STATISTICAL ANALYSIS PLAN – EFFICACY OF NITRIC OXIDE IN STROKE TRIAL 2 (ENOS-2)

SECTION 1. ADMINISTRATIVE INFORMATION

1 Title and trial registration

1a Title: Efficacy of Nitric Oxide Trial 2
Acronym: ENOS-2
1b Registration: ISRCTN17654248; IRAS project number: 281728

2 SAP version: 1.0 (02 February 2023)

3 Protocol version: 3.0 (14 September 2022)

4 Roles and responsibilities

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5 Signatures

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5a Authors:	Iris Mhlanga		
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ABBREVIATIONS

Abbreviation	Full Text
ACS	Acute Coronary Syndrome
ASU	Acute Stroke Unit
BLR	Binomial Logistic Regression
CI	Confidence Interval
CPHR	Cox Proportional Hazard Ratio
cSVD	Cerebral small vessel disease
CT	Computed Tomography
CTA	Computed Tomographic Angiography
COVID-19	Coronavirus
DBP	Diastolic Blood Pressure
DMC	Data Monitoring Committee
DNR	Do Not Resuscitate
DOAC	Direct Oral Anticoagulants
DSRS	Dysphagia Severity Rating Scale
ENOS-2	Efficacy of Nitric Oxide Trial 2
EQVAS	EuroQol-Visual Analogue Scale
EQ-5D-5L	EuroQoL- 5 Dimension- 5 Level quality of life questionnaire
FF	Fresh Frozen
GCS	Glasgow Comma Scale
GTN	Transdermal glyceryl trinitrate
HDU	High Dependency Unit
HT	Haemorrhagic Transformation
HSUV	Health Status Utility Value
ICH	Intracerebral Haemorrhage
IHD	Ischaemic Heart disease
IMP	Investigational Medicinal Product
IMPD	Investigational Medicinal Product Dossier
IS	Ischaemic Stroke
IV	Intra-venous
ITT	Intention to treat
ITU	Intensive Therapy Unit
LAD	Large Artery Disease
LVO	Large Vessel Occlusion
MEB	Major Extracranial Bleeding);
MHRA	Medicines and Healthcare products Regulatory Agency
MLR	Multiple Linear Regression
MMSE	Mini-Mental State Examination
MRI	Magnetic Resonance Imaging
mRS	modified Rankin Scale
NG	Nasogastric
NIH	National Institutes Health
NHS	National Health Service
NIHSS	National Institutes Health Stroke Scale
NO	Nitric oxide
NTG	Nitroglycerin
NUH	Nottingham University Hospitals NHS Trust
OLR	Ordinal Logistic Regression

PACS	Partial anterior circulation syndrome
PAD	Peripheral Arterial Disease
PEG	Percutaneous Endoscopic Gastrostomy
PICH	Primary intracerebral haemorrhage
POCS	Posterior Circulation Syndrome
PP	Per protocol
QOL	Quality Of Life
RTI	Respiratory Tract Infections
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SAR	Serious Adverse Reaction
SAS	Statistical Analysis System
SBP	Systolic Blood Pressure
SC	Subcutaneous Fluids
SRU	Stroke Rehabilitation Unit
SVD	Small vessel disease
TACS	Total Anterior Circulation Syndrome
TGA	Transient Global Amnesia
TIA	Transient Ischaemic Attack
TICS-M	Telephone Interview for Cognitive Scale-Modified
TSC	Trial Steering Committee
UoN	University of Nottingham
UTI	Urinary Tract Unit
ZDS	Zung Depression Scale

SECTION 2. INTRODUCTION

6 Background and rationale

Stroke: Is common (life-time risk 1/5-6) and devastating (death 25%, dependency 40% at 1 year). [4] Acute treatment is limited to alteplase,[5] mechanical thrombectomy,[6] aspirin, hemicraniectomy and stroke unit care. Anticoagulation is ineffective,[7] neuroprotection unproven,[8,9] and there is no widely-agreed treatment for intracerebral haemorrhage (ICH) although lowering blood pressure (BP) may be beneficial.[10] Developing new interventions in hospitals has failed, in part, due to delayed treatment beyond the 'golden' hour after stroke. The management of physiological disequilibrium - BP,[11] oxygen, glucose,[12] cerebral oedema - remains unclear, and it is reasonable to hypothesise that their treatment, if warranted, should start rapidly after stroke onset.

High BP: Is common (80%) in patients with acute IS and ICH, and is associated independently with increased early recurrence and late death or dependency.[13-15] Whether lowering BP improves outcome through reducing expansion and recurrence, or worsens it through reducing cerebral blood flow (due to dysfunctional autoregulation) remains unclear, in part because most trials started treatment several hours after onset. When assessing functional outcome, trial results have varied from a strong positive trend (INTERACT-2: SBP 14 mmHg lower with intensive treatment in ICH [10]) through neutral effect (IMAGES: BP 4/3 mmHg lower with intravenous magnesium in mixed IS/ICH;[8] CATIS: systolic BP 9 mmHg lower with intensive treatment in IS;[16] ENCHANTED-BP SBP 5.5 mmHg lower with intensive BP-lowering in IS [17]) to strong negative trend (BEST: oral propranolol or atenolol in mixed IS/ICH;[18] INWEST: intravenous nimodipine in IS;[19,20] SCAST: BP 5/2 mmHg lower with oral candesartan in mixed IS/ICH [11]). Meta-analysis, and meta-regression of trial outcomes versus BP change, have not identified benefit.[21,22]

Nitric oxide (NO) and donors such as glyceryl trinitrate (GTN): Are candidate treatments for acute stroke and multiple mechanisms exist by which they might be effective; taken together, these actions may 'buy time' for the brain, protect it and prime patients for arterial reperfusion therapies:

- NO/GTN lowers BP in acute/subacute stroke [23] and so may 'move' patients down the epidemiological curve relating high BP and poor outcome.[13] This mechanism may be of particular relevance in ICH.
- NO dilates cerebral arteries (e.g. middle cerebral) so could increase 'front door' cerebral blood flow (CBF) and peri-lesional perfusion, as seen in the GTN-3 pilot trial.[24]
- NO dilates pial arteries (shown experimentally [25]) so might increase CBF via the 'back-door'.
- NO donors are neuroprotective in preclinical stroke,[26] especially if given early.
- Endogenous NO levels are low in acute stroke;[27] hence, administration will supplement low [NO].
- GTN may 'prime' patients for rt-PA by lowering their BP so that more can be treated, and more rapidly after hospital arrival. RIGHT showed non-significant trends for these.[28]
- GTN, through cerebral vasodilation, may increase access of alteplase to obstructing clot and therefore increase the effectiveness of thrombolysis, i.e., GTN might be additive to rt-PA.

Several NO donors are licensed in the UK and are used widely in patients with ischaemic heart disease, heart failure and severe hypertension; these include intravenous sodium nitroprusside and transdermal glyceryl trinitrate. One uncontrolled pilot study found that sodium nitroprusside lowered BP without altering cerebral perfusion (assessed using CT SPECT), and attenuated platelet function, in patients with recent ischaemic stroke.[29] Four phase II and two phase III randomised controlled trials have assessed transdermal glyceryl trinitrate in patients with acute stroke (IS and ICH) (Table A).

Table A. Characteristics of completed trials of transdermal glyceryl trinitrate.

	GTN-1 [30]	GTN-2 [31]	GTN-3 [24]	RIGHT [28,32,33]	ENOS [2,34-36]	RIGHT-2 [3,37-40]
Setting	Hospital	Hospital	Hospital	Pre-hospital	Hospital	Pre-hospital
Time window (hr)	<120 hours	<72 hours	<120 hours	<4 hours	<48 hours	<4 hours
Stroke type	IS/ICH	IS/ICH	IS/ICH	IS/ICH	IS/ICH	IS/ICH
SBP range (mmHg)	No limits	100-230	140-220	>140	140-220	>120
Treatment blinding	Double-blind	Open-label	Single-blind	Single-blind	Single-blind	Single-blind
GTN dose (mg)	5	5/10	5	5	5	5
Thrombolysis given	N/A	N/A	N/A	After GTN	Before GTN	After GTN
Sample size						
Intended	38	90	18	80	>3500	850
Achieved	37	90	18	41	4011	1149

ICH: intracerebral haemorrhage; IS: ischaemic stroke; N/A: not applicable; SBP: systolic blood pressure

GTN lowered BP, increased heart rate, and did not alter cerebral blood flow (assessed using xenon CT) or platelet function.[24,28,30-33] The safety and efficacy of GTN in patients with acute stroke (IS, ICH) has been assessed in two large phase III trials.

International hospital-based MRC ENOS trial:[2] In comparison with no GTN, transdermal GTN was not associated with any difference in functional outcome (mRS), disability/Activities of Daily Living (BI), cognition (tMMSE, TICS, animal naming), mood (ZDS) or quality of life (EQ-5D/HUS, EQ-VAS) (Table B). No safety issues were found with GTN.[2]

In the subgroup of patients randomised within 6 hours of ictus (average time from event to randomisation 263 minutes) (identified here as 'ENOS-early'), treatment with GTN was associated with reduced death, and improved functional outcome (mRS, BI) and quality of life (Table B[1])

UK ambulance-based BHF RIGHT-2 trial: The average time to treatment was 70 minutes. In comparison with sham, transdermal GTN was not associated with any differences in functional outcome (mRS), disability/Activities of Daily Living (BI), cognition (tMMSE, TICS, animal naming), mood (ZDS) or quality of life (EQ-5D/HUS, EQ-VAS).

Table B. Outcomes in patients treated with GTN versus no GTN/sham
Data are number (%), median [interquartile range] or mean (standard deviation). Analysis with binary logistic regression (odds ratio), ordinal logistic regression (odds ratio) or multiple regression (mean difference).

Outcome measure	GTN	No GTN	OR/MD (95% CI)	p
ENOS (OTR 0-48 hours) [2]				
Modified Rankin Scale (/6)	3 [3]	3 [3]	1.01 (0.91, 1.13)	0.83
Barthel Index (/100)	66 (38)	63 (39)	2.18 (-0.23, 4.59)	0.11
EQ-VAS (/100)	57 (31)	56 (32)	0.8 (-1.3, 2.9)	0.70
Death (%)	233 (12)	263 (13)	0.89 (0.72, 1.10)	0.27
ENOS-early (OTR 0-6 hours) [1]				
Modified Rankin Scale (/6)	3 [3]	3 [3]	**0.53 (0.34, 0.82)**	**<0.001**
Barthel Index (/100)	74 (34)	60 (41)	**14 (4.6, 22.5)**	**0.01**
EQ-VAS (/100)	62 (29)	53 (35)	**9.6 (1.8, 17.5)**	**0.03**
Death (%)	11 (8)	26 (20)	**0.35 (0.13, 0.96)**	**0.04**
RIGHT-2 (OTR 0-4 hours) [3]				
Modified Rankin Scale (/6)	3 [3]	3 [3]	1.25 (0.97, 1.60)	0.083
Barthel Index (/100)	56 (45)	58 (44)	-3 (-8, 2)	0.29
EQ-VAS (/100)	45 (34)	47 (32)	-0.9 (-5.1, 3.2)	0.66
Death (%)	97 (23)	79 (19)	1.24 (0.91, 1. 86)	0.17

CI: confidence intervals; EQ-VAS: EuroQoL-Visual Analogue Scale; ICH: intracerebral haemorrhage; IS: ischaemic stroke; MD: mean difference; N/A: not applicable; OR: odds ratio; SBP: systolic blood pressure.

The following summary observations can be made based on data from the four phase II and two phase III GTN trials:

- Transdermal administration is advantageous since oral treatment is confounded by dysphagia in 50% of patients with acute stroke, whilst intravenous therapy requires intensive monitoring. Additionally, treatment can be stopped and restarted according to need.
- Peak concentrations of GTN are achieved by 1-2 hours.[41]
- Transdermal GTN lowers systolic BP significantly by 15, 60 and 120 minutes.[24,28]
- Transdermal GTN lowers central and peripheral SBP, DBP and pulse pressure; peak systolic BP and augmentation index; and 24-hour BP in both dipping and non-dipping patients.[24,28,31,42,43]
- Transdermal GTN is feasible to administer, well tolerated, and safe when given early after acute stroke.[2,3,24,28,31,42,44]
- GTN does not alter platelet function; hence, it can be given in ICH as well as IS.[42]
- GTN does not reduce cerebral blood flow.[24,31]
- GTN is safe in patients with severe carotid stenosis.[45]

Further, the effect of GTN on functional outcome appears to be time-dependent when administered after stroke:

- <2.0 hours: No effect; indeed, GTN may cause harm in ICH in this ultra-acute period (interpretation from RIGHT-2 [3,40])
- 2.5 to 5.5 hours: May improve functional outcome (interpretation from ENOS-early [1] and meta-analysis, Figures A , B).

- >6 hours: No effect (interpretation from GTN 1/2/3 and ENOS and an individual patient data meta-analysis [46]).

(The positive effect of GTN on functional outcome in the phase II RIGHT trial is ignored here because it was small.)

Figure A. Meta-analysis of GTN versus no GTN on end of trial functional outcome in patients treated within 6 hours of stroke onset (from [3])

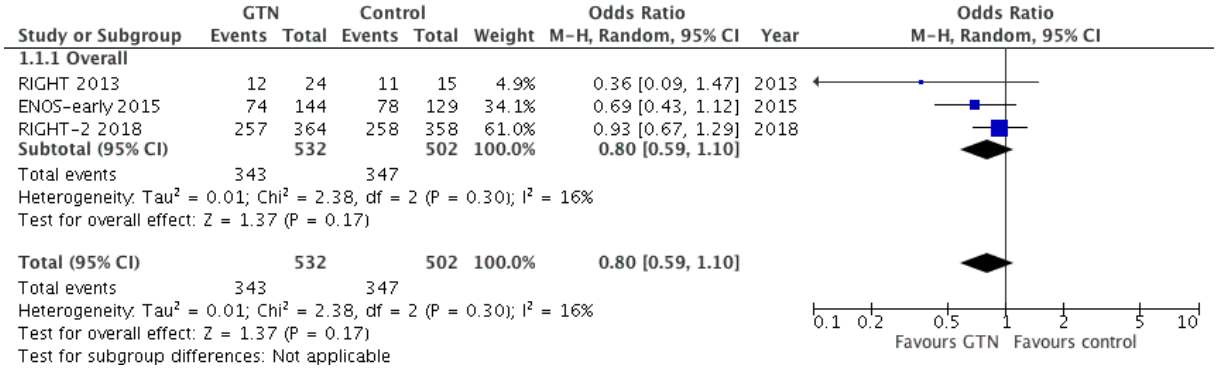
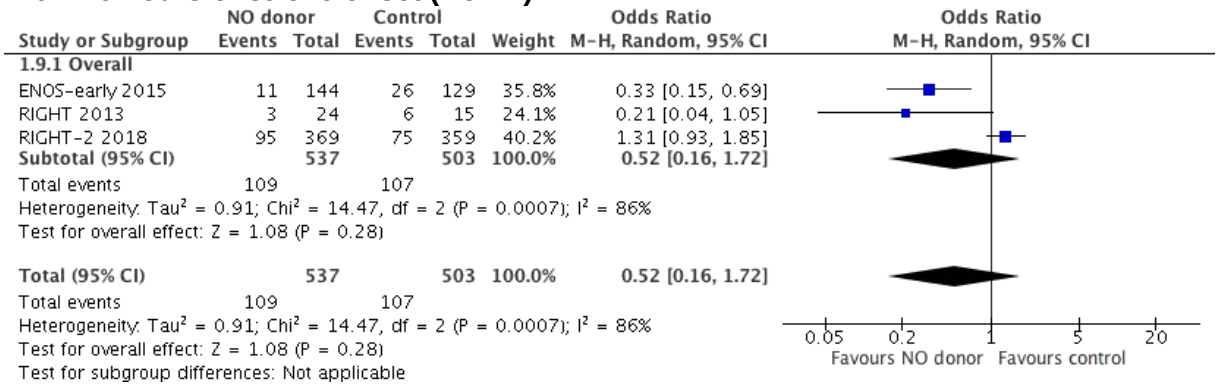


Figure B. Meta-analysis of GTN versus no GTN on end of trial death in patients treated within 6 hours of stroke onset (from [3])



In summary, GTN has been associated with beneficial effects on function and other clinical outcomes in hospitalised patients treated up to 6 hours after ictus. However, benefit was not seen in patients treated prior to hospital admission and there was weak evidence that ultra-acute treatment within 2 hours was associated within harm. As a result, ENOS-2 will assess, in a novel design, the feasibility of recruiting patients between 3 and 5 hours after ictus; if feasible, a larger safety and efficacy trial will be performed in this group of patients.

ENOS-2 will assess the feasibility, safety, and efficacy of hospital-delivered GTN when administered hyper-acutely after stroke. Five of the six GTN trials had a lower limit for systolic blood pressure of ≥100 mmHg,[31] >120 mmHg [3] or ≥140 mmHg;[2,24,28] the first study had no lower limit.[42] Since several potential mechanisms by which GTN might work are BP-independent, ENOS-2 will follow the RIGHT-2 protocol and include patients with high-normal BP as well as high BP, with a lower limit set to ≥ 120 mmHg. This has the advantage that the results will apply to a wider population of stroke patients and include more patients with severe stroke (some of whom have SBP <140 mmHg).

7 Objectives

The purpose of the study is to determine whether it is feasible to recruit patients to transdermal glyceryl trinitrate (GTN) administered between 3-5-hours after the onset of stroke to inform a definitive trial.

7a Primary Objective:

To assess the feasibility of recruiting, randomising, and treating patients with GTN vs sham and whether GTN lowers blood pressure.

7b Secondary Objectives:

i. To identify potential proof of concept on whether GTN reduces disability, low mood, poor cognition, and low quality of life.
ii. Potential genetic markers include nitric oxide synthase polymorphisms, but others will be studied as relevant in searches of the scientific literature.
iii. To investigate whether there is a difference in blood biomarkers between the two groups and whether biomarkers may be associated with outcome. Potential biomarkers include S-100 / nitric oxide (NO_x) but others will be studied as relevant in searches of the scientific literature.

SECTION 3. STUDY METHODS

8 Trial design

ENOS-2 is a prospective parallel-group randomised sham-controlled masked-endpoint phase II feasibility and safety trial of GTN versus sham with randomisation between 3 and 5 hours after stroke.

9 Sample size/power considerations

ENOS-2 is a feasibility study hence there is no formal sample size calculation. However, recruitment of 100 participants would allow an adjusted common odds ratio, acOR 0.51 to be detected, assuming mRS distribution as in[1]; alpha 0.05, power 0.80. The number of targeted participants will be 100 with ischaemic stroke (50 randomised to GTN, 50 randomised to sham) and 20 with intracerebral haemorrhage for safety (10 randomised to GTN, 10 randomised to sham).

10 Recruitment Setting

Adult patients with hyperacute stroke presenting at emergency departments and hyperacute stroke units in England. The trial setting will be in secondary care at NIHR Clinical Research Network sites with dedicated Research coordinators who will facilitate recruitment and follow up discharge or death.

11 Randomisation

Patients will be randomised 1:1 to receive either 5 mg transdermal GTN versus sham transdermal DuoDERM hydrocolloid dressing placed on the back or shoulders for two days. Randomisation will be performed by the Stroke Trials Unit (STU) in Nottingham and involve computerised stratification by stroke type (IS or not known; ICH) and minimisation on age, severity, time, systolic blood pressure and candidate for or received reperfusion therapy. Patients, relatives, researchers, and outcome assessors will be masked to treatment allocation.

12 Statistical interim analyses and stopping guidance.

12a Interim analyses:

An independent Data Monitoring Committee (DMC) will be established. The DMC will receive safety reports twice yearly, or more if requested, and perform unblind reviews of safety and efficacy data. The DMC have responsibility to monitor SAEs and neurological deterioration and outcome in IS and ICH separately. No interim analysis will be performed.

12b Adjustments of significance level:
There is no planned adjustment.

12c Stopping rules:
There are no formal stopping rules since the main aim of the study is to study feasibility. The trial will not be discontinued unless the safety of the participants is likely to be compromised. The trial may be terminated by either the TSC, the sponsor or the funder if there is overwhelming evidence of major safety concerns, new information becomes available that makes the trial unsafe or irrelevant or there is issues with trial conduct (e.g., loss of resources).

13 Timing of final analyses

Prior to each database lock, the trial managers/trial co-ordinators will chase outstanding data queries and the lock will take place in accordance with the documented data lock procedure once notification has been given to the trial programmer by the chief investigator.

Both interim and final locks will be documented and primarily consist of the creation of a read-only copy of the live database, with each copy available to the trial statistician via the online data extract process.

14 Table C.-Timing of outcome assessments

Assessments	Screen	Day 0 Baseline Pre-treatment	Day 0 Post-treatment	Day 1 During treatment	Day 2 After treatment	Discharge, Death	Day 90 Telephone
Face-to-face							
Clinical assessment	X[1]				X	X	
Eligibility screening	X						
CT Scan		X[1]	or X[1]				
Consent		X		or X	or X		
Randomisation		X					
GTN/Sham patch			X	X			
mRS		X[2]					
U & E FBC		X[1]					
Blood pressure		X[1]	X	X			
NIHSS		X[1]			X		
DSRS		X			X		
Blood biomarkers		X[3]			X[3]		

Hospital utilisation						X[4]	
Listed events				X[5]	X[5]		
SAEs, non-fatal			X	X	X		
SAEs fatal				X	X	X	X[6]
mRS							X[6]
Barthel Index							X[6]
EuroQoL							X[6]
Cognition (t-MMSE, TICS)							X[6]
Mood (ZDS)							X[6]

CT: Computerised tomography
NIHSS: National Institutes of Health stroke scale
MMSE: Mini-Mental State Examination
mRS modified Rankin scale
SAEs: Serious Adverse Events
1. Routine as part of clinical practice
2. Pre-morbid mRS
3. Biomarkers: soluble markers, genetics
4. Open-label blood pressure lowering, intravenous thrombolysis, mechanical thrombectomy, hyperacute stroke unit, Stroke Rehabilitation Unit, physiotherapy, occupational therapy, speech & language therapy, surgery for IS – hemicraniectomy, surgery for ICH, days in intensive/critical care unit.
5. Hypotension, hypertension, headache, infection
6. mRS [0 to 5, death=6], NIHSS [0 to 42, death=43], DSRS [0 to 12, death=13], GCS [3 to 15, death=2], BI [0 to 100, death=-5], EQ-5D [-0.5 to 1.0, death=0], EQ-VAS [0 to 100, death=-1], t-MMSE [0 to 22, death=-1], TICS-m [0 to 39, death=-1] and ZDS [≥70, death=102.5].

SECTION 4. STATISTICAL PRINCIPLES

15 Statistical significance

15a Level of statistical significance and p values
The results of analyses and comparisons will be shown with p<=0.05.

15b Confidence intervals
The results of analyses and comparisons will be shown with 95% confidence intervals.

16 Adherence
Compliance will be assessed by examining the participant's drug chart and recording evidence of treatment administration. Adherence will be recorded on the case report forms after treatment has been completed (day 2).

16a Definition:
Adherence will be considered sufficient if >=90% of participants receive their first active/sham treatment (**see Table 6**).

17 Protocol deviations*:*
All protocol deviations and violations must be reported immediately to the Chief Investigator, via the online electronic case report form. The CI will notify the Sponsor if a deviation or violation has an impact on participant safety or integrity of the trial data. The Sponsor will advise on appropriate measures to address the occurrence, which may include reporting of a serious GCP breach, internal audit of the trial and seeking counsel of the trial committees.

17a Protocol deviation presentation:
Listing of violations and deviations.

18 Analysis populations
All available data will be used, including overall patients presenting in clinic and screening data where available. Missing data will be reported.
1. Intention-to-treat population (ITT): summaries by treatment group will be provided according to the treatment the participant was randomised to.
2. Safety population: All participants who were randomised will be summarised according to the treatment they received irrespective of treatment.

SECTION 5. TRIAL POPULATION

19 Screening data
Overall patients presenting in clinic and screening data where available will be kept on screening logs and will be presented.

20 Eligibility

20a Inclusion
1. 120 adults (age ≥ 18 years) with presentation compatible with stroke
2. Treatment 3-5 hours post ictus (for patients with wake-up stroke, treatment no more than 5 hours after patient awakens)
3. One or more of the following symptoms present at time of enrolment:
   a. Dysphasia.
   b. Neglect (NIHSS 1-2)
   c. Hemianopia (NIHSS 1-3)
   d. Limb weakness (NIHSS on affected arm and/or leg 1-4)
   e. Systolic BP (≥ 120 mmHg)
4. If a CT/MR scan has already been performed, then it shows acute intracerebral haemorrhage or ischaemic stroke, or is normal.
5. Waiver of consent for treatment to ensure GTN given in 3-5-hour time window (and thrombolysis not delayed if ischaemic stroke).

20b Exclusion
1. mRS ≥ 4
2. Glucose (BM stix or equivalent) < 3 mmol/l
3. Glasgow coma scale ≤ 8
4. Witnessed seizure at presentation.
5. Known life expectancy <6 months.
6. Patient presenting with sensory symptoms only.

7. Known stroke mimic, aneurysmal subarachnoid haemorrhage, or haemorrhage due to venous thrombosis.
8. Systolic blood pressure <120 mmHg
9. Known allergy to glyceryl trinitrate (Transiderm-Nitro) patch.
10. Known sensitivity to Duoderm hydrocolloid dressing.
11. Planned for palliative care only.
12. Recent use of phosphodiesterase type 5 (PDE5) inhibitors, e.g., sildenafil
13. If a CT/MR scan has already been performed, then it shows a non-stroke lesion that explains the acute presentation.
14. Known previous enrolment in ENOS-2

21 Recruitment
Recruitment will be summarised in a CONSORT flow diagram.

22 Withdrawal/follow-up
Withdrawals, and missed follow-ups and their timing will be summarised in the CONSORT flow diagram.

23 Baseline patient characteristics
These will comprise demographics and clinical measures (**see Table 1**).
Summarisation of data will be shown as number (%), median [interquartile range] or mean (standard deviation), as appropriate.

SECTION 6. ANALYSIS

24 Outcome definitions

24a Primary endpoint
The primary end point of the study is the feasibility of recruiting and treating 120 patients (100 IS, 20 ICH) between 3 and 5 hours after stroke.

24b Secondary endpoints
1. Hospital admission:
    - Neurological impairment (NIHSS)
    - Systolic and diastolic blood pressure, heart rate.
    - Proportion of participants with systolic blood pressure <185 mmHg.
    - Feeding and dysphagia (dysphagia severity rating scale)
    - Stroke lesion size on brain scan (non-contrast CT or T2 MR).
    - Amount of cerebral arterial patency on brain scan (CT or MR angiography).
2. Hospital utilisation:
    - Open-label blood pressure lowering.
    - Intravenous thrombolysis.
    - Mechanical thrombectomy.
    - Hyperacute stroke unit.
    - Stroke Rehabilitation Unit.
    - Physiotherapy.
    - Occupational therapy.
    - Speech & language therapy.
    - Surgery for IS - Hemicraniectomy.
    - Surgery for ICH.
    - Days in intensive/critical care unit.
3. At day 2:
    - Systolic and diastolic blood pressure, heart rate.

4. At day 2: Biomarkers
    - Blood biomarkers (exact measures to be determined by literature review prior to measurement but examples include S-100, NOx).
    - Genetic markers (exact measures to be determined by literature review prior to measurement but examples include NO synthase polymorphisms).
5. At day 2 (or discharge if sooner):
    - Neurological impairment (NIHSS).
    - Stroke recurrence.
    - Neurological deterioration from baseline (NIHSS ≥4 points, or ≥2-point increase in any domain).
    - Feeding and dysphagia (DSRS).
6. At discharge/death
    - Length of stay in hospital.
    - Patient disposition.
7. At day 90 by telephone (or post):
    - Dependency – modified Rankin Scale (primary clinical endpoint at Day 90).
    - Disability/activities of daily living - Barthel Index (BI).
    - Quality of life - health utility status (HUS, derived from EuroQoL-5D), EQ-Visual Analogue Scale (EQ-VAS).
    - Cognition - telephone-MMSE, telephone interview cognition scale (TICS), animal naming.
    - Mood - Zung depression scale.
    - Patient disposition (died, institution/in hospital, home).

24c Safety endpoints

1. By day 2
    - Any serious adverse event.
    - Headache.
    - Infection (pneumonia/chest, urinary tract, other).
    - Hypotension requiring clinical intervention.
    - Hypertension requiring clinical intervention.
2. From day 3 to day 90
    - Any fatal serious adverse event.

Data on stroke recurrence and acute coronary syndrome, termed safety outcome events, will be collected up to day 90.

24d Units: Units will be shown in tables.

24e Calculations/transformations: Quality of life using UK weightings.

25 Analysis methods

25a Method *analyses of outcomes*

ENOS-2 is a feasibility trial, and the main analyses will present descriptive statistics and comparisons between treatment groups. Counts will be summarised using frequencies N and percentages %, ordinal variables will be summarised using median [interquartile range] and continuous variables using means (standard deviation). Central tendency, comparisons and regressions will be summarised and analysed as follows (***Table D***).

Table D. Descriptive and analytical statistics

	Nominal	Binary	Ordinal	Continuous	Time to Event
Central tendency	N (%)	N (%)	Median [interquartile	Mean (standard	N (%)

and distribution			range]	deviation)	
Comparisons	Chi-square (2x2, or rxc)	Chi-square (2x2)	Mann-Whitney U	t-test (pooled) change from baseline-Repeat Measures (RM)	
Regression	-	Binary logistic regression (BLR)	Ordinal logistic regression (OLR)	Multiple linear regression (MLR)	Cox proportional hazards regression (CPHR)

25b Analyses of Primary endpoint
Recruitment, treatment and follow-up rates will be estimated using tabulation and graphical presentation.
It is likely that a large definitive trial would be feasible if at least 70 participants were recruited into this study, that compliance with randomised treatment was high and that a high proportion of follow-up data was available.

25c Analyses of secondary outcomes
Mechanistic, clinical and safety outcomes will be compared between the treatment groups using binary logistic regression, Cox proportional hazards regression (death), ordinal logistic regression (mRS) or multiple linear regression (BI, TICS, t-MMSE, ZDS, EQ-5D, EQ-VAS, as appropriate (***Table A***), in exploratory analyses.

25d Covariate adjustment: Analyses will be adjusted for minimisation covariates including age, stroke severity (NIHSS), time after stroke, systolic blood pressure, and received reperfusion therapy.

25e Assumption checking:
The assumption of proportionality will be tested using the likelihood test.

25g Sensitivity analyses:
In addition to assessment of raw data, the primary clinical outcome (mRS) will be analysed using additional statistical approaches in sensitivity analyses using:
- Comparison using unadjusted ordinal logistic regression
- Comparison using adjusted multiple linear regression

26 Subgroup analyses:
The primary clinical outcome will be analysed in subgroups consisting of the minimisation factors:
- Age (years): <=70, >70
- Stroke severity, NIHSS (/42): <=10, >10
- Time after stroke (minutes): <=220, >220
- Systolic blood pressure (mmHg): <=160, >160
- Received reperfusion therapy: yes, no

27 Missing data
Missing data will not be imputed.

28 Harms
These will be presented in Table describing serious adverse events.

29 Statistical software
Statistical Analysis System (SAS) version 9.4 (or later), SAS Institute Incorporation, Cary, North Carolina.

SECTION 7. ADDITIONAL INFORMATION

30 Governance
The trial is funded by the Nottingham University Hospitals NHS Trust Charity and sponsored by the University of Nottingham. The trial is managed by a Trial Management Committee (TMC), supervised by a Trial Steering Committee (TSC) and overseen by an independent Data Monitoring Committee (DMC).

31 Minimising bias
Multiple approaches will be taken to minimise bias: central data registration with real-time on-line validation; minimisation at randomisation; stratification by stroke type (IS or not known; ICH); inclusion of patients enrolled in other studies (co-enrolment) where feasible; blinded central telephone (or postal) assessment of outcomes; and analysis by intention-to-enrol (i.e. all participants) and in pre-specified subgroups.

32 Publications, published and planned
1. Protocol: On trial website
2. SAP and baseline data: On trial website
3. Primary results paper: To be published.

33 Data sharing
Data will be shared with the VISTA Collaboration.

REFERENCES

1. Woodhouse L, Scutt P, Krishnan K, et al. Effect of hyperacute administration (within 6 hours) of transdermal glyceryl trinitrate, a nitric oxide donor, on outcome after stroke: subgroup analysis of the Efficacy of Nitric Oxide in Stroke (ENOS) trial. *Stroke* 2015; **46**: 3194-201.
2. Bath PM, Woodhouse L, Scutt P, et al. Efficacy of nitric oxide, with or without continuing antihypertensive treatment, for management of high blood pressure in acute stroke (ENOS): a partial-factorial randomised controlled trial. *Lancet* 2015; **385**(9968): 617-28.
3. RIGHT-2 Investigators. Prehospital transdermal glyceryl trinitrate in patients with ultra-acute presumed stroke (RIGHT-2): an ambulance-based, randomised, sham-controlled, blinded, phase 3 trial. *Lancet* 2019; **393**(10175): 1009-20.
4. Stroke Association. State of the nation: Stroke statistics. 2018.
5. Wardlaw JM, Murray V, Berge E, et al. Recombinant tissue plasminogen activator for acute ischaemic stroke: an updated systematic review and meta-analysis. *Lancet* 2012.
6. Goyal M, Menon BK, van Zwam WH, et al. Endovascular thrombectomy after large-vessel ischaemic stroke: a meta-analysis of individual patient data from five randomised trials. *Lancet* 2016; **397**(10029): 1723-31.
7. Bath PM, Lindenstrom E, Boysen G, et al. Tinzaparin in acute ischaemic stroke (TAIST): a randomised aspirin-controlled trial. *Lancet* 2001; **358**(9283): 702-10.
8. Intravenous Magnesium Efficacy in Stroke (IMAGES) Study Investigators. Magnesium for acute stroke (Intravenous Magnesium Efficacy in Stroke trial): randomised controlled trial. *Lancet* 2004; **363**: 439.
9. Shuaib A, Lees KR, Lyden P, et al. NXY-059 for the treatment of acute ischemic stroke. *New England Journal of Medicine* 2007; **357**: 562-71.
10. Anderson CS, Heeley E, Huang Y, et al. Rapid blood-pressure lowering in patients with acute intracerebral hemorrhage. *N Engl J Med* 2013; **368**(25): 2355-65.
11. Sandset EC, Bath PM, Boysen G, et al. The angiotensin-receptor blocker candesartan for treatment of acute stroke (SCAST): a randomised, placebo-controlled, double-blind trial. *Lancet* 2011; **377**(9767): 741-50.
12. Gray CS, Hildreth AJ, Sandercock P, et al. Glucose-potassium-insulin infusions in the management of post-stroke hyperglycaemia: the UK Glucose Insulin in Stroke Trial (GIST-UK). *Lancet Neurology* 2007; **6**: 397-406.
13. Leonardi-Bee J, Bath PM, Phillips SJ, Sandercock PA. Blood pressure and clinical outcomes in the International Stroke Trial. *Stroke* 2002; **33**(5): 1315-20.
14. Willmot M, Leonardi-Bee J, Bath PM. High blood pressure in acute stroke and subsequent outcome: a systematic review. *Hypertension* 2004; **43**(1): 18-24.
15. Sprigg N, Gray LJ, Bath PM, et al. Relationship between outcome and baseline blood pressure and other haemodynamic measures in acute ischaemic stroke: data from the TAIST trial. *J Hypertens* 2006; **24**(7): 1413-7.
16. He J, Zhang Y, Xu T, et al. Effects of Immediate Blood Pressure Reduction on Death and Major Disability in Patients With Acute Ischemic Stroke: The CATIS Randomized Clinical Trial. *JAMA* 2013.
17. Anderson CS, Huang Y, Lindley RI, et al. Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (ENCHANTED): an international, randomised, open-label, blinded-endpoint, phase 3 trial. *Lancet* 2019; **393**(10174): 877-88.
18. Barer DH, Cruickshank JM, Ebrahim SB, Mitchell JR. Low dose beta blockade in acute stroke ("BEST" trial): an evaluation. *British Medical Journal* 1988; **296**: 737-41.

19. Wahlgren NG, MacMahon DG, de Keyser J, Indredavik B, Ryman T, INWEST Study Group. Intravenous Nimodipine West European Stroke Trial (INWEST) of nimodipine in the treatment of acute ischaemic stroke. *Cerebrovascular Diseases* 1994; **4**: 204-10.
20. Ahmed N, Nasman P, Wahlgren NG. Effect of intravenous nimodipine on blood pressure and outcome after acute stroke. *Stroke* 2000; **31**: 1250-5.
21. Geeganage C, Bath PMW. Relationship between therapeutic changes in blood pressure and outcomes in acute stroke. A metaregression. *Hypertension* 2009; **54**: 775-81.
22. Geeganage C, Bath PM. Vasoactive drugs for acute stroke. *Cochrane Database Syst Rev* 2010; (7): CD002839.
23. Gray LJ, Sprigg N, Rashid PA, Willmot M, Bath PMW. Effect of nitric oxide donors on blood pressure and pulse pressure in acute and sub-actue stroke. *Journal of Stroke and Cerebrovascular Diseases* 2006; **15**(6): 245-9.
24. Willmot M, Ghadami A, Whysall B, Clarke W, Wardlaw J, Bath PM. Transdermal glyceryl trinitrate lowers blood pressure and maintains cerebral blood flow in recent stroke. *Hypertension* 2006; **47**: 1209-15.
25. Morikawa E, Rosenblatt S, Moskowitz MA. L-arginine dilates rat pial arterioles by nitric oxide-dependent mechanisms and increases blood flow during focal cerebral ischaemia. *Br J Pharmacol* 1992; **107**(4): 905-7.
26. Willmot M, Gray L, Gibson C, Murphy S, Bath PM. A systematic review of nitric oxide donors and L-arginine in experimental stroke; effects on infarct size and cerebral blood flow. *Nitric Oxide* 2005; **12**(3): 141-9.
27. Rashid PA, Whitehurst A, Lawson N, Bath PMW. Plasma nitric oxide (nitrate/nitrite) levels in acute stroke and their relationship with severity and outcome. *JStroke CerebrovascDis* 2003; **12**(2): 82-7.
28. Ankolekar S, Fuller M, Cross I, et al. Feasibility of an ambulance-based stroke trial, and safety of glyceryl trinitrate in ultra-acute stroke: the rapid intervention with glyceryl trinitrate in Hypertensive Stroke Trial (RIGHT, ISRCTN66434824). *Stroke* 2013; **44**(11): 3120-8.
29. Butterworth RJ, Cluckie A, Jackson SH, Buxton-Thomas M, Bath PM. Pathophysiological assessment of nitric oxide (given as sodium nitroprusside) in acute ischaemic stroke. *Cerebrovascular diseases (Basel, Switzerland)* 1998; **8**(3): 158-65.
30. Bath PM, Pathansali R, Iddenden R, Bath FJ. The effect of transdermal glyceryl trinitrate, a nitric oxide donor, on blood pressure and platelet function in acute stroke. *Cerebrovasc Dis* 2001; **11**(3): 265-72.
31. Rashid P, Weaver C, Leonardi-Bee J, Bath F, Fletcher S, Bath P. The effects of transdermal glyceryl trinitrate, a nitric oxide donor, on blood pressure, cerebral and cardiac hemodynamics, and plasma nitric oxide levels in acute stroke. *J Stroke Cerebrovasc Dis* 2003; **12**(3): 143-51.
32. Ankolekar S, Sare G, Geeganage C, et al. Determining the Feasibility of Ambulance-Based Randomised Controlled Trials in Patients with Ultra-Acute Stroke: Study Protocol for the "Rapid Intervention with GTN in Hypertensive Stroke Trial" (RIGHT, ISRCTN66434824). *Stroke Res Treat* 2012; **2012**: 385753.
33. Ankolekar S, Parry R, Sprigg N, Siriwardena AN, Bath PM. Views of Paramedics on Their Role in an Out-of-Hospital Ambulance-Based Trial in Ultra-Acute Stroke: Qualitative Data From the Rapid Intervention With Glyceryl Trinitrate in Hypertensive Stroke Trial (RIGHT). *Ann Emerg Med* 2014.
34. The ENOS Trial Investigators. Glyceryl trinitrate vs. control, and continuing vs. stopping temporarily prior antihypertensive therapy, in acute stroke: rationale and design of the Efficacy of Nitric Oxide in Stroke (ENOS) trial (ISRCTN99414122). *International Journal of Stroke* 2006; **1**: 245-9.

35. Bath PM, Houlton A, Woodhouse L, Sprigg N, Wardlaw J, Pocock S. Statistical analysis plan for the 'Efficacy of Nitric Oxide in Stroke' (ENOS) trial. *Int J Stroke* 2014; **9**(3): 372-4.
36. ENOS Investigators. Baseline Characteristics of the 4011 patients recruited into the "efficacy of Nitric Oxide in Stroke (ENOS) trial. *International Journal of Stroke* 2014; **9**(6): 711-20.
37. Scutt P, Appleton JP, Dixon M, et al. Statistical analysis plan for the 'Rapid Intervention with Glyceryl trinitrate in Hypertensive stroke Trial-2 (RIGHT-2)'. *Eur J Stroke* 2018; **3**(2): 193-6.
38. Appleton JP, Scutt P, Dixon M, et al. Ambulance-delivered transdermal glyceryl trinitrate versus sham for ultra-acute stroke: Rationale, design and protocol for the Rapid Intervention with Glyceryl trinitrate in Hypertensive stroke Trial-2 (RIGHT-2) trial (ISRCTN26986053). *Int J Stroke* 2019; **14**(2): 191-206.
39. Bath PM, Scutt P, Appleton JP, et al. Baseline characteristics of the 1149 patients recruited into the Rapid Intervention with Glyceryl trinitrate in Hypertensive stroke Trial-2 (RIGHT-2) randomized controlled trial. *Int J Stroke* 2019; **14**(3): 298-305.
40. Bath PM, Woodhouse LJ, Krishnan K, et al. Prehospital Transdermal Glyceryl Trinitrate for Ultra-Acute Intracerebral Hemorrhage: Data From the RIGHT-2 Trial. *Stroke* 2019; **50**(11): 3064-71.
41. Wolff M, Luckow V. In Vitro and In Vivo-release of nitroglycerin from a new transdermal therapeutic system. *Pharmaceutical Research* 1985; **1**: 23-9.
42. Bath PMW, Pathansali R, Iddenden R, Bath FJ. The effect of transdermal glyceryl trinitrate, a nitric oxide donor, on blood pressure and platelet function in acute stroke. *Cerebrovascular diseases (Basel, Switzerland)* 2001; **11**: 265-72.

Main paper tables

Table 1. **Baseline characteristics by treatment group transdermal glyceryl trinitrate (GTN).**
Data are number (%), median [interquartile range] or mean (standard deviation).
† Minimisation variables include age, severity, time, systolic blood pressure, and candidate for or received reperfusion therapy.

Characteristic	Statistic	N	All	GTN	Control
Patients randomised	**N**		**xxx**	**xxx**	**xxx**
Age (years) †	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Sex (males)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Onset to randomisation (minutes) †	Median [IQR] (range)	xxx	xxx [xxx] (xxx)	xxx [xxx] (xxx)	xxx [xxx] (xxx)
Weight	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Ethnicity					
...White	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Black	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Asian	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Other	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Wake-up stroke	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
From Nursing home	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Medical history					
Diabetes mellitus	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Atrial fibrillation	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Previous stroke	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Previous TIA	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
IHD	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hyperlipidaemia	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
PAD	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Family stroke	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Smoking (%)					
…Current	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
…Past	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Never	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Alcohol intake (%)					
…High (over 21 units per week)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Moderate (1-21 units per week)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…None	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Existing medications prior to stroke n (%)					
Nitrate therapy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Lipid lowering	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Antihypertensives					
Antihypertensives	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Diuretic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Beta Blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Alpha-blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Calcium channel blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Angiotensin converting	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Angiotensin-II receptor antagonist	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Renin	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Centrally acting	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Labetalol	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Other antihypertensives	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Antiplatelets					
Aspirin	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Dipyridamole	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Clopidogrel	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Cilostazol	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Prasugrel	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
Ticagrelor	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Anticoagulants					
…Warfarin	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…DOACs	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…None	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Anticoagulants reversal drugs					
None	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Vitamin K	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Idarucizumab	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Prothrombin	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
FF Plasma	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Admission					
Systolic blood pressure (mmHg) †	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Diastolic blood pressure (mmHg)	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Heart rate (bpm)	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Received alteplase†	Yes (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Received mechanical thrombectomy	Yes (%)				
Glasgow coma scale, GCS (/)	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
DSRS [/12]	Median [IQR] (range)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
NIHSS	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Modified Rankin Scale, mRS (pre-stroke)	Median [IQR] (range)	xxx	xxx [xxx] (xxx)	xxx [xxx] (xxx)	xxx [xxx] (xxx)
mRS scores					
…0	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…1	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…2	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…3	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
…4 (protocol deviation)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…5	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Clinical findings					
Facial weakness	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Arm weakness	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Leg weakness	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hand side weakness					
…left	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…right	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…both	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
… cannot access	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hands: Motor power on affected side					
… normal strength	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
… reduced strength	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
… some movement, fingertips do not reach palm	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…paralysis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Homonymous hemianopia/ quadrantnopia	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Neglect	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Brainstems signs	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
Nutrition					
Feeding ability					
…Normal diet	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Soft diet	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…NG feed	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…PEG feed	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…IV/SC fluids	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Nothing					
Baseline scan information					
Type of scan					
…CT	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…MRI	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…No scan	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Onset to scan (minutes)	Mean (SD) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Baseline Scan result					
...Normal	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...IS no HT	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...IS with HT	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...PICH	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Other	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Presence of compatible lesion	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Mass effect	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Cerebral atrophy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Periventricular white matter lucency	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hyperdense artery	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Previous strokes	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
CT angiography (CTA)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
...CTA show large vessel occlusion (LVO)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hospital Utilisations Day 0 and 1					
Received any antihypertensives other than trial patch 1	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Received any antihypertensives other than trial patch 2	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Treatments given					
Aspirin	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Clopidogrel	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Dipyridamole	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Ticagrelor	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Diuretics	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Beta-blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Alpha-blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Calcium channel blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Angiotensin-converting enzyme inhibitor	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Angiotensin-II receptor antagonist	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Renin inhibitors	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Centrally acting agent	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Labetalol	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Other antihypertensive agent	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hemicraniectomy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Mannitol	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Intensive care unit admission	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Other neurosurgery	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Blood pressure lowering used in addition to the trial patch					

Characteristic	Statistic	N	All	GTN	Control
Number of blood lowering drugs used	Median (IQR) (range)	xxx	xxx (xxx) (xxx)	xxx (xxx) (xxx)	xxx (xxx) (xxx)
Intravenous BP lowering drugs used	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Combined Alpha- and Beta- blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Calcium channel blocker	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Glyceryl trinitrate	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Diuretic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Sodium nitroprusside	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Hydralazine	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Transdermal glyceryl trinitrate	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Other BP lowering drugs	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Main/first reason for blood pressure lowering					
…Preparation for thrombolysis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Preparation for thrombectomy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…restart pre-stroke antihypertensives	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…cerebral oedema	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…intracerebral haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…stroke mimic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…other reasons	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Thrombolysis					
Intravenous Thrombolysis					
…Alteplase	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Reteplase	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Streptokinase	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Tenecteplase	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Urokinase	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Reason for <u>not</u> giving thrombolysis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

Characteristic	Statistic	N	All	GTN	Control
…Cerebral haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Ischaemic stroke too mild					
…Ischaemic stroke too severe	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Out of time window	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Refused	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Non-stroke/stroke mimic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Other reasons	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Neurosurgical Intervention					
Mechanical thrombectomy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
Reason for not receiving mechanical thrombectomy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…ASPECT score too low	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…No large artery occlusion	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Cerebral haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Ischaemic stroke- too mild	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Ischaemic stroke – too severe	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Out of time window	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Refused	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…Non-stroke/stroke mimic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
…other reasons	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
COVID-19 diagnosis					
...Definite	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Possible	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)
...Unlikely	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)

CT (computed tomography); CTA (computed tomography angiography); DSRS (dysphagia rating scale); FF (frozen fruits); GCS (Glasgow severity index); HT (haemorrhagic transformation); ICH (intracerebral haemorrhage); IHD (ischaemic heart disease); IS (ischaemic stroke); IV (intravenous); LVO (large vessel occlusion); MRI (magnetic resonance image); NG (nasogastric); NIHSS (NIH stroke severity scale); PAD (peripheral arterial disease); PEG (percutaneous endoscopic gastrostomy); PICH (peripheral intracerebral haemorrhage); SC (subcutaneous fluids); TIA (transient ischaemic stroke).

Table 2. Feasibility measures.

Data are number (%).

Recruitment of 100 IS patients, 20. We will assess the feasibility of recruiting, treating, and following up patients from 2 centres over three years. We will estimate a recruitment rate, treatment rate and follow-up rate. It is likely that a large definitive trial would be feasible if at least 70 participants were recruited into this study, that compliance with randomised treatment was high and that a high proportion of follow up data was available.

Measure	Metric	N	Achieved
Recruitment			
Overall			xxx/120 (xxx)
Target patients	100 with IS patients		xxx/100 (xxx)
	20 with ICH		xxx/20(xxx)
Primary			
Retention of enrolees at Day 90 (follow-up)	>70 participants	xxx	xxx /100 (xxx)
Treatment compliance/ adherence			
Received GTN	All 2 days treatment	xxx	xxx (xxx)
	First day treatment	xxx	xxx (xxx)
Co-enrolment			
MAPS-2	n(%)		xxx (xxx)
PhEAST	n(%)		xxx (xxx)
ReCAST-3	n(%)		xxx (xxx)
TICH-3	n(%)		xxx (xxx)
MACE-ICH	n(%)		xxx (xxx)
TRIDENT	n(%)		xxx (xxx)
ABC-ICH	n(%)		xxx (xxx)
ENRICH-AF	n(%)		xxx (xxx)
ATTEST-I	n(%)		xxx (xxx)
TASTE	n(%)		xxx (xxx)
OPTIMAS	n(%)		xxx (xxx)
ProFATE	n(%)		xxx (xxx)
Mrna-1273-P305	n(%)		xxx (xxx)
COMMITS	n(%)		xxx (xxx)
OPTIMIST	n(%)		xxx (xxx)

Table 3. Clinical Outcomes at Day 90.

Data are number (%), median [interquartile range] or mean (standard deviation). Analyses performed using binary logistic regression (BLR), ordinal logistic regression (OLR) or multiple linear regression (MLR) with adjustment for age, time from stroke onset to randomisation, systolic blood pressure, stroke severity (NIHSS), and received reperfusion therapy. Mean (SD) and MLR will be used instead of median [IQR] and OLR for ordinal scales with more than 7 levels (central limit theorem/large sample). Cox proportional hazards regression (CPHR) will be used for death.

Variable	Statistic	N	All	GTN	Sham	aOR/aHR/aMD (95%CI)	Difference (p)
Patients randomised	**N**	xxx	xxx	xxx	xxx		
Outcomes							
Functional, modified Rankin scale (/6)							
	Median [IQR]	xxx	xxx [xxx]	xxx [xxx]	xxx [xxx]	xxx (xxx, xxx)	aOLR
							OLR
	Mean (SD)	xxx	xxx [xxx]	xxx [xxx]	xxx [xxx]	xxx (xxx, xxx)	aMLR
0	n(%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
1	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
2	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
3	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
4	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
5	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
6	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Death	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	CPHR
Disability							
Barthel index, BI (/100)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Cognition							
Telephone, MMSE (/22)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
TICS-M (/39)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Animal naming	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Mood							
Zung depression scale, ZDS(/102.5)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR

Variable	Statistic	N	All	GTN	Sham	aOR/aHR/aMD (95%CI)	Difference (p)
Quality of Life							
EQ-5D HSUV (/1)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	OLR
EQ-VAS (/100)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	OLR
Disposition						xxx (xxx, xxx)	OLR
…Home	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Institution/in hospital	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Died	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Events							
Recurrent Stroke	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Acute Coronary Syndrome	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Elective rehabilitation admission	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Viral Infections	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR

BI: Barthel Index, TICS-m: Telephone interview cognitive scale- modified; t-MMSE: telephone Mini-Mental State Examination ; ZDS: Zung depression scale; quality of life (EQ-5D HSUV, EQ-VAS).
Death scores: mRS (6); BI (5); TICS-m (-1); ZDS (102.5); EQ-5D HSUV (0); EQ-VAS (-1)

Table 4. Clinical Outcomes at Day 2.
Data are number (%), median [IQR], or mean (standard deviation).

Variable	Statistic	N	All	GTN	Sham	aOR/aMD (95%CI)	Difference (p)
Patients randomised	**N**	xxx	xxx	xxx	xxx		
Mechanistic Outcomes							
Blood Pressure							
SBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
DBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Heart rate (bpm)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Clinical Outcomes							
Impairment							
NIHSS (/100)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Neurological deterioration NIHSS change from baseline (point increase)							
NIHSS ≥ 4	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
NIHSS ≥ 2	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Hand weakness						xxx (xxx, xxx)	OLR
…Normal strength	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Reduced strength	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Some movement	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Paralysis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Feeding ability						xxx (xxx, xxx)	OLR
…Normal diet	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Soft diet	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…NG feed	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…PEG feed	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…IV/SC fluids	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Nothing	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Dysphagia, DSRS (/12)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Events up to day 2							
Death	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Symptomatic ICH	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
ICH expansion	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Symptomatic Recurrent Stroke						xxx (xxx, xxx)	OLR

Variable	Statistic	N	All	GTN	Sham	aOR/aMD (95%CI)	Difference (p)
…IS	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…haemorrhagic	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…unknown type	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…none	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Clinical neuro-deterioration	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
MEB	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Acute coronary Syndrome (ACS)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
…STEMI							
…NSTEMI							
…Unstable angina							
…None							
Symptomatic pulmonary embolism	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Headache	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Hypotension	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Hypertension	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR

Please note:
ACS (Acute coronary syndrome); DPB diastolic blood pressure); DSRS (dysphagia rating scale); IV (intravenous); MEB (major extracranial bleeding); NIHSS (NIH Stroke Scale); NG (nasogastric); NSTEMI; PE (Pulmonary embolism); PEG (percutaneous endoscopic gastrostomy); SC (subcutaneous); SPB (systolic blood pressure); STEMI.

Table 5. Blood pressure and heart rate up to Day 2.
Data are mean (standard deviation).

Variable	Statistic	N	All	GTN	Sham	aMD (95%CI)	Difference (p)
Patients randomised	**N**	xxx	xxx	xxx	xxx		
Admission							
SBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
DBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Heart rate (bpm)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Day 0-2hrs post randomisation							
SBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
DBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Heart rate (bpm)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Day 1							
SBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
DBP (mmHg)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Heart rate (bpm)	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Overall Comparison							
SBP (mmHg)						xxx (xxx, xxx)	RM
DBP (mmHg)						xxx (xxx, xxx)	RM
Heart rate (bpm)						xxx (xxx, xxx)	RM

Please note
RM- Comparison by repeated measures with baseline adjustment

Table 6. Outcomes at Discharge.

Data are number (%) and mean (standard deviation).

Variable	Statistic	N	All	GTN	Control	aOR/aMD (95%CI)	Difference (p)
Patients randomised	**N**	xxx	xxx	xxx	xxx		
Disposition Outcomes							
Discharge destination						xxx (xxx, xxx)	OLR
…Home	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Institution	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Hospital	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Died	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
…Other	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)		
Length of stay in hospital, days	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Hospital Interventions							
Acute stroke unit, ASU	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Length of stay in ASU, days	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Intensive therapy/high dependency unit (ITU/HDU)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Length of stay in ITU/HDU, days	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Stroke rehabilitation unit (SRU)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Length of stay in SRU, days	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Non-stroke ward	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Length of stay in non-stroke ward	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Physiotherapy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Occupational therapy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Speech and language therapy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Dietician	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Intermittent pneumatic compression stockings	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Results of carotid ultrasound (stenosis)							

Variable	Statistic	N	All	GTN	Control	aOR/aMD (95%CI)	Difference (p)
Left stenosis	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Right stenosis	Mean (SD)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	MLR
Do not resuscitate (DNR)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	BLR
Final Diagnosis of the randomising event							
Stroke diagnosis							OLR
…Ischaemic stroke	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…PICH	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…TIA	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Subarachnoid haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Stroke type, unknown							
Mimic							
Haemorrhage							
Non-stroke diagnosis							
…Traumatic brain injury	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Seizure/fit	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Migraine/headache	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Tumour	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Abscess	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Vestibular/labyrinthine	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Neuropathy	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Dementia	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Subdural haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
….Extradural haemorrhage	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Encephalitis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Meningitis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
….Transient global amnesia (TGA)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Syncope/faint	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Urinary tract infection (UTI)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	

Variable	Statistic	N	All	GTN	Control	aOR/aMD (95%CI)	Difference (p)
..Respiratory tract infection (RTI)	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Hypoglycaemia	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
….Functional	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Alcohol related	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Drug related	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Other non-CNS sepsis	n (%)	xxx	xxx (xxx)	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx)	
…Other diagnosis							

Please Note:
Home = (Home- independent; Warden-aided flat; Residential home, Home- needing care)
Institution = (Carer's home; Respite care; Nursing home)
Hospital=(Rehabilitation hospital; In hospital with readmission; Still in hospital after admission; Transfer to another hospital- ICU/ITU; Transfer to another hospital- repatriation)
Other diagnosis means not listed above.

Table 7. **Compliance by randomised group: GTN and Control.**
Data are number (%).

Period	**GTN**	**Control**	**Difference (p)**
Received first day of treatment	xxx (xxx)	xxx (xxx)	Chi-sq
Received all 2 days treatment	xxx (xxx)	xxx (xxx)	Chi-sq
Received some treatment	xxx (xxx)	xxx (xxx)	Chi-sq

Table 8. Serious adverse events for GTN in relationship to the study.
Data are number (%).

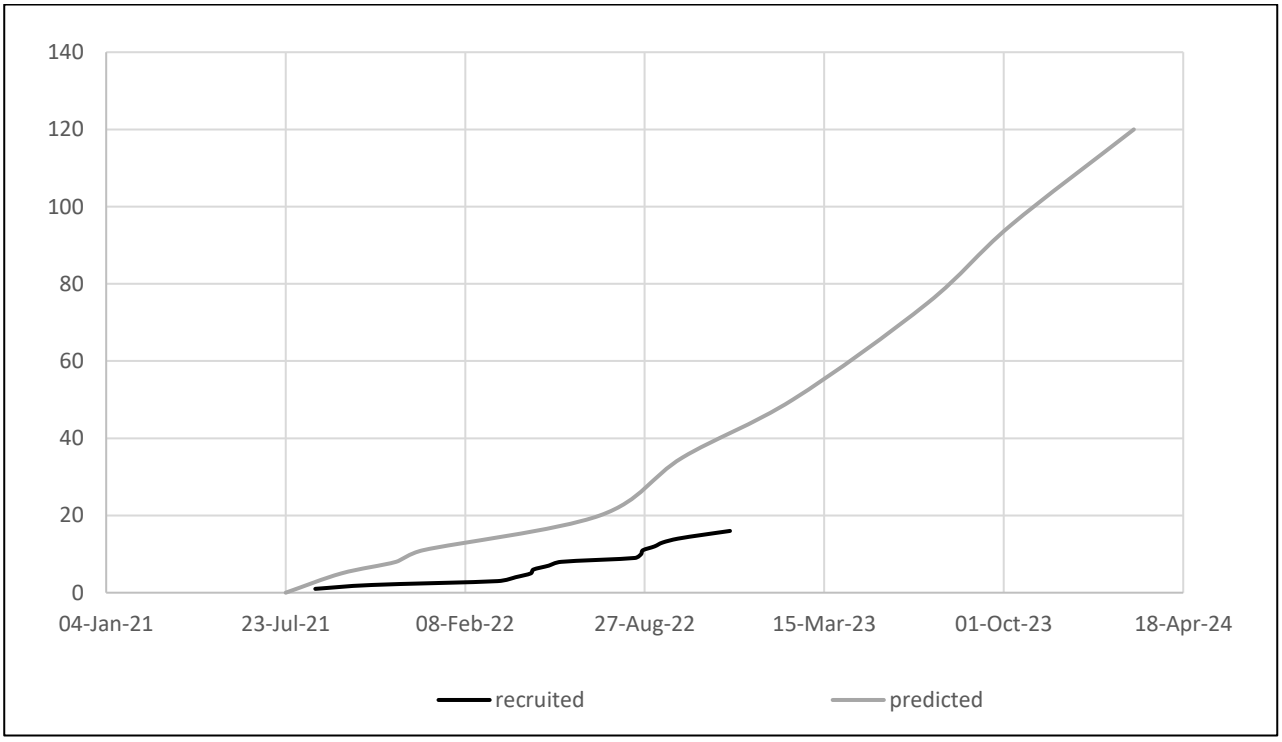
SAEs up to day 90					Fatal SAEs up to day 90				
	All	**GTN**	**Sham**	**Difference (p)**		**All**	**GTN**	**Sham**	**Difference (p)**
Number SAEs	xxx	xxx	xxx		Number SAEs	xxx	xxx	xxx	
Treatment time									
…Before		xxx	xxx (xxx)	BLR	Before		xxx	xxx (xxx)	BLR
…During	xxx	xxx (xxx)	xxx (xxx)	BLR	During	xxx	xxx (xxx)	xxx (xxx)	BLR
…After	xxx	xxx (xxx)	xxx (xxx)	BLR	After	xxx	xxx (xxx)	xxx (xxx)	BLR
Relationship to treatment									
…Not related	xxx	xxx (xxx)	xxx (xxx)	Chi-sq/FET	Not related	xxx	xxx (xxx)	xxx (xxx)	Chi-sq/FET
…Improbable	xxx	xxx (xxx)	xxx (xxx)	-	Improbable	xxx	xxx (xxx)	xxx (xxx)	-
…Possible	xxx	xxx (xxx)	xxx (xxx)	-	Possible	xxx	xxx (xxx)	xxx (xxx)	-
…Probable	xxx	xxx (xxx)	xxx (xxx)	-	Probable	xxx	xxx (xxx)	xxx (xxx)	-
…Definite	xxx	xxx (xxx)	xxx (xxx)	-	Definite	xxx	xxx (xxx)	xxx (xxx)	-

Table 9. **Participants with at least one serious adverse event by organ randomised to GTN versus Sham control.**
Data are number (%), and comparison by binary logistic regression.

	All			Fatal		
Number	**GTN**	**Control**	**Difference (p)**	**GTN**	**Control**	**Difference (p)**
Cardiovascular	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Hypotension	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Nervous system	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Haemorrhagic transformation	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Transient ischaemic attack	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Respiratory	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Gastrointestinal	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Genitourinary	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Haematological/ immunological	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Metabolic/endocrine	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Musculoskeletal/ cutaneous	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Miscellaneous	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Other	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx
Total	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx	xxx (xxx)	xxx (xxx)	xxx (xxx, xxx), p=0.xxx

Figures

Figures 1: Recruitment Figure



Figures 2: CONSORT flowchart diagram

Form	GTN	Control
Randomisation	XX (XX.X)XXX (XXX)	XXX (XXX)
Baseline	XXX (XXX)	XXX (XXX)
Day 2 form		
Completed	XXX (XXX)	XXX (XXX)
...Died	XXX (XXX)	XXX (XXX)
Not complete: Withdrawn	XXX (XXX)	XXX (XXX)
Death/Discharge form		
Completed	XXX (XXX)	XXX (XXX)
...Died	XXX (XXX)	XXX (XXX)
Not complete: Withdrawn from follow up	XXX (XXX)	XXX (XXX)
Not complete: Lost to follow up	XXX (XXX)	XXX (XXX)
Day 90		
Completed	XXX (XXX)	XXX (XXX)
...Died	XXX (XXX)	XXX (XXX)
Not complete: Lost with mRS	XXX (XXX)	XXX (XXX)
Not complete: Lost without mRS	XXX (XXX)	XXX (XXX)
Not complete: Refused	XXX (XXX)	XXX (XXX)
Not complete: Withdrawn with known vital status	XXX (XXX)	XXX (XXX)
Not complete: Withdrawn with unknown vital status	XXX (XXX)	XXX (XXX)

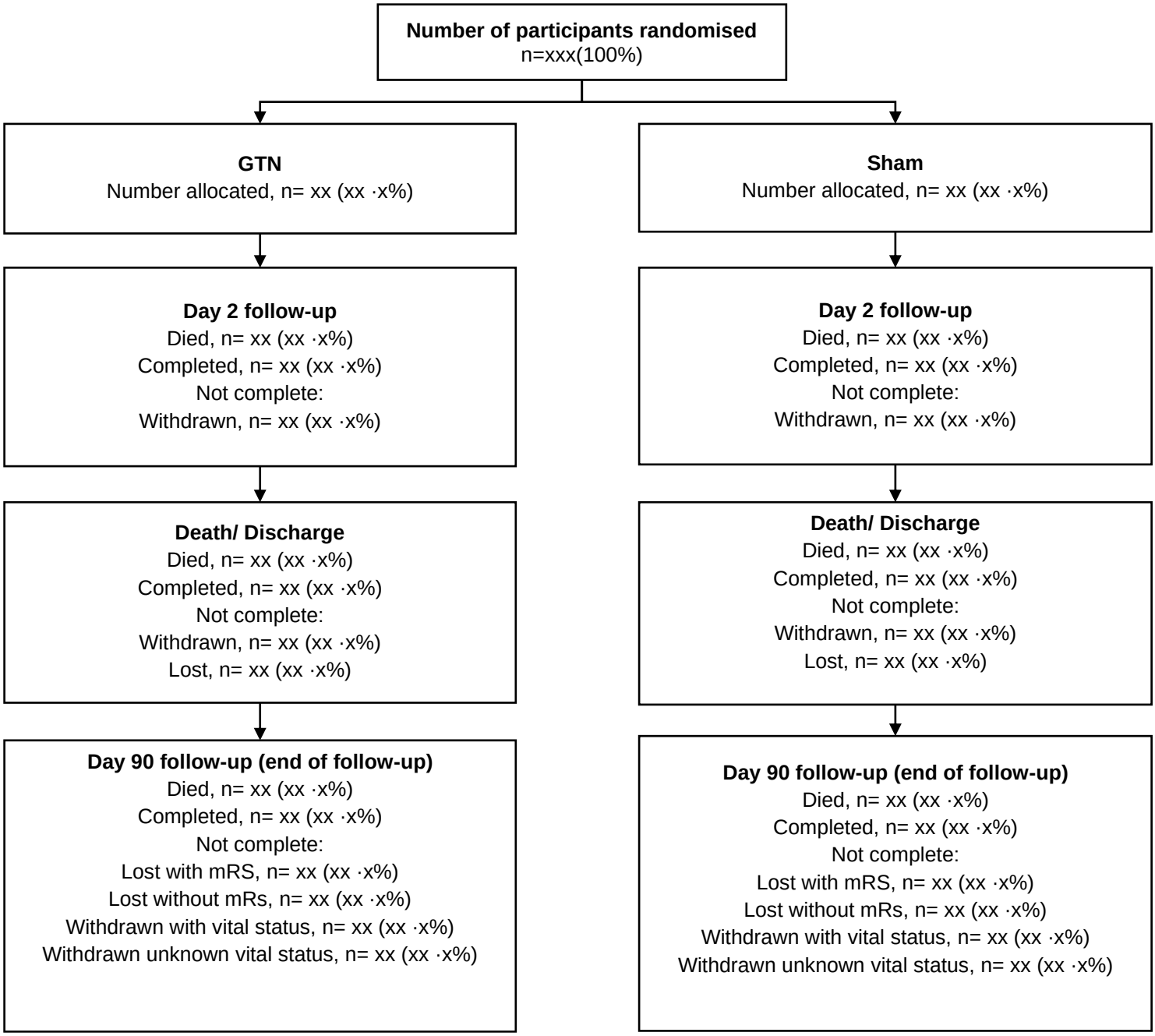


Figure 3. Shift diagram of mRS at Day 90

Figure 4. Shift diagram of the DSRS a Day 2

Figure 5. Trend in systolic and diastolic blood pressure measurements over the first 2 days

Figure 6. Trend in the heart rate over the first 2 days