Prospective Evaluation of Blood Coagulability and Effect of Treatment in Patients with Stroke Using Rotational Thromboelastometry

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Background: Stroke is the second largest cause of death worldwide. Abnormalities in hemostasis play an important role in the pathophysiology of ischemic stroke (IS). These hemostatic defects can be detected using rotational thromboelastometry (ROTEM) as a global method of measuring coagulation.This study assessed the effects of IS on blood hypercoagulability using ROTEM method, before and subsequent to therapeutic interventions. *Methods:* In a prospective observational cohort study, whole blood coagulation using ROTEM, along with full blood count and standard coagulation tests, were compared between patients with IS and an age-matched control group of healthy volunteers. Further assessment took place at 2-4 hours and at 24 hours in the stroke group after therapy to assess the effects of therapeutic intervention. *Results:* Seventy-two patients with IS were age-matched to 71 healthy subjects. Clotting time (CT) INTEM ($P = .01$) and maximum clot firmness (MCF) INTEM ($P = .02$) were significantly different between stroke patients at baseline and healthy subjects, but this difference disappeared when controlled for by smoking status. There was no association between ROTEM parameters and time from stroke symptom onset or stroke severity as reflected in The National Institute of Health Stroke Scale score. Significant but small changes in the values of MCF-EXTEM, clot formation time (CFT) EXTEM, and alpha-EXTEM CT were observed after therapeutic intervention (thrombolysis or aspirin treatment). *Conclusions:* ROTEM testing does not seem to detect a hypercoagulable state in patients with IS. Nonetheless, some ROTEM parameters had a small change after antiplatelet therapy or thrombolysis. **Key Words:** Cerebrovascular disease—stroke—ROTEM—aspirin—thrombolysis—hypercoagulability.

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Stroke is the second leading cause of death worldwide with ischemic stroke (IS) accounting for more than 80% of all strokes.[1] Such ischemic cerebrovascular events have been identified as being associated with a hypercoagulable state,[2-4] which explains the role of the 2 main treatments available, antiplatelet therapy and thrombolysis.[5,6] Although the efficacy of these treatments has been proven in large clinical trials, this does not take into account individual variation in terms of patient response to these therapies or risk of bleeding.[7,8]

Viscoelastic techniques, which include thromboelastography (TEG; Haemoscope Corporation, Niles, IL) and rotational thromboelastometry (ROTEM; Pentapharm GmbH, Munich, Germany), measure the dynamics of clot formation, strength, and lysis and therefore may provide information on hypercoagulability and the potential effects of therapeutic interventions. They have been used in the assessment of hemostasis in various clinical settings,[9-12] and only 2 studies have investigated hypercoagulability in stroke using TEG[13,14] with that of Elliott also reporting on the response to thrombolysis in a small number of patients. However, there appear to be no studies of ROTEM to assess hypercoagulability in stroke. It has been argued that ROTEM is superior to TEG because it is more resistant to vibration and has an integrated 4-channel automated system with autopipette and easy to follow on-screen instructions.[15] This would then potentially lend itself to be a useful test in the diagnosis of IS and assessing the effects of therapeutic interventions. Hence, this is the first study to investigate ROTEM in IS and aims to compare ROTEM parameters between patients with first-ever IS and age-matched healthy subjects. Second, it aims to assess the effect of therapeutic intervention (both thrombolysis and aspirin) on ROTEM parameters.

Methods

Study Design

A prospective observational cohort study to compare blood coagulability in patients with IS versus age-matched healthy volunteers using ROTEM.

Patient Population

Patients with a first-ever stroke were recruited on presentation to the emergency department of a large teaching hospital (ABMU Health Board, Swansea, United Kingdom). Once a provisional diagnosis of stroke was made, strict inclusion criteria were applied: adults (≥18 years) with first-ever IS based on clinical history, examination and neuroradiology, and checked by a member of the research team against World Health Organization diagnostic criteria[16] gave written fully informed consent. For those unable to consent because of lack of mental capacity, assent was sought from personal or professional legal representatives. Those who had previous stroke, on anticoagulant therapy, with disease states that are likely to affect coagulation (eg, liver disease, malignancy, renal failure) or imminent death were excluded. Clinicians were blinded to the result of ROTEM testing. ROTEM parameters have wide reference range that could be affected by various conditions[17]; we therefore decided to age match these patients with a control group generated from healthy local population. These healthy subjects were tested at the Hemostasis Biomedical Research Unit and were recruited via various advertising means including posters, internal, email, or direct invitations of staff and patients' relatives.

Blood Sampling and Data Collection

Once written informed consent was obtained, clinical and demographic data were collected. Baseline and 24 hours National Institute of Health Stroke Scale (NIHSS) scores (performed by a trained member of the stroke team) and ischemic time calculated from time of symptom onset to the time of blood collection were recorded. Symptom onset was decided as reported by patients when they were aware of the first symptom including those who woke up with symptoms. A baseline venous blood sample was collected for ROTEM analysis and standard clotting tests including full blood count (FBC), activated partial thromboplastin time (aPTT), prothrombin time (PT), and plasma fibrinogen. A further 2 samples of blood were taken at 2-4 and 24 hours to assess the effects of therapeutic intervention on blood coagulation.

ROTEM Analysis

Evolving global clot thickness was assessed using the standard method of ROTEM (ROTEM delta; Tem Innovations GmbH). Two ROTEM tests were performed to assess extrinsic (EXTEM) and intrinsic (INTEM) coagulation pathways. Measurement was undertaken according to the manufacturer's instructions in a disposable 8 mm diameter cup by addition of 300 μL of citrated blood (BD Vacutainer, Belliver Industrial Estate, Plymouth, UK; Ref: 367691) to 20 μL Star-tem (.2 mol/L CaCl2; Tem Innovations GmbH, Germany Ref: 503-10) followed by 20 μL of EXTEM (recombinant tissue factor and phospholipids; Tem Innovations GmbH, Germany; Ref: 503-05) or 20 μL INTEM, (partial thromboplastin; Tem Innovations GmbH, Munich, Germany; Ref: 503-10) for activation of the extrinsic and intrinsic pathways, respectively. Cups were then lifted onto a measurement channel containing a plastic pin 6 mm in diameter, which was immersed into the blood sample. The following measures were recorded as shown in Figure 1: clotting time (CT), clot formation time (CFT), maximum clot firmness (MCF), alpha angle (α), and maximum lysis.