DASH

Statistical Analysis Plan

Version 1.0
26 July 2022

Based on Protocol version 2.1 5 May 2022

Trial registration: ISRCTN67038373

The following people have reviewed the Statistical Analysis Plan and are in agreement with the contents				
Name	**Job Title**	**Trial role**	**Signature**	**Date**
Lisa J Woodhouse	Medical Statistician	Trial Statistician (Author)		27/07/2022
Trish Hepburn	Senior Medical Statistician	Senior Trial Statistician		27/07/2022
NIKOLA SPRIGG	Professor Stroke Medicine	Chief Investigator	DR. NIKOLA SPRIGG.	27/07/2022
Michael J R Desborough	Consultant Haematologist	Deputy Chief Investigator		27/07/2022

Table of Contents

1 INTRODUCTION AND PURPOSE

This document details the rules proposed and the presentation that will be followed, as closely as possible, when analysing and reporting the main results from the NIHR RfPB funded DASH trial.

The purpose of the plan is to:

1. Ensure that the analysis is appropriate for the aims of the trial, reflects good statistical practice, and that interpretation of a priori and post hoc analyses respectively is appropriate.

2. Explain in detail how the data will be handled and analysed to enable others to perform or replicate these analyses.

This analysis plan will be made available if required by journal editors or referees when the main papers are submitted for publication. Additional analyses suggested by reviewers or editors will be performed if considered appropriate. This should be documented in a file note.

Amendments to the statistical analysis plan will be described and justified in the final report of the trial and where appropriate in publications arising from the analysis.

2 SYNOPSIS OT TRIAL DESIGN AND PROCEDURES

Title	Desmopressin for reversal of Antiplatelet drugs in Stroke due to Haemorrhage (DASH)
Acronym	DASH
Short title	Desmopressin for reversal of Antiplatelet drugs in Stroke due to Haemorrhage (DASH)
Chief Investigator	Prof Nikola Sprigg Deputy Chief Investigator: Dr Michael Desborough
Objectives	To assess the feasibility of screening, checking the eligibility, approaching, randomising, administering the intervention, and completing follow-up for patients treated with desmopressin or placebo to inform a definitive trial.
Trial Configuration	A phase II double blind randomised placebo controlled feasibility trial
Setting	Secondary care
Sample size estimate	This is a feasibility study so there is no formal sample size calculation. It is likely that a large definitive trial would be feasible if at least 50 participants were recruited into this study, that adherence to study drug was high and that a high proportion of follow up data was available. Lower recruitment would not preclude progression if there was some evidence that the barriers to recruitment identified could be overcome.
Number of participants	50

Eligibility criteria	**Inclusion:** Adults (≥18 years); confirmed intracerebral haemorrhage on imaging; less than 24 hours from onset of symptoms [or from when last seen free of stroke symptoms]; prescribed and thought to be taking a daily oral antiplatelet drug in the preceding seven days (cyclooxygenase inhibitors, phosphodiesterase inhibitors or $P2Y_{12}$ inhibitors); signed consent (or waiver of consent). **Exclusion:** Aneurysmal subarachnoid haemorrhage known at time of enrolment; haemorrhage suspected to be due to transformation of ischaemic stroke; haemorrhage known to be due to thrombolytic drug; haemorrhage known to be due to venous thrombosis; risk/s of fluid retention associated with desmopressin judged clinically significant by the attending physician (for example patients with pulmonary oedema and/or cardiac failure); significant hypotension (systolic blood pressure <90mmHg); known drug-eluting coronary artery stent in previous three months; allergy to desmopressin; pregnant or breast-feeding; life expectancy less than four hours, or planned for palliative care only; Glasgow coma scale less than 5, mRS >4.
Description of interventions	Intravenous desmopressin: 20µg in 50mls Sodium Chloride 0.9% infused over 20 minutes. Comparator – placebo (Sodium Chloride 0.9% intravenous infusion) administered by identical regimen.
Duration of study	12 months. Participants will be followed up for 90 days.
Randomisation and blinding	Patients will be randomised (1:1) to receive either desmopressin or matching placebo (Sodium Chloride 0.9%) via intravenous injection. Randomisation will be

	performed by the Stroke Trials Unit (STU) and involve computerised minimisation on key prognostic factors: age; sex; time since onset; systolic blood pressure; and presence of intraventricular haemorrhage. Patients, relatives, researchers and outcome assessors will be masked to treatment allocation.
Outcome measures	***Feasibility outcomes*** Number of eligible patients who receive allocated treatment Rate of eligible patients randomised Proportion of eligible patients approached; proportion of eligible patients randomised and reasons for non-randomisation; adherence to intervention; proportion of participants followed up to 90 days and reasons for loss to follow up; proportion of randomised participants with full outcome data available, and reasons for non-availability ***Secondary outcomes*** Hyponatraemia at 24 hours; early case fatality <28 days; case fatality at day 90; serious adverse events (including thromboembolic events) up to day 90; change in intracerebral haemorrhage volume at 24 hours; discharge destination; disability (Barthel index, day 90); quality of life (EuroQol, day 90); cognition (telephone MMSE day 90); length of hospital stay; health economic assessment (EQ-5D); assessment of baseline platelet dysfunction (P-selectin) and correlation with response to desmopressin; change in factor VIII, VWF antigen and VWF activity, one hour after administration of desmopressin.

Statistical methods	This is a feasibility trial and the main analysis will be with descriptive statistics only. Counts will be summarised using N and %, and continuous variables will be summarised using means and standard deviations or medians and interquartile ranges depending on their distribution. Whilst some variables will be summarised by treatment group, no formal statistical comparisons will be made and any analyses will be considered purely exploratory.

2.1 Sample size and justification

Since this is a feasibility study with one of the objectives being to determine potential recruitment rates, a formal sample size calculation is not appropriate. If more than 50 participants were randomised from 10 sites in a 12 month period, it is likely that, assuming similar recruitment rates in additional sites, a larger study recruiting approximately 1200 participants in around 50 sites recruiting for 60 months would be feasible. Depending on the final sample size calculation for the definitive study, the number of sites and recruitment period could be determined using the information from the rates and patterns observed in the feasibility study. Information about set up times for sites will also better inform recruitment projections for a larger study.

TICH-2 and other studies indicate approximately 25% of all people presenting with intracerebral haemorrhage are taking antiplatelet drugs.

2.2 Blinding and breaking of blind

Clinicians, patients and outcome assessors (clinical, radiological and haematological assessors) will be blinded to treatment allocation. The blinding status of individuals involved in the trial is given below:

	Blinding status	**Comments**
Participants	Blinded	Participants will be blinded.
Principal Investigator and other site staff	Blinded	Site staff will be blinded.
Chief Investigator	Blinded	The CI will not have access to any participant data with the potential to unblind.
Trial management staff at Stroke Trials Unit	Blinded	Trial Management staff will not have access to any participant data with the potential to unblind.
Trial Statistician	Blinded	The trial statistician will not have access to any participant data with the potential to unblind. Provision of any summaries required by allocation will be carried out by an independent statistician.

Database Programmer at Stroke Trials Unit	Not Blinded	The database programmer will have access to this information in order to maintain the database and manage queries.
Independent statistician	Not blinded	The independent statistician will have access to the treatment codes to provide tables and listings to the Data Monitoring Committee. Output will be stored in an access restricted area.

In general there should be no need to unblind the allocated treatment. If some contraindication to desmopressin develops after randomisation (e.g. clinical evidence of thrombosis), the trial treatment should simply be stopped. Unblinding should be done only in those rare cases when the doctor believes that clinical management depends importantly upon knowledge of whether the patient received desmopressin or placebo. In those few cases when urgent unblinding is considered necessary, the emergency telephone number should be telephoned, giving the name of the doctor authorising unblinding and the treatment pack number. The caller will then be told whether the patient received desmopressin or placebo. The rate of unblinding will be monitored and audited. The emergency contact details will be given to the investigators at participating sites.

In the event of breaking the treatment code this will normally be recorded as part of managing a SAE (see below for more details) and such actions will be reported in a timely manner. The Chief Investigator (delegated the sponsor's responsibilities) shall be informed immediately (within 24 hours) of any serious

adverse events and shall determine seriousness and causality in conjunction with any treating medical practitioners.

2.3 Trial Committees

Trial Management Committee

The TMC consists of the Chief Investigator, Deputy Chief Investigator, Senior Trial Manager, Trial Manager, Database Programmer, Trial Administrator and Trial Statistician. The TMC are responsible for the day-to-day management of the trial and will monitor all aspects of the conduct and progress of the trial, ensure that the protocol is adhered to and take appropriate action to safeguard participants and the quality of data collected in the trial. The TMC will report to the independent Trial Steering Committee (TSC). Trial co-ordination will be through the Stroke Trials Unit, in conjunction with the Nottingham Clinical Trials Unit (NCTU). This group, based at the Stroke Trials Unit, will meet regularly, at least every four weeks.

Trial Steering Committee

The TSC provides overall supervision, monitors progress against targets, receives reports on safety from the Data Safety Monitoring Committee, and provides advice to the Chief Investigator and TMC. The TSC met initially to review and agree the protocol, and then approximately every six months. The TSC will provide independent assessment of whether they feel a definitive trial will be feasible.

Data Monitoring Committee

An independent Data Monitoring Committee (DMC) will monitor safety of participants. The DMC will receive safety reports approximately every six months, or more frequently if requested and perform unblinded reviews of safety data. These reports will be provided by an independent statistician who has access to the treatment codes.

The DMC will report their assessment to the independent chair of the TSC.

2.4 Assessments

Clinical assessment; Baseline (Day 1), end of treatment (within 1 hour), Follow-up: 24 hours, and day 90 – telephone interview (or postal questionnaire if participant preference or unable to complete via phone).

Radiological – Follow-up: 24 hours CT brain scan to assess for haematoma expansion.

Safety check – Follow-up: Blood pressure: during infusion and post infusion; 24 hours serum sodium

Haematological assessments:

1. P-selectin; Pre-treatment blood test.
2. VWF (antigen and activity), Factor VIII; Pre and post-treatment (within one hour).

2.5 Outcome measures

Feasibility outcomes (primary)

Outcomes include:

- Proportion of eligible patients who are enrolled and receive randomised treatment.
- Proportion of patients who were screened who were eligible.
- Proportion of eligible patients approached who were randomised.
- Rate of participant recruitment per month per site.
- Time to randomisation following hospital admission.
- Adherence to allocated treatment.

- Proportion of participants followed up to 90 days and reasons for loss to follow up.
- Proportion of randomised participants with the proposed primary and secondary outcome data available.

Secondary outcomes

The following variables are expected to be outcomes in a subsequent definitive trial and are being collected in this trial:

- Haematological assessment of baseline platelet dysfunction (P-selectin) and correlation with response to desmopressin
- Change in factor VIII, VWF antigen and VWF activity, one hour after administration of desmopressin
- Hyponatraemia at 24 hours
- Change in intracerebral haemorrhage volume at 24 hours
- Early mortality <28 days
- Discharge destination
- Length of hospital stay
- Death or dependency at day 90 (modified Rankin scale)
- Mortality up to day 90
- Disability (Barthel index, day 90)
- Quality of life (EuroQol, day 90)
- Cognition (TICS-m, day 90)
- Serious adverse events (including thromboembolic events) up to day 90

3 INTERIM ANALYSIS

There was no planned interim analysis

4 GENERAL ANALYSIS CONSIDERATIONS

4.1 Analysis samples

The screened population will include all participants screened for this trial. All analyses will be of descriptive nature. No formal between group comparisons are planned and no statistical testing is required.

The Intention to treat (ITT) dataset will include all randomised participants. They will be summarised overall and according to their randomised treatment group. Missing data will not be imputed.

4.2 Timing of final analysis

All outcomes will be analysed collectively after database lock.

4.3 Statistical software

Analyses will be performed in SAS version 9.4 (SAS Institute, Cary, NC, United States).

4.4 Derived outcomes

Glasgow Coma Scale (GCS) score is the sum of 3 individual items and ranges from 3 to 15, with 15 meaning fully awake.

National Institutes of Health Stroke Scale (NIHSS) score is the sum of 15 questions and ranges from 0 to 42, with 0 meaning no stroke symptoms.

Barthel Index total score is the sum of ten individual items and ranges from 0 to 100, with a score of 100 meaning functional independence in activities of daily living.

EQ5D health status utility value (HSUV) is derived from five individual items and ranges from -0.59 to 1 with 1 being perfect health. The UK version of the EQ-5D index algorithm using the time trade off value set will be used for all patients.[1]

EQ-5D index scoring algorithm (Time trade-off: UK version)

Score	Weight
Full health (11111)	1
At least one 2 or 3	-0.081
At least one 3	-0.269
Mobility	
Score 2	-0.069
Score 3	-0.314
Self-care	
Score 2	-0.104
Score 3	-0.214
Usual activities	
Score 2	-0.036
Score 3	-0.094
Pain/discomfort	
Score 2	-0.123
Score 3	-0.386
Anxiety/depression	
Score 2	-0.071
Score 3	-0.236

For example, a health state of 32211 will convert to 0.196 (=1-0.81-0.269-0.314-0.104-0.071).

Telephone Interview for Cognitive Status-modified (TICS-m) score is the sum of 10 questions and ranges from 0 to 39, with 39 meaning no evidence of cognitive issues.

Zung depression scale total score is calculated by multiplying the summation of ten individual items by 2.5 and ranges from 25 to 100, with 25 meaning no evidence of poor mood.

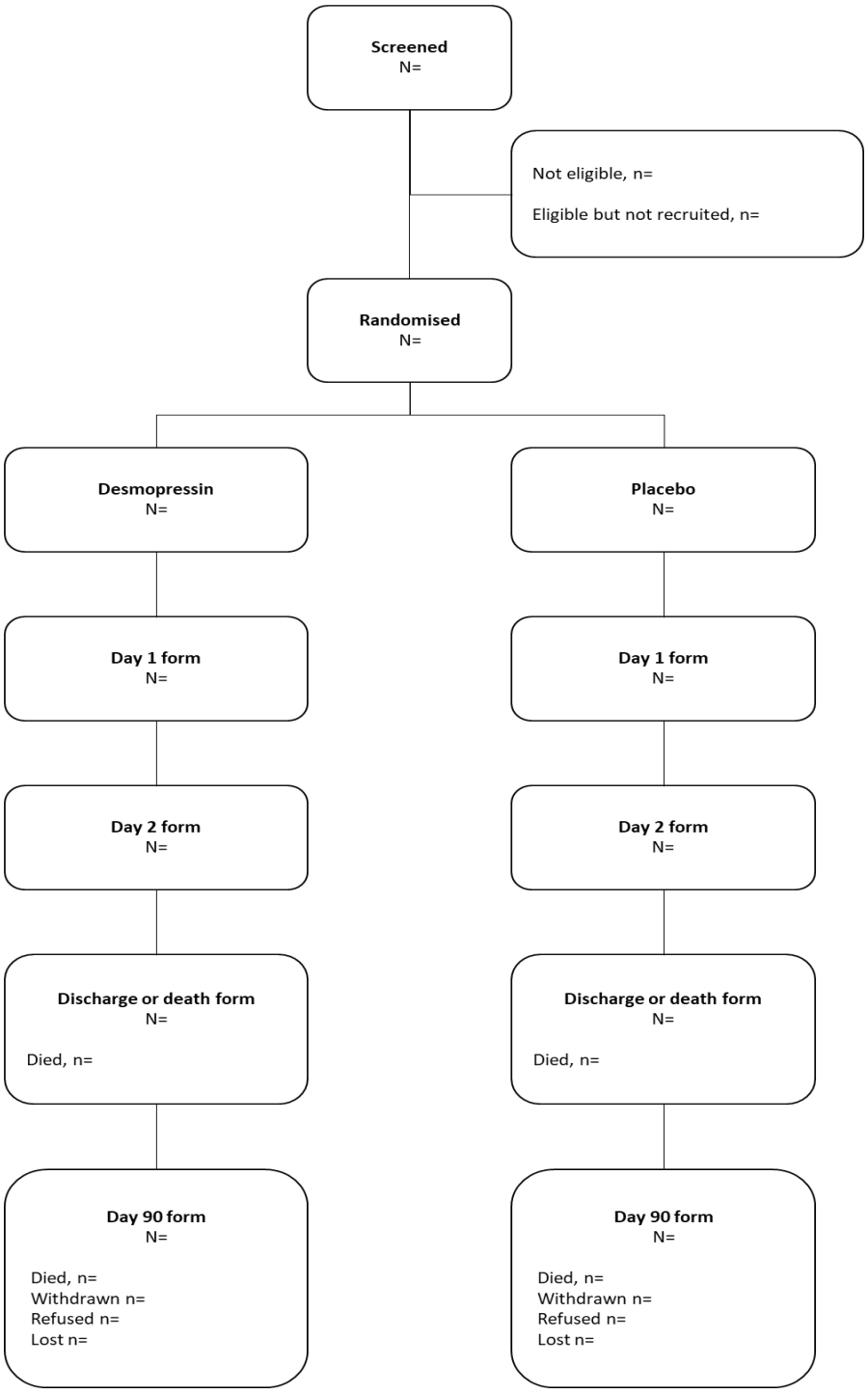
4.5 Procedures for missing data

Missing data will be reported. All analyses will be performed without imputation of missing data.

5 DESCRIPTION OF PARTICIPANT CHARACTERISTICS

5.1 Participant flow

The flow of participants from screening to randomisation and follow up is shown below.



5.2 Baseline characteristics

Continuous variables at baseline will be summarised for each trial arm and overall, in terms of the mean, standard deviation, median, lower and upper quartiles, minimum, maximum, and number of observations. Categorical variables will be summarised in terms of frequency counts and percentages.

6 ASSESSMENT OF STUDY QUALITY

6.1 Adherence

Adherence to randomised treatment will be summarised for both treatment arms. This will be summarised as the number of participants receiving full treatment and number of participants receiving some/no treatment with reasons for non- or partial receipt. Overall adherence will also be summarised.

6.2 Follow-up and discontinuations

The return rate of follow up questionnaire at 90 days will be summarised for both trial arms and overall. The number of discontinuations (withdrawals and losses to follow-up) will be presented in the participant flow diagram specified in Section 5.1.

6.3 Protocol violations and deviations

All protocol violations will be summarised by trial arm and overall. Deviations will also be listed but they will only be summarised overall, not by trial arm.

6.3.1 Temperature excursions

Temperature excursion information will be summarised by site and overall. Information presented will include; number of excursions, total excursion duration (hours), mean excursion duration, numbers of IMP packs exposed, quarantined, number of IMP packs destroyed, and the number of participants receiving exposed packs.

7 ASSESSMENT OF FEASIBILITY

This is a feasibility trial and the main analysis will be with descriptive statistics only. Counts will be summarised using N and %, and continuous variables will be summarised using means and standard deviations or medians and interquartile ranges depending on their distribution. Whilst some variables will be summarised by treatment group, no formal statistical comparisons will be made and any analyses will be considered purely exploratory. The only between treatment group observation with the potential to impact on the assessment of whether or not it would be feasible to run a definitive trial would be if concerning differences were noted in safety.

7.1 Analyses to evaluate the feasibility of a larger study

Feasibility of recruitment and retention

A CONSORT flow diagram will show the numbers of participants screened in hospital, randomised and having completed follow-up at 90 days. Numbers of participants screened, eligible, recruited, eligible but not recruited and excluded (including reasons) will also be summarised by recruitment centre separately from CONSORT diagram.

Completeness of data

Completeness of the case report forms at each time point will be summarised in the CONSORT flow diagram.

Completeness of data items, including baseline characteristics and outcomes, will be summarised overall within the relevant tables.

7.2 Assessment of efficacy

This is a feasibility trial and as such will have no formal assessment of efficacy. The proposed primary efficacy outcome in a definitive trial would be death or dependency at day 90, measured using the modified Rankin scale. Shifts in this scale will be summarised for this trial but no formal confirmatory statistical analyses will be performed. Similarly, all other efficacy variables will be summarised using descriptive statistics.

Other efficacy variables that will be summarised include Barthel Index (disability), EQ-5D (quality of life, using health status utility values), TICS-m (cognition), Verbal fluency (cognition) and Zung depression score (mood).

The number of participants with completed scores for each outcome will also be reported.

8 ASSESSMENT OF SAFETY

Data regarding safety will be summarised using descriptive statistics according to the treatment the participant received. Safety outcomes will include:

- Death
- Adverse events collected at the Day 2 follow-up.
- Serious adverse events occurring within the first 7 days will be assessed for seriousness, expectedness and causality. In addition, fatal serious adverse events and safety outcome events (fluid overload, hyponatraemia) will be reported until day 90. Serious adverse events will be categorised in accordance with the medical dictionary for regulatory authorities.
- Hyponatraemia: All <135 mmol/L, Severe <125 mmol/L

9 EXPLORATORY ANALYSES

Exploratory analysis data will be summarised using descriptive statistics according to the treatment the participant received.

There will be an assessment of the data collected from the pre and 24 hour post enrolment scans. Outcomes collected include:

1. Pre-enrolment scans: time from onset to scan (hours), ICH location, side of brain affected, cause of ICH, evidence of finger-like projections (FLP), evidence of subarachnoid haemorrhage (SAH), Blend sign, Island sign, Black hole sign, evidence of hypodensity, Barras shape, Barras Density,

evidence of Intraventricular haemorrhage (IVH), Graeb score, modified Graeb score, evidence of atrophy, evidence of periventricular lucency, evidence of old vascular lesions, haemorrhage volume, IVH volume, Peri-haematomal oedema (PHE) volume, size of midline shift.

2. Post enrolment scans: time from randomisation to scan (hours), haemorrhage volume, IVH volume, Peri-haematomal oedema (PHE) volume, size of midline shift.

Evidence of any haematoma expansion will also be determined using data from both the pre and 24 hour post enrolment scans. Haematoma expansion is defined as an increase of >6 mL or a growth of >33%.

The baseline platelet dysfunction assessment will include the proportion of patients taking aspirin with impaired baseline platelet function and the proportion of patients taking P2Y12 inhibitors with impaired baseline platelet function.

The assessment of bloods will include calculated means for factor VIII, Von Willebrand Factor antigen and Von Willebrand Factor activity taken prior to and at one hour post administration of desmopressin. Mean change between the pre and post results will also be provided for these measurements.

10 REFERENCES

1. Whynes DK, S.N., Selby J, Bath PMW, *Testing for differential item functioning within the EQ-5D.* Medical Decision Making, 2013. **33**: p. 252.