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)

The ENOS Trial Investigators*

Abstract **High blood pressure (BP) is common in acute stroke and is independently associated with a poor outcome. Many patients with acute stroke are taking antihypertensive medications. To test the safety and efficacy of 7 days of transdermal glyceryl trinitrate (GTN, 5 mg/day) vs. no GTN in patients with acute stroke; patients taking antihypertensive therapy immediately before their stroke are also randomised to continue vs. stop this temporarily. ENOS is a prospective international multicentre single-blind randomised-controlled trial in 5000 patients with acute (<48 h of onset) ischaemic or haemorrhagic stroke. The primary outcome is combined death and dependency (modified Rankin scale >2) at 90 days measured by blinded central telephone follow-up. Secondary outcomes include: BP over the 7 days of treatment; death, impairment (Scandinavian stroke scale), recurrence, and neuroimaging at 7 days; discharge disposition, disability (Barthel index), cognition (mini-mental status examination) and quality of life (EuroQoL). The sample size will allow an absolute difference in death/dependency of 5% to be detected with 90% power at 5% significance for GTN versus no GTN. Randomisation and data collection are performed over a secure Internet site with real-time data validation. Neuroimaging and serious adverse events are adjudicated blinded to treatment.**

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Introduction

High blood pressure (BP) is common in both acute ischaemic stroke (IS) (1) and primary intracerebral haemorrhage (PICH). Patients with high BP have a worse outcome, whether judged as early recurrence, death within a few weeks, or combined death and dependency after several months (1–3). Lowering BP might therefore reduce these events and improve functional outcome. However, cerebral autoregulation is dysfunctional in acute stroke (4) and lowering BP might instead lead to hypoperfusion, stroke extension, and a worse outcome. This equipoise has been debated for more than 20 years (5, 6) and still no large trials of lowering BP have been completed (7, 8). Enhancing this state of equipoise, several small studies in patients with IS have reported conflicting results. Trials of oral β-receptor antagonists and intravenous calcium channel blockers (nimodipine) found that outcome was worse in those patients randomised to active treatment (9, 10). In the latter study, the worsening of outcome paralleled the treatment-induced decline in BP (11). In contrast, treatment with an oral angiotensin receptor antagonist (candesartan) in patients with a severely elevated BP after there is was associated with a reduction in combined death and vascular events, although no effect on BP or functional outcome was detected (12).

Nitric oxide (NO) donors are candidate treatments for acute stroke (13); NO is a cerebral and systemic vasodilator, modulates vascular and neuronal function, and inhibits apoptosis. Preclinical studies of cerebral ischaemia have found that NO donors reduce stroke lesion size, and improve regional cerebral blood flow and functional outcome (14). Four small clinical studies of NO donors have been performed in acute stroke. Intravenous sodium nitroprusside reduced BP without altering CBF, and exhibited antiplatelet effects (thereby precluding its use in PICH) (15). Three small trials of transdermal glyceryl trinitrate (GTN) found that it lowered peripheral and central BP and pulse

pressure, had no effects on platelet activity (so it can be given in PICH) or middle cerebral artery blood flow velocity, and improved aortic compliance (16–18). The latter study also found no adverse effects of GTN on cerebral blood flow or intracranial pressure (18). No safety concerns were present in these studies (16–19). Furthermore, GTN could be given by skin patch irrespective of the presence of dysphagia, a frequent complication of stroke that limits oral drug administration.

Many patients with acute stroke are admitted on antihypertensive medication and it remains unclear whether this should be continued or stopped temporarily (20). Continuing drugs such as β-receptor antagonists or calcium channel blockers might be hazardous; alternatively, stopping prior therapy might lead to rebound hypertension.

The 'Efficacy of Nitric Oxide in Acute Stroke' (ENOS) trial is testing the safety and efficacy of transdermal GTN, and of continuing or stopping temporarily prior antihypertensive medication, in patients with acute ischaemic or haemorrhagic stroke (21).

Methods

Design

ENOS is a prospective, international, multicentre, single-blind, parallel-group, partial-factorial, randomised-controlled trial. The study was conducted according to the principles of the Declaration of Helsinki and 'International Conference on Harmonisation of Good Clinical Practice'. Study approval by national (UK) and local research ethics committees (all centres) has been obtained. The management of personal data adheres to the UK Data Protection Act 1998.

Patient population

Hospitalised patients with a clinical syndrome of stroke are eligible for the trial if they are aged 18 or over, have a motor deficit in arm and/or leg, have a systolic BP between 140 and 220 mmHg, can be treated within 48 h of onset, and have given written informed consent; assent from a relative is acceptable if patients are semi conscious, dysphasic, or confused, in accordance with the practice of the local research ethics committee.

Patients are excluded if they have one or more of the following:

- coma (Glasgow Coma Scale <8)
- pure sensory stroke; preceding moderate or severe disability (modified Rankin scale, mRS 3–5)
- confounding neurological or psychiatric disease
- a condition mimicking stroke (e.g. hypoglycaemia, Todd's paresis)
- severe liver dysfunction or renal dysfunction
- severe concomitant medical conditions
- pregnancy or breast feeding
- previous participation in ENOS
- planned surgical intervention (e.g. carotid endarterectomy), or
- a definite need for, or contradiction to, nitrates and/or prestroke antihypertensive therapy.

Patients may be coenrolled into nondrug stroke trials.

Baseline measures

Baseline demographic (age, sex), clinical details (syndrome (22), severity (23, 24), BP, heart rate) are determined after consent/assent and before randomisation. BP and heart rate are measured three times over 15 min using validated automatic clinical monitors (Omron HEM-705CP or HEM-757, Illinois, USA) (25).

Neuroimaging [computed tomography (CT) or magnetic resonance (MR) scanning] is performed to diagnose stroke type (ischaemic, PICH) or nonstroke lesions; scanning is undertaken, ideally, before treatment but otherwise during the 7-day treatment period. Scan interpretation by the local investigator is recorded; scans are also adjudicated centrally over the Internet NeuroGrid by independent assessors blinded to treatment and using a validated structured classification system (26, 27). Investigators are also encouraged to perform carotid ultrasound examination in patients with IS to facilitate safety analyses in those with severe carotid stenosis; such scanning may be performed at any time during the hospital admission and is not necessary before randomisation.

Randomisation

Patients are randomised equally to treatment with GTN or no GTN, with stratification by prior antihypertensive treatment and country, and minimisation on key prognostic baseline variables: sex, age (<70, $\geqslant 70$ years), stroke severity [SSS $\leqslant 40$, >40 (23)], time to treatment ($\leqslant 24$, >24 h), and total anterior circulation syndrome (22). Those patients receiving prior antihypertensive therapy are also randomised to continue or temporarily stop this. Randomisation, all data collection, and SAE and CT adjudication are performed over a secure password-protected and data-encrypted Internet website (www.enos.ac.uk) (28); indeed, ENOS is the world's first acute stroke trial to use the Internet in this way.

Treatment

Treatment is given daily for 7 days, or until discharge if earlier, and consists of a GTN patch (5 mg, usually placed between 08·00 and 09·00 hours) or no patch, and continuing or temporarily stopping prestroke antihypertensive therapy if relevant. Patients taking prestroke antihypertensive therapy will in essence be randomised into a factorial trial nested within the overall study investigating GTN (Table 1), a design

used by other trials such as ASCOT (29, 30). Patients are blinded to GTN by placing a gauze dressing over the patch or a similar area of skin if randomised to no GTN. The doses of GTN and antihypertensive therapy are not adjusted during treatment. Treatment is given on top of standard best medical care, including thrombolysis (if licensed and routine at the centre), any other licensed treatment for acute stroke, and aspirin.

Study agents can be stopped if the patient withdraws consent, for safety reasons, or if unacceptable adverse events develop. Nontrial nitrates and antihypertensive agents should not be given during the treatment period. Systematic use of long-term oral antithrombotic and lipid-lowering agents is recommended for secondary prevention in patients with IS; antihypertensive therapy is encouraged once the 7-day treatment period is over.

Primary outcome

The primary outcome is combined death and dependency, measured using the mRS (24, 31) with score 3–6. mRS is determined centrally by telephone by a trained assessor (blinded to treatment assignment and baseline clinical characteristics) in each country at day 90 (+10 days). The primary outcome will be analysed in prespecified subgroups: sex, age (<70, >70 years), stroke severity (SSS<30, 30–40, >40), time to treatment (<12, 12–24, >24 h), type of stroke (ischaemic, PICH), presumed aetiology by modified TOAST criteria (cardioembolism, large vessel, small vessel (24, 32)), and clinical syndrome by Bamford classification (22, 24).

Secondary outcomes

Investigators reassess patients after randomised interventions have ceased at day 7 (or earlier if discharged) and at discharge from hospital. Outcomes assessed at the end of treatment include frequency of patients deteriorating neurologically (a decrease in SSS of >5 points or a decrease in the consciousness part of SSS of >2 points (24)), having a recurrent stroke (classified as ischaemic, haemorrhagic, or unknown type), and death. A posttreatment follow-up scan may be performed in those patients whose baseline scan is undertaken before treatment; comparison of the posttreatment and baseline scans will facilitate safety assessments, e.g. infarct swelling/treatment interaction. Hospital-related events include discharge disposition (death, discharge to institution, discharge home) and length of stay. Prespecified secondary outcomes at 90 days include median mRS, disability (Barthel index, BI<60; BI<95; median BI), death, quality of life [EuroQoL (33)], cognition [modified mini-mental state examination (MMSE) (34)], and mood [Zung depression score (35)]. As with the primary outcome, these are determined centrally by telephone, with the assessor blinded to randomised treatment and baseline and hospital clinical details.

Investigator-reported serious adverse events are grouped by time of onset (before, during, or after treatment) and validated and categorised blindly by independent adjudicators.

Table 1 Partial factorial design of the 'Efficacy of Nitric Oxide in Stroke' (ENOS) trial with estimated numbers of patients in each treatment group

	Prestroke Antihypertensive Medication			
	None	Stop	Continue	Total
GTN	1250	625	625	2500
No GTN	1250	625	625	2500
Total	2500	1250	1250	5000

All patients are randomised to GTN or no GTN; those taking antihypertensive medication immediately before their stroke are also randomised to continue or stop this temporarily. The interventions are given for seven days starting within 48 h of stroke onset.
GTN, glyceryl trinitrate.

Data-Monitoring Committee (DMC)

The DMC review unblinded data twice yearly in respect of safety and efficacy, and consider the study in the context of other trials of altering BP in stroke. In addition to assessing the effect of the two interventions (GTN vs. no GTN, continue vs. stop prestroke antihypertensive therapy) across all patients, they are monitoring safety measures in particular subgroups of patients including those who present very early, those with severe stroke (TACS (22)), and those who turn out to have significant carotid disease (ipsilateral stenosis of the internal carotid artery >50%).

Sample size

The study requires the enrolment of 5000 patients to detect an absolute reduction in combined death or dependency (mRS 3–6) of 5% (relative reduction 10%) assuming a frequency of 50% in the control group, with 90% power, an overall significance of $P<0{\cdot}049$ (which includes an interim analysis at approximately 2200 patients with significance set at $P<0{\cdot}001$), and losses to follow-up of 3%. It is assumed that approximately 50% of patients (~2500) will be taking antihypertensive therapy immediately before their stroke; this sample size allows an absolute reduction in death or dependency of 7% to be detected between continuing and stopping therapy with 90% power.

Statistical analyses

Both efficacy and safety analyses will include all randomised patients. Per-protocol analyses will be performed on patients who receive at least 4 days of treatment. The effect of treatment on serious adverse events will be assessed at both the end of treatment (day 7) and the end of follow-up (day 90).

Logistic regression will be performed on the dichotomised primary outcome measure; ordinal measures such as SSS, MMSE, and EuroQoL Zung will be analysed using the Mann–Whitney *U*-test. Death will be analysed using the Kaplan–Meier and Cox regression models with adjustment for baseline variables, including age, sex, impairment (SSS), syndrome (TACS), and BP. Analyses will first compare the effect of GTN vs. no GTN, and continue vs. stop prestroke antihypertensive medication; they will then test for any interaction between the interventions. Analyses will be two sided ($P < 0{\cdot}05$ with no adjustment for multiple comparisons) and 95% confidence intervals will be presented.

Study organisation and funding

ENOS is an independent academic trial performed by an international collaborative group. The study is supervised by a Trial Steering Committee and International Advisory Committee, and run by a Trial Management Committee based at the ENOS Trial Coordinating Centre in Nottingham. Independent and blinded Adjudicators classify serious adverse events for a separate and independent DMC. The trial is funded by BUPA Foundation (start up phase) and Medical Research Council (main phase), with additional support from The Stroke Association, The Hypertension Trust, Reichstadt bequest, and University of Nottingham. A major substudy utilising magnetic resonance imaging is supported by the Biomedical Research Council of Singapore.

Summary

ENOS is addressing two key questions in the management of patients with acute stroke, namely the safety and efficacy of lowering BP with transdermal GTN vs. no GTN, and of continuing vs. stopping temporarily prestroke antihypertensive therapy. The large sample size of 5000 patients means that a modest but worthwhile clinical effect (5% absolute risk reduction) can be detectable with high statistical power (90%). ENOS is the world's first trial in acute stroke to use the Internet for randomisation, data registration, and adjudication of SAEs and neuroimages. A positive trial would mean that transdermal GTN could be rapidly introduced into clinical practice around the world as it is readily available, easy to administer, and inexpensive (~ £5/patient). We invite centres from around the world to join this important collaborative international venture.

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