Clinical trial results:
A pilot, open label, phase II clinical trial of nebulised recombinant tissue-Plasminogen Activator (rtPA)in patients with COVID-19 ARDS: The Plasminogen Activator COVID-19 ARDS (PACA) trial

Summary

EudraCT number	2020-001640-26
Trial protocol	GB
Global end of trial date	28 September 2021
Results information	
Result version number	v1 (current)
This version publication date	30 December 2022
First version publication date	30 December 2022

Trial information

Trial identification	
Sponsor protocol code	132151
Additional study identifiers	
ISRCTN number	-
ClinicalTrials.gov id (NCT number)	NCT04356833
WHO universal trial number (UTN)	-

Notes:

Sponsors	
Sponsor organisation name	University College London
Sponsor organisation address	Gower Street, London, United Kingdom, WC1E 6BT
Public contact	Joint Research Office, University College London , ctimps@ucl.ac.uk
Scientific contact	Professor Pratima Chowdary , Katharine Dormandy Haemophilia & Thrombosis Centre, Royal Free London NHS Foundation Trust, +44 020 7472 6835, p.chowdary@ucl.ac.uk

Notes:

Paediatric regulatory details	
Is trial part of an agreed paediatric investigation plan (PIP)	No
Does article 45 of REGULATION (EC) No 1901/2006 apply to this trial?	No
Does article 46 of REGULATION (EC) No 1901/2006 apply to this trial?	No

Notes:

Results analysis stage

Analysis stage	Final
Date of interim/final analysis	14 September 2021
Is this the analysis of the primary completion data?	Yes
Primary completion date	18 March 2021
Global end of trial reached?	Yes
Global end of trial date	28 September 2021
Was the trial ended prematurely?	No

Notes:

General information about the trial

Main objective of the trial:

The overall objective of the pilot study is to investigate the potential for clinical efficacy and safety of nebulised recombinant tissue plasminogen activator (rtPA) in patients hospitalised with Severe COVID-19 complicated by mild to severe ARDS as assessed by an improvement in oxygen saturation and incidence of major bleeding events.

Protection of trial subjects:

The study recruited patients requiring either invasive mechanical ventilation (IMV) or non-invasive respiratory support (NIV/NIRS). Eligible patients (or if patients lack capacity, their legal representative) were provided with an information sheet and informed consent was sought. Re-consenting was sought when the patient recovered capacity sufficiently. If the patient refused consent after recovery, any further assessments were stopped.

Background therapy:

The study was designed to provide an additional intervention, to act alongside standard of care practices on the wards and intensive care units (ICU) within the hospital.

Evidence for comparator: -

Actual start date of recruitment	22 April 2020
Long term follow-up planned	No
Independent data monitoring committee (IDMC) involvement?	Yes

Notes:

Population of trial subjects

Subjects enrolled per country

Country: Number of subjects enrolled	United Kingdom: 53
Worldwide total number of subjects	53
EEA total number of subjects	0

Notes:

Subjects enrolled per age group

In utero	0
Preterm newborn - gestational age < 37 wk	0
Newborns (0-27 days)	0

Infants and toddlers (28 days-23 months)	0
Children (2-11 years)	0
Adolescents (12-17 years)	0
Adults (18-64 years)	28
From 65 to 84 years	23
85 years and over	2

Subject disposition

Recruitment

Recruitment details:

Cohort 1: 9 patients receiving IMP and SoC. 6 patients on invasive mechanical ventilation (IMV), 3 receiving non-invasive respiratory support (NIRS). 18 matched controls, 2 controls for every 1 treatment patient. Recruitment: Apr - Jul 2020
Cohort 2: 26 patients receiving IMP and SoC. 12 on IMV, 14 receiving NIRS. Recruitment: Jan - Mar 2021

Pre-assignment

Screening details:

1. Patients with COVID-19 confirmed by PCR or radiologically
2. Patients on IMV:
PaO2/FiO2 of ≤ 300
Intubated > 6 hrs but less than seven days
3. Patients on NIRS:
PaO2/FiO2 ≤ 300 (or equivalent calculation from SpO2/FiO2)
In-patient >6 hours and being actively treated
On non-invasive ventilator support

Period 1

Period 1 title	Overall Study (overall period)
Is this the baseline period?	Yes
Allocation method	Not applicable
Blinding used	Not blinded

Blinding implementation details:

N/A

Arms

Are arms mutually exclusive?	No
Arm title	Cohort 1 - Nebulised rt-PA

Arm description:

For patients in the rt-PA group, 10 mg of rt-PA will be given in addition to standard of care for COVID-19 acute respiratory distress syndrome (ARDS).

Arm type	Experimental
Investigational medicinal product name	Alteplase
Investigational medicinal product code	
Other name	Recombinant tissue plasminogen activator (rt-PA), Actilyse (Brand name)
Pharmaceutical forms	Powder and solution for solution for injection
Routes of administration	Inhalation use

Dosage and administration details:

Actilyse 10 mg powder and solvent for solution for injection and infusion or Actilyse 20 mg powder and solvent for solution for injection and infusion. Actilyse is authorised as an intravenous formulation, Avtilyse is not authorised for administration via a nebulised route.

In routine clinical practice, rtPA is administered intravenously. For this study, 10mg rtPA was administered by nebulisation every 6 hours for a maximum of 14 treatment days in cohort 1.
In cohort 2, patients requiring NIV or standard oxygen therapy, received a loading dose of 20 mg three times a day for 2 days followed by 20 mg twice a day. Patients requiring IMV received 20 mg every 8 hrs. Treatment was administered for a maximum of 14 treatment days.

Arm title	Cohort 1- historical matched controls

Arm description:

No Intervention: Historical matched controls
Historical matched controls were recruited at a ratio of 2 controls to every 1 rtPA + SOC arm patient, and were matched according to the following characteristics:

1. Ventilation and oxygen type (IMV and non-invasive oxygen support)
2. Severity as determined by PaO2/FiO2 ratio
3. Gender
4. Age (+/- 2 years, up to a maximum of 10 years)
5. Ethnicity

Arm type	No intervention

No investigational medicinal product assigned in this arm

Arm title	Cohort 2- Nebulised rt-PA

Arm description:

In cohort 2, fewer timepoints were collected, which will allowed for more rapid recruitment while at the same time not compromising safety monitoring. A more flexible dosing regimen for rtPA was utilised. Patients on IMV received 60mg daily over three doses for up to 14 days. Patients on NIV received 60mg daily for over 3 doses for two days, followed by 12 days receiving 40mg
daily over two doses

Arm type	Experimental
Investigational medicinal product name	Alteplase
Investigational medicinal product code	
Other name	Recombinant tissue plasminogen activator (rt-PA), Actilyse (Brand name)
Pharmaceutical forms	Powder and solvent for solution for injection/infusion
Routes of administration	Inhalation use

Dosage and administration details:

Actilyse 10 mg powder and solvent for solution for injection and infusion or Actilyse 20 mg powder and solvent for solution for injection and infusion. Actilyse is authorised as an intravenous formulation, Avtilyse is not authorised for administration via a nebulised route.

In routine clinical practice, rtPA is administered intravenously. For this study, 10mg rtPA was administered by nebulisation every 6 hours for a maximum of 14 treatment days in cohort 1.
In cohort 2, patients requiring NIV or standard oxygen therapy, received a loading dose of 20 mg three times a day for 2 days followed by 20 mg twice a day. Patients requiring IMV received 20 mg every 8 hrs. Treatment was administered for a maximum of 14 treatment days.

Arm title	Cohort 2- Nebulised rt-PA - IMV

Arm description:

Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) IMV

Arm type	Experimental
Investigational medicinal product name	Alteplase
Investigational medicinal product code	
Other name	Recombinant tissue plasminogen activator (rt-PA), Actilyse (Brand name)
Pharmaceutical forms	Powder and solvent for solution for injection/infusion
Routes of administration	Inhalation use

Dosage and administration details:

Actilyse 10 mg powder and solvent for solution for injection and infusion or Actilyse 20 mg powder and solvent for solution for injection and infusion. Actilyse is authorised as an intravenous formulation, Avtilyse is not authorised for administration via a nebulised route.

In routine clinical practice, rtPA is administered intravenously. For this study, 10mg rtPA was administered by nebulisation every 6 hours for a maximum of 14 treatment days in cohort 1.
In cohort 2, patients requiring NIV or standard oxygen therapy, received a loading dose of 20 mg three times a day for 2 days followed by 20 mg twice a day. Patients requiring IMV received 20 mg every 8 hrs. Treatment was administered for a maximum of 14 treatment days.

Arm title	Cohort 2- Nebulised rt-PA - NIV

Arm description:

Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) NIV

Arm type	Experimental

Investigational medicinal product name	Alteplase
Investigational medicinal product code	
Other name	Recombinant tissue plasminogen activator (rt-PA), Actilyse (Brand name)
Pharmaceutical forms	Powder and solution for solution for injection
Routes of administration	Injection

Dosage and administration details:

Actilyse 10 mg powder and solvent for solution for injection and infusion or Actilyse 20 mg powder and solvent for solution for injection and infusion. Actilyse is authorised as an intravenous formulation, Avtilyse is not authorised for administration via a nebulised route.

In routine clinical practice, rtPA is administered intravenously. For this study, 10mg rtPA was administered by nebulisation every 6 hours for a maximum of 14 treatment days in cohort 1.
In cohort 2, patients requiring NIV or standard oxygen therapy, received a loading dose of 20 mg three times a day for 2 days followed by 20 mg twice a day. Patients requiring IMV received 20 mg every 8 hrs. Treatment was administered for a maximum of 14 treatment days.

Number of subjects in period 1	Cohort 1 - Nebulised rt-PA	Cohort 1- historical matched controls	Cohort 2- Nebulised rt-PA
Started	9	18	26
Completed	9	18	26

Number of subjects in period 1	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV
Started	12	14
Completed	12	14

Baseline characteristics

Reporting groups	
Reporting group title	Cohort 1 - Nebulised rt-PA
Reporting group description:	
For patients in the rt-PA group, 10 mg of rt-PA will be given in addition to standard of care for COVID-19 acute respiratory distress syndrome (ARDS).	
Reporting group title	Cohort 1- historical matched controls
Reporting group description:	
No Intervention: Historical matched controls Historical matched controls were recruited at a ratio of 2 controls to every 1 rtPA + SOC arm patient, and were matched according to the following characteristics: 1. Ventilation and oxygen type (IMV and non-invasive oxygen support) 2. Severity as determined by PaO2/FiO2 ratio 3. Gender 4. Age (+/- 2 years, up to a maximum of 10 years) 5. Ethnicity	
Reporting group title	Cohort 2- Nebulised rt-PA
Reporting group description:	
In cohort 2, fewer timepoints were collected, which will allowed for more rapid recruitment while at the same time not compromising safety monitoring. A more flexible dosing regimen for rtPA was utilised. Patients on IMV received 60mg daily over three doses for up to 14 days. Patients on NIV received 60mg daily for over 3 doses for two days, followed by 12 days receiving 40mg daily over two doses	
Reporting group title	Cohort 2- Nebulised rt-PA - IMV
Reporting group description:	
Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) IMV	
Reporting group title	Cohort 2- Nebulised rt-PA - NIV
Reporting group description:	
Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) NIV	

Reporting group values	Cohort 1 - Nebulised rt-PA	Cohort 1- historical matched controls	Cohort 2- Nebulised rt-PA
Number of subjects	9	18	26
Age categorical			
Analysis of Cohort1 and Cohort2 was undertaken separately.			
Units: Subjects			
In utero Preterm newborn infants (gestational age < 37 wks) Newborns (0-27 days) Infants and toddlers (28 days-23 months) Children (2-11 years) Adolescents (12-17 years) Adults (18-64 years) From 65-84 years 85 years and over			
Age continuous Units: years			
arithmetic mean	65.1	66.8	63.6
standard deviation	± 9.9	± 11.8	± 12.14

Gender categorical			
Analysis of Cohort1 and Cohort2 was undertaken separately			
Units: Subjects			
Female	5	9	7
Male	4	9	19
PaO2/FiO2 ratio			
Cohort 1: All available P/F ratio values were extracted per day and summarised every 4 hours (± 2h). Time 0 is baseline. Cohort 2: Up to six P/F ratio values were extracted per day including the worst P/F ratio over the preceding one day; however, the analysis for Cohort 2 includes only the lowest value for the day. Analysis of C1 and C2 was undertaken separately.			
Units: mmHg (rounded)			
arithmetic mean	154	149	123
standard deviation	± 53	± 72	± 35

Reporting group values	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV	Total
Number of subjects	12	14	53
Age categorical			
Analysis of Cohort1 and Cohort2 was undertaken separately.			
Units: Subjects			
In utero			
Preterm newborn infants (gestational age < 37 wks)			
Newborns (0-27 days)			
Infants and toddlers (28 days-23 months)			
Children (2-11 years)			
Adolescents (12-17 years)			
Adults (18-64 years)			
From 65-84 years			
85 years and over			
Age continuous			
Units: years			
arithmetic mean	64.2	63.1	
standard deviation	± 8.89	± 14.7	-
Gender categorical			
Analysis of Cohort1 and Cohort2 was undertaken separately			
Units: Subjects			
Female	5	2	21
Male	7	12	32
PaO2/FiO2 ratio			
Cohort 1: All available P/F ratio values were extracted per day and summarised every 4 hours (± 2h). Time 0 is baseline. Cohort 2: Up to six P/F ratio values were extracted per day including the worst P/F ratio over the preceding one day; however, the analysis for Cohort 2 includes only the lowest value for the day. Analysis of C1 and C2 was undertaken separately.			
Units: mmHg (rounded)			
arithmetic mean	120	126	
standard deviation	± 28	± 42	-

End points

End points reporting groups

Reporting group title	Cohort 1 - Nebulised rt-PA

Reporting group description:

For patients in the rt-PA group, 10 mg of rt-PA will be given in addition to standard of care for COVID-19 acute respiratory distress syndrome (ARDS).

Reporting group title	Cohort 1- historical matched controls

Reporting group description:

No Intervention: Historical matched controls
Historical matched controls were recruited at a ratio of 2 controls to every 1 rtPA + SOC arm patient, and were matched according to the following characteristics:

1. Ventilation and oxygen type (IMV and non-invasive oxygen support)
2. Severity as determined by PaO2/FiO2 ratio
3. Gender
4. Age (+/- 2 years, up to a maximum of 10 years)
5. Ethnicity

Reporting group title	Cohort 2- Nebulised rt-PA

Reporting group description:

In cohort 2, fewer timepoints were collected, which will allowed for more rapid recruitment while at the same time not compromising safety monitoring. A more flexible dosing regimen for rtPA was utilised. Patients on IMV received 60mg daily over three doses for up to 14 days. Patients on NIV received 60mg daily for over 3 doses for two days, followed by 12 days receiving 40mg
daily over two doses

Reporting group title	Cohort 2- Nebulised rt-PA - IMV

Reporting group description:

Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) IMV

Reporting group title	Cohort 2- Nebulised rt-PA - NIV

Reporting group description:

Cohort 2- Nebulised recombinant tissue-Plasminogen Activator (rt-PA) NIV

Primary: Efficacy - PaO2/FiO2 Ratio

End point title	Efficacy - PaO2/FiO2 Ratio[1][2]

End point description:

PaO2/FiO2 measured at multiple timepoints over the study period: baseline, during treatment, end of treatment, 3 days post end of treatment and 5 days post end of treatment. For Cohort 1, all available P/F ratio values were extracted per day and summarised every 4 hours (± 2h). For Cohort 2, up to six P/F ratio values were extracted per days including the worst P/F ratio over the preceding one day; however, the analysis for Cohort 2 included only the lowest value for the day; Limited data was observed due to patient discharge or death: Day 14 is presented below, and the last value available on treatment regardless of the duration of treatment (death or discharge may have occurred within 14 days) was summarised post-hoc.
Summarized values were rounded to the nearest integer

End point type	Primary

End point timeframe:

Across 28 days - Limited data was observed due to patient discharge or death: Day 14 is presented below

Notes:

[1] - No statistical analyses have been specified for this primary end point. It is expected there is at least one statistical analysis for each primary end point.
Justification: Only descriptive statistics are presented

[2] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data presented for all patients in Cohort 2

End point values	Cohort 1 - Nebulised rt-PA	Cohort 1- historical matched controls	Cohort 2- Nebulised rt-PA	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	2[3]	4	13[4]	
Units: PaO2 / FiO2 ratio				
arithmetic mean (standard deviation)				
Day 14	227 (± 83)	209 (± 49)	191 (± 105)	

Notes:

[3] - for the post-hoc analysis of Last On-Treatment Day, n=9, mean=218 and SD =73

[4] - for the post-hoc analysis of Last On-Treatment Day, n=26, mean=207 and SD =110

Statistical analyses

No statistical analyses for this end point

Primary: Safety- major bleeding events directly attributable to study drug

End point title	Safety- major bleeding events directly attributable to study drug[5][6]

End point description:

Incidence and severity of major bleeding events assessed as related to the study drug summarised in each group.
Bleeding events were not recorded in historical matched controls (cohort 1).

End point type	Primary

End point timeframe:

28 days

Notes:

[5] - No statistical analyses have been specified for this primary end point. It is expected there is at least one statistical analysis for each primary end point.
Justification: Only descriptive statistics are presented

[6] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	9	26		
Units: Count of participants				
number (not applicable)				
Incidence and severity of major bleeding events	0	1		

Statistical analyses

No statistical analyses for this end point

Primary: Safety- Serious adverse events causally related to treatment

End point title	Safety- Serious adverse events causally related to

End point description:

Incidence of serious adverse events causally related to treatment. Adverse events (other than bleeding

events) were not recorded in the study.

End point type	Primary

End point timeframe:

28 days

Notes:

[7] - No statistical analyses have been specified for this primary end point. It is expected there is at least one statistical analysis for each primary end point.
Justification: Only descriptive statistics are presented

[8] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	9	26		
Units: Participants				
number (not applicable)				
Incidence of serious adverse events	0	0		

Statistical analyses

No statistical analyses for this end point

Primary: Safety- Fibrinogen levels

End point title	Safety- Fibrinogen levels[9][10]

End point description:

Decrease in fibrinogen levels to < 1gm/L during the treatment period and 48 hours after the last dose. Fibrinogen levels were not recorded for the historical matched controls (cohort 1).

End point type	Primary

End point timeframe:

28 days

Notes:

[9] - No statistical analyses have been specified for this primary end point. It is expected there is at least one statistical analysis for each primary end point.
Justification: Only descriptive statistics are presented

[10] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	9	26		
Units: Count of participants				
number (not applicable)				
Decrease in fibrinogen levels to < 1gm/L	0	0		

Statistical analyses

No statistical analyses for this end point

Secondary: Lung Compliance

End point title	Lung Compliance[11]

End point description:

Changes in lung compliance (defined as tidal volume / (peak inspiratory pressure – PEEP) from baseline (same day as start of treatment but prior to start of treatment) was a secondary outcome as defined in the protocol. Limited data was observed due to patient discharge or death, therefore the last value available on treatment regardless of the duration of treatment (death or discharge may have occurred within 14 days) was summarised post-hoc and presented below. For cohort 2, the daily measurement coinciding with the worst PF ratio (as close as possible in date and time) was used for the summaries. Lung compliance not recorded in the historical matched controls or in patients in the NIV group.

End point type	Secondary

End point timeframe:

across 28 days (last on-treatment presented)

Notes:

[11] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Lung compliance not recorded in the historical matched controls or in patients in the NIV group

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA		
Subject group type	Reporting group	Reporting group		
Number of subjects analysed	6	12		
Units: mL/cmH2O				
arithmetic mean (standard deviation)				
Lung compliance	40.4 (± 22.5)	37.6 (± 42.8)		

Statistical analyses

No statistical analyses for this end point

Secondary: Clinical Status as Determined by a 7-Point WHO Ordinal Scale

End point title	Clinical Status as Determined by a 7-Point WHO Ordinal

End point description:

Clinical status as assessed by a 7-point WHO ordinal scale at baseline and daily up to 5 days post end of treatment and at day 28, discharge or death (whichever comes first).

1. Limitation of activities
2. Hospitalized, no oxygen therapy
3. Oxygen by mask or nasal prongs
4. Non-invasive ventilation or high-flow oxygen
5. Intubation and mechanical ventilation
6. Ventilation + additional organ support (vasopressor, RRT, ECMO)
7. Death

This outcome measure was modified between cohort 1 and 2, and the 7-point WHO scale was only collected from patients in cohort 2. Limited data was observed due to patient discharge or death, therefore only data for day 14 for cohort 2 are presented below.

End point type	Secondary

End point timeframe:

28 days (Day 14 presented)

Notes:

[12] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.

Justification: This outcome measure was modified between cohort 1 and 2, and the 7-point WHO scale was only collected from patients in cohort 2

End point values	Cohort 2- Nebulised rt-PA			
Subject group type	Reporting group			
Number of subjects analysed	26			
Units: 7 Point Ordinal Scale				
1. Limitation of activities	0			
2. Hospitalized, no oxygen therapy	2			
3. Oxygen by mask or nasal prongs	3			
4. Non-invasive ventilation or high-flow oxygen	0			
5. Intubation and mechanical ventilation	1			
6. Ventilation + additional organ support	6			
7. Death	0			
Data Missing	14			

Statistical analyses

No statistical analyses for this end point

Secondary: Sequential Organ Failure Assessment (SOFA) Score

End point title	Sequential Organ Failure Assessment (SOFA) Score[13]

End point description:

Mean daily Sequential Organ Failure Assessment (SOFA) score at baseline and daily up to 5 days post end of treatment. Limited data was observed due to patient discharge or death, therefore day 14 is presented below.

End point type	Secondary

End point timeframe:

28 days (day 14 presented)

Notes:

[13] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.

Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 1- historical matched controls	Cohort 2- Nebulised rt-PA	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	7[14]	2[15]	10	
Units: Score on a scale				
arithmetic mean (standard deviation)	7.71 (± 4.82)	7 (± 7.07)	8.3 (± 4.35)	

Notes:

[14] - Limited data was observed due to patient discharge or death, therefore day 14 is presented below.

[15] - Limited data was observed due to patient discharge or death, therefore day 14 is presented below.

Statistical analyses

No statistical analyses for this end point

Secondary: Number of Oxygen Free Days

End point title	Number of Oxygen Free Days[16]

End point description:

Number of oxygen free days, up to 28 days or death or discharge, whichever occurs first was a secondary outcome as defined in the protocol. However, a post-hoc exploration of the data recalculated the number of Oxygen free days, taking into account the number of days post-discharge. For this new calculation, days post-discharge were assumed to be days when the patient did not receive oxygen or ventilation. Number of oxygen free days was not recorded in historical matched controls (cohort 1)

End point type	Secondary

End point timeframe:

28 days

Notes:

[16] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	9	12	14	
Units: Days				
arithmetic mean (standard deviation)	6.1 (± 9.6)	4.42 (± 8.1)	13.43 (± 11.1)	

Statistical analyses

No statistical analyses for this end point

Secondary: Number of ventilator free days

End point title	Number of ventilator free days[17]

End point description:

Number of ventilator free days, up to 28 days or death or discharge, whichever occurs first was a secondary outcome as defined in the protocol. However, a post-hoc exploration of the data recalculated the number of ventilator free days, taking into account the number of days post-discharge. For this new calculation, days post-discharge were assumed to be days when the patient did not receive oxygen or ventilation. Ventilator free days were not recorded in the historical matched controls (cohort 1)

End point type	Secondary

End point timeframe:

28 days

Notes:

[17] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	9	12	14	
Units: Days				
arithmetic mean (standard deviation)	11.78 (± 12.98)	5.75 (± 9.94)	21.4 (± 9.68)	

Statistical analyses

No statistical analyses for this end point

Secondary: New Oxygen Use (Relapse)

End point title	New Oxygen Use (Relapse)[18]

End point description:

Incidence and number of days of new oxygen use, non-invasive ventilation or high flow oxygen devices in the first 28 days. This was a secondary outcome as defined in the protocol, however a post-hoc exploration of the data recalculated the number of days of new oxygen use and "New Oxygen use (relapse)" was defined as any patient requiring oxygen support after being on room air for a whole day.

End point type	Secondary

End point timeframe:

28 days

Notes:

[18] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	9	12	14	
Units: Subjects	0	1	0	

Statistical analyses

No statistical analyses for this end point

Secondary: New IMV (Deterioration)

End point title	New IMV (Deterioration)[19]

End point description:

Incidence and number of days of new mechanical ventilation use during in the first 28 days was a secondary outcome measure as defined in the protocol. However, a post-hoc exploration of the data

recalculated the number of days of "New IMV (deterioration)" which was defined as any patient requiring mechanical ventilation (IMV) after a period of non-mechanical ventilation after receiving r-tPA.

End point type	Secondary

End point timeframe:

28 days

Notes:

[19] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data not available for the matched Historical Control Group, in cohort 1

End point values	Cohort 1 - Nebulised rt-PA	Cohort 2- Nebulised rt-PA - IMV	Cohort 2- Nebulised rt-PA - NIV	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	9	12	14	
Units: Subjects	0	1	2	

Statistical analyses

No statistical analyses for this end point

Secondary: In-hospital Mortality

End point title	In-hospital Mortality[20]

End point description:

In-hospital Mortality

End point type	Secondary

End point timeframe:

28 days

Notes:

[20] - The end point is not reporting statistics for all the arms in the baseline period. It is expected all the baseline period arms will be reported on when providing values for an end point on the baseline period.
Justification: Data presented for all patients in Cohort 2

End point values	Cohort 1 - Nebulised rt-PA	Cohort 1- historical matched controls	Cohort 2- Nebulised rt-PA	
Subject group type	Reporting group	Reporting group	Reporting group	
Number of subjects analysed	9	18	26	
Units: Subjects	1	10	8	

Statistical analyses

No statistical analyses for this end point

Adverse events

Adverse events information

Timeframe for reporting adverse events:

Maximum of 28 days per patient, starting from dosing day 1

Adverse event reporting additional description:

Only SAEs related to the IMP, and all bleeding events were reported. Not reported for historical controls (C1) Bleeding events were identified as adverse events of special interest (AESI). The ISTH classification for bleed severity as used for anticoagulation studies was implemented. Only serious adverse reactions were reported.

Assessment type	Systematic

Dictionary used

Dictionary name	CTCAE
Dictionary version	4

Reporting groups

Reporting group title	Nebulised rt-PA - Cohort 1

Reporting group description:

Experimental: Nebulised recombinant tissue-Plasminogen Activator (rt-PA) - cohort 1

Initial dosing regime: 10 mg of rt-PA dissolved in 5 ml of diluent given every 6 hrs (resulting in a total daily dose of 40mg) for a maximum of 66 hrs, in addition to standard of care for COVID-19 acute respiratory distress syndrome (ARDS). Following protocol amendment, dosing duration was increased from 3 days to up to 14 days of rt-PA treatment. Six patients were receiving Invasive mechanical ventilation and 3 were receiving non-invasive ventilation

Reporting group title	Nebulised Rt-PA - Cohort 2

Reporting group description:

Experimental: Nebulised recombinant tissue-Plasminogen Activator (rt-PA) - cohort 2

In cohort 2, fewer timepoints were collected, which will allowed for more rapid recruitment while at the same time not compromising safety monitoring. A more flexible dosing regimen for rtPA was utilised. 26 patients were recruited in total, of these, 12 patients were on IMV and 14 patients were on non-invasive oxygen support.

Patients on IMV received 60mg daily over three doses for up to 14 days

Patients on NIV received 60mg daily for over 3 doses for two days, followed by 12 days receiving 40mg daily over two doses..

Serious adverse events	Nebulised rt-PA - Cohort 1	Nebulised Rt-PA - Cohort 2	
Total subjects affected by serious adverse events			
subjects affected / exposed	0 / 9 (0.00%)	1 / 26 (3.85%)	
number of deaths (all causes)	1	8	
number of deaths resulting from adverse events	0	0	
Nervous system disorders			
Cerebral bleed			
subjects affected / exposed	0 / 9 (0.00%)	1 / 26 (3.85%)	
occurrences causally related to treatment / all	0 / 0	0 / 1	
deaths causally related to treatment / all	0 / 0	0 / 1	

Frequency threshold for reporting non-serious adverse events: 0 %

Non-serious adverse events	Nebulised rt-PA - Cohort 1	Nebulised Rt-PA - Cohort 2	
Total subjects affected by non-serious adverse events			
subjects affected / exposed	4 / 9 (44.44%)	13 / 26 (50.00%)	
Surgical and medical procedures			
Bleed: Central venous catheters access site			
subjects affected / exposed	2 / 9 (22.22%)	0 / 26 (0.00%)	
occurrences (all)	2	0	
Blood-stained tracheobronchial secretion			
subjects affected / exposed	1 / 9 (11.11%)	8 / 26 (30.77%)	
occurrences (all)	1	14	
Tracheostomy site bleed			
subjects affected / exposed	2 / 9 (22.22%)	1 / 26 (3.85%)	
occurrences (all)	2	1	
Bleed: chest-drain related			
subjects affected / exposed	0 / 9 (0.00%)	1 / 26 (3.85%)	
occurrences (all)	0	1	
General disorders and administration site conditions			
Other			
subjects affected / exposed	1 / 9 (11.11%)	3 / 26 (11.54%)	
occurrences (all)	1	3	
Ear and labyrinth disorders			
Epistaxis			
subjects affected / exposed	0 / 9 (0.00%)	1 / 26 (3.85%)	
occurrences (all)	0	3	
Gastrointestinal disorders			
Gastro-intestinal bleed			
subjects affected / exposed	1 / 9 (11.11%)	2 / 26 (7.69%)	
occurrences (all)	1	2	

More information

Substantial protocol amendments (globally)

Were there any global substantial amendments to the protocol? Yes

Date	Amendment
30 April 2020	How the diagnosis of COVID-19 will be made – this will be either by PCR or radiologically. Certain sections have been clarified regarding fibrinogen monitoring and the action to be taken. In particular, it was unclear whether replacement therapy should be given when fibrinogen levels are <1.5 g/L or <1.0 g/L. This has been clarified. A drop in fibrinogen <1.5 g/L is not a stopping criteria for the trial and accordingly this has never been listed in the Stopping Criteria section (7.10) as the intention has always been that this was a reason for treatment discontinuation in an individual patient. The wording in the protocol has been clarified around this. Updated exclusion criteria no 7 as follows: 'Patients considered inappropriate for critical care (prior decision re ceiling of care established e.g. being considered for palliative care)'. The new example given is more in line with the exclusion criteria. Information has been added around the rationale for not excluding patients who are on anticoagulants and to clarify and provide rationale as to why we will not exclude patients on anticoagulation at higher than standard doses. We have removed the following from the ISTH definition of major bleeding in the stopping criteria (so it will no longer be a stopping criteria for the trial): 'Bleeding causing a fall in haemoglobin level of 20 g/L or more, or leading to transfusion of two or more units of whole blood or red cells.' This is because the patients on ICU are regularly receiving blood transfusions, and even minor bleeding may cause a drop in Hb because of other complications. We have updated the stopping criteria around bleeding to add 'that occur between start of treatment and 30 h after the last dose' as events past that timepoint would not be considered related to the trial treatment and so would not be considered a reason to stop the trial.

22 May 2020	Updated dosing schedule to allow dosing for a maximum duration of 14 days. The duration of therapy can be extended from five days to a maximum of 14 treatment days in total. This decision can only be made in the absence of any dose-limiting toxicity. Treatment discontinuation: Treatment may be stopped at any point in a patient if any of the following rules are met: - If plasma fibrinogen levels fall to <1.5 g/L, the further dosing of rtPA in the patient will be stopped. - Patient maintains saturation on room air that is either normal or normal for them for 48 hours as assessed by the investigator. - A trial stopping rule has been met. Re-treatment with rtPA after treatment discontinuation due to improvement in patient saturation: If there is a recurrence of COVID-19 ARDS related symptoms or a worsening of PaO2/FiO2 ratio, which the investigator considers could be related to treatment discontinuation, treatment can be restarted. The decision to re-treat will be made within 5 days (120 hrs) from the last dose of treatment. Treatment may not be restarted more than once. Treatment cannot be given for more than 56 doses. The rationale for restarting of treatment must be documented in the notes, along with the absence of toxicity. Re-treatment of patients previously consented and treated on the trial: For patients that have already been treated under the previous 3 day dosing schedule, if they remain eligible for the trial, they will be offered the option to continue treatment as above – the maximum number of days of extra treatment for these patients will be 11 days. Patients (or their legal representatives) must give consent to this re-treatment. The total duration of trial participation for these patients may exceed 28 days. Also updates made to the endpoints in relation to this extended treatment duration.
22 September 2020	Use of historical matched controls: The standard of care arm will be replaced with a historical matched control arm. Historical controls will instead be recruited at a ratio of two controls to every one treatment arm patient. The study team agreed that if controls are matched according to ventilation type, disease severity, age and gender, this should be sufficient to identify controls and also allow for some degree of underlying matching of comorbidities. The identification of historical controls will begin immediately as it is permitted under the notices relating to the Health Service (Control of Patient Information) Regulations 2002. Under this notice, prior consent is not required in order to carry out this process. However, in order to retain the data in line with clinical trial regulations, consent will be sought from patients. If consent is not obtained by the time the notice expires or patients explicitly ask to not be included in the trial as historical controls, their data will be removed. Use of deceased patients as historical use of controls: We would also like to potentially include deceased patients as historical controls if they are a suitable match. The data monitoring committee highlighted the importance of allowing this because if they are not included, it may bias the control arm towards having a more favourable outcome than what standard of care actually provides. Given that GDPR and the data protection act do not apply to deceased people, the study team do not feel that consent is required in order to include deceased patients as historical controls. In addition, it may not be appropriate to contact next of kin for assent given that the patient will have died within the last few months. We would appreciate the RECs views on this and whether they feel it is acceptable to include these patients in the trial without the need to approach their next of kin for assent.

12 November 2020	We have updated the consent section for historical control patients (section 6.4) following a discussion with CAG. Consent will not be sought from any historical controls patients because the use of their personal identifiable information is covered by the COPI notice. We therefore no longer plan to use the patient information sheet and consent form for historical controls (version 1.0 dated 18 Sep 20) as previously approved by REC. We have also made updates to the data to be collected for historical control patients to allow for a more complete comparison between treatment patients and control patients and to clarify that the data will be collected where available for 28 days from the date of admission to hospital, rather than 'at a frequency that where possible matches the treatment arm patient that they have been matched to' as it is not possible to collect for a total of 31 days due to the set-up of the database. We have also updated the criteria to be used for matching of control patients to allow for better matching and made some further clarifications. Other corrections/clarifications: Correction of WHO 7 point score criteria. To clarify that lung compliance will be calculated using tidal volume / (peak inspiratory pressure – PEEP) rather than 'mL/cm HO VT/(PIP – PEEP)'. Driving pressure removed from list of ventilation details to be collected for IMV patients as it is not required. Added the SOFA score to also be collected for control patients The Glasgow coma scale will be recorded based on the output data collected for the historical controls as this is required to calculate the SOFA. Clarification added around the medical history and COVID-19 admission history that is of particular relevance. Also clarified that PaO2/FiO2 will be a calculated field on the trial database derived from the fields for PaO2 and FiO2.
08 January 2021	The major change is the addition of cohort 2, in which we plan to enrol patients for treatment with nebulised recombinant tissue plasminogen activator to firmly establish the safety profile. Preliminary review of the data has not demonstrated any significant drop in fibrinogen or systemic absorption of administration of 40 mg tPA over 24 hrs by inhalation for 3 to 14 days. As most clinicians are used to intravenous preparation, accrual of additional data was felt to be vital in preparation for a randomised control study. Cohort 2 will include up to 30 patients, with a minimum of 10 ventilated and 10 non ventilated patients receiving the intervention. The remainder 10 can be in either group. The dosing has been changed for pragmatic considerations. Ventilated patients will receive 20 mg three times a day instead of 10 mg four times a day. This is to account for the dead space created by the ventilator circuit and we consider will provide a more comparable dose delivery to the non-ventilated patients. Non-ventilated patients will receive 20 mg three times a day for 48 hrs to help build up the dose of tPA, followed by 20 twice a day. Patients can receive the drug for a maximum of 14 days with termination of treatment when patients achieve normal saturation on room air for 24 hrs. The collection of data for cohort 2 is similar to cohort 1, except the frequency of data points will be decreased to once a day. Multiple time points will be retained for the ratio of inspired oxygen and blood oxygen as it is a primary endpoint. Cohort 1 and cohort 2 will also be analysed separately. Co-enrolment into other clinical trials will be allowed, and documented. We have reduced the number of exclusion criteria for patients in Cohort 2 compared to the first cohort. We will also stop treatment in an individual patient if fibrinogen levels fall below 1.0gm/L (previously 1.5g/L) as we will allow patients with a lower baseline fibrinogen level to enter the trial.
15 February 2021	PI change at Barnet Hospital

Notes:

Interruptions (globally)

Were there any global interruptions to the trial? No

Limitations and caveats

Limitations of the trial such as small numbers of subjects analysed or technical problems leading to unreliable data.

Open label, mall sample size, missing data and absence of a concurrent control group. Intensive care stay, a secondary outcome, was not reported due to protocol amendments, differences in design between cohorts and resulting limited comparability.

Notes: