research Associate
CRO	Contract Research Organization
CTCAE	Common Terminology Criteria for Adverse Events
DB	Data Base
DSUR	Development Safety Update Report
EC	Ethics Committee
ECG	Electrocardiogram
e-CRF/CRF	Electronic/Case Report Form
EDC	Electronic Data Capture
EOS	End of Study
ER	Estrogen Receptors
EU	European Union
FAS	Full Analysis Set
FAV	Favipiravir
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GP	General Practitioner
HRT	Hormone replacement therapy
eICF	electronic Informed Consent Form
ICH	International Conference on Harmonisation
IP/IMP	Investigational Product/ Investigational Medicinal Product
IRB	Institutional Review Board
IUD	Intrauterine Device

NEWS	National Early Warning Score
OVX	Ovariectomised
PCR	Polymerase Chain Reaction
PD	Protocol Deviation
PI	Principal Investigator
PP	Per Protocol
PR	Pulse Rate
PT	Preferred Terms
SAE	Serious Adverse Event
SAF	Safety Analysis Set
SAP	Statistical Analysis Plan
SD	Standard Deviation
SERMs	Selective Estrogen Receptor Modulators
SO	Standard Of Treatment
SOC	System Organ Class
SPC / SmPC	Summary of Product Characteristics
SpO_2	Oxygen saturation
SUSAR	Suspected Unexpected Serious Adverse Reaction
TEAE	Treatment-emergent adverse events
TESAE	Treatment-emergent serious adverse events
TRHP	Total Raloxifene in Hydrolysed Plasma
VTE	Venous Thromboembolism

1. STUDY SYNOPSIS

Title: Multicenter, adaptive, randomized, placebo-controlled, double blind, parallel-group Phase 2/3 trial, to study efficacy and safety of two doses of raloxifene in adult paucisymptomatic COVID-19 patients.
Protocol number: Sponsor code RLX0120
EudraCT Number: 2020-003936-25
Clinical phase: Phase 2/3 adaptive trial
Timelines Enrollment period: 12 weeks Screening period: ≤ 1 week FPI-FPO: 4 weeks TX period: 2 weeks FU period: 2 weeks Post study QoL assessment: 3 months
Rationale: At present, there are no standard approaches to care paucisymptomatic patients with COVID-19 related symptoms. Currently intensive care life-support therapies represent the only effective intervention to reduce mortality for patients with critical COVID-19. Several antivirals have been currently proposed as potential effective therapies against COVID-19. Among the most promising compounds there are a broad-spectrum inhibitor of viral RNA polymerase, Favipiravir (FAV) and Remdesivir. The proven antiviral efficacy of this drugs is generally limited in COVID-19 patients by the low bioavailability in lungs. Recently through the Exscalate supercomputing platform were found several promising molecules able to fight the SARS-CoV-2, that shown low serum concentrations and high lung concentrations. Among these, raloxifene was found as a promising molecule to treat mild to moderate COVID-19 patients due to its dual activity: modulate the replication and activity of Sars-CoV-2 and interact with the estrogen receptors (that would seem to play a key role in the protection against the virus via a cascade of events including an anti-inflammatory effect). The aim of this study is to assess the efficacy of two different doses of raloxifene (60 mg and 120 mg) for preventing the evolution of COVID-19 towards severe and critical disease and their capability to reduce the mean time of viral shedding in COVID-19 patients who did not show yet severe symptoms.
Study design: Multicenter, adaptive, double-blind, randomized, placebo controlled, double blind, parallel-group, to study efficacy and safety, with the following adaptive components: - Parallel multi-arms (2 interventional arms and 1 placebo control arm); - A 2-stage sequential design (1 *interim* analysis + 1 final analysis); - Sample size re-calculation at interim stage; - Stopping rule for efficacy or futility at interim stage.
Planned nr. of countries: 3: Italy, France, Spain
Coordinating centre: Prof. Emanuele Nicastri, MD, Direttore UOC Divisione di Malattie Infettive ad Alta Intensità di Cura e Altamente Contagiose, INMI Lazzaro Spallanzani, Via Portuense, 292, 00149 Roma, Italy.
Investigational product:

TEST (T):	Hard gelatine capsule containing 60 mg of raloxifene, for oral administration.
REFERENCE (R):	Hard gelatine capsule containing placebo, for oral administration.

Dose regimen: After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), a single daily oral dose of raloxifene (60 mg Group 1, 120 mg Group 2 – treatment groups) or placebo (Group 3 - control group) will be taken on by the patients for two weeks. The patients will be randomly (1:1:1) assigned to receive either raloxifene treatment or placebo.

Treatment groups:

- Group 1:
  will receive one capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2:
  will receive two capsules of raloxifene 60 mg.

Control group:

- Group 3:
  will receive two capsules of placebo.

Objectives:

The objective of this study is to evaluate the efficacy and safety of two different doses of raloxifene orally administered compared to placebo in patients with early diagnosis of paucisymptomatic COVID-19. Efficacy will be assessed based on the proportion of patients with undetectable SARS-CoV-2 at day 7 after randomization and the proportion of patients who requires supplemental oxygen therapy and/or mechanical ventilation by day 14 after randomization. Safety will also be assessed.

Primary objectives:

- evaluation of the effectiveness of therapy in reducing the proportion of subjects who still have viruses in the upper airways after 7 days of therapy;
- evaluation of the effectiveness of therapy in reducing the proportion of subjects who requires supplemental oxygen therapy and/or mechanical ventilation within 14 days of starting therapy.

Secondary objectives:

- evaluation of the effectiveness of therapy in reducing the proportion of subjects who still have viruses in the upper airways after 14 and 28 days of therapy;
- evaluation of the effectiveness of therapy in reducing the proportion of subject patients who requires supplemental oxygen therapy and/or mechanical ventilation within 7 or 28 days of starting therapy;
- 7, 14 and 28 days drug safety and tolerability profile;
- Assessment of body temperature, blood and biochemical parameters between T0 and T28.

Endpoints:

Primary endpoints:
Virologic outcome. Proportion of participants with undetectable SARS-CoV-2 at PCR at day 7 after randomization.

Clinical outcome. Proportion of participants who does not require supplemental oxygen therapy (NEWS ≤ 2) and/or mechanical ventilation at day 14 after randomization.

Secondary endpoints:

- Proportion of participants with undetectable SARS-CoV-2 at PCR at day 14 after randomization at day 28 after randomization;

➢ Proportion of participants who does not require supplemental oxygen therapy (NEWS ≤ 2) and/or mechanical ventilation at day 7 and 28 after randomization; ➢ Proportion of patients in each NEWS category at time 7, 14 and 28 after randomization; ➢ Mean value of NEWS category at time 7, 14 and 28 after randomization; ➢ Proportion of participants with any adverse event with grade ≤ 2 according to CTCAE at day 7, 14 and 28 after randomization; ➢ Proportion of participants with any severe adverse events (grade ≥ 3 according to CTCAE) at day 7, 14 and 28 after randomization; ➢ Proportion of hospitalized participants who at the beginning of the study were at domicile isolation at day 7, 14 and 28 after randomization; ➢ Proportion of participants admitted to intensive care at day 7, 14 and 28 after randomization; ➢ Proportion of survivors at day 7, 14 and 28 after randomization; ➢ Mean variation of value of the following biomarker parameters, from base line to day 7, 14, 21 and 28 after randomization: o Complete blood cell counts; o Hepatic function (ALT, AST and bilirubin); o Coagulation (PT, aPTT and INR); o Other markers including (D-dimer, CPK, LDH); ➢ Quality of life questionnaire 3 months after the randomization.
Safety and tolerability assessments: All patients will daily self-monitor the following parameters and may call a doctor if needed.Treatment-emergent adverse events (TEAEs); vital signs (blood pressure [BP], pulse rate [PR], body temperature [BT]), transdermal oxygen saturation (SpO_2). **(see appendix 1)**.
Sequential design procedures: The group sequential design includes 3-arm 2-stage procedure allowing for early stopping rule according to efficacy and futility of at least one active arm. In case of continuation after interim analysis, the actual number of new participants will be calculated according to the observed efficacy of the best favourable and promising arm at the interim analysis. Pocock's spending functions and conditional power will be used to control the type I and II errors, considering that interim analysis will be performed when half of the initial planned patients have been reached the primary endpoints (i.e. assessments available). Bonferroni method is used to adjust for multiple endpoint and treatment comparisons.
Sample size: Sample size will significantly change according to the observed effect within the trial sample (in case of no early stop, expected sample size is between 250 and 450 randomized participants). In particular, it is plan to randomize 50 participants per arm at the interim stage (N=150) and then, in case of continuation of the study to the final stage, it is expected to randomize on average additional 174 patients, depending on sample size reassessment and possible drop of an uneffective treatment arm. Drop-outs of randomized subjects will not be replaced.
Main selection criteria: **Inclusion criteria:**

1. Subject autonomously provides informed consent prior to initiation of any study procedures;

2. Males and females ≥ 40 years old at time of enrolment;

3. Understands and agrees to comply with planned study procedures, has the availability of an email address as well as an Internet connection at domicile location;

4. Agrees to the collection of nasopharyngeal swabs and venous blood samples per protocol;

5. Has laboratory-confirmed SARS-CoV-2 infection as determined by an approved molecular test (PCR) in Europe within 10 days at the screening time;

6. Patient paucisymptomatic who complains at the screening time at least one of the following symptoms mild to moderate: fever, dyspnea, headache, cough, dysgeusia, conjunctivitis, vomiting, diarrhea, anosmia, muscle or body aches or other symptoms which in the opinion of the Investigator are part of the COVID-19 clinical picture;

7. No need of supplemental oxygen therapy, mechanical ventilation;

8. Females of child-bearing potential and with an active sexual life must not wish to get pregnant within 30 days after the end of the study and must be using at least one of the following reliable methods of contraception:
   a) Hormonal contraception, systemic, implantable, transdermal, or injectable contraceptives for at least 2 months before the screening visit until 30 days after final visit
   b) A non-hormonal intrauterine device [IUD] or female condom with spermicide or contraceptive sponge with spermicide or diaphragm with spermicide or cervical cap with spermicide for at least 2 months before the screening visit until 30 days after final visit
   c) A male sexual partner who agrees to use a male condom with spermicide
   d) A sterile sexual partner

   Female participants of non-child-bearing potential or in post-menopausal status for at least 1 year will be admitted. For all female subjects, with child-bearing potential, pregnancy test result must be negative before first drug intake on T7 and T14.

Exclusion criteria:

Patients who meet any of the following criteria are NOT eligible for inclusion in the study.

1. Being totally asymptomatic at the screening time;

2. Requires supplemental oxygen therapy or mechanical ventilation;

3. Being already under raloxifene or other SERM treatment for another medical condition at the time of randomization;

4. Being concurrently involved in another trial with IP or participation in any clinical trial with IP for 1 months before this study. The 1-month interval is calculated as the time between the last visit of the previous study and the first day of the present study (date of the informed consent signature);

5. Clinically significant abnormal physical findings which could interfere with the objectives of the study;

6. *Diseases*:
   a) history of stroke and/or venous thromboembolism;
   b) known moderate / severe renal impairment: Chronic Kidney Disease (CKD) stage 3 or higher;
   c) known liver disease (Child-Pugh Class A or higher);
   d) presence of known hypoalbuminemia;
   e) endometrial bleeding;
   f) signs or symptoms of endometrial cancer;

7. Autoimmune diseases receiving therapy at the time of randomization;

8. Risk of venous thrombosis or any condition/disease that could bring to an extended period of immobilization;

9. Ascertained or presumptive hypersensitivity to the active principles (raloxifene) and/or excipients or allergic reactions in general, which the Investigator considers may affect the outcome of the study;

10. *Medications*: in particular cholestyramine (or any ion exchange resin), medications used in treatment of early or advanced breast cancer (including adjuvant therapy), warfarin, any drug that cannot be co-administered with the experimental compound;

11. *Pregnancy:*

    a) positive or missing pregnancy test before first drug intake or day 1;
    b) pregnant or lactating women;

12. Women of childbearing potential and fertile men who does not agree to use at least one primary form of contraception for the duration of the study.

Statistical plan and data analysis:

This is a multi-arm parallel randomized controlled clinical trial with the aim to show superiority of raloxifene orally administered compared to placebo in terms of either virologic or clinical outcome.

The sample size of the study has been based on the following assumptions:

- **Virologic assumption**: Early treatment with antivirals increases the proportion of patients with undetectable SARS-CoV-2 in upper respiratory tract at day 7 after therapy from 25% to 50%;
- **Clinical assumption**: Early treatment with antivirals increases the proportion of participants who recover without need of mechanical ventilation and/or supplemental oxygen therapy by the day 14 after therapy from 50% to 75%.

Based on these assumptions, the planned sample size will allow to achieve a power greater than 80% to show superiority of raloxifene vs placebo in terms of either one primary endpoint, controlling the one-sided alpha below 0.025.

Summary statistics have been defined for quantitative variables (number of observations, mean, standard deviation, median, minimum and maximum) and qualitative variables (number and percentage per category).

The proportion of participants with undetectable SARS-CoV-2 at day 7 after randomization, and the proportion of participants who not requires mechanical ventilation and/or supplemental oxygen therapy (NEWS $\leq$ 2) at

day 14 after randomization will be analysed and compared by treatment (raloxifene vs placebo) by means of logistic regression.

All secondary endpoints will be analysed at each available timepoint by means of descriptive statistics and by appropriate parametric tests. Change from baseline value and shift tables versus baseline will be summarized for all post-baseline visits, if applicable.

AEs will be presented in terms of the number of AEs and incidence. Other safety parameters will be summarized by treatment at each available visit by means of descriptive statistics.

The Safety and the Full Analysis Set population will consist of all patients who will be randomized and received at least one dose of the investigational product. Safety population will be analysed according to the actual treatment received; Full Analysis Set population will be analysed according to ITT principle, i.e. by treatment allocation. Primary and secondary efficacy analyses will be conducted on the FAS population while the SAF population will be used for safety analyses.

A Statistical Analysis Plan (SAP) will be issued with more technical and detailed elaboration of the principal features of statistical analyses. Any deviation from the original statistical plan will be described in the Clinical Study Report.

Appendix 1

Biochemistry (T0, T7, T14, T21, T28)
Sodium
Potassium
Chloride
Urea
Uric acid
Creatinine
Calcium
Inorganic phosphorus
Fasting Glucose
Total and direct bilirubin
Total protein
Albumin
Alpha-1 Globulins
Alpha-2 Globulins
Beta Globulins
Alkaline phosphatase
AST
ALT
Gamma glutamyl transferase
Total Cholesterol
Triglycerides

Haematology (T0, T7, T14, T21, T28)
Haemoglobin
Red cell count
Packed cell volume
Mean cell volume
Mean cell haemoglobin
Mean cell haemoglobin concentration
Platelet count
White cell count
Neutrophils
Lymphocytes
Monocytes
Eosinophils
Basophils

Coagulation (T0, T7, T14, T21, T28)
Prothrombin time
Activated partial thromboplastin time
INR

Virology (T7, T14, T28)
PCR

Other markers (T0, T7, T14, T21, T28)
D-dimer;
Creatine phosphokinase (CPK);
Lactate dehydrogenase (LDH).

2. SCHEDULE OF EVALUATIONS

Study procedures	**Pre-screening**	**Screening**[1] (from day -7 to day 0)	**Baseline (T0)**	**T7**	**T14** (End of treatment)	**T21**	**T28**	**ETDV**[5]	**3 Months**
				Week 1	**Week 2**	**Week 3**	**Week 4**		
Laboratory-confirmed SARS-CoV-2 infection	X*								
Electronic Informed Consent signature		X							
Demographic data and life style recording		X							
Medical/surgical History and underlying disease		X							
Vital signs daily measurement (BP, PR, BT) and transdermal oxygen saturation (SpO2)			X						
Pregnancy Test[2] (female only)			X	X	X				
Laboratory analyses[3]			X	X	X	X	X		
NEWS score evaluation			X	X	X		X		
Inclusion/Exclusion Criteria		X							
Randomization		X							
IMP daily administration				X**					
Nasopharyngeal swabs				X	X		X		
Supplemental oxygen therapy (NEWS ≥2) and/or				X	X		X		

Study procedures	**Pre-screening**	**Screening**[1] (from day -7 to day 0)	**Baseline (T0)**	**T7**	**T14** (End of treatment)	**T21**	**T28**	**ETDV** [5]	**3 Months**
				Week 1	**Week 2**	**Week 3**	**Week 4**		
mechanical ventilation (Yes/No)									
ICU admission (Yes/No)				X	X		X		
Study drug dispensation			X						
Verify study medication dosing compliance				X	X				
AEs / SAEs / CMs Record and assessment[4]		X						X	
QoL questionnaire (EQ5D5L)									X

* Confirmation of SARS-CoV-2 infection must be available before the screening visit. Only diagnosed COVID-19 paucisymptomatic patients will be evaluated for the trial participation

** After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), patiens will take on a single daily oral dose, for a total of 14 days of treatment

1. Screening visit will be carried out within 7 days from the diagnosis of SARS-CoV-2 infection
2. Urine pregnancy test will be done at patient's domicile
3. Laboratory Analyses:

Biochemistry (T0, T7, T14, T21, T28)
Sodium
Potassium
Chloride
Urea
Uric acid
Creatinine
Calcium
Inorganic phosphorus
Fasting Glucose
Total and direct bilirubin
Total protein
Albumin
Alpha-1 Globulins

Haematology (T0, T7, T14, T21, T28)
Haemoglobin
Red cell count
Packed cell volume
Mean cell volume
Mean cell haemoglobin
Mean cell haemoglobin concentration
Platelet count
White cell count
Neutrophils
Lymphocytes
Monocytes
Eosinophils
Basophils

Alpha-2 Globulins
Beta Globulins
Alkaline phosphatase
AST
ALT
Gamma glutamyl transferase
Total Cholesterol
Triglycerides

Coagulation (T0, T7, T14, T21, T28)
Prothrombin time
Activated partial thromboplastin time
INR

Virology (T7, T14, T28)
PCR

Other marker (T0, T7, T14, T21, T28)
D-dimer;
Creatine phosphokinase (CPK);
Lactate dehydrogenase (LDH).

4. AEs will be monitored from the screening visit, immediately after electronic informed consent, up to the final visit.
5. In case of discontinuation, subjects will undergo an Early Trial Discontinuation Visit

2.1. BACKGROUND INFORMATION

In December 2019, a new identified coronavirus (SARS-CoV-2) outbreak in Wuhan causes public health crisis in China and spreads worldwide. On February 11, 2020, the World Health Organization officially named the disease caused by the new coronavirus "COVID-19". The Chinese Government takes stronger and harsher measures to control the progression of its outbreak. Meanwhile, five editions of "Diagnosis and Treatment for Novel Coronavirus-Infected Pneumonia" has been timely and countinuosly issued, which play extremely important roles in guiding the clinical management of COVID-19 nationwide in China. Common symptoms of COVID-19 include fever, cough, shortness of breath and dyspnea that may eventually progress towards acute respiratory distress syndrome and death. About 80% of patients have mild to moderate disease, 14% have severe disease and 6% are critical (namely, they develop respiratory failure, septic shock, and/or multiple organ dysfunction/failure)[1].The first known case of SARS-CoV occurred in Foshan, China in November 2002 and new cases emerged in mainland China in February 2003. The first emergence of MERS-CoV occurred in June 2012 in Saudi Arabia. These events demonstrated that the threats of CoVs must not be underestimated and that it is important to advance the knowledge on these viruses, their replication and interactions with the hosts to develop new pharmacological treatments and vaccines.

In this situation, new strategies that can mitigate the impact of COVID-19 on the National Health System are urgently needed. Specifically, these strategies should be aimed at: a) preventing patients with mild COVID- 19 to progress towards severe/critical disease, for lightening the impact on hospitals; b) shortening time of viral shedding in order to curb individual infectivity and reduce the average number of secondary cases. At present, there are neither standard approaches nor approved drugs to care patients with COVID-19 and intensive care life-support therapies represent the only effective intervention to reduce mortality for patients with critical COVID-19. In addition, there are no standard approaches to care patients with mild to moderate COVID-19 related symptoms. Several antivirals have been currently proposed as potential effective therapies against COVID-19. Among the most promising compounds there are a broad-spectrum inhibitor of viral RNA polymerase, Favipiravir (FAV) and Remdesivir. The proven antiviral efficacy of this drugs is generally limited in COVID-19 patients by the low bioavailability in lungs.

Recently through the Exscalate supercomputing platform were found several promising molecules able to fight the SARS-CoV-2, that shown low serum concentrations and high lung concentrations. Among these, raloxifene was found as a promising molecule to treat mild to moderate COVID-19 patients due to its dual activity: modulate the replication and activity of Sars-CoV-2 and interact with the estrogen receptors (that would seem to play a key role in the protection against the virus via a cascade of events including an anti-inflammatory effect).

The aim of this clinical study is to assess the efficacy of the well-known drug, Raloxifene, already marketed in other indications, emerged from a repurposing approach as a promising agent for preventing the evolution of COVID-19 towards severe and critical disease.

2.2. RELEVANT NON-CLINICAL PHARMACOLOGY

Several preclinical animal studies to investigate effects of raloxifene on bone were performed. These studies, which have been conducted mainly in rats and cynomolgus monkeys, have used ovariectomised (OVX) or sexually immature animals as a model of estrogen deficiency. Several analytical methods

have been applied to evaluate different aspects of the effects on bone including analysis of bone mineral density, biomechanical strength, histomorphometry, and biochemical markers of bone metabolism.The doses used (0.001-30 mg/kg/day in rats and 1-5 mg/kg/day in monkeys) result in an exposure consistent with that in women receiving the proposed daily dose of 60 mg.
The duration of some studies (particularly the two chronic studies of 12 months in OVX rats and 2 years in OVX cynomolgus monkeys) is adequate to demonstrate the beneficial effects on bone turnover.

There is sufficient preclinical evidence that raloxifene has a tissue-selective estrogen-receptor (ER) agonist or antagonist profile. Concerning the mimetic actions, there is sufficient preclinical evidence supporting the anti-osteoporotic properties of raloxifene in conditions of estrogen deficiency.
Furthermore, the studies also demonstrated that raloxifene lowers serum cholesterol levels in OVX rats, rabbits and monkeys mimicking actions of estrogens (however raloxifene is less potent than estrogens in reducing cholesterol levels and does not prevent the development of atherosclerotic alterations).
On the other hand, raloxifene behaves in reproductive sites, such as mammary tissue and uterus, like an ER antagonist. Its lack of uterine stimulation distinguishes raloxifene from other antiestrogens, like tamoxifen, which have partial agonist activity on the endometrium.[2]

General pharmacodynamics studies

A wide range of studies has been performed *in vitro* and *in vivo* (rabbit, mouse, guinea pig). At oral doses up to 600 mg/kg/day raloxifene showed no marked cardiovascular, renal, gastrointestinal or smooth muscle activity.

Pharmacokinetics/Toxicokinetics studies

The majority of the pharmacokinetic studies were performed in rats, mice and monkeys, with a few studies performed in dogs. Many studies relied on ^{14}C-raloxifene; authentic raloxifene was also measured by HPLC.

The data have shown good oral absorption, extensive “first-pass” metabolism in the intestinal mucosa and liver to form glucuronide conjugates, a high level of plasma protein binding and faecal excretion of raloxifene and its metabolites via the bile.

A wide range of oral doses was used in pharmacology studies (1-5 mg/kg) with much higher doses in toxicology studies (up to 600-1700 mg/kg). In general, plasma concentrations of raloxifene increased with dose in mice, rats and monkeys, but increases were not always proportional, especially at high doses. A similar pattern was observed in postmenopausal women given doses up to 600 mg orally.

The results seen in the animal models are consistent with the higher exposure levels in animals compared to humans. Systemic exposure to raloxifene after 1 year or more of daily dosing was approximately 41, 505 and 13 times greater in mice, rats and monkeys, respectively, than in postmenopausal women given 60 mg/day for two years. It can be concluded that animals were appropriately exposed to the compound in these studies[2].

2.3. A SUMMARY OF TOXICOLOGY DATA

Raloxifene was evaluated for toxicity in two rodent (mice and rats) and two non-rodent (Beagle dogs and rhesus monkeys) species, by oral and intraperitoneal (in rats) route. Acute toxicity was low. No mortality was observed with doses up to 5000 mg/kg orally to mice and rats. Few rats showed toxicity after i.p. injection of a 2000 mg/kg dose of raloxifene.

Repeated dose toxicity was investigated in mice (3 months), rats (up to 1 year), dogs (6 months) andcynomolgus monkeys (up to 1 year) following oral administration. In general, raloxifene was welltolerated and observations from these studies were considered to reflect the pharmacodynamic actionsof raloxifene on reproductive tissues and estrous cycle due to the SERM-like activity. A no-effectlevel was not determined.

Reproductive toxicity studies was performed in rats andrabbits. The observed effects were all consistent with the known action of raloxifene on the estrogenreceptor. At doses of 0.1 to 10 mg/kg/day in female rats, raloxifene disrupted estrous cycles of femalerats during treatment, but did not delay fertile matings after treatment termination and only marginallyreduced litter size, increased gestation length, and altered the timing of events in neonataldevelopment. When given during the preimplantation period, raloxifene delayed and disrupted embryoimplantation resulting in prolonged gestation and reduced litter size but development of offspring toweaning was not affected. Teratology studies were conducted in rabbits and rats. In rabbits, abortionand a low rate of ventricular septal defects (≥ 0.1 mg/kg) and hydrocephaly(≥ 10 mg/kg) were seen. In rats, retardation of foetal development, wavy ribs and kidney cavitationoccurred (≥ 1 mg/kg).
Results from the standard battery of *in vitro* and *in vivo* mutagenicity tests show that raloxifene is devoid of genotoxic potential.

Carcinogenicity studies to assessthe oncogenic/carcinogenic potential of raloxifene has been performedin mice (21 month study) and in rats (24 month), with appropriate exposure multiples over human doses. An increased incidence of ovarian neoplasias was observed in both species, as well as an increase of serum ALT levels. Treatment of female rodents with raloxifene throughout their lives produced specific hormonal imbalances. Such imbalances are known to result in ovarian tumours in rodents, which have not been observed in women who have received raloxifene[2].

2.4. PHARMACOKINETICS AND PRODUCT METABOLISM

Prevention indication

The pharmacokinetics of raloxifene has been studied in healthy volunteers, postmenopausal women, and patients with hepatic impairment (5 subjects with cirrhosis).

The mean oral bioavailability of raloxifene was low, approximately 2%. However, the median estimate of absorption of total drug-related substance is much higher and the absolute oral bioavailability is dependent on metabolic interconversion between raloxifene and its glucuronides. Most of the raloxifene in the systemic circulation appears to be derived from deconjugation of the glucuronides.

Raloxifene is a high clearance drug, with a clearance approximately equal to liver blood flow. It undergoes significant enterohepatic circulation.

Following single oral doses (study GGGI), the T_{max} for raloxifene was approximately 6 hours (ranging from 1-24 hours), $t_{1/2}$ was 33 hours (23-92 hours). According to a study with ^{14}C-raloxifene, peak concentrations of total raloxifene in hydrolysed plasma (TRHP) represented more than 99% of total radioactivity in plasma, indicating rapid glucuronidation. The terminal half-lives of raloxifene, TRHP and glucuronides were similar, ranging from 15.6 to 21.8 hours. After repeated oral doses, raloxifene kinetics (e.g. elimination rate constant, clearance and volume of distribution) was linear with respect to time. The raloxifene concentration-dose relationship was linear, but not dose-proportional.

Raloxifene and its glucuronides are highly bound to plasma proteins, including both albumin (>95%) and alpha-1-glycoprotein (89%). Neither raloxifene nor the glucuronides are distributed into the cellular component of blood.

The only metabolites detected in plasma and urine are glucuronide conjugates (raloxifene-4'-glucuronide, -6'-glucuronide and –6,4'-diglucuronide). No oxidative metabolites have been found.

Raloxifene and its metabolites are primarily excreted in the faeces, less than 6% is recovered in urine. The amount of unchanged raloxifene in urine is negligible.

The effect of food on the pharmacokinetic parameters of raloxifene is not significant.

Population pharmacokinetic analyses (NONMEM model) confirmed the results from individual pharmacokinetic studies. The pharmacokinetics of raloxifene and TRHP are characterised by large intra- and inter-subject variability. Pharmacokinetics was not affected by age, dose or ethnic factors. Self-reported alcohol consumption (>3 drinks per week) did not influence raloxifene pharmacokinetics. In smokers, raloxifene clearance was approximately 20% greater than in nonsmokers, but this difference is less than the 30% within-subject variability.

A formal study in patients with renal impairment has not been performed, as renal excretion of raloxifene is a minor pathway. Dose adjustment does not appear necessary in subjects with mild renal impairment on the basis of population pharmacokinetic analysis. In the absence of data, raloxifene is contraindicated in severe renal impairment.

In subjects with hepatic cirrhosis and mild hepatic impairment (Child-Pugh Class A), the C_{max} and AUC for plasma raloxifene, TRHP and raloxifene-glucuronide (single dose of raloxifene) were significantly higher than in healthy control subjects. A statistically significant association was observed between raloxifene-glucuronide, TRHP and raloxifene AUC versus serum total bilirubin. Raloxifene is contraindicated in hepatic impairment, including cholestasis.

A number of tablet formulations have been used in clinical trials. The different formulations and the market-image tablets have been shown to be bioequivalent.

Treatment indication

Population pharmacokinetic analyses from patients (postmenopausal women with osteoporosis) in Study GGGK demonstrated that the pharmacokinetics of raloxifene were not clinically significantly affected by age, body weight or BMI, cigarette smoking, chronic alcohol use or decreased renal function. However, in the absence of data, raloxifene is contraindicated in severe renal impairment.

Interaction studies

In *in vitro* studies, significant plasma protein binding interactions were not observed with warfarin, phenytoin, tamoxifen or testosterone.

In *in vivo* studies, an approximately 40% reduction in systemic raloxifene exposure was observed after cholestyramine, therefore cholestyramine should not be co-administered with raloxifene. Calcium carbonate and aluminium hydroxide/magnesium hydroxide antacid did not modify the absorption and exposure to raloxifene and metabolites. Oral ampicillin therapy resulted in lower raloxifene concentrations, but the interaction is probably not of significant consequence during short-term antibiotic treatment. Raloxifene reduces the efficacy of warfarin treatment. A significant decrease in the peak and AUC of the prothrombin time response to warfarin was observed after multiple doses of raloxifene; careful monitoring of prothrombin time is necessary over several weeks. Concomitant administration of raloxifene resulted in increase in C_{max} of digoxin (by less than 5%), but did not affect overall exposure.

In the population pharmacokinetic analyses, histamine H1-receptor antagonists, laxatives and fibre were associated with slight decreases in observed average raloxifene concentrations.

For other frequently co-administered drugs such as paracetamol, non-steroidal anti-inflammatory drugs (e.g. acetylsalicylic acid, ibuprofen and naproxen), oral antibiotics, H1 antagonists, H2 antagonists and benzodiazepines, no clinically relevant effects of co-administration on raloxifene plasma concentrations were identified.

The Phase I study GGIP, which assessed the pharmacokinetic interaction of raloxifene after multiple administration of methilprednisolone in postmenopausal women with no concomitant hormone replacement therapy, showed that raloxifene has no effect on the pharmacokinetics of methylprednisolone given as a single dose[2].

2.5. A SUMMARY OF CLINICAL DATA

The core clinical documentation for the prevention of osteoporosis in postmenopausal womenindication consisted of three main Phase III studies (randomised, multicentre, double-blind, placebocontrolled parallel groups) and several Phase II studies.The core clinical documentation for the treatment of osteoporosis in postmenopausal women indication was based on 3-year data from a single, very large Phase III study (randomised, multicentre, double-blind, placebo-controlled parallel group) (Study GGGK) involving 7705 women with osteoporosis.

2.5.1. Main Phase III Efficacy Studies: Prevention of Osteoporosis

These main studies enrolled a total of 1754 postmenopausal women for long-term double-blind treatment with placebo or raloxifene (30, 60 or 150 mg/day). Study GGGH enrolled subjects who had undergone hysterectomies, and included an unopposed conjugated estrogen treatment arm (0.625 mg/day). Two-year interim reports with intention-to-treat and per protocol analyses have been provided.

The study population was women (mean age of 54 years) who were 2 to 8 years postmenopausal, except for the study GGGH in which women who were 0 to 15 years postmenopausal were included. The

inclusion criteria comprised subjects with T-scores for lumbar spine BMD ranging from -2.5 to +2.0 inclusive. The osteopenic population, with T-scores from -2.5 to -1, is considered most relevant for the claimed indication; approximately 50% of the study population had osteopenia.

The primary efficacy criteria in these studies were change and percent change from baseline to endpoint in lumbar spine (L1-L4) and total hip BMD. Secondary efficacy criteria included markers of bone metabolism (serum osteocalcin, serum total and bone-specific alkaline phosphatase, urinary type I collagen fragment C-telopeptide), serum lipids, measures of coagulation and fibrinolytic activity and assessment of endometrial thickness[2].

Effects on bone

GGGF was a European study to establish the effects of long-term therapy with three doses ofraloxifene compared to placebo on BMD of the lumbar spine and total hip. Randomised patientsreceived placebo (n=150) or raloxifene 30 mg (n=152), 60 mg (n=152), or 150 mg (n=147)daily.
After 24 months of treatment, the mean percentage changes from baseline for lumbar spine BMD were –0.8% (placebo), +1.3% (30 mg), +1.6% (60 mg) and +2.2% (150 mg). At the level of the total hip, the changes were –0.8% (placebo), +1.0% (30 mg), +1.6% (60 mg) and +1.5% (150 mg). Increases in BMD were statistically significant compared to placebo at all measurements sites (except the distal and ultradistal radius) for all doses of raloxifene. The results are clinically significant and relevant with respect to the proposed indication. The biochemical markers of bone metabolism decreased significantly in all raloxifene groups versus placebo, consistent with decreased bone turnover.

GGGG was a US study to establish the effects of long-term therapy with three doses of raloxifene compared to placebo on BMD of the lumbar spine and total hip. Randomised patients received placebo (n=136) or raloxifene 30 mg (n=136), 60 mg (n=134), 150 mg (n=138) o.d.
Over 24 months of treatment, the mean percentage changes from baseline for lumbar spine BMD were –1.2% (placebo), +0.4% (30 mg), +0.8% (60 mg) and +0.8% (150 mg). At the level of the total hip, the changes were –0.8% (placebo), +1.0% (30 mg), +1.2% (60 mg) and +1.6% (150 mg). Although the effects of raloxifene on lumbar spine and total hip BMD (except for the radius and total body BMD) and bone markers (except for the specific alkaline phosphatase in the raloxifene 30 mg group) were clearly distinguishable from placebo, the effects on lumbar spine BMD were weaker compared to study GGGF. Nevertheless, the effect of raloxifene 60 mg/day was statistically significant compared to placebo. Raloxifene 30 mg appeared to be less effective than 60 mg and 150 mg.

GGGH was a study to establish the effects of long-term therapy with two doses of raloxifene or unopposed conjugated estrogen compared to placebo on BMD of the lumbar spine and total hip. Randomised patients received placebo (n=152) or raloxifene 60 mg (n=152), 150 mg (n=157), or conjugated equine estrogen 0.625 mg (n=158) o.d.
After 24 months of treatment, the mean percentage changes from baseline for lumbar spine BMD were –1.6% (placebo), +0.2% (60 mg), +0.4% (150 mg) and +3.8% (estrogen). At the level of the total hip, the changes were –0.5% (placebo), +0.8% (60 mg), +0.5% (150 mg) and +2.4% (estrogen).
At both doses, raloxifene had a weak, but statistically significant effect compared to placebo on lumbar and total hip BMD. However, the magnitude of the effect of estrogen was significantly greater. The actual “mean benefit” from raloxifene and estrogen compared to placebo was 1.8% and 5.0% respectively. A slight downward slope in the lumbar spine and total hip BMD curves was noted during the second year of raloxifene, but not in the estrogen groups. This was only observed at the 150-mg/day

doses, not at 30 mg/day, 60 mg/day or in other phase III studies. However, the mean BMD values remained at, or slightly above baseline for up to 24 months. Raloxifene and estrogen had similar effects on biochemical markers of bone metabolism, but the magnitude of the effect ofestrogen was also clearly more pronounced than that of raloxifene.

Raloxifene has a well-documented effect compared with placebo on lumbar spine and total hip BMD. Although qualitatively the effects of raloxifene on BMD and biochemical markers of bone metabolism are similar to those of estrogen, their magnitude is less. Hence, evidence of fracture benefit in patients with osteoporosis was required, even for the proposed osteoporosis prevention claim.
A 2-year interim analysis of an ongoing double blind, placebo-controlled phase III study in osteoporotic postmenopausal women was provided. This study enrolled a total of 7705 patients randomly assigned to one of three treatment groups: raloxifene 60 mg, 120 mg or placebo. Both postmenopausal women with osteoporosis and established osteoporosis (prevalent vertebral fractures) were included. The diagnostic criteria and evaluation for incident fractures are appropriate.
Both raloxifene doses significantly reduced the proportion of patients having at least one incident vertebral fracture during 24 months of treatment compared to vitamin D and calcium supplemented placebo. The reduction in the risk of experiencing at least one incident fracture was in the range of 35% to 55% in patients with and without prevalent vertebral fracture at baseline, showing the same range of figures as those reported in observational studies evaluating the anti-fracture efficacy of estrogen (HRT).
The 3-year analysis of this study was submitted in 1999 to support the extension of the indication to include treatment of postmenopausal osteoporosis. In contrast to estrogen, no data are currently available on the ability of raloxifene to prevent non-vertebral fractures.
Similar efficacy related subgroup analyses were carried out in studies GGGF and GGGG. Raloxifene was observed to improve BMD regardless of BMI (Body mass index). Population pharmacodynamic analyses for the main efficacy studies supported the proposed dose of raloxifene 60 mg/day[2].

Effects on serum lipid profile

Raloxifene has clear effects on serum lipid profile. At 60 mg/day, raloxifene decreases total cholesterol by approximately 3-6%, LDL cholesterol by 4-10%, HDL cholesterol by 1-4%, LDL/HDL ratio by 7% and total cholesterol/HDL ratio by 5% over 24 months. However, HDL cholesterol has decreased slightly more during placebo than raloxifene. Raloxifene does not affect serum triglyceride levels, which may increase during estrogen treatment. It also slightly decreased lipoprotein A and apolipoprotein B levels. Overall, the effects of raloxifene (60 mg/day) on serum lipids were similar to those of estrogen, but weaker. Raloxifene did not have adverse effects on lipid profile. In contrast to estrogen, no data are yet available to demonstrate the benefit of raloxifene on atherosclerotic cardiovascular disease. Similar efficacy related subgroup analyses carried out in studies GGGF and GGGG indicated that in smokers, the reduction in LDL cholesterol was less pronounced than in nonsmokers[2].

2.5.2. Main Phase III Osteoporosis Treatment Efficacy Study (GGGK)

GGGK was a Phase 3, multicentre, double-blind, placebo-controlled, randomised clinical study which compared raloxifene 60 mg/day, raloxifene 120 mg/day and placebo in the treatment of postmenopausal women with osteoporosis or established osteoporosis. It was designed as a completed 36-month core treatment phase and a 12-month extension phase. It randomly assigned 7705 eligible patients to one of the three treatment groups (placebo, N=2576; raloxifene 60 mg, N-2557 and raloxifene 120 mg,

N=2572). All patients were supplemented with calcium and vitamin D. The study population was postmenopausal women (mean age 66.5 years) who had osteoporosis.

GGGK was designed as two separate substudies. Substudy I included patients with osteoporosis by BMD criteria (BMD T-score <-2,5 in either the femoral neck or lumbar spine). Substudy II included patients who were osteoporotic by BMD and also had at least one moderate or 2 mild prevalent vertebral fractures or at least 2 moderate prevalent vertebral fractures regardless of BMD. Approximately twice as many patients were enrolled into Substudy I (n=5064) as into Substudy II (n=2641)[2].

Primary efficacy objectives of this study and each substudy were to assess the effects of raloxifene 60 mg and 120 mg compared with placebo on incidence of new vertebral fractures and change and percentage change in lumbar spine (L1-L4) and femoral neck BMD. Additional secondary efficacy objectives included the effects of raloxifene compared with placebo on nonvertebral fractures, on total body and radial BMD, on biochemical markers of bone turnover, and on serum lipids and other markers of cardiovascular risk.

Effects on bone

Based on the three-year analysis of data from Study GGGK, each dose of raloxifene (60 mg and 120 mg per day) in each substudy statistically and clinically significantly decreased the proportion of women with at least one adjudicated new incident vertebral fracture vs placebo. In patients in Substudy I, raloxifene 60 mg-day was associated with a 47% reduction (RR 0.53, 95% CI 0.35, 0.79) in the risk of new vertebral fractures and raloxifene 120 mg/day was associated with a 38% risk reduction (RR 0.62, CI 0.44, 0.93). In patients in Substudy II, there was a 31% risk reduction (RR 0.69, CI 0.56, 0.86) with raloxifene 60 mg and a 49% risk reduction (RR 0.51, CI 0.40, 0.65) with raloxifene 120 mg. The proportion of women with at least one adjudicated incident vertebral fracture was statistically significantly lower in the 120-mg group than in the 60-mg group in Substudy II, but not in Substudy I. However, no statistically significant differences were observed between the raloxifene doses for the reduction of new clinical or multiple vertebral fractures in patients in Substudy II or I. Overall, raloxifene 60 mg/day was associated with a 41% reduction and raloxifene 120 mg/day was associated with a 52% reduction in the risk of new clinically apparent vertebral fractures. The CPMP has agreed that the overall risk/benefit assessment favours the 60-mg dose over the 120-mg dose for the treatment of postmenopausal osteoporosis regardless of baseline disease severity. No effect on hip fractures has been demonstrated.

Each dose of raloxifene in each substudy statistically significantly increased BMD at the lumbar spine and femoral neck by 2-3% vs placebo at 36 months. Total body and ultradistal radius BMD increased 1-2% vs placebo at 24 months. The doses were equivalent in their effects on BMD in both patients with and without prevalent vertebral fractures. Raloxifene treated patients had 15 to 25% greater reductions in markers of bone turnover (osteocalcin, bone specific alkaline phosphatase and Urinary Type I Collagen Fragment/Cr) than placebo treated patients who had 10 to 20% reductions. Each dose of raloxifene in each study was effective in reducing skeletal turnover[2].

Effects on serum lipids and markers of cardiovascular risk

In the osteoporosis treatment population, raloxifene also has clear effects on serum lipids and other markers of cardiovascular risk. Compared with placebo, raloxifene treated patients had consistent

decreases of 6 to 7%, 10 to 12%, 10 to 13% and 7 to 9% in total cholesterol, LDL-cholesterol, fibrinogen and Apolipoprotein B, respectively at 36 months. There were no effects of raloxifene on HDL-cholesterol or glycosylated hemoglobin. While median percentage reductions in triglycerides were observed in raloxifene treated patients, the reductions were greater for the placebo group. A 3% increase in Apolipoprotein A was observed with raloxifene treatment compared with placebo. However, no data are available to demonstrate benefit of raloxifene on cardiovascular disease[2].

2.5.3. Safety

Raloxifene is registered in EU with Centralized Procedures since 1998 for the treatment and prevention of osteoporosis in postmenopausal women with a recommended dose of 60 mg/day and is intended for long term use. This means more than 20 years in post-marketing exposure, in addition to extensive clinical studies in supporting the indication approved.

Raloxifene safety profile is supported by a huge number of data from long-term treatments; more than 7.700 post-menopausal women with osteoporosis have been exposed to raloxifene (the MORE Study) with a long follow-up period.

In addition, as reported in the European SmPC, raloxifene therapy for 8 years did not significantly affect the risk of cardiovascular events in patients enrolled in the osteoporosis treatment study. Similarly, in the RUTH study (multinational, double-blind, randomized, placebo controlled trial conducted in postmenopausal women at risk for major coronary events) in which a total of 10,101 women were enrolled and randomized to one of two therapy groups, raloxifene did not affect the incidence of myocardial infarction, hospitalization acute coronary syndrome, stroke or overall mortality, including overall cardiovascular mortality, compared to placebo. The relative risk of venous thromboembolic events observed during raloxifene treatment was 1.60 (CI 0.95, 2.71) when compared to placebo, and it was 1.0 (CI 0.3, 6.2) when compared to estrogen or hormonal replacement therapy. The risk of a thromboembolic event was greater in the first four months of therapy.

Then, other 19,747 postmenopausal women at increased risk of invasive breast cancer were included into STAR trial and the safety results were combined with the above-mentioned studies and enclosed in the current European SmPC[3]. Therefore, in this population the safety profile of Raloxifene is very well known and described into Patient Information approved by Regulatory Agencies.

Even if not authorized for use in men, male subjects have been exposed and Raloxifene is well tolerated in men[4]. Given at the dose of 120 mg/day for 6 months to male healthy subjects, raloxifene increases levels of LH, FSH and total testosterone[5].

The safety profile was assessed also in special populations of patients (renal impairment, hepatic dysfunction). Risk of venous thromboembolic events is considered negligible for short term treatments and concomitant use of antiplatelet therapy. Regarding higher doses of raloxifene doses up to 600 mg/day were used in clinical studies in healthy postmenopausal women.

In several dose-ranging clinical studies (raloxifene 200 mg and 600 mg), no significant difference between active treatment groups was observed. The two doses of raloxifene demonstrated comparable efficacy, but the higher dose (600 mg) resulted in a greater frequency of hot flushes[3].

Raloxifene up to 150 mg once-a-day or twice-a-day was used in clinical studies in postmenopausal women with osteoporosis to establish the effects of long-term therapy and prevention indication.

In osteoporosis prevention the proportion of subjects treated with 150 mg/day Raloxifene reported accidental injuries[3].

Lastly, raloxifene (60 mg/day) + current standard of care (SOC) treatment at the time [i.e.pegylated interferon (PegIFN) α2a plus ribavirin (RBV)], has been used in Japan for the treatment of postmenopausal women with genotype 1b chronic hepatitis C. It improved the efficacy of SOC in the treatment of postmenopausal women with chronic hepatitis C and didn't show an increase of the risks. Only one raloxifene -treated patient discontinued the drug because of a systemic rash following 2 weeks of treatment[6].

The assessment of safety should take the difference in the target population into consideration. Raloxifene is approved for the treatment and prevention of osteoporosis in postmenopausal women. It has been suggested that women with osteoporosis are at greater risk of venous thromboembolism (VTE) than non-osteoporotic patients[7]. On the contrary, this is not the case for paucisymptomatic out-patients with COVID-19. The lack of risk for VTE is reflected in the recommendations made for the management of such patients[8]: "In non-hospitalized patients with COVID-19, there are currently no data to support the measurement of coagulation markers (e.g., D-dimers, prothrombin time, platelet count, fibrinogen)" "For non-hospitalized patients with COVID-19, anticoagulants and antiplatelet therapy should not be initiated for prevention of venous thromboembolism (VTE) or arterial thrombosis, unless there are other indications".

In conclusion, raloxifene has been extensively studied and administered in the post marketing setting since 22 years, in post-menopausal women (long term use) with a resulting positive safety profile in the authorised conditions for use. The drug has also been studied in men, and no additional safety risk was highlighted.

Although the safety profile of raloxifene in the treatment of COVID-19 is not known, considering the planned treatment duration (see the overall study design), the dosage and the target population (paucisymptomatic), no significant major risk could be anticipated. Safety will be closely monitored through the reporting and collection of adverse event and the timely evaluation of potential safety signal.

The table below gives the adverse reactions and frequencies observed in treatment and prevention studies involving over 13,000 postmenopausal women along with adverse reactions arising from postmarketing reports. The duration of treatment in these studies ranged from 6 to 60 months. The majority of adverse reactions have not usually required cessation of therapy.
The following convention has been used for the classification of the adverse reactions: very common (≥1/10), common (≥1/100 to <1/10), uncommon (> 1/1,000 to < 1/100), rare (> 1/10,000 to < 1/1,000), very rare (<1/10,000)[3].

Blood and lymphatic system disorders *Uncommon*: Thrombocytopenia [a]
Nervous system disorders

*Common:*Headache, including migraine [a] *Uncommon:* Fatal strokes
Vascular disorders *Very common*: Vasodilation (hot flushes) *Uncommon:* Venous thromboembolic events, including deep vein thrombosis, pulmonary embolism, retinal vein thrombosis, superficial vein thrombophlebitis, Arterial thromboembolic reactions [a]
Gastrointestinal disorders *Very common*: Gastrointestinal symptoms a such as nausea, vomiting, abdominal pain, dyspepsia
Skin and subcutaneous tissue disorders *Common:* Rash [a]
Musculoskeletal and connective tissue disorders *Common*: Leg cramps
Reproductive system and breast disorders *Common:* Mild breast symptoms a such as pain, enlargement and tenderness
General disorders and administration site conditions *Very common:* Flu syndrome *Common:* Peripheral oedema
Investigations *Very common:* Increased blood pressure [a]

[a] Term(s) included based on postmarketing experience[3].

2.6. DISEASE REVIEW AND STUDY RATIONALE

Coronaviruses (Covs) are a large family of viruses belonging to the family Coronaviridae. The limited number of coronaviruses known to be circulating in humans were considered to cause mild infections and they were regarded in the past as relatively harmless respiratory human pathogens. However, in the last years the emergence of the severe acute respiratory syndrome coronavirus (SARS-CoV) and the Middle East Respiratory Syndrome (MERS) virus revealed that some coronaviruses can cause severe and sometimes fatal respiratory tract infections in humans[9].

Investigations of the epidemiological and clinical characteristics, and outcomes of patients infected by SARS-CoV-2 demonstrated that the infection causes clusters of severe respiratory illness similar to the known SARS-CoV. The symptoms of human infection with SARS-CoV-2 are generally fever, fatigue, dry cough and dyspnea. Noteworthy, a considerable percentage of COVID-19 cases rapidly progress to severe and critical types, among which acute lung injury (ALI) and acute respiratory distress syndrome (ARDS) are the most common complications, resulting in a large number of pneumonia hospitalized patients requiring supplemental oxygen, mechanical ventilation, or even ECMO. Pulmonary edema is a detrimental feature as well as a key causal factor of ALI/ARDS.

In this situation, new strategies that can mitigate the impact of COVID-19 on the National Health System are urgently needed. Specifically, these strategies should be aimed at: a) preventing patients with mild COVID-19 to progress towards severe/critical disease, for lightening the impact on hospitals; b) shortening time of viral shedding in order to curb individual infectivity and reduce the average number of secondary cases. At present, there are neither standard approaches nor approved drugs to care patients with COVID-19 and intensive care life-support therapies represent the only effective intervention to reduce mortality for patients with critical COVID-19. In addition, there are not standard approaches to care patients with mild to moderate COVID-19 related symptoms. Several antivirals have been currently proposed as potential effective therapies against COVID-19. Among the most promising compounds

there are a broad-spectrum inhibitor of viral RNA polymerase, Favipiravir (FAV) and Remdesivir. The proven antiviral efficacy of this drugs is generally limited in COVID-19 patients by the low bioavailability in lungs.

From a clinical development perspective, the identification and development of new drugs is a lengthy process not adequate to face the emergency of the immediate global challenge generated by the COVID-19 outbreak, so the repurposing approach of drugs already tested, "safe in man", or already approved for different therapeutic uses is considered a rapid solution, since the pharmacokinetic, toxicological, and manufacturing data are available and validated for immediate application in clinical settings[10].

To this aim, the analysis of a compound library containing commercial drugs and clinical candidates "safe in man" or characterized up to late clinical stage was conducted to identify a number of molecules able to inhibit SARS-Cov-2 replication by using Computer Associated Drug Design (CADD) techniques, and in vitro screening and testing.

In particular, E4C (Exscalate4CoV) is a public-private consortium supported by the European Commission's Horizon 2020 programs for projects to counter the Coronavirus pandemic and improve the management and care of patients. At the core of E4C is Exscalate (EXaSCalesmArtpLatform Against paThogEns), at present the most powerful and cost-efficient intelligent supercomputing platform in the world able to screen extremely huge "chemical libraries" of 500 billion molecules, and process more than 3 million molecules per second, with the aim to predict and select the best molecules for further characterization. The combination of advanced CADD (Computer Associated Drug) with high throughput biochemical and phenotypic screening allows rapid evaluation and shortening of discovery time of new drugs, in an approach extremely useful in case of pandemic viruses and other pathogens, where an immediate response and the identification of effective treatments are of paramount importance. Exscalate supercomputing platform were found several promising molecules able to fight the SARS-CoV-2, that shown low serum concentrations and high lung concentrations. Among these, raloxifene was found as promising molecule to treat paucisymptomatic COVID-19 patients.

Raloxifene is the generic name of 1-[6-Hydroxy-2-(4-hydroxyphenyl)benzo[b]thien-3-yl]-1-[4-[2-(1-piperidinyl)ethoxy]phenyl]methanone and is a selective benzothiophene Estrogen Receptor Modulator (SERM) with lipid lowering effects and activity against osteoporosis, approved for the treatment and prevention of osteoporosis in postmenopausal women.

Three main reasons support the rationale of using raloxifene against Convid19 infection.

The first resides in the observed gender disparity in COVID-19 severe cases and in fatality rate that has been reported in China, where the infection rate among males and females was similar, but the death rate among males was 4.7% compared with 2.8% for females. Likewise, in Italy a higher death rate in male patients (14.8%) than in female patients (8.2%) was reported by the Italian National Institute of Health.

Several factors, genetic, hormonal and behavioural, can contribute to the observed gender disparity, although experimental data are lacking so far. It is well-known that male and female subjects respond differently to many viral infections, or to pathogens in general due to a more intense and stronger immune responses, either innate and adaptive, to viral infection, that favours viral clearance. Sex hormones have shown to influence sex-specific response to viral infection by directly modulating

immune responses[11]. A recent paper reports results of a cross-sectional study of COVID-19 patients in China[12] demonstrating that females have a better prognosis than males, non-menopausal women have shorter length of hospital stays, and AMH and estradiol are negatively correlated with COVID-19's severity. Further, there is a negative correlation between estradiol and the levels of IL-6, IL-8, IL-2R and TNF-α, which are significantly correlated with disease severity or composite endpoint.

It has to be noted that estrogen is able to upregulate the expression of ACE2, the functional receptor for SARS-CoV as well as for the recently identified SARS-CoV-2. A recent publication potentially correlates sex hormones to disease severity by relating pre-existing chronic diseases and insulin resistance to defective ER signalling in humans[13].

In animal models, ERα knockout mice of both sexes present insulin resistance, glucose tolerance and obesity, all comorbidities associated with COVID-19 aggravation[14].

Previous results in literature (both animals and humans) highlight sex-specific differences in susceptibility to SARS-CoV and perhaps other coronavirus infections. Experimental studies in male and female mice infected with SARS-CoV, have shown that male mice have a higher susceptibility to SARS-CoV infection and a higher mortality, compared to females, consistently with the human disease. Ovariectomy in female mice or treatment with estrogen antagonists increased the death rate of females, which could be rescued by treatment with agents belonging to SERM class. Those experiments in animal models emphasized an evident protective role of estrogens in coronavirus infection, and a protective action of SERM in a relevant in vivo model. The results are consistent with the sex-bias observed in human SARS-CoV-2 infections and provide mechanistic insights into the differences in disease severity in men and women[15].

Scientific literature highlights sex differences in immune response and its influence on the incidence and severity of diseases. Considering trauma, shock, and infection, the female sex is associated with advantageous outcomes. Since the hormonal context exerts an important influence on the homeostasis control and the defense mechanisms, it is important to consider the beneficial effects and pleiotropic role of estrogens with regard to providing better control of defense and the different immune cells involved, in addition to cardiovascular system protection and flow maintenance[16].

Direct and indirect mechanisms underlying the effects of these compounds were investigated, both in relevant preclinical models[17] and in clinical trials (NCT04359329), and the results show a possible positive effect in viral infections suggesting that estradiol and estrogen-related compounds could play a major role in antiviral therapies for SARS-CoV-2 as potential new agents to treat COVID-19 patients.

The second resides in the well described and documented antiviral activity of raloxifene. In particular the antiviral activity of SERMs (Estrogen Receptor Modulator) were mainly focused on three infections: human immunodeficiency virus (HIV), hepatitis C virus (HCV), and Ebola virus (EBOV). Tamoxifen, first generation SERM, was found active against HIV, HCV, and herpes simplex virus 1 (HSV-1)[18]. The antiviral rationale has been further confirmed in models of SARS-CoV and MERS-CoV where classical SERMs showed the capacity to delay virus infection, in a similar way to what was seen with Ebola virus infection[19] for which hypotheses of mechanism of action were also generated[20], thus highlighting the evidence that estrogen and estrogen-related compounds can play a relevant role in antiviral therapies targeting SARS-CoV-2.

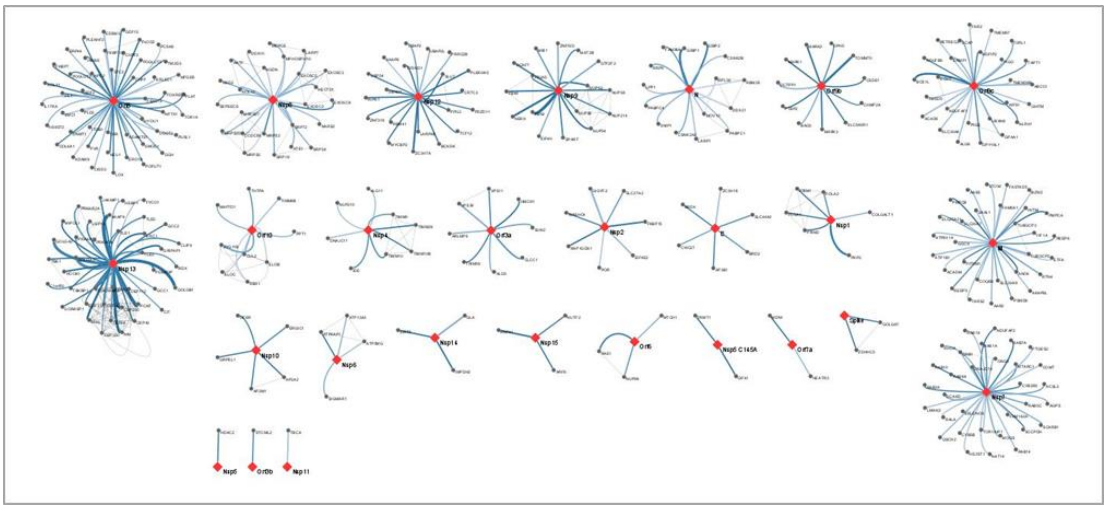
The third resides in data obtained so far through the repositioning programs, with specific deepenings on Raloxifene interaction with Covid proteases, the validation of a poly-pharmacology hypothesis, and, finally, the in vitro characterization.

Indeed, E4C (Excalate for Covid) selected, by using an integrated approach between the EXSCALATE platform and the virtual screening protocols, raloxifene as a clinical candidate against SARS-CoV-2, predicting the high probability of the drug to interact with the main SARS-CoV-2 protease, the 3C-like protease, as main result of the parallel screening on different SARS-CoV-2 target's proteins.

Moreover, the potential of raloxifene in the treatment of COVID-19 with a specific focus on the mechanisms underlying the observed drug effect and complex interactions within biological systems involving SERMs has been explored. Specifically, raloxifene and its activity against SARS-CoV-2 have been evaluated with the aim to validate a poly-pharmacology hypothesis.

Two systems were considered:

a. Genes identified in GWAS (Genome-Wide Association Study) as relevant for viral response. They are: ANKRD32, CDRT4, PSMD13, ERO1L, LZTFL1, XCR1, FYCO1, IFNAR2, CXCR6, CCR9, AP000295.9, AK5
b. Molecular networks so far identified of SARS-CoV-2-host interactome (359 elements) and consensus on the modulated expression during SARS-CoV-2 infection (14052 elements) (in the figure below the SARS-CoV-2-host interactome is reported; in red the viral proteins).



The following 3 molecular pathways are believed to be mainly involved and attributable to raloxifene:

1. A pathway including the genes modulated by raloxifene molecular target, specifically ESR2, and connected with inflammation;

2. A pathway including the genes expressed in the lungs, modulated by raloxifene molecular target, specifically ESR that, when deregulated, are linked to severe asthma, in agreement with the genes enrichment in GWAS studies whose unfavourable variants cause worse respiratory consequences;
3. A pathway directly modulated by the virus, both during the cell entry phase and replication phase, where the virus not only binds some cell proteins, but they are upstream of other proteins modulated by expression in the 14052 element consensus network, and downstream of the raloxifene-controlled pathways.

Finally, Raloxifene was tested on SARS-CoV 2 VeroE6 cells for antiviral assay. This assay is based on viral infection of VeroE6 cells followed by monitoring of cytopathic and antiviral effects. Data were generated by two different groups and in both cases an IC50 value in 1-5 microMolar (μM) range was determined.

This value is the best value so far found within the class of SERMs, and is in line with in vitro outcomes from previous experiments conducted with raloxifene on other viruses. To be noted that the range of activity of Raloxifene (1-5 μM) perfectly suits with the distribution of raloxifene in tissue (in particular lungs). Indeed, despite a low serum bioavailabily, raloxifene concentrated at tissues level and exerts its cytopathic and antiviral effects, overcoming some of the limitations showed by other antiviral drugs currently in use as remdesivir, that reaches low levels in lungs.

2.6.1. Alternative treatments

Nowadays there are no approved drugs for the treatment of paucysimptomatic COVID-19 patients.

2.6.2. Risk - benefit evaluation

The clinical pharmacology of raloxifene has been adequately studied. The minimum effective dose was not reliably established by the short-term dose ranging studies.

In 3 well conducted, double blind clinical trials, the primary and secondary criteria of efficacy are relevant. Raloxifene was statistically significantly superior to placebo (calcium supplementation only) with respect to the primary efficacy criteria. Although the three-raloxifene doses examined (30, 60 and 150 mg) were not statistically different and the dose response relationship was weak, the population pharmacodynamic analyses support the proposed raloxifene dose of 60 mg/day. Qualitatively the effects of raloxifene on bone mineral density and resorption are similar to those of estrogen, but of lesser magnitude. Moreover, a reduction in the risk of incident vertebral fractures has been demonstrated in women with osteoporosis compared to placebo.

In studies involving over 2000 women with a treatment duration ranging from 2-24 months, the majority of undesirable effects did not usually require cessation of therapy. Discontinuation of therapy due to any clinical adverse experience occurred in 10.7% of 581 raloxifene-treated patients and 11.1% of 584 placebo-treated patients. The treatment-emergent events associated with the use of raloxifene that occurred with a significant difference between raloxifene and placebo treatment were: venous thromboembolic events (occurred in a frequency of 0.8%), superficial vein thrombophlebitis, modest increase of vasodilatation (hot flushes), leg cramps and peripheral oedema.

When comparing raloxifene patients (n=317) with continuous combined HRT (n=110) or cyclic HRT (n=205) patients in some clinical trials, the incidence of breast symptoms and uterine bleeding in raloxifene treated women was significantly lower than in women treated with either form of HRT. Raloxifene does not have adverse effects on the lipid profile. No clinically significant changes in vital signs and safety laboratory tests have been seen.

The Phase III study GGGK showed that at 36 months raloxifene 60 mg/day and 120 mg/day statistically and clinically significantly decreased the proportion of women with at least one adjudicated new incident vertebral fracture vs placebo regardless of baseline disease severity. Both raloxifene 60 mg and 120 mg effectively increased lumbar spine, femoral neck and total body BMD and decreased markers of bone turnover. Raloxifene significantly decreased LDL cholesterol and other intermediate markers of cardiovascular disease (fibrinogen) without a concomitant rise in serum triglycerides.

Raloxifene, in the osteoporosis treatment population, did not induce endometrial proliferation or vaginal bleeding and was not associated with an increased risk of uterine or endometrial malignancy. Raloxifene did not cause breast pain and was associated with a reduction in the risk of breast cancer (median duration 40 months). Vasodilatation and leg cramps were side effects also observed in thispopulation. The only serious risk was that of VTE and the risk continues to be similar to that reported for estrogen use. Current or past history of VTE is a contraindication to raloxifene therapy.

The CPMP agreed that the benefit/risk was positive for the proposed indications. It was also concluded that the overall risk/benefit assessment favours the 60 mg dose over the 120 mg dose. Therefore the CPMP considered that the data presented were sufficient to recommend that the indications prevention and treatment of postmenopausal osteoporosis should be granted.

In March 2003, the sections 4.4 and 5.1 of the SPC were updated on the basis of the cumulative four years results on new clinical safety and efficacy data of the MORE (Comparison of Raloxifene Hydrochloride and Placebo in the Treatment of Postmenopausal Women With Osteoporosis) study. The study confirmed that treatment with raloxifene for 4 years decreased the rate of new vertebral fractures by 46% in osteoporotic patients. In addition, in the 4th year alone, raloxifene reduced the new vertebral fracture risk by 39% (during the 4th year, patients were permitted the concomitant use of bisphosphonates, calcitonin and fluorides). There was no significant difference in the number of nonvertebral fractures. A cumulative analysis also showed a reduction of clinical vertebral fractures.

No indicators for an increased risk of malignant endometrial tumours were shown. The overall rate of breast cancer was reduced in the raloxifene groups as compared to the placebo group. raloxifene treatment compared to placebo reduced the risk of total breast cancer by 62%, the risk of invasive breast cancer by 71% and the risk of invasive estrogen receptor (ER) positive breast cancer by 79%. The decrease was explained by the decrease of ER-positive (invasive) tumours.A significant increase in the number of venous thromboembolic events was seen in the raloxifene groups, especially in the early phase of the treatment. However, this risk is already acknowledged in the SPC. No significant differences between the groups were seen in the ECG and in the reporting of clinical cardiac and cerebrovascular events. There were favourable changes in the lipid profile, except for a small increase in serum triglycerides.

Hormone replacement therapy aggravates certain complications of pelvic floor relaxation, but safety information following 3 years of raloxifene treatment supported that raloxifene treatment did not increase pelvic floor relaxation and pelvic floor surgery.

Potential benefit of Raloxifene in COVID-19 patients treatment

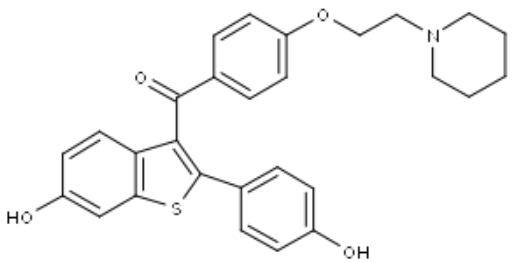
Distribution of oral raloxifene is concentrated into body tissue with relevant levels in lungs, kidneys and liver, where they are higher than in serum[21]. Raloxifene exerts its pharmacological activity at very low circulating concentrations (nM) but with wide distribution in the tissues[22] and high concentrations reached in non-reproductive tissues highly sensitive to viral infections, like lungs where the drug has a better distribution than Remdesivir, thus suggesting that a targeted distribution and concentration of raloxifene into the lungs can be achieved together with parallel minimization of side effects due to distribution in unwanted organs. High need exists of anti-COVID-19 drugs with good distribution into lungs[23] to optimize tissue distribution and maximize the inhibition of viral replication.

Taken together, these data suggest that ER modulation may be a suitable pharmacological approach for preventing/attenuating the cytokine storm and inflammation associated with COVID-19 and in particular the use of SERMs, specifically raloxifene, may represent a promising pharmacological option. Such a therapeutic approach would be particularly useful for treatment of both male and female patients in early phase with mild/moderate symptoms of the disease, in order to prevent or mitigate the possible evolution towards more serious and dangerous forms of the disease, due to the onset of the cytokine storm.

Furthermore, given the encouraging data obtained with raloxifene so far, if the ability of raloxifene to reduce the mean time of viral shedding in COVID-19 patients who did not show yet severe symptoms (paucisymptomatic patients), is confirmed, it could certainly help to reduce the risk of a second epidemic wave.

2.6.3. Description of the Investigational Product

Raloxifene is the generic name of 1-[6-Hydroxy-2-(4-hydroxyphenyl)benzo[b]thien-3-yl]-1-[4-[2-(1-piperidinyl)ethoxy]phenyl]methanone, having the below chemical structure:



The compound is a selective benzothiophene Estrogen Receptor Modulator (SERM) with lipid lowering effects and activity against osteoporosis, approved for the treatment and prevention of osteoporosis in postmenopausal women. Raloxifene is the active ingredient of branded (Optruma®-Eli Lilly or Evista®-Daiichi Sankyo) or generic drugs (RaloxifeneTeva), and is marketed as 60 mg oral tablets. The drug is registered in Europe since 1998 for the treatment and prevention of osteoporosis in postmenopausal women, and in the US is registered since 1997 for treatment and prevention of osteoporosis and reduction of risk of invasive breast cancer in postmenopausal women.

Mechanism of action

Raloxifene is a second-generation SERM with a double activity as estrogen agonist in some tissues (bone, lipid metabolism) and estrogen antagonist in others, like endometrium and breast, producing some of the estrogen's beneficial effects without producing its adverse events. Raloxifene decreases bone resorption and overall bone turnover, thus increasing bone density with little or no effect on the endometrium, resulting in no predisposition to uterine cancer. The drug is able to lower total cholesterol LDL in the serum, with no effect on HDL.

Raloxifene, like Estradiol (E2), crosses the cytoplasmic membrane and the nuclear membrane and binds to the Estrogen Receptor (ER) in the nucleus. The benzothiophene ring of raloxifene binds to the ER with a comparable affinity to E2. The rationale for the estrogen antagonist effects of the drug on uterine and breast tissues could reside in the ER blockade mediated by raloxifene.

In bone and in other non-reproductive tissues, raloxifene bound to ER (RLX-ER) should activate a specific sequence of DNA known as the Raloxifene Responding Element (RRE), a group of genes that regulates the synthesis of specific cell proteins responsible for the estrogen agonist effect of the drug in these non-reproductive tissues. In addition, raloxifene exerts two other direct actions on bone tissue: 1) decrease up to 50% of interleukin-6 (IL-6) production and 2) decrease up to 30% of the production of tumor necrosis factor α (TNF-α).

3. OVERALL STUDY DESIGN AND INVESTIGATIONAL PLAN

3.1. STUDY OBJECTIVES

The objective of this study is to evaluate the efficacy and safety of two different doses of raloxifene orally administered compared to placebo in patients with early diagnosis of paucisymptomatic COVID-19. Efficacy will be assessed based on the proportion of patients with undetectable SARS-CoV-2 at day 7 after randomization and the proportion of patients who requires supplemental oxygen therapy and/or mechanical ventilation by day 14 after randomization. Safety will also be assessed.

3.2. STUDY ADMINISTRATIVE STRUCTURE

This study will be performed in clinical centers located in Europe (Italy, France, Spain). At each study center, the Principal Investigator (PI) will be responsible for ensuring that the investigation is conducted according to the signed Investigator agreement, the protocol, GCP guidelines, and local regulations.

The PI at each study center will be responsible for the management of the study, which will consist of maintaining the study file and the patient records, corresponding with the IRB/IEC, and completing the electronic case report form (eCRF) and reporting SAEs within 24 hours of initial awareness.

The PI is responsible for supervising any individual or party to whom the investigator delegates trial related duties and functions conducted at the trial site. If the investigator/institution retains the services of any individual or party to perform trial-related duites and functions, the investigator/institution should ensure this individual or party is qualified to perform those trial-related duties and functions and should implement procedures to ensure the integrity of the trial-related duties and functions performed and any data generated.

3.3. OVERALL STUDY DESIGN

This clinical study will be a multicenter, adaptive, double-blind, randomized, placebo controlled, parallel-group, to study efficacy and safety, with the following adaptive components:

- Parallel multi-arms (2 interventional arms and 1 placebo control arm);
- A 2-stage sequential design (1 interim analyses + 1 final analysis);
- Sample size re-calculation at interim stage;
- Stopping rule for efficacy or futility at interim stage.

The study will be a total of 5 weeks in duration: a screening period of 7 days, followed by 2 weeks of double-blind treatment and a 2-week follow-up period. At the end of the screening period, patients meeting the entry criteria for this study will be randomized (1:1:1) to 1 of 3 treatment groups (group 1, group 2 or group 3) and instructed by a physician on the correct self-administration of the treatment and completion of the patient diaries.

After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), a single daily oral dose of raloxifene (60 mg Group 1, 120 mg Group 2 – treatment groups) or placebo (Group 3 - control group) will be take on by the patients for two weeks.

Treatment groups:

- Group 1: will receive one capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2: will receive two capsules of raloxifene 60 mg.

Control group:

- Group 3: will receive two capsules of placebo.

Following the completion of the double-blind treatment or premature discontinuation during the double-blind phase, patients will be followed up for safety assessments until the planned last study visit. In case of early treatment termination an End of Follow Up will be seeked at the same timepoint.

3.3.1. Rationale for Selection of dose, control group and treatment schedule in the study

A double-blind study design was adopted to minimize systematic bias. Randomization is expected to minimize patient selection bias and increase baseline comparability between treatment groups. The use of placebo control is critical to the study design for, providing an accurate estimate of the additive benefit of pharmacotherapy.

Even if the actual dosage form of raloxifene approved for the treatment of osteophorosis is 60 mg, due to its well assessed safety profile (for 60 mg and 120 mg), for the execution of this study will be used also the raloxifene 120 mg, for a total of 3 treatment arms (see section 2.5).

The group sequential design includes 3-arm 2-stage procedure allowing for early stopping rule according to efficacy and futility of at least one active arm. In case of continuation after interim analysis, the actual number of new participants will be calculated according to the observed efficacy of the best favourable and promising arm at the interim analysis.

Patients with insufficient therapeutic response, tolerability issues, or worsening of symptoms may be discontinued at any time during the study.

4. SELECTION OF STUDY POPULATION

For the purpose of the study the following definition are applied.

A case of COVID-19 is a person with SARS-CoV-2 infection as determined by an approved molecular test (PCR) from an adequate sampling of upper respiratory tract irrespective of clinical signs and symptoms.

A paucisymptomatic case of COVID-19 who does not need immediate admission to hospital and has a national early warning score (NEWS) ≤ 2.

NEWS score has been revised on 2019 by England and Wales National Health Service as following[24]:

Element	Score						
	3	2	1	0	1	2	3
Respiratory rate	≤ 8		9-11	12-20		21-24	≥ 25
SpO_2	≤ 91	92-93	94-95	≥ 96			
Oxygen		Yes		NO			
Systolic Blood Pressure	≤ 90	91-100	101-110	111-219			≥ 220
Pulse	≤ 40		41-50	51-90	91-110	111-130	≥ 131
ACVPU				A			C,V,P,U
Temperature, ºC	≤ 35.0		35.1-36.0	36.1-38.0	38.1-39.0	≥ 39.1	

ACVPU: Alert, Confusion, Voice, Pain, Unresponsive

4.1. INCLUSION CRITERIA

To be eligible for inclusion into this study, each patient must fulfil the following inclusion criteria.

1. Subject autonomously provides informed consent prior to initiation of any study procedures;
2. Males and females ≥ 40 years old at time of enrolment;
3. Understands and agrees to comply with planned study procedures, has the availability of an email address as well as an Internet connection at the domicile location;
4. Agrees to the collection of nasopharyngeal swabs and venous blood samples per protocol;
5. Has laboratory-confirmed SARS-CoV-2 infection as determined by an approved molecular test (PCR) in Europe within 10 days at the screening time;
6. Patient paucisymptomatic who complains at the screening time at least one of the following symptoms mild to moderate: fever, dyspnea, headache, cough, dysgeusia, conjunctivitis, vomiting, diarrhea, anosmia, muscle or body aches or other symptoms which in the opinion of the Investigator are part of the COVID-19 clinical picture;

7. No need of supplemental oxygen therapy, mechanical ventilation;

8. Females of child-bearing potential and with an active sexual life must not wish to get pregnant within 30 days after the end of the study and must be using at least one of the following reliable methods of contraception:
    a. Hormonal contraception, systemic, implantable, transdermal, or injectable contraceptives for at least 2 months before the screening visit until 30 days after final visit
    b. A non-hormonal intrauterine device [IUD] or female condom with spermicide or contraceptive sponge with spermicide or diaphragm with spermicide or cervical cap with spermicide for at least 2 months before the screening visit until 30 days after final visit
    c. A male sexual partner who agrees to use a male condom with spermicide
    d. A sterile sexual partner

    Female participants of non-child-bearing potential or in post-menopausal status for at least 1 year will be admitted. For all female subjects, with child-bearing potential, pregnancy test result must be negative before first drug intake, on T7 and T14.

4.2. EXCLUSION CRITERIA

Patients who meet any of the following criteria are NOT eligible for inclusion in the study.

1. Being totally asymptomatic at the screening time;

2. Requires supplemental oxygen therapy or mechanical ventilation;

3. Being already under raloxifene or other SERM treatment for another medical condition at the time of randomization;

4. Being concurrently involved in another trial with IP or participation in any clinical trial with IP for 1 months before this study. The 1-month interval is calculated as the time between the last visit of the previous study and the first day of the present study (date of the informed consent signature);

5. Clinically significant abnormal physical findings which could interfere with the objectives of the study;

6. *Diseases*:
    a) history of stroke and/or venous thromboembolism;
    b) known moderate / severe renal impairment: Chronic Kidney Disease (CKD) stage 3 or higher;
    c) known liver disease (Child-Pugh Class A or higher);
    d) presence of known hypoalbuminemia;
    e) endometrial bleeding;

f) signs or symptoms of endometrial cancer;

7. Autoimmune diseases receiving therapy at the time of randomization;

8. Risk of venous thrombosis or any condition/disease that could bring to an extended period of immobilization;

9. Ascertained or presumptive hypersensitivity to the active principles (raloxifene) and/or excipients or allergic reactions in general, which the Investigator considers may affect the outcome of the study;

10. *Medications*: in particular cholestyramine (or any ion exchange resin), medications used in treatment of early or advanced breast cancer (including adjuvant therapy), warfarin, any drug that cannot be co-administered with the experimental compound;

11. *Pregnancy:*

    a) positive or missing pregnancy test before first drug intake or day 1;
    b) pregnant or lactating women;

12. Women of childbearing potential and fertile men who do not agree to use at least one primary form of contraception for the duration of the study.

4.3. ASSIGNMENT OF SUBJECT NUMBER

All the subjects will sign the electronic informed consent, via GENIUS ENGAGE for the present study.
Genius ENGAGE is the EXOM solution for the electronic Informed Consent (eICF) with multimedia tools like audio, video, and links, accessible through a any device like a mobile, tablet or laptop, fully integrated with the the eCRF.
All the patients will be coded with “unique subject identifiers”. The unique subject identifier consists of the sponsor study code (i.e. study code), the 3-digit site number (i.e. 101), the 4-digit screening number (e.g. S001, S002, etc.).
Study code, site number and screening number are separated by slashes (“/”). Randomization numbers will be assigned automatically via EXOM GENIUS IWRS^{TM}.

CONFIDENTIAL

5. STUDY MEDICATION

5.1. PRESENTATION, STORAGE, PACKAGING AND LABELING OF THE INVESTIGATIONAL MEDICINAL PRODUCT

After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), a single daily oral dose of raloxifene (60 mg Group 1, 120 mg Group 2 – treatment groups) or placebo (Group 3 - control group) will be take on by the patients for two weeks. The patients will be randomly (1:1:1) assigned to receive either raloxifene treatment or placebo.

Treatment groups:

- Group 1:
  will receive one capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2:
  will receive two capsules of raloxifene 60 mg.

Control group:

- Group 3: will receive two capsules of placebo.

5.1.1. Presentation of Investigational Medicinal Product

The investigational product is in the form of oral 60 mg capsules containing the active ingredient raloxifene. Raloxifene 60 mg capsules are orange hard gelatine capsules, size 0.

Raloxifene capsules contain the following excipients: sodium starch glycolate, citric acid monohydrate, microcrystalline cellulose, dibasic calcium phosphate, poloxamer 407, magnesium stearate, hypromellose, lactose monohydrate, titanium dioxide (E171) and macrogol / PEG 4000.

Placebo capsules are orange hard gelatine capsules, size 0 containing the inert excipient microcrystalline cellulose.

5.1.2. Manufacturing, Packaging and Labelling of IMP

Raloxifene and Placebo capsules are packaged in PVC-ALU blisters in the form of patient kits, numbered to maintain blinding. Raloxifene and placebo capsules do not require special storage conditions.

Raloxifene and placebo capsules are manufactured by STM PHARMA PRO S.r.l. according to current Good Manufacturing Practice requirements.

Medication labels will comply with the Competent Authority requirements and will be printed in a Multilanguage format where needed.

The formulation labelling will report all the information requested according to the Annex 13 to the Good Manufacturing Practice (published by the Commission in The rules governing medicinal products in the European Community, Volume 4) as follows:

a. Name, address and telephone number of the sponsor, contract research organisation or investigator (the main contact for information on the product, clinical study and emergency unblinding)
b. Pharmaceutical dosage form, route of administration, quantity of dosage units
c. The batch and/or code number to identify the contents and packaging operation
d. A study reference code allowing identification of the study, site, investigator and sponsor if not given elsewhere
e. The study subject identification number/treatment number and where relevant, the visit number
f. The name of the investigator (if not included in (a) or (d))
g. Directions for use (reference may be made to a leaflet or other explanatory document intended for the study subject or person administering the product)
h. "For clinical study use only" or similar wording
i. The storage conditions
j. Period of use (use-by date, expiry date or re-test date as applicable), in month/year format and in a manner that avoids any ambiguity
k. "Keep out of reach of children"

Labels will be in local language.

The hospital pharmacist (Investigators in those countries where it is required that shipment is made directly to them) is responsible for receipt, proper storage and delivery of study drug to qualified and duly authorized study nurse. Partially used or unused study drug boxes should be destroyed on site (and documentation of destruction provided to Dompé farmaceutici s.p.a.) or returned to Dompé farmaceutici s.p.a., at the end of the study. The Hospital Pharmacist or Investigator, who will keep a cumulative inventory and dispensing records, will maintain all supplies under adequate security. Adequate record of receipt, use or loss of drug will be retained.

5.1.3. Supply, Storage and Handling of IMP

An appropriate number of packages will be initially sent to the Hospital Pharmacy as soon regulatory/ethics approvals have been obtained and a financial agreement has been signed with the site. IMP re-supply will be planned on demand, according to enrolment rate.

The IMP must be kept at a temperature not exceeding 30°C and must not be frozen.

A temperature probe will accompany the drug on shipment. Temperature range reached during shipment will be verified on receipt, so that potential stability concerns during shipment can be investigated and appropriate action taken.

Once received at the Hospital Pharmacy, the Pharmacist (or designee) will check the package and acknowledge the receipt; any deviations from expected package content (inconsistency, damages)

should be immediately reported to Dompé (or designee) and the use of the drug suspended until authorization for its continued use has been given by Dompé (or designee).

The IMP must be stored in a secure location, in a temperature controlled room. Temperature records must be available for the CRA to review; any deviations from the recommended storage conditions should be immediately reported to Dompé (or designee) and the use of the drug suspended until authorization for its continued use has been given by Dompé (or designee).

Treatment Box will be brought to the patient's domicile by a properly qualified and duly authorized study nurse once randomized. Each patient will receive the Treatment Box matching his/her randomization number.

5.1.4. Blinding

Appearance, including packaging and labelling, of the IMP (capsules, packaging) will not allow to recognize actual treatment (either raloxifene or placebo).

As described in Section 8.8, during the trial, blinding may be broken by the Investigator for emergency purposes only, where knowledge of the blinded treatment could influence further patient care. Unblinding will be performed by Sponsor Pharmacovigilance for safety reasons and in case of Serious Adverse Events qualifying for regulatory reporting (SUSAR), in line with regulatory requirements.

Study blind will be broken after database lock.

5.2. DOSE, ROUTE AND SCHEDULE OF IMP ADMINISTRATION

The experimental treatment will be orally self-administered by the patients who signed the electronic informed consent.

After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), a single daily oral dose of raloxifene (60 mg Group 1, 120 mg Group 2 – treatment groups) or placebo (Group 3 - control group) will be taken on by the patients for two weeks.

Treatment groups:

- Group 1: will receive one capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2: will receive two capsules of raloxifene 60 mg.

Control group:

- Group 3: will receive two capsules of placebo.

5.3. ACCOUNTABILITY OF THE IMP

All IMP supplies will be accurately and adequately accounted for, from receipt at the depot up to deliver to the patients; each step will be recorded in study-specific documents, as detailed below, that will be available for verification by the CRA at each monitoring visit.

Drug accountability records at the depot level will include:

1. Shipment/receipt confirmation of the IMP at the depot of the logistic provider, as per documentation provided by or on behalf of Dompé.
2. IMP stored at the depot at any time as per IMP Accountability Log, to include IMP received, delivered to the patients, accidentally destroyed, or disposed of at expiration or at the end of the trial.
3. IMP delivered to each patient/treatment cycle, as per the eCRF.

The administration of the IMP (date/time for each administration) will be recorded by the patient on an electronic patient Diary.

eDiary data will be collected via dedicated tele-medicine platform named EXOM Genius ROSA.

This is a dual way (Investigator - Patient) private, study specific, fully encrypted, multichannel application for remote patient visits and monitoring. It is 21 CFR Part 11 & GDPR compliant and ISO 27001 Class IIA medical device.

eDiary data will be then transferred directly to the eCRF and will be checked in a timely manner by the Investigator.

All the records will include, as appropriate: dates, quantities (kits, treatment boxes, blisters, capsules), batch numbers, expiration dates (if applicable), and any unique code numbers assigned to the IMP and/or patients, so that the following can be adequately documented:

- the patients were provided with the doses specified by the protocol/amendment(s),
- the IMP provided to the site has been fully reconciled at the site.

The CRA will review the IMP records above and check actual content of IMP boxes/blisters (both unused and used) and will reconcile quantities up to the level of capsules, prior to making arrangements for their disposal.

Any IMP supply remaining at the end of the trial will be returned to Sponsor or its delegate or disposed of on site.

5.4. CONCOMITANT MEDICATION

5.4.1. Prior and concomitant medications

All concomitant medications taken by the patients during the Screening will be recorded by the Investigator in the eCRF.

All the concomitant medications (administration started after the first dose of IMP up to the end of trial participation) will be recorded by the patient on the electronic patient Diary and reported in the appropriate section of the eCRF.

All the details as per the eCRF fields (sequential number, drug name, indication, starting dose, start/stop date, route of administration) will be recorded. No change in dose will be tracked.

The following agents do not need to be recorded: homeopathic medications; elective vitamins and minerals; osmotic laxatives and locally acting antacids; topical medication.

6. STUDY PROCEDURE AND ASSESSMENTS

A schedule for the tests and evaluations to be conducted in this study is found in the Schedule of Evaluations (See section 2). Details are reported below. Any deviation from these protocol procedures will be reported in the study-specific Protocol Deviation (PD) form.

For all measurements, the actual date and time of assessment, including date of sampling and self-monitoring assessment, will be recorded in the eCRF. Where a time window is acceptable, this is clearly indicated in the following sections.

6.1. SCREENING AND RANDOMIZATION VISITS

6.1.1. Randomization

Patients will be randomized in a 1:1.1 fashion to either:

GROUP 1 (1 capsule of raloxifene 60 mg and 1 capsule of placebo)

GROUP 2 (2 capsules of raloxifene 60 mg)

GROUP 3 (2 capsules of placebo)

The randomization groups will be generated with a computer procedure by the method of random permuted blocks. The randomization system GENIUS IWRS will be prepared and implemented by the CRO and the platform will be part of the eCRF. The randomization platform includes also a special functionality for electronically obtaing the code breaking in case of an urgent medical need.

Moreover as a backup unblinding procedure, each site will be provided with sealed evenvelopes containing the name of the assigned treatment.

Similarly, the randomization list will be provided to the facility responsible for IMP packaging/labelling for the purpose of IMP preparation.

The randomization code will be broken at study completion, i.e. when the last patient has completed his/her last follow-up visit, and once the database has been locked.

6.2. STUDY VISITS AND FOLLOW-UP ASSESSMENTS

Since all paucisymptomatic patients with COVID-19 must remain in trust isolation at home or at an appropriate location, a specifically trained and authorized team of study nurses will take care of collecting blood samples and nasopharyngeal swabs at patient's domicile.
For each patient the study will be a total of 5 weeks in duration: a screening period of 7 days, followed by 2 weeks of double-blind treatment and an 2-week follow-up period. Three months after the randomization patients will be asked to fill in a Quality of life questionnaire.

At the end of the screening period, patients meeting the entry criteria for this study will be randomized (1:1:1) to 1 of 3 treatment groups (group 1, group 2 or group 3) and instructed on the correct self-administration of the treatment, self measurements of blood pressure, heart rate, transdermal oxygen and body temperature through individual medical devices and completion of the patient diaries.

After an administration of two oral doses in the first day of treatment (one dose in the morning and one dose in the evening, each dose administered with 2 capsules containing 60 mg of the active substance or placebo), a single daily oral dose of raloxifene (60 mg Group 1, 120 mg Group 2 – treatment groups) or placebo (Group 3 - control group) will be taken on by the patients for two weeks.

After the treatment period patients will record in the patient diary their parameters self measured (blood pressure, heart rate, transdermal oxygen and body temperature) for an 2-week follow up period.

Three months after the randomization patients will be asked to fill in a Quality of life questionnaire.

6.2.1. Treatment arms

Treatment groups:

- Group 1: will receive one capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2: will receive two capsules of raloxifene 60 mg.

Control group:

- Group 3: will receive two capsules of placebo.

6.2.2. Standard Patient monitoring

All patients will daily self-monitor their parameters and may call the Investigator if needed. In case the enrolled person cannot perform the daily self-monitoring, a close relative may support him, provided that all possible preacautions and safety devices are used. Patients will be provided with individual electronic device for measuring blood pressure, heart rate, body temperature and transdermal SpO2.
Patients will be also provided with an electronic patient diary in which report the information about the daily drug self-administration, the above mentioned parameters recorded, the AEs (if any) and the concomitant medications (if any).

The Investigator will remotely regularly assess NEWS and evaluate the need for immediate visit or hospitalization.

The Investigator will contact the General Practitioner who will decide to directly involve emergency service for immediate visits for critical patients presenting:

A. SpO2 ≤ 95%;
B. Systolic blood pressure >180 mmHg or <100 mmHg
C. Body temperature >39°C or less than 35.0 °C;
D. Heart rate >110 bpm or less than 40 bpm;
E. Sensorial deterioration according to doctor's judgment.

Patients will receive a Study Nurse visit at the day 0, 7, 14, 21 and 28.

At day 7, 14, 28 patients will be tested for viral shedding. At day 0, 7, 14 patients will undergo pregnancy test (female only). At day 0, 7, 14, 21, 28 patients will undergo blood test including hepatic function, renal function, coagulation, LDH, CPK and complete blood counts.

Venous blood samples (~ 20 mL) will be collected from a forearm vein.

Healthcare workers approaching COVID-19 patients will use personal protective equipment including a FFP3 (or FFP2) mask, gloves, gown and googles. FFP3 should be used always in case of any procedure on respiratory tract (including nasopharyngeal swab).

6.2.3. Other therapies allowed

Any concomitant medication that the participant receives during the participation in this study will also be recorded (see Sections 5.4.1 and 6.2.2.). Specifically, dose, posology, frequency of administration, start and end date and reason of use will be required and collected.
As for SmPC the following medications are not allowed as concomitant medications: cholestyramine (or any ion exchange resin), medications used in treatment of early or advanced breast cancer (including adjuvant therapy), any antiretroviral medication, warfarin or other coumarin derivatives (the combination with raloxifene may cause modest decreases in the prothrombin time).

6.2.4. Safety monitoring and individual stopping rules

Safety data to be recorded:

- vital signs: blood pressure (systolic and diastolic) and heart rate will be measured daily the patient him/herself using an electronic device. BP and HR measurements will be done with the subject comfortably seated on a chair, with the middle of the cuff on the upper arm at the level of the right atrium (the midpoint of the sternum). Also the body temperature will be measured by 10:00 am using an electronic device.
- SpO2 will be measured by transdermal oximetry to be collected once daily concomitantly with the BP / HR measurement.
- laboratory tests will be performed on venous blood samples taken from the patient at domicile (drawings performed by a nurse sent to the patient's domicile and delivered to the study site's laboratory).The lab results should be reviewed by the Investigator and clinically relevant changes recorded as AEs. Training to the personell in charge of the samples collection and delivering to the laboratory, and instructions to the laboratory personnel for reporting the lab assessments will be performed at the initial site visit.

Any sign or symptom associated to adverse events will be reported by the patient in an electronic patient diary EXOM GENIUS ROSA and reported during the telemedicine consult. The eDiary should be filled in daily by the patient by 09:00 pm.
All patients will be withdrawn from the study when criteria for supplemental oxygen therapy and/or mechanical ventilation are met. Another stopping rule (patient's withdrawal from study) includes drug related adverse events grade ≥ 3 according to Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 [Common Terminology Criteria for Adverse Events (CTCAE) Last Updated: 03/01/18 available at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm]
Patients are free to withdraw from participation in the study at any time upon request, without any consequence. Patients should be listed as having withdrawn consent only when they no longer wish to participate in the study and no longer authorize the Investigators to make efforts to continue to obtain their outcome data. Every effort should be made to encourage patients to remain in the study for the

duration of their planned outcome assessments. Patients should be educated on the continued scientific importance of their data, even if they discontinue the study. In the case of a patients becoming lost to follow-up, attempts to contact the patient should be made and documented in the patient's medical records.

6.3. EARLY WITHDRAWAL FROM THERAPY OR ASSESSMENT

Patients will be informed that they have the right to withdraw from the study at any time (withdrawal of consent), without prejudice to their medical care, and are not obliged to state their reasons.

Likewise if a patient declares his/her wish to discontinue from the study e.g. for personal reasons, an attempt should be made to establish that the true reason is not a SAE (bearing in mind the patient is not obliged to state his/her reasons).

Patients who discontinue the treatment with the IMP will not be withdrawn from the study, but will be asked to complete observations as per the protocol, unless otherwise they withdraw their consent. It is important that any randomized patient remains in the study and is followed for both efficacy and safety outcomes, regardless he/she has completed or discontinued the study treatment. Investigators will be trained about the importance of patient retention through the duration of the trial.

Any withdrawal must be fully documented in the eCRF.

Patients who will have a negative swab test result during the treatment phase will stop the treatment, and will be followed for the following two weeks for safety reason only.

Premature discontinuation

A premature discontinuation will occur when a patient who signed the ICF ceases participation in the study, regardless of circumstances, before the completion of the study protocol procedures. Patients can be prematurely discontinued from the study for one of the following reasons:

- Failure to meet inclusion/exclusion criteria
- Hypoalbuminemia at a level considered clinically relevant by the investigator
- Withdrawal of consent
- Lost to follow-up (Every effort must be made to contact the patient; a registered letter must be sent)
- Study terminated by the Sponsor
- Other reasons, such as administrative reasons or pregnancy
- Documented disease progression
- Severe protocol violations, such as an incorrect treatment administration, or a concomitant use of not permitted medications. Before removal, these cases should first be discussed with Dompé farmaceutici s.p.a.

- Development of AE or unacceptable toxicity, precluding further therapy with the study drug.

The investigator should advise patients that prematurely discontinue on any therapies or treatments for their condition and refer them for further treatment as appropriate.

In case of discontinuation, subjects will undergo a remote Early Trial Discontinuation Visit aimed to assess physical abnormalities, vital signs and transdermal oxygen saturation measurement.

Replacement Procedures

No patient who has been randomized and withdraws from the study for any reason will be replaced.

6.4. END OF STUDY (EOS)

The EOS is defined as the last day the last patient completes the last study assessment, or retracts the consent to participate in the study, or withdraws from the study, or is deceased or otherwise lost to follow-up.

7. ENDPOINTS

7.1. STUDY ENDPOINTS

7.1.1. Primary endpoints

Virologic outcome. Proportion of participants with undetectable SARS-CoV-2 at PCR at day 7 after randomization.

Clinical outcome. Proportion of participants who not requires supplemental oxygen therapy (NEWS ≤2) and/or mechanical ventilation at day 14 after randomization.

7.1.2. Secondary endpoints

- Proportion of participants with undetectable SARS-CoV-2 at PCR at day 14 after randomization at day 28 after randomization;
- Proportion of participants who does not require supplemental oxygen therapy (NEWS ≤ 2) and/or mechanical ventilation at day 7 and 28 after randomization;
- Proportion of patients in each NEWS category at time 7, 14 and 28 after randomization;
- Mean value of NEWS category at time 7, 14 and 28 after randomization;
- Proportion of participants with any adverse event with grade ≤ 2 according to CTCAE at day 7, 14 and 28 after randomization;
- Proportion of participants with any severe adverse events (grade ≥ 3 according to CTCAE) at day 7, 14 and 28 after randomization;
- Proportion of hospitalized participants who at the beginning of the study were at domicile isolation at day 7, 14 and 28 after randomization;
- Proportion of participants admitted to intensive care at day 7, 14 and 28 after randomization;
- Proportion of survivors at day 7, 14 and 28 after randomization;
- Mean variation of value of the following biomarker parameters, from base line to day 7, 14, 21 and 28 after randomization:
    - Complete blood cell counts;
    - Hepatic function (ALT, AST and bilirubin);
    - Coagulation (PT, aPTT and INR);
    - Other marker including (D-dimer, CPK, LDH);
- Quality of life questionnaire 3 months after the randomization.

8. EVALUATION OF ADVERSE EVENTS AND SAFETY INFORMATION

8.1. DEFINITIONS

Adverse Event

An **Adverse Event** (**AE**) is defined as any untoward medical occurrence in a patient or clinical investigation subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavourable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not related to the medicinal product.

Adverse Drug Reaction

An **Adverse Drug Reaction (ADR)** is defined as an adverse experience which is a reasonably likely to have been caused by the drug. Adverse events are to be considered unsuspected if the relationship to the study drug as described in the table in section 8.2.1 is none or unlikely; whereas any AE reported in the study having a possible, probable or highly probable relationship to study drug will be considered as an ADR. The definition covers also medication errors and uses outside what is foreseen in the protocol, including misuse and abuse of the product.

Serious Adverse Event

A **Serious Adverse Event** (**SAE**) is defined as any untoward medical occurrence that at any dose:

- results in death,
- is life-threatening (i.e. the patient was at risk of death at the time of the event. It does not refer to an event which hypothetically might have caused death if it were more severe),
- requires inpatient hospitalization or prolongation of existing hospitalization,

  NOTE: In general, hospitalization means that the individual remained at the hospital or emergency ward for observation and/or treatment (usually involving an overnight stay) that would not have been appropriate in the physician's office or an out-patient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred, the event should be considered serious.
- results in persistent or significant disability/incapacity,

  NOTE: This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, or accidental trauma (e.g., sprained ankle) which may interfere or prevent everyday life functions, but do not constitute a substantial disruption.
- is a congenital anomaly/birth defect,
- is medically significant or important medical condition, i.e.an important medical event that based upon appropriate medical judgment, may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed above.

An important medical condition is an event that may not result in death, be life-threatening, or require hospitalization but may be considered a SAE when, based upon appropriate medical judgment, it may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in patient hospitalization, or the development of drug dependency or drug abuse.

Pre-planned hospitalization or hospitalization for routine treatment or monitoring of the studied indication, not associated with any deterioration in condition are not considered to be SAEs (see Par. 8.3.2).
These events must be recorded in the AE page of the eCRF where a variable will be ticked to indicate that they are not SAEs.

Death shall always be reported as SAE and the cause of death shall always be specified when known. Death due to progression of disease may be unrelated to study treatment and causality shall be assessed by the Investigator. The investigator should report the event immediately to the sponsor and the sponsor shall comply with regulatory reporting, unless there is no reasonable possibility that the drug may have caused the fatal outcome of the disease progression.

Expectedness

Despite raloxifene has been on the market for many years with an authorised Summary of Product Characteristics, considering that this trial is investigating raloxifene in a new condition and patient population, all events (AE and SAE) will be assessed as unexpected by the Sponsor. The authorised SmPC may be considered the reference document for safety information, but shall not be used for the evaluation of the expectedness.

Suspected serious unexpected adverse reaction (SUSAR)

A suspected serious unexpected adverse reaction is defined as an adverse reaction that is both unexpected (not consistent with the applicable product information) and also meets the definition of a Serious Adverse Reaction.

Adverse event of special interest (AESI)

An adverse event of special interest (serious or non-serious) is an event thought to be [potentially] associated with the investigational compound or disease under study and for which close monitoring is advised. Reporting on Adverse Events of Special Interest is an emerging and ever more critical aspect related to characterizing the safety profile of a compound, for which ongoing monitoring and communication by the investigator to the sponsor as SAE is requested.

8.2. MONITORING FOR ADVERSE EVENTS

All patients will daily self-monitor their parameters, AEs (if any) and concomitant medications (if any), and fill in an electronic study diary GENIUS ROSA. All patients may call the Investigator if needed.

During the study visit, the subject shall have the opportunity to spontaneously mention any problem and the investigator will search for possible adverse events by asking generic questions to the patient without pointing to specific events which may have occurred.

AEs should be reported for any clinically relevant change in concomitant condition(s) that is the result of an untoward (unfavorable and unintended) change in a subject's medical health. Changes in any protocol-specific systemic parameter evaluated during the study are to be reviewed by the Investigator. In addition, the subject's responses to any questionnaire utilized during the study are to be reviewed by the Investigator. Any untoward (unfavorable and unintended) change in a protocol-specific parameter or questionnaire response that is clinically relevant is to be reported as an AE. These clinically relevant changes will be reported regardless of causality.

8.3. RECORDING

AEs will be collected and recorded for any untoward event that occurs in a patient from the time he or she signs the ICF for the trial up to the end of the follow up period. Thus, any untoward medical occurrences or unfavorable and unintended signs, symptoms, or diseases that occur in the pretreatment, in treatment, or post treatment period are to be considered AEs and/or SAEs, and consequently recorded and reported as such. Should a non-serious AE become serious, the Investigator will then follow the same reporting procedures as for SAEs.
Each AE will be described by:

- Its duration (start and stop dates).
- Its seriousness.
- Its relationship to the study drug (suspected/unsuspected).
- Severity grade: The severity of AEs and SAEs will be graded using the Common Terminology Criteria for Adverse Events (CTCAE), version 5.0 (Appendix 3).
- Action(s) taken.
- Outcome.

Medical conditions/diseases and related signs/symptoms present before starting study treatment shall be documented in the medical history section of the eCRF; these conditions are considered AEs only if they increase either in frequency or severity once informed consent has been signed.

8.3.1. Relationship of AEs to the Investigational Product

The Investigator will assess the possible relationship between the AE and the investigational medication, according to the criteria in **Table** below:

Relationship of the Adverse Event to the IMP

None (Intercurrent Event)	An event that is not and cannot be related to the Investigational Product, e.g. patient is a passenger in a road traffic accident.
Unlikely (remote)	Relationship is not likely e.g. a clinical event including laboratory test abnormality with temporal relationship to drug administration which makes a causal relationship improbable and in which other drugs, chemicals or underlying disease provide more plausible explanations
Possible	Relationship may exist, but could have been produced by the patient's condition or treatment or other cause
Probable	Relationship is likely, the AE abates upon discontinuation of Investigational Product and cannot be due to the patient's condition
Highly Probable	Strong relationship, the event abates upon discontinuation of Investigational Product and, if applicable, re-appears upon repeat exposure

8.3.2. Severity of AEs

The Investigator will grade the severity of any AE using the classification according the CTCAE dictionary, as reported in Appendix 3.

The severity of AEs and SAEs will be graded using the CTCAE version 5.0. Any AE not listed in the CTCAE will be graded as follows:

Grade Definition:

1. Mild
2. Moderate
3. Severe
4. Life-threatening or disabling
5. Death.

8.3.3. Follow-Up of patients with Adverse Events

The Investigator is responsible for adequate and safe medical care of subjects during the trial and for ensuring that appropriate medical care and relevant follow-up procedures are maintained after the trial. All AEs should be followed-up to determine outcome of the reaction. The Investigator should follow up the event until resolution or stabilization of the condition. It is the Investigator's responsibility to ensure that the subjects experiencing AEs receive definite treatment for any AE, if required.

If subject was hospitalized due to a SAE, a copy of the discharge summary is to be forwarded to the Study Safety Contact as soon as it becomes available.

In addition, a letter from the Investigator that summarizes the events related to the case as well as results of any relevant laboratory tests also may be requested. Further, depending upon the nature of the SAE, Dompé may request copies of applicable segments of the patient's medical records. In case of death, a copy of the autopsy report, if performed, should also be provided.

The Investigator shall inform the Study Safety Contact with an appropriate written communication, whenever he becomes aware of new available information regarding the SAE, once the condition is resolved or stabilized and when no more information about the event is expected. Follow-up SAE information should be processed as initial SAE notification (see Par. 8.4.1).

For pharmacovigilance purposes, all SAEs should be followed-up in order to clarify as completely as possible their nature and/or causality and until all queries have been resolved. All SAEs will be followed up until the events resolve or the events or sequelae stabilize, or it is unlikely that any additional information can be obtained after demonstration of due diligence with follow-up efforts (i.e. subject or Investigator is unable to provide additional information, or the subject is lost to follow up), unless subject has withdrawn his/her consent.

8.4. SERIOUS ADVERSE EVENT REPORTING

8.4.1. Reporting Procedure for Investigators

The investigator must report all SAEs, regardless of presumed causal relationship to Study Safety Contact (i.e. Dompé Pharmacovigilance and Contract Research Organization Pharmacovigilance expert) recording the SAE data on the AEs section of the eCRF within 24 hours of learning of the event. The paper SAE forms are only intended as a back-up option when the eCRF system is not accessible. In this case, the original paper forms are to remain on site and copies are to be transmitted via email or confirmed facsimile (fax) transmission to:

- e-mail address: rlx0120-safety@exomgroup.com
- Fax number: +39 02 36026913

Contact details for SAE reporting are provided in the section "Contact Information".

The investigator should also report information on SAEs that continue after patient has completed his/her participation in the study (whether study completion or withdrawal), unless patient has withdrawn his/her consent.

Information on SAEs will be recorded on the AEs section form in the eCRF. Follow-up reports (as many as required) should be completed following the same procedure; the system will mark the SAE form as "Follow up no. XX"

Whenever more than one SAE is observed, the Investigator should identify which is the primary adverse event, i.e. the most relevant one. If other events are listed in the same report, the Investigator, along with their relatedness to the Investigational Product, should identify which adverse events are serious and which are non-serious. In any case, the Investigator is requested to record his/her opinion about the relatedness of the observed event(s) with the investigational medication.

In line with CT3 Detailed Guidance and ICH E2A provisions, although the Investigator does not usually need to actively monitor patients for AEs once the trial has ended, if the Investigator becomes aware of

a SAE occurring to a patient after that patient has ended his/her participation in the study (whether study completion or withdrawal), the SAE should be reported by the Investigator using the following email address: farmacovigilanza@dompe.com or fax: +39 02 36026913. Such "post-study cases" should be regarded for expedited reporting purposes by Dompé, as though they were study reports. Therefore, a causality assessment and determination of expectedness are needed for a decision on whether or not expedited reporting is required.

8.4.2. Adverse Events of Special Interest (AESI)

During the study all AESI will be considered equivalent to a SAE and will be reported following the procedures described above for the reporting of a SAE. The following events will be considered adverse events of special interest:

- Venous thromboembolic events (including deep vein thrombosis)
  - pulmonary embolism
  - retinal vein thrombosis
  - superficial vein thrombophlebitis
- Arterial thromboembolic reactions
- Thrombocytopenia

8.4.3. Conditions that should not be reported as serious adverse events

The conditions listed below, that may require hospitalization of a patient, are not considered to be SAE and shall not be reported as such, but only need to be recorded in the eCRF:

- Hospitalization for routine treatment or monitoring of the studied indication, not associated with any deterioration in condition.
- Trial end points
- Abnormal lab values or test results that do not induce clinical signs and/or symptoms and require intervention/therapy, i.e. are clinically significant.

8.4.4. Reporting Procedure to Ethics Committee (EC) and to Regulatory Authorities

In addition to informing the Study Safety Contact, the Investigator shall notify SAE to his/her EC as applicable (in particular in case of death); in addition, for reported deaths of a subject, the Investigator shall supply Study Safety Contact and the Ethics Committee with any additional information requested. Copies of all correspondence relating to reporting of any SAEs to the EC should be maintained in the Investigator's Files.

Dompé Pharmacovigilance shall submit any suspected unexpected serious adverse reaction (SUSAR) to the concerned EC and Regulatory Authority (via Eudravigilance), as soon as possible and in no event later than:

- seven calendar days after becoming aware of the information if the event is fatal or life threatening; to be followed by any relevant information within eight days.

- fifteen calendar days after becoming aware of the information if the serious event is neither fatal nor life threatening.

Dompé Pharmacovigilance shall report any relevant updated follow-up safety information as soon as available.
If the results of an investigation show that an ADR not initially determined to be reportable is reclassified as reportable, Dompé Pharmacovigilance shall report such reaction in a written safety report as soon as possible, within the timeframes defined by current law requirements.

Dompé shall be responsible to prepare and submit annual safety reports (Development Safety Update Report – DSUR) to relevant Regulatory Authorities, as applicable. In addition, Investigator will receive from Dompé Pharmacovigilance appropriate periodic safety updates, as per applicable local requirements and regulations.

Dompé will ensure ongoing safety evaluation to be conducted periodically during the study on the safety data received, including monitoring of AESI.

8.5. EXPOSURE TO INVESTIGATIONAL PRODUCT DURING PREGNANCY

Women of childbearing potential are not excluded from the study as long as adequate birth control methods are being utilized. Women of childbearing potential are defined as all women physiologically capable of becoming pregnant. Prior to enrolment in the clinical trial, female patients of childbearing potential and their partners must be advised of the importance of avoiding pregnancy during the entire course of the study treatment and for the 30 days after the study treatment period ends and of the potential risks associated with an unintentional pregnancy. During the trial, female patients are to be instructed to contact the Investigator immediately if they suspect they might be pregnant. In the same way, male patients who become aware that the partner might be pregnant, are to be instructed to contact the Investigator immediately.

The Investigator must report every pregnancy on a Pregnancy Report Form as soon as possible (within 24 hours of learning of the pregnancy to the Study Safety Contact (contacts specified in the section "Contact Information"), even if no AE has occurred, and follow it up to term. If, however, the pregnancy is associated with an SAE (e.g., if the mother is hospitalized for dehydration), in addition to the Pregnancy Report Form, a separate SAE Report Form must be filed as described in Section 8.4.1 with the appropriate serious criterion (e.g., hospitalization) indicated on the SAE Report Form. Miscarriage, stillbirth and any malformation/disease must be reported as a SAE.

Any pregnancy leads to the immediate exclusion from the trial.

8.6. ADVERSE EVENTS CAUSING TREATMENT DISCONTINUATION

If a patient is withdrawn from the study as a consequence of an AE, this must be recorded in the eCRF, and the patient must be followed up until the resolution of the AE or as instructed by the medical monitor.

8.7. OVERDOSE

Cases of overdose (accidental or intentional) which may or may not result in serious adverse reactions are to be reported to Study Safety Contact, following the same procedure for SAE, within 24 hours from

the Investigator's knowledge of its occurrence. The Medical Expert should be contacted to discuss corrective treatment, if necessary.

An overdose of the study drug is defined as:

- Capsules: The administration of more than 50% of the daily dose

Overdose includes reports related to drug intake through different routes (e.g. ingestion) or with suicidal intentions and consequent drug overdose.
The Investigator shall provide in the SAE form information about symptoms, corrective treatment and outcome of overdose.

8.8. UNBLINDING

During the trial, blinding may be broken by the Investigator for emergency purposes only, when knowledge of the treatment identity is essential for treating the subject, or by Sponsor Pharmacovigilance for safety reasons and in case of Serious Adverse Events qualifying for regulatory reporting (SUSAR), in line with regulatory requirements.

If the treatment code needs to be broken in the interest of patient's safety for a medical emergency, the Investigator is allowed to break the treatment code for the specific patient, even before informing the Sponsor. Investigator will be allowed to unblind study medication electronically through a specific procedure within the eCRF (GENIUS IWRS). The system will notify the unmasking via e-mail to the Study Safety Contact and Dompé Medical Expert without revealing the treatment identity, but only referring to the kit number involved in the unmasking in order to avoid a dissemination of unmasked information. Proper training as well a specific user's manual is provided to investigators for the use of GENIUS IWRS and of the unblinding functionality. As a backup solution, sealed envelopes containing the individual treatment codes will be available at sites and must be kept by the Investigator in a secure location accessible only to designated staff in order to avoid dissemination of treatment identification to personnel involved in study conduct who must remain blind. During the study, the integrity of the envelopes will be regularly checked by the Site Monitor, during the visits at site. At the end of the study, all the individual envelopes must be returned to Dompé.

Dompé Drug Safety may need to unmask a patient's treatment for safety reason or if a reported SAE meets criteria of a SUSAR in order to fulfil expedited regulatory reporting requirements. Dompè Drug Safety will be allowed to unblind study medication electronically through the specific procedure within the eCRF (GENIUS IWRS). Additionally, a copy of the individual envelopes identified with the subject assignment number/ kit number will be also provided to the Dompé Drug Safety before study start, together with access to the GENIUS IWRS. The identity of the treatments will remain unknown to the subject, Investigator, site staff, CRO and Dompé Development personnel until the study completion and formal unmasking.

The randomization code will be broken when the last enrolled patient has completed therapy, and once the database has been locked.

9. STATISTICS

9.1. SAMPLE SIZE

The sample size of the study is calculated based on the following assumptions:

- Virologic assumption: Early treatment with antivirals increases the proportion of patients with undetectable SARS-CoV-2 in upper respiratory tract at day 7 after therapy from 25% to 50%;
- Clinical assumption: Early treatment with antivirals increases the proportion of participants who recover without need of mechanical ventilation and/or supplemental oxygen therapy by the day 14 after therapy from 50% to 75%.

Based on these assumptions and considering a randomization ratio 1:1:1, the sample size will be adaptively determined to achieve a power greater than 80% to show superiority of raloxifene vs placebo in terms of either one primary endpoint (section 7.1.1) and controlling the one-sided alpha below 0.025.

Sample size will be determined throught the following steps:

- an interim analysis will take place when 50 participants per arm (N=150) have reached their primary enpdoints (i.e. assessments available).
    - In case of efficacy of at least one treatment arm or futily of both arms (see criteria in section 9.3) the study enrollement will be stopped,
    - otherwise the actual number of new participants in each arm may be blindly reassessed according to observed efficacy of the best favourable and promising arm;
- in case of continuation of the study to the final stage, it is expected to randomize on average additional 174 patients, depending on sample size reassessment and possible dropping of an uneffective treatment arm. Expected sample size will be between 250 and 450 randomized participants;
- drop-outs of randomized subjects will not be replaced.

This approach allows either to minimize the number of enrolled participants if the experimental assumptions are too conservative or to have a good power level if they are too optimistic.

Pocock's spending functions and conditional power (CP) will be used to control the type I and II errors, considering that interim analysis will be performed when half of the initial planned patients have been reached the primary endpoints (i.e. assessments available).

Bonferroni method is used to adjust for multiple endpoint vs treatment comparisons.

Sample size calculation has been performed through simulation (100000 replications). The statistical software used for simulation was SAS®, Version 9.4.

9.2. RANDOMIZATION

Eligible Patient will be randomized in a 1:1:1 fashion to either:

- Group 1: 1 capsule of raloxifene 60 mg and 1 capsule of placebo.
- Group 2: 2 capsules of raloxifene 60 mg.

- Group 3: 2 capsules of placebo.

Randomization will be performed through IRS. Each Patient Kit number will be randomly associated with a treatment group. The randomization list will be provided to the facility responsible for IMP packaging/labelling for the purpose of IMP preparation. Each randomized patient will be allocated with randomization number according to the stratified randomization list. Dropouts after randomization will not be replaced.

Randomization will be stratified by site and gender to ensure balanced assignment across treatment groups. The stratified permuted block randomization list will be generated with a computer procedure by an independent statistician not involved in the conduct of the study.

Access to individual patient treatment code will be allowed only in the event of a medical emergency where the knowledge of patient treatment is required to provide the patient with appropriate care. The investigator will be provided with a protected access to the randomization system to allow opening of the treatment allocation for a specific patient in case of a medical emergency.

Dompé Pharmacovigilance contact person will be provided with a protected access to the randomization information in case of unblinding for safety procedures.

Unblinding events will be recorded and reported in the final study report.

The randomization codes will be accessible to the independent statistician who will perform the interim analysis. The treatment assignment information will be kept confidential and will not be disclosed to any other person than those ones involved in these intermediate analysis.

The randomization code will be broken according to study procedures after the last enrolled patient has completed his/her follow-up visit.

9.3. STATISTICAL METHODOLOGY

An interim analysis is planned when half of the planned evaluable patients has reached the primary endpoints at 7 and 14 days (i.e. assessments available). The intent of the interim analysis is either for identification of early superiority of raloxifene (early efficacy), or for an early stop of one or both treatment arms for futility, or for blind sample size reassessement.

Pocock's spending functions and CP will be used to control the type I and II errors for analyses of primary endpoints. Pocock's boundaries (on standardized Z scale) for efficacy and futility at interim and final analyses are reported in Table 1.

Table 1: Pocock spending functions boundaries for primary endpoints

Analysis	Sample Size	Standardized Z Scaleboundaries for primary endpoints	
		Alpha	Beta
Interim	50%	2.69640	1.59184
Final	100%	2.69640	2.69640

The analysis will be conducted by an independent statistician who will communicate to the Sponsor the result of the interim analysis and the consequent decisions on the continuation of the study, drop of an unefficacious treatment arm and on the reassessment of sample size, if required.

On the basis of Pocock's boundaries and CP at interim analysis, each treatment arm will be classified and ranked as "Effective", "Favorable", "Promising", or "Unfavorable". According to the most effective up-ranked arm, a decisions on the continuation of the study and on the reassessment of sample size will taken.

In case only one treatment arm is defined as "Unfavorable" at the interim analysis, this arm will be dropped from the continuation of study and no additional subjects will allocated to it. Already randomized subjects will continue the follow-up as planned.

Criteria for classification and ranking of treatment arms and possible consequent actions are reported in Table 2.

Table 2: Criteria for classification of treatment arms and actions post-interim analysis

Arm Classification	Criteria		Action based on the most up-ranked treatment arm
	Pocock's Alpha	Higher Conditional Power between Virologic and Clinical endpoint	
1 - Effective	$Z > 2.69640$ AND CP>90%		Stop for efficacy
2 - Favorable	-	CP>80%	No sample size reassessment
3 - Promising	-	35%<CP≤80%	Sample size reassessment
4 - Unfavorable	$Z \leq 1.59184$ OR CP≤35%		Stop for futility

9.4. PATIENT POPULATION

The following population will be defined:

- The Safety (SAF) population will consist of all randomized patients who received at least one dose of the investigational product. Safety population will be analyzed according to the actual treatment received. The SAF population will be used to present results on safety data.
- The Full Analysis Set (FAS) population will consist of all randomized patients who received at least one dose of the investigational product. FAS population will be analyzed according to ITT principle, i.e. by treatment allocation regardless happening of intercurrent events. The FAS population will be used for the primary analyses of the study and to present results on efficacy data.

The Per Protocol (PP) population will consist of all randomized patients who received at least one dose of the investigational product and do not have Major Protocol Deviations. The PP population will be used for sensitivity analyses.

9.5. STATISTICAL METHODOLOGY

All patient data collected on the eCRF will be listed by patient and center.

Appropriate descriptive statistics will be produced, according to the variable:

- for continuous data n, mean, standard deviation (SD), median and range (minimum and maximum) will be presented;
- for categorical data, frequency distributions and percentages will be presented;
- for time-to-event variables, cumulative freedom from event will be evaluated using Kaplan-Meier method. The degree of uncertainty will be expressed with 95% confidence limits (calculated per the method proposed by Greenwood). Comparison of curves among arms will be performed with the log-rank test. Kaplan-Meier graphs will be presented along with the number of patient-at-risk at exact time points. Subjects ongoing and who are free from event at the time of DB lock will be censored at the DB lock date. Subjects who have discontinued without an event will be censored at the date of discontinuation.

Unless otherwise specified, the significance level used for statistical testing will be 0.05 and two-sided tests will be used.

9.5.1. Demographic and baseline characteristics

Demographic and baseline characteristics will be summarized for all patients in the FAS population, by treatment group.

9.5.2. Analysis of efficacy variables

9.5.2.1. Primary analyses

The proportion of subjects with undetectable SARS-CoV-2 at PCR at day 7 after randomization (virologic endpoint) and the proportion of subjects who not requires supplemental oxygen therapy (NEWS ≤2) and/or mechanical ventilation at day 14 after randomization (clinical endpoint) will be separately analyzed by means of logistic regression models adjusting by pre-defined factors as center, age, gender and post-menopausal status. Additional covariates may be defined in the SAP. Each treatment arm will be compared independently with the placebo.

The null hypothesis for the virologic endpoint is that proportion of treated subjects with undetectable SARS-CoV-2 at PCR at day 7 after randomization is lower or equal the placebo one ($\pi_{PLACEBO}$). Two sets of null and alternative hypotheses are provided, one for each treatment dosage:

$$H_{01\text{-}60mg}: \pi_{RALOXIFENE\text{-}60mg} \leq \pi_{PLACEBO} \qquad H_{01\text{-}120mg}: \pi_{RALOXIFENE\text{-}120mg} \leq \pi_{PLACEBO}$$

$$H_{11\text{-}60mg}: \pi_{RALOXIFENE\text{-}60mg} > \pi_{PLACEBO} \qquad H_{11\text{-}120mg}: \pi_{RALOXIFENE\text{-}120mg} > \pi_{PLACEBO}$$

The null hypothesis for the clinical endpoint is that proportion of treated subjects who not requires supplemental oxygen therapy (NEWS ≤2) and/or mechanical ventilation at day 14 after randomization is lower or equal the placebo one ($T_{PLACEBO}$). Two sets of null and alternative hypotheses are provided, one for each treatment dosage:

$H_{02\text{-}60mg}$: $T_{RALOXIFENE\text{-}60mg} \leq \pi_{PLACEBO}$ $H_{02\text{-}120mg}$: $T_{RALOXIFENE\text{-}120mg} \leq \pi_{PLACEBO}$

$H_{12\text{-}60mg}$: $T_{RALOXIFENE\text{-}60mg} > \pi_{PLACEBO}$ $H_{12\text{-}120mg}$: $T_{RALOXIFENE\text{-}120mg} > \pi_{PLACEBO}$

Superiority of raloxifene will be claimed if at least one null hypothesis is rejected, at interim or at final analysis according to criteria shown in section 9.3.

Sensitivity analyses will be defined in the SAP to assess the robustness of results on primary endpoints versus the presence of missing data (unexpected) and the presence of protocol deviations.

9.5.2.2. Secondary analyses

All secondary endpoints will be analyzed at each available timepoints by means of descriptive statistics and by appropriate parametric tests depending on the nature of the variable and its distribution. Data transformation might be used in order to satisfy the assumption of normality requested by parametric statistical tests. In case such assumptions are not met, non-parametric counterpart tests will be used. Change from baseline value (for continuous variables) and shift tables versus baseline (for categorical variables) will also be summarized for all post-baseline visits.

The incidence of participants with undetectable SARS-CoV-2 at PCR, the incidence of participants who requires supplemental oxygen therapy (NEWS ≤ 2) and/or mechanical ventilation, and mortality will be analyzed as proportion at each time point and as time-to-event.

Details will be provided in the SAP.

9.5.3. Analysis of exploratory variables

In addition to descriptive statistics at each available timepoint, explorative variables will be analyzed by means of inferential tests depending on their nature and distribution (all confidence intervals and statistical tests on explorative endpoints are of descriptive nature). Change from baseline value (for continuous variables) and shift tables versus baseline (for categorical variables) might be reported for all post-baseline visits.

9.5.4. Analysis of safety variables

TEAEs, ADRs and TESAE will be presented by treatment arms in terms of number of AEs and incidence by System Organ Class (SOC) and Preferred Terms (PT) using MedDRA. Analyses will be provided also by severity and relationship to the study drug. In addition, time-to-event methods will be used to summarize the overall survival.

Vital signs and laboratory tests will be presented using descriptive statistics at each available visit. Additionally, the frequency of subjects reporting an abnormal or abnormal clinically significant laboratory value will be presented for each laboratory parameter. Shift tables versus baseline will compare abnormal laboratory findings at each post-baseline visit.

9.5.5. Missing data

All reasonable efforts will be made to reduce the rate of missing data. Investigators will be trained about the importance of patient retention and full data capture. Also, any reasonable attempts should be made by the Investigators to emphasize continued subject's participation for the full duration of the trial.

9.5.6. Specification of subgroups for analysis

Statistical tests for interaction (between subgroup and treatment arm) will be performed to decide about the need to further investigate subgroups of the trial population based on the following variables: age group, gender and Country.

Subgroup analysis will be performed if interaction tests are statistically significant at 15% nominal level. Statistical details and potential new subgroups definitions will be reported in the SAP.

9.5.7. Study termination

The End of Study (EOS) is defined as the last day the last patient completes the last study assessment in the hospital, or retracts the consent to participate in the study, or withdraws from the study, or is deceased or otherwise lost to follow-up.

9.5.8. Changes to the statistical plan

The Statistical Analysis Plan will be issued before the interim analysis with more technical and detailed elaboration of the principal features of statistical analyses. Additional post-hoc analysis may be produced to further allow comparison between treatment and placebo, according to the results obtained.

Any deviations from the original statistical plan (including unplanned analyses) will be clearly documented in the Clinical Study Report.

10. ETHICAL CONSIDERATIONS

10.1. INSTITUTIONAL REVIEW BOARD AND INDEPENDENT ETHICS COMMITTEE

This study will be carried out in full compliance with the guidelines of ethics committee (EC) and government agencies of each respective country as well as the European Union (EU) Clinical Trial Directive (Directive 2001/20/EC)and any subsequent applicable law, where applicable. Before the study begins, the study centers will require approval from an EC and Regulatory agency. During the course of the study, the Sponsor or authorized contract research organization (CRO) representative will provide timely and accurate reports to the EC on the progress of the study, at intervals not exceeding 1 year (or as appropriate), and will notify the EC of SAEs or other significant safety findings. The study protocol, ICF, information sheet advertisements, and amendments (if any) will be approved by the EC at the study centers in conformance with the EU Clinical trial directive (Directive 2001/20/EC), and any subsequent applicable law and local regulations.

10.2. ETHICAL CONDUCT OF THE STUDY

The procedures outlined in this clinical trial protocol are designed to ensure that the Sponsor and the Investigators from the Clinical Sites perform their activities throughout the set-up, conduct, evaluation, documentation and analysis of the study, in accordance to the principles of the Good Clinical Practice (GCP) guidelines of the International Conference on Harmonization (ICH) and the Declaration of Helsinki. The study will be carried out adhering to local legal requirements and the applicable national laws, whichever represents the greater protection for the individuals.
Study protocol, patient information and supportive materials for the electronic informed consent will be submitted to the appropriate Ethics Committee for approval. The Sponsor will be responsible to inform in a timely manner the appropriate Ethics Committee about any changes in the study protocol which could interfere with the patient's safety.

10.3. PATIENT INFORMATION AND CONSENT

The investigators, sub-investigators and General Practitioner(GP) involved in this clinical trial who are responsible for treating the patient for COVID-19 are responsible for providing all necessary information about the participation in the study to their patients, and consequently to obtain the signature of the Informed Consent.

Since patients can't go to clinical sites for discussing their participation to the study and eventually sign the informed consent, the consent procedure will be managed remotely (Genius remote ENGAGE) through an electronic application that allows the interaction between investigator and patient as well as the electronic signature of the consent form in the full respect of the enforced legal and regulatory requirements.
The same procedure applies to the information of the patient and providing of consent to the processing of personal data according to the European Regulation n. 679/2016 on the Protection of Personal Data,

the Personal Data Protection Code (Legislative Decree 196/03) and subsequent amendments and additions, and to the provisions, guidelines and general authorizations of the National Guarantor for personal Data Protection.

10.4. CONFIDENTIALITY AND DATA PROTECTION

By signing this protocol, the investigator agrees to keep all the information provided by the sponsor in strict confidentiality and to request the same confidentiality from his/her staff. Study documents provided by the sponsor (protocols and other materials) will be stored appropriately to ensure confidentiality. The information provided by the sponsor to the investigator cannot be disclosed to others without direct written authorisation from the sponsor, except for the extent necessary to obtain the electronic informed consent from the subjects wishing to participate in the study.

Data on subjects collected in the source documents during the study will be transferred to the Sponsor in an anonymized way. If, as an exception, for safety or regulatory reasons identification of a subject becomes necessary, the sponsor and the investigator will be bound to keep this information confidential.

On the eCRF(and*patient diaries)*, patients will be identified ONLY by the assigned patient number. If patient names are included on copies of documents submitted to Sponsor or its delegate, the names will be obliterated or masked and the assigned patient number added to the document.

The Investigator should keep a separate log (Patient Master List) of patient's codes, names and addresses.

10.5. ADMINISTRATIVE ASPECTS

The investigational medicinal products required for the conduct of this study will be provided free of charge by the Sponsor to the participating clinical sites.

Coverage for any damage resulting from the participation of the subjects in the clinical trial is warrented. In addition to the general insurance of the individual participating clinical centers, an insurance cover will be issued in favour of the subjects participating in this clinical study. The insurance is in compliance with the local regulation and with the requirements of the Health Authorities.

11. DATA HANDLING AND RECORD KEEPING

11.1. CASE REPORT FORMS

An eCRF customized for the trial will be provided by the CRO in compliance with EU regulations.

An eCRF is required and should be completed for each patient enrolled. Entry in the eCRF will be progressed as soon as the data are available. The Investigator will be responsible for the accuracy and timeliness of the data entered in the eCRFs. All "free field" comments must be entered in ENGLISH.

Source documents are the originals of any documents used by the Investigator or hospital/institution that allow verification of the existence of the patient and substantiate the integrity of the data collected during the trial. Source documents should be available to support all the data recorded in the eCRF; location of source documents, including those for which the eCRF might be accepted as being the sole source document, will be specified and listed in the "Source Document Location" log at the Site Initiation Visit. The Investigator is responsible for ensuring that data are reliably and correctly generated, recorded, processed and reported, in agreement with the ALCOAC principles (Attributable-Legible-Contemporaneous-Original-Accurate-Complete).

The data are the sole property of Dompé farmaceutici s.p.a. and should not be made available in any form to third parties, except for authorized representatives of the CRO, or appropriate Health/Regulatory Authorities, without written permission from Dompé farmaceutici s.p.a.

11.2. PATIENT DIARY

An eDiary (will be supplied to each randomized patient by the CRO on a pre-programmed electronic device (e.g. smartphone or tablet) directly connected to the eCRF. The eDiary must be used for the purpose of trial RLX0120 and should not be used or made available in any form to third parties, except for authorized Dompé designee or representatives of appropriate Health/Regulatory Authorities.

The patient will report in the eDiary the details (date, time, number of capsules) of each administration of the IMP and vital signs self-monitored, AEs, CMs. It is responsibility of the Investigator/Nurse to explain to each patient how to enter the data in the eDiary; similarly, it is responsibility of the Investigator to check in a timely manner administration data.

11.3. PROTOCOL DEVIATION

Any deviations from the protocol procedures described in relevant sections will be reported in the study-specific PD form. The form will provide a detailed self-explaining narrative of the PD to include identification of the procedure that has been deviated, the reason why the deviation occurred and measures taken to prevent recurrence of the deviation, as appropriate.

11.4. DATA MANAGEMENT

Data management of the eCRFs, eDiaries and PD forms will be performed by the CRO appointed by Dompé. The PD will be recorded during the study and the final list of PDs will be available before database lock.

All data will be verified in a timely manner for missing information, inconsistencies, and for any necessary medical clarifications. Queries arising from the edit checks (either programmed or manual) will be sent to the Investigator for response.

Once all data queries have been resolved, and comments/changes arisen from the Data Review Meeting incorporated, the study data will be declared to be "clean", and the study database will be locked ready for analysis.

A Data Management Plan will be issued, detailing the flow of data handling from entry in to the eCRF to final database audit.

11.5. DOCUMENTATION REQUIRED PRIOR TO INITIATION OF AND DURING THE STUDY

The following will be required from the Investigator prior to the initiation visit:

- Current, signed and dated Curriculum Vitae of Principal Investigator and any Sub-Investigators/co-workers. Updates should be provided at least every two years.

- Normal ranges of all laboratory tests to be performed at the study site and a recent certification or accreditation of established quality control (or other documentation of established quality control or external quality assessment or other validation). Updates should be provided as soon as any reference value has changed.

- A signed page of the final protocol and any amendments.

- A signed copy of the study Financial Agreement/Clinical Study Agreement with Sponsor or its delegate, including all study specific costs.

- List and any updates of delegated responsibility (Study Team Signature List / Delegation of Responsibilities form).

- A financial disclosure agreement completed and signed by the PI and all Sub-Investigators. If applicable, the PI will provide an updated financial disclosure agreement to the Sponsor 1 year after the completion of the study

11.6. ESSENTIAL DOCUMENT RETENTION

The investigator must ensure that the clinical data required by the study protocol are carefully

reported in the subject's source documents detailing the unique identification number and the date and time of the study procedures performed. Any correction to the source data entries must be carried out by the investigator or a designated member of staff. Incorrect entries must not be covered with correcting fluid, or obliterated, or made illegible in any way. A single stroke must be drawn through the original entry. Corrections have to be dated and initialled. The investigator must provide a reasonable explanation for all missing data. The source documents will be completed, signed by the investigator, the sensitive data will be obscured (i.e. only randomization number will be clearly legible) and the source document will be made available to the Sponsor for data management procedures.

The Investigator will retain copies of all the essential documents (as defined by ICH-GCP) until at least 2 years after the last approval of a marketing application in an ICH region, and until there are no pending or contemplated marketing applications in an ICH region, or at least 2 years have elapsed since the formal discontinuation of clinical development of the Investigational Product. These documents should be retained for a longer period however if required by the applicable regulatory requirements. The Investigator should take measures to prevent accidental or premature destruction of these documents.

The essential documents include at least: the signed protocol, copies of the completed CRFs, signed electronic Informed Consent Forms from all patients who consented, hospital records and other source documents, and all other documentation included in the Investigator Site File and Pharmacy/Dispensing File.

The Investigator will inform Sponsor representative or its delegate of the storage location of these essential documents and must contact Sponsor representative or its delegates before disposing of any. If the Investigator wishes to assign the files to someone else or to remove them to another location, he/she should consult with Sponsor representative or its delegates about this change.

Sponsor representative or its delegates will inform the Investigator in writing when these documents no longer need to be retained.

12. STUDY MANAGEMENT

The study will be performed in accordance with the protocol, the Declaration of Helsinki. The approval and/or the acknowledgment of the study protocol, the SmPC and all other relevant documentation by the National Competent Authority and local EthicsCommittee competent for the study site will be obtained before the start of the study,according to the current regulations. The present clinical study will be carried out according to the current revision of GoodClinical Practice (GCP), ICH topic E6 (R2), and the applicable local law requirements.

12.1. MONITORING

Monitoring will be carried out by appropriate staff of the CRO appointed by Dompé.

If for healthy or logistics reasons on-site monitoring visits will not be possible at the planned time intervals, such visits will be conducted in a virtual mode. A purpose-built source document workspace (Genius SITE VAULT) under the full and unique control of the site staff and completely separated from the study database will be implemented. Site staff will upload certified electronic copies of the source documents and will allow the study monitor to conduct the verification of the source data against the eCRF data. Encription protocols as well audit trails assure full security and regulatory compliance.

The purpose of the monitoring visit is to verify that the rights and the wellbeing of the patient are protected, that the reported data are accurate, complete and verifiable from source documents and that the conduct of the trial complies with the currently approved protocol and any amendments, with ICH GCP (E6-R2), and with regulatory requirements.

Monitoring will also include support to site for handling of samples for the laboratories and electronic device for patients' self-monitoring, IMP accountability and reconciliation, as well as follow-up with site as to SAEs. The CRA will also follow-up resolution of clinical/safety queries. Specific monitoring activities will be detailed in the Monitoring Plan.

Prior to study start, the Investigator will be informed of the anticipated frequency of the monitoring visits. (S)He will also receive a notification prior to each monitoring visit during the course of the study. It is expected that the Investigator and/or his/her sub-Investigator(s) and other appropriate staff, including the pharmacist(s) and responsible of the local laboratory, will be available on the day of the visit to discuss study conduct and to cooperate with the monitor to ensure that any problems detected during the course of these monitoring visits are resolved.

Before any patient enters the study, a representative of Sponsor or its delegate, will meet with the PI and his or her staff to review the procedures to be followed during the study and to train them on recording the data in the eCRF using the electronic data capture (EDC) system. After the first patient is enrolled, the Sponsor/CRO representative, a monitor, will periodically monitor the progress of the study by conducting on-site visits. This CRA will also be able to review query statuses remotely, possibly warranting more frequent communication with the PI and his or her staff. The PI will make available to the CRA the eCRF, source documents, signed consent forms, and all other study-related documents. The PI and his or her staff will be responsible for reviewing eCRF, resolving data queries generated by the CRA via the system, providing missing or corrected data, approving all changes performed on his

or her data, and endorsing the patient data within theeCRF.This approval method will include applying an electronic signature, a uniquely assigned username and password that together will represent a traditional handwritten signature.

12.2. ACCESS TO RECORDS

The Investigator will allow designated Dompefarmaceutici S.p.A. representatives, including staff from the appointed CRO, and regulatory/ethics bodies to have direct access to the source documents to verify the data reported in the eCRF. Source documents are the originals of any documents used by the Investigator or hospital/institution that allow verification of the existence of the patient and substantiate the integrity of the data collected during the trial.

12.3. AUDIT AND INSPECTION

The study site may be audited by Sponsor representativeor its delegate or inspected by a regulatory agency on one or more occasions The Investigator may be informed in advance of such a visit.

12.4. PROTOCOL AMENDMENTS

Any amendment to this protocol will be provided to the PI in writing by Dompé farmaceutici s.p.a. No protocol amendment may be implemented (with the exceptions noted below) before it has been approved by the IRB/IEC and the signature page, signed by the PI, has been received by Dompé farmaceutici s.p.a. If the protocol is amended to eliminate or reduce the risk to patients, the amendment may be implemented before IRB/IEC review and approval. However, the IRB/EC must be informed in writing of such an amendment, and approval must be obtained within reasonable time limits. Deviating from the protocol is permitted only if absolutely necessary for the safety or clinical management of the patients and must immediately be reported to Dompé farmaceutici s.p.a.

12.5. DISCONTINUATION OF THE STUDY

Dompé farmaceutici s.p.a. reserves the right to terminate the study in its entirety or at a specific study center at any timeon the basis of new information regarding safety or efficacy, or if study progress is unsatisfactory, or for other valid administrative reasons.

12.6. PUBLICATIONS

Study results will be communicated in full to the competent Health Authorities by the submission of a complete clinical study report.
The sponsor agrees that the study results may be published by the investigator, and the investigator agrees to submit any manuscript (abstract, publication, paper, etc.) to the sponsor before any public disclosure.
This will be done in order to ensure that clinical study results are reported in an objective, accurate and balanced manner. The sponsor reviews the proposed manuscripts, before submission, within a reasonable period of time (30-90 days in relation with the complexity of the work).

The investigator will also be provided by the sponsor with the clinical study report and the results of any additional analysis, tables, figures, etc. undertaken for the purposes of the article, in order to take responsibility for the content of the publication(s). On an exceptional basis, the sponsor may temporarily delay registration of certain data elements (e.g. compound, name, outcome, measures, etc.) to seek necessary intellectual property protection. This is because early disclosure of such data could, in some circumstances, prevent or negatively impact patentability.

13. APPENDICES

13.1. APPENDIX 1 - SPONSOR APPROVAL PAGE

Multicenter, adaptive, randomized, placebo-controlled, double blind, parallel-group Phase 2/3 trial, to study efficacy and safety of two doses of raloxifene in adult patients with early diagnosis of paucisymptomatic COVID-19 positive to SARS-CoV-2 and mild to moderate symptoms.

Sponsor Medical Expert: _____, CMO Date: 22 / 12 / 2020
Printed Name, Title

Sponsor Clinical Trial Manager: _____ Date: 22 / 12 / 2020
Printed Name, Title
CLINICAL OPERATIONS SPECIALIST

Sponsor Head of Clinical Operations _____ Date: 22 / 12 / 2020
Printed Name, Title

13.2. APPENDIX 2 - COORDINATING INVESTIGATOR AND INVESTIGATOR'S SIGNATURE PAGE

Coordinating Investigator	Prof. Emanuele Nicastri
Signature:	
Date:	15/2/2021

Investigator's Statement

I have read study protocol *RLX0120 - Multicenter, adaptive, randomized, placebo-controlled, double blind, parallel-group Phase 2/3 trial, to study efficacy and safety of two doses of raloxifene in adult paucisymptomatic COVID-19 patients* and agree to conduct the study as outlined in the protocol, and in accordance with the Declaration of Helsinki, ICH-GCP and any local regulations, being responsible for personally supervise the study conduct and ensure study staff complies with protocol requirement.

Name of Principal Investigator (block letters): EMANUELE NICASTRI

Signature: _____ Date: 15/2/2021

13.3. APPENDIX 3 - COMMON TERMINOLOGY CRITERIA FOR ADVERSE EVENTS (CTCAE) VERSION 5.0

The CTCAE version 5.0 can be accessed via the following link:

https://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/CTCAE_v5_Quick_Reference_8.5x11.pdf

14. REFERENCES

[1] European Centre for Disease Prevention and Control. Outbreak of novel coronavirus disease 2019 (COVID-19): increased transmission globally – fifth update, 2 March 2020. ECDC: Stockholm; 2020; Guan WJ et al., N Engl J Med. 2020 doi: 10.1056/NEJMoa2002032

[2] Scientific Discussion held for the approval of Evista (EPAR), 1 October 2003.

[3] Summary of Product Characteristics (Annex 1) of Evista – 60mg.

[4] Doran PM et al., *Journal of Bone and Mineral Research* 2001; 6: 2118-2125.

[5] Duschek EJ et al., *European Journal of Endocrinology* 2004; 150: 539–546.

[6] Furusyo N et al., *Journal of Hepatology.* 2012; 57: 1186-1192.

[7] Breart G et al., *Osteoporosis International* 2020; 21: 1181-1187.

[8] COVID-19 Treatment Guidelines Panel. Coronavirus Disease 2019 (COVID-19) Treatment Guidelines. National Institutes of Health.

[9] Pereira, H et al., 1989 *Coronaviridae. Andrewes 'Viruses of Vertebrates*, 5th ed.pp. 42-57; Holmes, K.V. et al., 1996 *Virology* 1, 1075-1093.

[10] Anand K et al., *Science* 2003; 300: 1763-7.

[11] Strope JD et al., *Are sex discordant outcomes in COVID-19 related to sex hormones? Sem. Oncol*. 2020.

[12] Ding T et al., *A Multi-hospital Study in Wuhan, China : Protective Effects of Non-menopause and Female Hormones on SARS-CoV-2 infection,* preprint 2020.

[13] Hussain A et al., *COVID-19 and diabetes: Knowledge in progress,* Diabetes Res. Clin. Pract. 2020.

[14] Finucane FM et al., *Coronavirus and Obesity: Could Insulin Resistance Mediate the Severity of Covid-19 Infection?*, Front. Public Health 2020; 8:184.

[15] Rudragouda et al., *Journal of Immunology* 2017; 198: 4049-4053.

[16] Breithaupt-Faloppa AC et al., *17β-Estradiol, a potential ally to alleviate SARS-CoV-2 infection*, Clinics 2020.

[17] Dyall J et al., *Antimicrob Agents Chemother* 2014; 58: 4885-4893; Channappanavar R et al., *J Immunol* 2017; 198: 4046–4053.

[18] Montoya MC et al., *Repurposing Estrogen Receptor Antagonists for the Treatment of Infectious Disease*, mBio 2018;9:e02272-18; Laurence J et al. *Blood* 1990; 75: 696-703; Watashi K et al. *J BiolChem* 2007; 282: 32765-32772.

[19] Johansen LM et al., *FDA-Approved Selective Estrogen Receptor Modulators Inhibit Ebola Virus Infection*, Science Translational Medicine 2013;5:190ra79.

[20] Zhao Y et al., *Toremifene interacts with and destabilizes the Ebola virus glycoprotein*, Nature 2016;535:169-172.

[21] Jeffrey A et al., *Journal of Steroid Biochemistry and Molecular Biology* 1997;61:97-106.

[22] Murthy AN et al., American Association of Pharmaceutical Scientists (AAPS) Annual Meeting 2012.

[23] Sun D, *American Association of Pharmaceutical Scientists Journal* 2020 26;22-77.

[24] Barker RO et al., *Distribution of the National Early Warning Score (NEWS) in care home residents*, Age and Ageing. 2019 Dec 1;49(1):141-145.