PROGNOSTIC FACTORS FOLLOWING ISCHAEMIC STROKE

A thesis by

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Submitted for the degree of Doctor of Medicine

to

The University of Glasgow

from

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September 1999

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Acknowledgements

I am greatly indebted to Professor K. R. Lees for the opportunity to perform the studies described, and for his expert advice and encouragement.

I should also like to acknowledge the invaluable support and advice offered by Professor J.L. Reid. Dr P.F. Semple and Dr G.T. McInnes.

Dr C.J. Weir provided assistance in the preparation of the more complex statistical analyses described, for which I am very grateful. I would also like to express my gratitude to Dr J. Foster whose patient tuition allowed me to grasp the basics of magnetic resonance physics.

The tireless efforts of Mrs Karen Shields and Mr Iain Sim in the acquisition of the carotid and transcranial Doppler data are gratefully acknowledged.

Declaration

The work described in this thesis was carried out while I was employed as a clinical research fellow in the University Department of Medicine and Therapeutics of the Western Infirmary. The Doppler ultrasound studies presented in chapter two were performed by Mrs Karen Shields and Mr Iain Sim. Programming of the magnetic resonance scanner was performed by Dr John Foster of the Department of Clinical Physics. The more complex statistical analyses discussed in chapter five were supervised by Dr Chris Weir of the Robertson Centre for Biostatistics. Dr Alison Bolster provided technical expertise in the acquisition and analysis of the SPECT scans described in chapter two. I performed the assessment, recruitment and follow-up of all subjects studied in the work presented in chapters 2, 3 and 6. I was involved in the acute management of 12 of the 15 patients treated with thrombolytic therapy who are described in chapter 7. Unless stated above, all work described in the following chapters was carried out by myself. The writing of this thesis was entirely my own work.

Summary

The work presented for examination in this thesis concerns the investigation and management of patients with stroke. The underlying theme which unites the chapters is that of outcome following stroke, and how it may be influenced or predicted.

Chapter one is a broad overview of stroke disease, incorporating a brief summary of the socio-economic burden of stroke in the Western world. The clinical features and mechanisms of acute ischaemic stroke are discussed, and primary and secondary preventative measures are outlined. Each of the clinical, biochemical and radiological factors discussed later in the thesis is introduced, and for each topic a synopsis of earlier published work is presented. As chapter six deals with a novel magnetic resonance imaging sequence, chapter one also contains a summary of the basic physical principles of magnetic resonance.

Chapter two reports a randomised, double blind placebo-controlled study designed to investigate the effect of the angiotensin converting enzyme inhibitor perindopril upon cerebral and renal perfusion in hypertensive stroke patients with carotid artery disease. Control of hypertension in this group of patients is associated with the theoretical risk of reduction in cerebral perfusion distal to a stenotic lesion of the internal carotid artery; however failure to reduce elevated levels of blood pressure is associated with significant morbidity. Perindopril reduces blood pressure but not cerebral perfusion in hypertensive patients with recent ischaemic stroke, although the

mechanistic basis of this observation is unclear. I hypothesised that perindopril would lower blood pressure but not cerebral or renal perfusion in patients with ischaemic stroke accompanied by significant carotid arterial disease. Transcranial and duplex Doppler ultrasound were used to monitor global cerebral blood flow; regional cerebral perfusion was assessed using ^{99m}Tc hexamethyl propylene amine oxide single photon emission computed tomography (HMPAO SPECT) and dynamic HMPAO bolus imaging techniques. Glomerular filtration rate was measured using ^{51}Chromium EDTA. With a sample size of 24 patients we expected to detect a difference in internal carotid artery flow (as assessed by carotid Doppler) of 16% with 80% power.

Patients were identified at our cerebrovascular clinic, and after consent was obtained they were randomised to receive either perindopril 4mg daily for 14 days or placebo. Regular measurements of blood pressure, internal carotid artery velocity, internal carotid artery diameter and middle cerebral artery velocity were made before and at regular intervals for 24 hours after dosing. SPECT scanning was performed before dosing and at time of peak drug effect (6-8 hours after dose).

Blood pressure was effectively reduced in the treated group, and no fall in cerebral perfusion or drug associated neurological deterioration was observed. The glomerular filtration rate of one patient in the treated group fell after administration of perindopril; treatment was withdrawn and the patient

was well at the conclusion of the study. In the other patients, GFR was unchanged.

I conclude that although perindopril lowers blood pressure without affecting cerebral perfusion in this population, screening for renal artery stenosis should be considered before treatment is introduced.

Chapter three reports a study to investigate the feasibility of rigorous control of blood glucose in hyperglycaemic patients following stroke. Hyperglycaemia (blood glucose concentration greater than 8.0 mmol/L) at the time of ischaemic stroke has been identified as an independent poor prognostic factor, equivalent to adding 20 years to the patient's age. A randomised, controlled trial to investigate the potential benefit of blood glucose lowering in the context of acute ischaemic stroke is warranted, and we sought to investigate the feasibility of one approach.

After obtaining informed consent, 24 patients presenting within 12 hours of onset of acute ischaemic stroke with blood glucose greater than or equal to 8 mmol/lit and no requirement for insulin were randomised to receive either standard control of blood sugar or rigorous glycaemic control using intravenous insulin administered according to a predetermined sliding scale Treatment was maintained for 48 hours after admission. The study was performed on an open label basis due to practical difficulties in titrating a placebo infusion. Patients in both arms of the study received infusion of intravenous crystalloid at a rate determined by their fluid balance and

metabolic status; blood glucose was measured two-hourly in all patients for 48 hours. Normal diet was permitted if swallowing was safe, however no tube feeding was permitted for the duration of the infusion. Patients were reviewed at one month after admission, and clinical parameters were recorded.

The sliding scale regime was found to be well tolerated by patients, with only one episode of symptoms consistent with iatrogenic hypoglycaemia. Blood glucose concentration in the treated group was lower, with significant reduction in area under the glucose*time curve. A larger efficacy study to investigate the effect of intervention with insulin in such patients is warranted.

The prognostic significance of triglyceride concentration following ischaemic stroke is investigated in chapter four. Recent data have shown an unexpected association between poor acute stroke outcome and lower serum cholesterol. Triglyceride concentration has been linked to coronary heart disease and stroke; however there are currently no data on the relationship between triglyceride and stroke outcome. Such information may yield further mechanistic information on the relationship between lipids and stroke outcome.

I studied 1312 non-diabetic patients presenting to our acute stroke unit with computed tomography confirmed acute stroke. Fasting blood samples were drawn within 24 hours of admission for glucose, lipids and a standard battery of biochemistry and hematological tests. Information on age, stroke type, admission blood pressure, smoking status, presence of atrial fibrillation,

resolution time of symptoms and Oxfordshire Community Stroke Project clinical classification was collated. Stepwise proportional hazards regression was used to estimate the effect of the above variables on survival following stroke.

Increased age ($p<0.0001$), presence of atrial fibrillation ($p=0.009$), hyperglycemia ($p=0.001$), and lower triglyceride level ($p<0.0001$) independently predicted higher mortality; early resolution of symptoms ($p=0.004$) was independently associated with lower mortality. Although serum cholesterol level predicted outcome after adjusting for other prognostic factors, it did not remain significant when triglyceride level was entered in the model. The relative hazard per additional quartile of triglyceride was 0.84 (95% confidence interval 0.77 to 0.92).

I conclude that low triglyceride concentration strongly predicts higher mortality following stroke, whereas serum cholesterol level is not an independent predictor. Outcome following stroke thus relates more strongly to triglyceride-rich than to cholesterol-rich lipoprotein concentrations. The mechanisms which underlie this observation require further investigation.

In chapter five the prognostic significance of visible infarction on computed tomography in patients with lacunar stroke is examined. The functional outcome of patients with lacunar stroke is variable, and relatively few prognostic factors have been identified. All patients presenting to our unit with

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lacunar symptoms between June 1990 and February 1998 were identified. Patients with non-vascular causes for the symptoms, patients imaged with MRI and patients imaged within 12 hours of onset of symptoms were excluded from the analysis. 404 remaining patients were divided into two groups: "CT positive" if the scan showed an appropriately-placed area of cerebral infarction, and "CT negative" if the scan showed no such lesion. Survival times in patients with and without visible infarction on CT were compared using a log-rank test. A proportional hazards model was then used to assess the effect of visible infarction on survival after correcting for other clinical and biochemical factors known or suspected to influence prognosis.

No significant difference in duration of hospital stay, survival or functional outcome was seen between the two groups. I conclude that the presence of visible infarction on CT does not influence prognosis after lacunar stroke.

In chapter six the development and implementation of a novel diffusion-weighted magnetic resonance sequence is reported. Diffusion-weighted imaging (DW-MRI) is highly sensitive to changes in the apparent diffusion of water that occur in regions of cerebral ischaemia, allowing identification of ischaemic brain damage much earlier than was previously possible with conventional sequences. Previously reported diffusion-weighted sequences have required powerful (1.5T) magnets and Echo-planar capabilities or else have severe motion artefact sensitivity. We have developed a time-reversed steady state (PSIF) diffusion-weighted sequence which uses a 1.0T magnet and does not require echo-planar capabilities.

Nine consecutive patients with acute ischaemic stroke were scanned within 8 hours of onset of symptoms. Median age was 66.5 years range 50 - 83. All patients were scanned with the PSIF sequence, followed by conventional T2-weighted imaging. In all cases, lesions were seen on DWI that were not seen on the T2 images. The median time from symptom onset to scan was 5 hours 55 minutes (Range 2 hours 35 minutes to 7 hours 10 minutes). Localisation in each of these cases assisted medical management. In 2 cases, more than one lesion was present in each individual. The diffusion-weighted images allowed the new lesion to be discriminated from older lesions.

We conclude that the DW-PSIF sequence represents a clinically useful means of imaging acute ischaemic stroke at 1.0T, providing more diagnostic information than T2 weighted imaging alone. The sequence does not require high magnetic field strength or echo-planar capabilities, and has the potential to be widely applied in centres previously unable to use diffusion-weighted MRI due to technical limitations.

In chapter seven, preliminary experience with the use of recombinant tissue plasminogen activator in a UK stroke centre is reported. No treatment for acute ischaemic stroke is currently licensed for use in the United Kingdom, and the use of thrombolytic therapy is restricted to a small number of specialist centres, where it is employed outwith licence.

Patients were selected and treated in accordance with the American Heart Association guidelines. NIH, Barthel and Rankin scores were prospectively measured on admission, at three days and at three months for all patients with stroke treated with rt-PA in our unit between April 1997 and November 1998. Intracranial and systemic haemorrhagic complications were recorded.

Six women and nine men received thrombolytic treatment, mean age 74 years (58-93). Initial median NIH score was 19 (range 5-30). Nine patients had symptoms consistent with a total anterior circulation infarction. There were three cases of partial anterior circulation infarct, two lacunar infarcts and one posterior circulation infarct. One patient received two doses of rt-PA on separate occasions. Fourteen of the fifteen recipients survived until day 3. Among this group, median NIH score fell by six points (SD 7.9). Eleven of fifteen recipients survived until month 3. Median NIH score in this group fell by 11 points (SD 7.6). Two patients died of cerebral oedema complicating massive cerebral infarction, two of hypostatic bronchopneumonia. Bleeding complications consisted of one systemic episode and one asymptomatic intracranial haemorrhage. At three months, four patients had Barthel scores of 95-100, four had scores of 55-90 and three were below 50.

Our experience with rt-PA in a UK stroke unit is consistent with that of the NINDS study investigators. No inferences concerning efficacy can be made due to the small numbers involved; however despite initial severity of stroke, 47% of those treated were alive and at home at three months.

Chapter eight contains a synopsis of the work presented, together with a brief discussion of the results in the context of our current understanding of stroke disease. Future directions of study arising from the research are identified and discussed.

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Chapter One

Introduction

1.01 The burden of stroke

Stroke is among the most important causes of severe disability in the Western world[1] and is the third leading cause of death (after coronary heart disease and cancer) in most Western countries. The condition has a profound social and economic effect on the affected patient, the relatives of the patient, and on society as a whole. In the United Kingdom, stroke disease consumes a substantial proportion of health service resources directly, and is responsible for many less obvious costs. Although stroke patients account for only 2% of hospital discharges in Scotland, 4.6% of all Scottish health service costs are used in the care of patients with stroke,[2] and patients with stroke account for 7% of Scottish hospital bed-days. The considerable financial costs to the health service are relatively easy to measure; however the enormous burden borne by society as a whole is more difficult to estimate. Approximately one quarter of patients with stroke are below retirement age, leading to direct loss of income and to indirect costs such as reduced national productivity. In addition, much of the long-term care afforded to patients disabled by stroke disease is provided by family members, and to date little effort has been made to define the level and specific nature of the burden borne by relatives of stroke patients.[3] Recognition of the cost of stroke to society has raised the profile of stroke care in the United Kingdom, and emphasised the importance of the delivery of quality stroke care in a cost-effective manner.[4] In 1992 improved stroke care was identified as a priority in the improvement of the Health of the Nation.[5] A number of subsequent studies have demonstrated the benefit of organised hospital-based stroke services upon stroke mortality[6,7,8] and long-term dependency.[8] Unfortunately the heterogeneous

nature of the stroke units studied made it difficult to identify which facets of the package of care provided by stroke units were of particular benefit. It is likely that expert physical rehabilitation and secondary prevention with antiplatelet agents, anticoagulants and carotid endarterectomy contributed to the better outcome enjoyed by patients treated in dedicated stroke units; however it is likely that other less tangible benefits of the multidisciplinary approach exist and have yet to be identified.

The challenge of reducing the burden of stroke to society is huge, and will not be met solely by improving the initial and secondary preventative care offered to patients with stroke. Primary stroke prevention is potentially the most effective method of reducing overall stroke-related morbidity and mortality, and the importance of risk factor intervention such as blood pressure control, smoking cessation, and appropriate management of predisposing cardiac disease has become apparent over recent years. Although both primary and secondary preventative strategies have improved, the progressive rise in the number of elderly people in Western countries leads to the conclusion that the burden of stroke will remain a significant health and socio-economic problem for many years to come. The following chapters will consider a number of metabolic, radiological and therapeutic factors which may influence outcome following ischaemic stroke. The purpose of this introductory chapter is to provide a background to the main themes discussed later in the thesis.

1.02 Stroke diagnosis

The diagnosis of stroke usually begins with identification of a focal neurological deficit. The initial encounter with the patient should achieve several aims:

i) Confirmation of the vascular nature of the lesion.

ii) Identification of the location and extent of the cerebral damage.

iii) Institution of appropriate treatment to minimise ongoing cerebral damage.

iv) An indication of the likely aetiology of the event.

Subsequent interaction with the patient should involve:

i) Institution of appropriate secondary preventative measures to prevent subsequent vascular events.

ii) Recognition of existing co-morbidity and anticipation of development of known complications of stroke disease such as deep venous thrombosis.

The main differential diagnoses of cerebrovascular disease are seizure activity and migraine. In the elderly, systemic infections such as urinary tract sepsis may present with neurological symptoms which may be confused with cerebral ischaemia. Sufficient information to exclude these possibilities may usually be obtained from a detailed history from the patient or witnesses. Further alternative causes of focal neurological deficit include cerebral space occupying lesion such as subdural haemorrhage or tumour, metabolic disturbance such as hypo or hyperglycaemia, infections such as encephalitis or cerebral abscess, hypertensive encephalopathy, syncope, or

demyelination. In many cases further clarification with brain imaging or blood tests may be required before the vascular nature of the symptoms can be identified.

Having identified cerebrovascular disease as the likely underlying pathology, attention should be turned to the nature of the vascular lesion. It is important to distinguish cerebral haemorrhage from cerebral infarction at an early stage, due to profound differences in the management of these conditions. Many scales designed to allow differentiation of ischaemic stroke from cerebral haemorrhage on clinical grounds have been proposed;[9,10,11] however none has been shown to be reliable, and in some cases may be frankly misleading.[12] Brain imaging is necessary reliably to exclude cerebral haemorrhage. Conventional x-ray computed tomography will reliably detect intracerebral haemorrhage within two weeks of stroke; however if more than two weeks has elapsed since ictus, involutional change within the intracerebral haematoma makes haemorrhage hard to distinguish from infarct using this imaging modality, and magnetic resonance imaging may be required.

In most cases, clinical examination allows reasonable estimation of the location and extent of cerebral damage following stroke. The majority of symptoms arising as a result of stroke can be attributed to dysfunction of an identifiable area of brain, for example monoparesis following infarction of a discrete area of motor cortex. Other symptoms arising as a result of stroke may be less easy to categorise: isolated dysarthria, vertigo, confusion and

cognitive disturbance such as amnesia may all reflect focal neurological dysfunction but may also occur as a result of a more diffuse cerebral insult. Localisation of cerebral infarction may yield information concerning the underlying aetiology of the stroke, which in turn may influence further management. Patients with ischaemic stroke in the carotid territory and ipsilateral severe carotid artery stenosis should be considered for carotid endarterectomy. Patients with infarction in watershed areas should be investigated for underlying causes of severe hypotension such as paroxysmal dysrhythmias or silent myocardial infarction. The aetiological subtypes of cerebral infarction are discussed in more detail below.

1.03 Brain embolism

Embolism has been identified as a frequent cause of brain infarction. Many potential sources of emboli are known, however it is often difficult to implicate a particular embolic source following stroke as several potential predisposing factors may coexist within the same patient.

Cardiac embolism

Occlusion of cerebral arteries arising as a result of embolus from the heart was recognised more than a century ago.[13] however until recently it was thought to be an uncommon cause of stroke. This view was revised when several large pathological studies suggested that cerebral embolism was the likely mechanism in approximately one quarter of all ischaemic strokes, with this proportion rising slightly in young patients with stroke.[14,15]

Many cardiac problems can give rise to cerebral embolism. The commonest and most important disorder is atrial fibrillation (AF). Chronic AF of rheumatic or non-rheumatic origin is associated with a five fold increased risk of ischaemic stroke, independent of other factors such as age, sex and hypertension.[16] Other disturbances of cardiac rhythm such as the sick sinus syndrome also significantly increase the risk of ischaemic stroke. These rhythm disturbances are common, particularly in the elderly population,[16] and are easy to detect. Judicious use of anticoagulants in this group represents an effective means of primary stroke prevention.

In addition to disturbances of cardiac rhythm, structural cardiac disease may predispose to stroke. Left ventricular mural thrombus may arise as a result of recent myocardial infarction, left ventricular dilatation or left ventricular aneurysm. In autopsy series, left ventricular mural thrombus has been demonstrated in 20-60% of patients with myocardial infarction (MI), although systemic embolisation is recognised in only 3-10% of post-MI patients.

Paradoxical cerebral emboli may occur, usually through a patent foramen ovale (PFO). Screening of asymptomatic individuals reveals that physiological shunting through a PFO may occur in 10-20% of those screened, however this proportion rises to 40-50% when young patients with stroke are studied. Cardiac valvular disease may lead to cardioembolic stroke either as a result of valve-related rhythm disturbance or following thrombus formation on damaged native or prosthetic valves. Mitral valve prolapse (MVP) is the commonest form of adult valvular disease. Its significance in relation to cardiogenic

cerebral embolism remains unclear. It appears that the majority of patients with MVP are at no increased risk of ischaemic stroke unless there is associated endothelial damage or fibrinous deposits on the affected valve.

Artery-to-artery embolism

Many sites on the arterial tree may give rise to emboli which occlude vessels distal to the source. Emboli arising in this fashion may be composed of fibrin / platelet aggregates, cholesterol crystals, calcified particles or other debris from ruptured atherosclerotic plaques, or red fibrin dependent thrombus. Identification of artery-to-artery embolism is difficult, hence its true frequency is difficult to estimate.[17] Embolic sources within both the intracranial and extracranial portions of the internal carotid artery, the vertebral arteries, the basilar artery and the aortic arch have been reported.[17,18,19]

The clinical characteristics of embolic brain infarction are variable; however a number of features are helpful in the identification of embolic disease. The abrupt onset of symptoms which do not fluctuate and can be attributed to superficial or cortical cerebral infarction is highly suggestive of an embolic mechanism. A history of previous cerebral infarction in a different arterial territory, history of cardiac disease or abnormal findings on cardiac examination would all strengthen the suspicion of a central cardiac or arterial source.

Clinical suspicion of an embolic aetiology should lead to an exhaustive search for a potential source. In addition to full neurological and cardiovascular

examination and brain imaging, electrocardiography (including prolonged monitoring), carotid ultrasound, echocardiography and cerebral angiography should be considered. Trans-oesophageal echocardiography should be considered in patients with normal trans-thoracic echocardiography in whom a suspicion of central embolism remains as this technique affords superior views of the left atrium, mitral valve, atrial septum and ascending aorta.

Therapeutic intervention will depend upon the results of these investigations. Patients with cardiogenic embolism should be considered for systemic anticoagulation, while those with evidence of artery-to-artery embolism should be offered antiplatelet therapy and (if appropriate) surgical endarterectomy.

1.04 Large artery occlusive disease

The extracranial internal carotid artery supplies the ipsilateral cerebral hemisphere including most of the deep subcortical areas. Atherosclerosis involving this vessel most commonly affects the first 2 centimetres immediately distal to its origin. Atheroma at this site may lead to ischaemic stroke either due to artery-to-artery embolism or as a result of haemodynamic changes distal to the narrowed vessel. In the absence of adequate collateral circulation, complete occlusion of the internal carotid artery leads to extensive hemispheric infarction. When the vessel remains patent but is sufficiently narrowed to reduce cerebral perfusion, infarction of the "watershed" areas that lie at the boundaries of the territories of the major cerebral arteries may occur. Watershed infarction may occur in the anterior border zone (between the anterior and middle cerebral arteries) or the posterior border zone (between

middle and posterior cerebral arteries). Anterior watershed infarction is characterised by crural or brachial paresis with relative sparing of the face. If the dominant hemisphere is affected, transcortical motor aphasia may occur. Posterior watershed infarction presents with relatively little motor involvement, hemianopia or lower quadrantanopia, transcortical sensory aphasia and parietal lobe signs such as neglect in a non-dominant hemisphere lesion or dyscalculia if the dominant hemisphere is affected.

The intracranial part of the internal carotid artery may also be affected by atheromatous change, particularly in the cavernous portion at the carotid siphon. As with more proximal disease of the internal carotid artery, ischaemic stroke may occur as a result of propagation of emboli from plaque at this site, or as a result of haemodynamic compromise distal to the narrowed vessel. The features of stroke arising as a result of intracranial internal carotid artery disease are indistinguishable from those due to extracranial disease. It is possible that the significance of intracranial carotid disease as a cause of stroke has been underestimated due to the inaccessibility of this portion of the artery to non-invasive investigation.[20]

The middle cerebral artery is the larger of the two terminal branches of the internal carotid artery. This vessel supplies most of the lateral aspect of the hemisphere via its superior and inferior branches. It also supplies the basal ganglia and internal capsule through a number of small lenticulostriate end arteries which arise from the main stem. Infarction confined to the territory of the middle cerebral artery usually occurs as a result of cerebral embolism

from a proximal source, although atherosclerosis of the trunk of the vessel may occur and give rise to symptoms, particularly in Black and Asian populations.[21] Clinical features of middle cerebral artery infarction depend upon the site of the occlusion. Lesions proximal to the origin of the lenticulostriate arteries lead to extensive infarction involving cortical and subcortical regions of the affected hemisphere. The resulting clinical syndrome is characterised by dense hemiplegia in a pyramidal distribution, severe global dysphasia or neglect, hemianopia, hemisensory disturbance and conjugate eye deviation towards the side of the infarct. Neurological deficit imparted by more distal middle cerebral artery infarction is less severe and occurs in one of two patterns, depending upon which of the terminal branches is affected. Superior division infarction typically involves motor paresis and non-fluent language disorder with relative sparing of vision and sensation. Inferior division middle cerebral artery ischaemia is characterised by fluent language disorder with relatively little motor deficit.

The anterior cerebral artery is the smaller of the two terminal branches of the internal carotid artery. The artery runs forward from its origin, supplying the anterior and medial aspects of the hemisphere. Several deep branches run laterally to supply the anterior limb of the internal capsule and part of the head of the caudate nucleus. Infarction of the anterior cerebral artery is uncommon and usually due to embolism rather than intrinsic atherosclerosis. Clinical features of anterior cerebral artery occlusion comprise lower limb weakness, urinary incontinence and frontal lobe features such as abulia.

The vertebral arteries, three sets of paired cerebellar arteries, basilar artery and posterior cerebral arteries together comprise the posterior circulation, which supplies the brainstem, cerebellum, thalamus, occipital lobes and medial temporal lobes. Ischaemic damage to these regions may arise as a result of cerebral emboli or atheroma of the posterior circulation. As with the intracranial portion of the internal carotid artery, non-invasive demonstration of arterial disease in the posterior circulation is difficult without easy access to magnetic resonance angiography, hence the condition is under diagnosed and little is known of its natural history.[22] The clinical features of posterior circulation ischaemia vary, depending upon the precise site of the lesion. Basilar artery stenosis may present with recurrent stereotyped episodes of dizziness, dysarthria, hemi- or quadriparesis, diplopia or ataxia. In patients with thrombosis of the basilar artery, these symptoms may have a stuttering onset and may progress to coma.

Cerebellar infarction usually arises as a result of embolic occlusion of one of the cerebellar arteries. It is characterised by dysarthria, ataxia, nystagmus, vomiting and unsteady gait.[23] Thalamic infarction, when unilateral, is manifest as dense hemisensory loss. Medial thalamic lesions may cause cognitive disturbance such as memory loss and agitation. Disturbance of sleep / wake cycle may also occur.[24]

Posterior cerebral artery infarction is usually embolic in nature and is manifest as isolated homonymous hemianopia as a result of unilateral occipital lobe

infarction, or as colour or visual agnosias which may occur when the medial aspect of the temporal lobe is involved.

Diagnosis of posterior circulation ischaemia has been greatly improved with the advent of magnetic resonance imaging (MRI). MR techniques are not limited by the bony artefact which greatly reduces the usefulness of conventional x-ray computed tomography in posterior fossa imaging. The superior spacial resolution of MR techniques allows identification of very small lesions, and magnetic resonance angiography provides a non-invasive means of imaging vessels not amenable to insonation.

1.05 Small artery occlusive disease

Small intracranial arteries arise from two main sources. Arteries arising from the large vessels at the base of the skull penetrate the substance of the brain and supply the deep white matter of the cerebral hemispheres, the basal ganglia and the brainstem. A further group of small arteries originates from the pial arteries which lie in the subarachnoid space, closely related to the cerebral cortex. These penetrating arteries plunge from the hemispheric surface to supply subcortical regions of the hemispheres. Infarction of tissue supplied by these small arteries usually arises as a result of occlusion secondary to disorganisation of the vessel wall with disappearance of vascular smooth muscle, sclerosis, hyalinosis and lipid deposition. The main risk factor for this microangiopathy is hypertension. Following sustained, severe hypertension, fibrinoid necrosis, aneurysmal dilatation and intramural haemorrhage may occur in addition to the lipohyalinosis described above.

Occlusion of these small arteries leads to lacunar infarction.[25] small, well defined areas of cerebral infarction involving the deep white matter of the hemispheres the basal ganglia or brainstem. The clinical features of lacunar infarction are well recognised. Pure motor stroke arises as a result of lacunar infarction involving the corticospinal tract either in the corona radiata, internal capsule, mesencephalon, pons or medulla. Typically the face and limbs are equally affected; the deficit is not associated with other neurological symptoms and in 16-57% of patients may have been preceded by similar, transient symptoms.[25]

Lacunar infarction may affect the sensory pathways alone, causing pure sensory symptoms, or may involve both sensory and motor pathways as they run in apposition in the corona radiata, thalamocapsular area, pons or medulla. Less commonly encountered are the syndromes of ataxic hemiparesis and dysarthria / clumsy hand. Interruption of the cerebello-thalamo-cortico-ponto-cerebellar loop by a lacunar lesion in the corona radiata, internal capsule or pons has been proposed as the underlying mechanism.[27]

Isolated lacunar infarction causes well-recognised lateralised symptoms, however multiple events may lead to more diffuse symptoms, culminating in the "état lacunaire", characterised by the progressive development of abnormal gait ("marche à petits pas") incontinence, dysarthria and cognitive impairment. Only a minority of patients with lacunar stroke will progress to the

état lacunaire. The prognosis following lacunar infarction is better than for other stroke subtypes, with 5-10% mortality at one year.[28]

1.06 Stroke and hypertension

High blood pressure is the single most important risk factor for the development of cerebrovascular disease. In one report,[29] 70% of all patients presenting with acute ischaemic stroke were found to be hypertensive, and a history of chronic hypertension was obtained from approximately half of these patients. The role of antihypertensive treatment in primary prevention of stroke is well established. Meta-analysis[30] of a number of large prospective studies has shown that a reduction in diastolic blood pressure of 5 mmHg confers a 42% reduction in stroke risk. The relationship between blood pressure and incidence of stroke appears to be log-linear,[31] with no apparent threshold level below which no further reduction in stroke risk accrues. It has been suggested that this relationship may be even stronger than initially suggested due to sampling error, as the baseline blood pressure values took no account of random blood pressure fluctuation and errors inherent in blood pressure measurement. This phenomenon, termed "regression dilution bias"[32] results in underestimation of the strength of the relationship between blood pressure and stroke. It has been estimated that correction for regression dilution bias increases the gradient of the relationship by 60%.[33]

The benefits of antihypertensive therapy in patients with established cerebrovascular disease are less well established. Although a similar log-linear relationship between blood pressure and recurrent stroke incidence has

been reported,[34] other studies have postulated a J-shaped curve with progressive increase in the risk of stroke recurrence as diastolic blood pressure falls below 80 mmHg.[36] As a similar relationship has been demonstrated between blood pressure and myocardial infarction,[36] it has been suggested that patients with pathologically low blood pressures due to cardiac disease are responsible for the observed increase in vascular events as blood pressure falls.[37]

Cerebral autoregulation

Hypertension accelerates and contributes to many of the pathophysiological changes in the heart, intracranial and extracranial vasculature which predispose to the development of both ischaemic and haemorrhagic stroke. The mechanisms through which large vessel atheroma, arteriosclerosis, lipohyalinosis of small intracerebral vessels or coronary artery disease may cause stroke have been discussed; however in the hypertensive brain changes in cerebral autoregulation may confer additional risk of cerebral ischaemia. Under normal circumstances, the normotensive individual is able to regulate cerebral perfusion over a broad range of blood pressure. Cerebral blood flow remains constant at approximately 50 millilitres of blood per 100 grams of brain tissue per minute provided that mean arterial blood pressure remains between approximately 60 mmHg and 140 mmHg. In chronically hypertensive individuals, the autoregulatory mechanism becomes "reset" at a higher level of blood pressure. This confers the ability to withstand higher levels of blood pressure without the development of cerebral oedema, however the individual becomes less able to maintain cerebral perfusion when

a hypotensive episode occurs. This cerebral hypoperfusion is manifest clinically as watershed cerebral infarction in a hypertensive patient, occurring at relative / "normal" levels of blood pressure. Cerebral autoregulation has been extensively studied in animals. Normalisation of cerebral autoregulation has been demonstrated in hypertensive animals treated with antihypertensive therapy;[38] to date experiments in humans have failed to demonstrate similar findings.[39]

Primary prevention of stroke

Meta-analysis of prospective blood pressure lowering studies[31] in patients without pre-existing cerebrovascular disease suggests that considerable reduction in stroke risk is achievable with only modest blood pressure reduction. The proportional reduction in stroke risk seen in blood pressure trials with lower blood pressure criteria was comparable to the reduction in stroke risk seen in trials with higher blood pressure criteria. In terms of primary prevention of stroke, similar benefit is derived from treating all patients with relatively mild hypertension as is derived from treating a cohort of more severely affected individuals.

Assuming the group risk reductions seen in this meta-analysis are applicable to individuals, it would appear that the highest ratio of strokes prevented to patients treated will be seen in patients at highest risk. Risk of stroke increases with age; middle-aged hypertensives have an annual stroke risk of 0.5%,[30] whereas in the hypertensive elderly population this risk is trebled.[40]

Extrapolation from the conclusions of the meta-analysis,[31] over a treatment period of 10 years, the number needed to treat to prevent one stroke in the hypertensive elderly population would be 17, compared to 50 in the hypertensive middle-aged population.

Primary prevention of stroke in the elderly

The potential benefit of intervention to lower blood pressure in the hypertensive elderly population has been demonstrated in a number of large prospective studies. Reductions in vascular death and non-fatal coronary and cerebrovascular events have been reported in patients over 60 years of age with isolated systolic hypertension.[41] In this study, relative reduction in stroke risk was 36% indicating that in the elderly population treatment of isolated systolic hypertension confers benefit.

The epidemiological evidence linking hypertension to stroke recurrence is less clear than that linking hypertension to first stroke. Retrospective analysis of a large cohort of patients over a 30 year period failed to demonstrate a significant relationship between stroke recurrence and level of blood pressure.[42] A smaller prospective study of blood pressure reduction in normotensives following TIA or non-disabling ischaemic stroke demonstrated a direct and continuous relationship between blood pressure and the secondary incidence of stroke.[43] This and three other studies[31,44] examined the effect of intervention to lower blood pressure on risk of stroke recurrence in normotensives or hypertensives. Due to a combination of small numbers of participants and modest blood pressure reduction in the treated groups, all

four studies were inconclusive. Meta-analysis of these studies suggests reduction of stroke risk of 19%, but with wide confidence intervals (95% C I – 4 9%, + 11%)

More definitive evidence of the effect of blood pressure lowering on risk of stroke recurrence is expected when the perindopril protection against recurrent stroke study (PROGRESS)[45] reports in 2001. This study has recruited 6000 normotensive or mildly hypertensive stroke survivors and is following them up over five years. It aims to investigate the efficacy of a thiazide diuretic and / or an angiotensin converting enzyme inhibitor as secondary prevention of stroke in this population. Although the reduction in blood pressure achieved in the treated group is rather lower than initially predicted (12.1 / 4.8 mmHg in the dual therapy cohort), the event rate is higher than expected and thus the PROGRESS study should be adequately powered to assess the benefit of blood pressure lowering treatment in patients with cerebrovascular disease.

Blood pressure lowering acutely after stroke

There are a number of theoretical reasons why intervention to lower elevated blood pressure in the context of acute ischaemic stroke may have a deleterious effect. It is known that focal impairment of cerebral autoregulation may occur in the acute phase after stroke.[46] In the affected area, reduction in cerebral perfusion pressure may not be met with reflex arterial vasodilatation, resulting in cerebral hypoperfusion. This further ischaemic insult may increase cerebral damage and worsen outcome. A further potential hazard of blood

pressure lowering in acute stroke is the risk of reducing pressure across an undiagnosed stenotic lesion involving a large extra- or intracranial artery. Some evidence from prospective studies supports the theoretical concerns described above. The INWEST[47] study of intravenous nimodipine infusion after acute stroke demonstrated an association between early fall in blood pressure and worse clinical outcome. In a small pilot study of the ion channel blocker Lifarizine[48] a fall in blood pressure was associated with poor outcome in elderly female recipients of the drug. It should be emphasised that the deleterious effect of blood pressure lowering early following stroke does not apply to patients with severe or accelerated-phase hypertension. In these patients very high levels of blood pressure are associated with headache, seizure activity and focal neurological disturbance. Intervention to lower blood pressure over a matter of hours should be instituted to prevent worsening of cerebral oedema and to reduce the risk of potentially fatal hypertensive complications. Cautious intervention to lower blood pressure may also be warranted in stroke patients with co-existing illness such as severe cardiac failure, dissecting arterial aneurysm or acute myocardial infarction.

Blood pressure management following stroke is further confounded by physiological blood pressure changes which occur in patients admitted to hospital with stroke. An initial blood pressure rise and subsequent fall over 48-72 hours is well recognised.[49] This fluctuation appears to be related to psychosocial and physical stress associated with hospital admission, hence is not indicative of antecedent hypertension. Decisions regarding long term blood pressure treatment should therefore be deferred until a consistent

series of blood pressure readings have been obtained and evidence of pre-existing hypertension such as left ventricular hypertrophy on ECG or evidence of end organ hypertensive damage has been sought. The observed fluctuation in blood pressure tends to be more pronounced in patients with intracranial haemorrhage,[59] and as elevated blood pressure in these patients may predispose to further intracerebral bleeding, a more aggressive approach to the treatment of elevated blood pressure is probably justified although little prospective evidence is available.

There is relatively little published evidence to guide clinicians in their choice of antihypertensive agent in both acute and long term intervention to lower blood pressure after stroke. In the rare situations where urgent early intervention to lower blood pressure is required, the choice of agent may be restricted due to swallowing difficulty or other coexisting conditions such as severe heart failure. If parenteral therapy is necessary, short acting agents such as nitrates should be infused as this obviates any risk of prolonged and precipitous fall in blood pressure.

Due to the paucity of prospective evidence concerning secondary preventative blood pressure treatment, the choice of antihypertensive agent is usually determined by co-existing conditions and by the preference of the clinician. There is some evidence to suggest that, unlike older antihypertensive drugs, angiotensin converting enzyme inhibitors can reduce blood pressure without lowering cerebral blood flow[57] in patients with cerebrovascular disease. These agents have also been shown to reduce the

incidence of recurrent myocardial infarction in humans,[52] and cause regression of atheromatous plaque in primates.[53]

It is clear that intervention to reduce elevated blood pressure reduces risk of first stroke, and evidence is emerging that risk of stroke recurrence can be reduced with satisfactory blood pressure control. The optimal management of blood pressure acutely after stroke or in normotensive stroke survivors remains unclear. Further research is required in order to define which patients to treat, when to institute therapy and which agents to use.

1.07 Blood glucose after stroke

The significance of blood glucose concentration in the context of experimental neuronal ischaemia has long been appreciated. The role of hyperglycaemia in patients with acute stroke is less well defined. Animal work suggests that the detrimental effects of hyperglycaemia are mediated by a number of mechanisms and vary depending on the site and extent of cerebral ischaemic insult. There is some early evidence from clinical trials to suggest that observations in the animal model may be involved in human stroke. Published evidence of the effects of hyperglycaemia in both the animal model and humans will be briefly reviewed

Focal cerebral ischaemia

In the experimental model, hyperglycaemia during focal cerebral ischaemia exerts a variety of effects on infarct size, tissue pH and functional outcome. Investigation of the influence of serum glucose concentration on infarct size

following permanent feline MCA occlusion by de Courten-Meyers revealed significant increases in infarct size and mortality affecting animals rendered hyperglycaemic with IV infusion of 10% dextrose prior to onset of ischaemia.[54] Similar increases in infarct size were observed in hyperglycaemic rats (blood glucose 119-360 mg/dl) following thread occlusion of the MCA.[55] The authors of both of these studies concluded that serum glucose concentration at the time of large cerebral vessel occlusion influences stroke outcome. Chemical analysis of ischaemic neuronal tissue in monkeys has suggested that hyperglycaemia reduces pH of affected brain, possibly due to local accumulation of lactic acid.[56] Tissue acidosis has also been observed in the rat model of hyperglycaemic cerebral ischaemia and in addition, significantly elevated levels of cortical lactate have been consistently noted in hyperglycaemic animals.[57] This finding has been attributed to a relatively greater availability of substrate for anaerobic glycolysis within the ischaemic brain tissue of these animals. As lactic acid is thought to be a significant mediator in the final common pathway of ischaemic cell death (possibly due to promotion of free-radical formation), the authors postulated that increased glucose delivery to the ischaemic penumbra was the most likely mechanism for their observations.

The picture is somewhat clouded by the failure of attempts to reproduce the findings of de Courten-Meyers' group.[58] Zasslow et al suggested that hyperglycaemia decreased the extent and severity of acute neuronal ischaemic changes in the cat model of permanent MCA occlusion. The discrepancy between these two cat studies may in part be explained by minor

methodological differences (survival times of six hours[54] compared with two weeks[58]) and by relative differences in blood glucose concentration of 33%. Similar discrepancies were found when the studies in rats were repeated.[59] These differences were found not to be attributable to methodological differences and led to further, more detailed study of other factors which may have influenced outcome. It was found that hyperglycaemia adversely affects outcome in models of focal thrombotic infarction when the affected tissue receives collateral supply. Hyperglycaemia prior to and during infarction involving non-anastomosing arterial beds does not increase and may reduce the eventual infarct volume.[60]

The protective effect of hyperglycaemia following end-arterial infarction may possibly be explained by the stabilising effect it has on the membranes of neurones in the peri-infarct region. High levels of blood glucose prevent the spontaneous potassium depolarisation usually observed in the peri-infarct neurones of normoglycaemic animals.[60] This in turn prevents the acceleration in cellular metabolism that has been associated with selective neuronal death seen in the normoglycaemic model.[61]

Hyperglycaemia following human stroke

The effect of hyperglycaemia upon outcome following human stroke has been the subject of much interest, however, as in the experimental model, conflicting results have frequently been reported. Retrospective studies in America have revealed relatively poorer neurologic outcome following stroke in patients with diabetes.[62] In later studies of non-diabetic stroke patients,[63] an

association between hyperglycaemia at presentation, larger infarct volume (as assessed by CT) and poorer neurological outcome was observed. Other authors have noted an apparent association between raised blood glucose and incidence of cerebral oedema following stroke, independent of the presence of pre-existing diabetes.[64]

Studies of cerebral metabolism immediately following stroke using positron emission tomography[65] have revealed an association between hyperglycaemia and altered regional glucose metabolism within the brain, including lower glucose consumption within the ischaemic area, and asymmetries in glucose consumption when homologous areas of each hemisphere are studied. The significance of these findings has yet to be established.

There has been much debate over the interpretation of the results of these clinical studies. The elevated levels of blood glucose may have caused the larger infarcts and hence worsened outcome or may simply reflect a greater stress response as result of a relatively larger ischaemic insult. In an attempt to clarify whether the relationship between elevated blood glucose and poor outcome was one of cause or effect, a number of studies attempted to measure other markers of the physiological "stress response" in the context of acute cerebral ischaemia. Myers et al[66] reported abnormally high levels of sympathetic activity in patients with cerebral infarction. The degree of activation of the "stress response" was assessed by analysing plasma levels of noradrenaline, adrenaline and dopamine. Levels of these hormones were

significantly elevated in patients with cerebral infarction compared to control subjects. Patients with transient ischaemic attack had intermediate levels of plasma catecholamines. The investigators found no correlation between catecholamine level and stroke severity, type or location, nor did the differences correlate with age, blood pressure or heart rate. Other studies [67,68] have revealed significant activation of the cortisol axis following stroke. Higher levels of urinary cortisol excretion were found in patients with acute stroke than control subjects. The degree of activation of the cortisol axis correlated with elevation of plasma catecholamine levels. High cortisol excretion was also significantly associated with poorer functional outcome in a discriminant analysis. Significantly, these and other[69] studies did not show an association between plasma catecholamine levels or urinary cortisol and blood glucose concentration, irrespective of the presence or absence of diabetes. The authors concluded that hyperglycaemia observed during the acute phase of stroke cannot be explained by increased stress.

This finding has been substantiated by a recent long term follow up study.[70] The effect of hyperglycaemia on stroke mortality and morbidity was studied by assessing its effect on outcome after adjusting for known prognostic factors. Plasma glucose concentration above 8 mmol/lit predicted poor prognosis after correcting for age, stroke severity and stroke subtype. The authors concluded that raised plasma glucose is unlikely to be due to a stress response and should arguably be treated actively. Randomised, prospective trials to examine the effect of intervention to lower blood glucose in hyperglycaemic stroke patients are needed.

Global cerebral ischaemia

Animal models of global cerebral ischaemia (induced cardiac arrest, artificial hypotension, bilateral carotid occlusion) have demonstrated that acute hyperglycaemia prior to global cerebral ischaemia worsens neurologic damage,[71] impairs recovery of ATP,[72] and causes greater delayed hypoperfusion.[73] These findings have been observed in both rats and primates, and are consistently affected by the timing and duration of hyperglycaemia relative to the ischaemic insult. The most deleterious effects were seen when glucose was administered prior to onset of global ischaemia. This significant increase in cerebral injury was observed even when blood glucose had fallen to physiological levels at the time of induction of cerebral ischaemia.

Although these experimental findings have been largely reproducible, other published work[74] has observed differences in cortical lactate concentrations between hyper- and normoglycaemic rat models of global ischaemia with no associated difference in infarct size or functional outcome. Although the methodology of this study has been criticised,[75] the findings remain unexplained.

Glucose handling in the immature brain is known to differ from adult brain. Unsurprisingly, the effect of elevated glucose concentration on ischaemic brain injury has been shown to differ in immature animals.[76] Hyperglycaemic ischaemia in the young rat is associated with neither cortical lactate

accumulation nor alteration of whole-brain glucose metabolism. It has been postulated[77] that the anaerobic threshold is higher in immature brain tissue, hence the role of glucose in the generation of lactate is diminished.

Relatively few studies of hyperglycaemia in the context of global cerebral ischaemia exist, and the results are inconclusive. Premorbid hyperglycaemia may correlate with poorer neurologic recovery post arrest but the evidence is not compelling. Blood glucose measurements taken during cardiopulmonary resuscitation tend to be too closely related to the duration and difficulty of resuscitation to provide useful information on the effect of peri-arrest hyperglycaemia on neurologic recovery.

Glucose, diabetes and stroke

Patients with diabetes are at greater risk of stroke than the non-diabetic population. Following stroke, the prognosis for patients with diabetes is known to be poorer, and their recurrence risk is higher.[78] There are many potential reasons for these observations, and the relative significance of each is unclear. Patients with diabetes have a three-fold increase in the frequency of proliferative lesions characterised by basement membrane thickening and endothelial cell proliferation. An increased prevalence of extracranial carotid atherosclerosis is found within the diabetic population and other peripheral vascular disease is more common. In addition to vascular factors, other contributory factors to the increased incidence of stroke have been postulated. The erythrocytes of diabetic patients are less deformable and

blood viscosity tends to be more variable.[79] These factors may be implicated in the development of small-vessel cerebral infarction.

Studies in humans have suggested that chronic hyperglycaemia is associated with alteration of cerebral haemodynamics, specifically reduced MCA velocities and some loss of cerebral autoregulatory function.[80] Glucose handling within the brains of diabetics differs from that of non-diabetic individuals. The precise nature of the metabolic abnormality has yet to be identified; however it is known that neuronal glucose uptake in diabetic patients is similar to non-diabetics, and it is assumed that the observed differences in glucose metabolism are due to altered intracellular processes.

Similar abnormalities of intracranial haemodynamics have been observed in animal models of chronic hyperglycaemia,[81] however these findings are less consistent than published human data, and some authors have reported increased cerebral blood flow in the chronically hyperglycaemic rat.[81] Discrepancies are also found when studies of cerebral autoregulation and brain glucose metabolism in diabetic animals are examined. Relative failure[82] and preservation[83] of cerebral autoregulatory mechanisms have been reported.

The observed differences between the findings of human and animal studies may relate to the imperfect nature of the animal model of diabetes. It is apparent that the effects of chronic diabetes on the cerebrovascular system are many and complex, and are dependent upon a host of other factors such

as age, blood pressure, lipid status and cigarette consumption. Further research into this area is underway.

Intervention with insulin during cerebral ischaemia

As mentioned above, little is known of the effect of insulin upon outcome in hyperglycaemic patients with ischaemic stroke. Theoretically, intervention to lower blood glucose in ischaemic stroke should reduce the supply of substrate for anaerobic glycolysis within penumbral brain tissue, hence reducing neuronal death and improving eventual outcome. In addition to its hypoglycaemic effect, it has been suggested that insulin may act centrally at insulin-like growth factor receptors expressed within the CNS, exerting a neuromodulatory effect. This postulated neuroprotective effect independent of hypoglycaemic action is under investigation and some supportive evidence has been published[84] however the effect is thought unlikely to achieve clinical significance. In addition to its local effects following focal cerebral ischaemia, insulin may exert some beneficial systemic and haemodynamic effects. Intense insulin treatment may affect platelet function,[85] correct disturbed lipoprotein patterns[86] and decrease plasma activity of plasminogen activator inhibitor,[87] which has been shown to be elevated in diabetic individuals. When considering the potential benefit of insulin administration, it is important to be aware of the detrimental effects of excessive hypoglycaemia on the brain. These are described elsewhere.[88]

Comparison of hypo- and hyperglycaemic animals in experimental models of cerebral ischaemia has revealed higher intracellular pH, more favourable

cellular energy state and lower intracellular lactate levels in hypoglycaemic animals.[89]

Comparative studies observing the effects of pre-ischaemic glucose manipulation provide conclusive evidence of the beneficial effect of mild hypoglycaemia immediately prior to the ischaemic insult. Of more clinical relevance are the studies examining the potential benefit of glycaemic manipulation following ischaemia. In the rat model, intervention with insulin to reduce elevated levels of blood sugar to euglycaemic levels following the induction of ischaemia has been shown to reduce consequent cerebral ischaemic damage, improve functional outcome and effectively negate the detrimental effect of pre-ischaemic hyperglycaemia.[90] As expected, excessive postischaemic hypoglycaemia in the animal model is associated with increased seizure activity and poorer outcome.

Summary

The relationship between hyperglycaemia and outcome following cerebral ischaemia is not yet fully understood. There is reasonable evidence to suggest that hyperglycaemia immediately following cerebral ischaemia is not entirely attributable to a stress response and that it predicts a relatively poor prognosis. Experimental data would suggest that the effect of hyperglycaemia on neuronal tissue may depend upon the degree of collateral blood supply to the affected area. Although there is a paucity of prospective data at present, current evidence suggests that intervention to reduce high levels of blood

sugar while avoiding hypoglycaemia is justified until prospective data to guide clinicians become available.

1.08 Imaging of acute ischaemic stroke with x-ray computed tomography.

As discussed above, the diagnosis of acute stroke is based upon clinical observation and exclusion of other potential causes of focal neurological deficit such as seizure activity or metabolic disturbance. Unequivocal confirmation of the diagnosis and precise lesion localisation often require imaging with x-ray computed tomography or magnetic resonance scanning. At present, CT scanning is performed more commonly than MR scanning in patients with acute ischaemic stroke. Although magnetic resonance techniques are known to be more sensitive and specific than CT imaging in the context of cerebral ischaemia, CT is more widely available and can reliably exclude haemorrhage or tumour. Computed tomography of the brain is performed in the axial plane. A rotating x-ray tube projects a collimated beam of x-rays through the head parallel to the plane joining the lateral canthus to the external auditory meatus. The beam is attenuated to a variable extent, depending upon the density of the materials through which it passes. Beam attenuation is measured using a detector, and the information is processed by computer to generate an image of the brain in the transaxial plane. Images are usually acquired at 5 millimetre intervals through the posterior fossa and at 10 millimetre intervals through the cerebral hemispheres although these intervals may be changed for certain applications.

In the first hours following ischaemic stroke, the pathophysiological processes which characterise acute ischaemic injury do not change the electron density of the affected tissue, hence during this time up to 60% of CT scans are normal.[91] Early changes in CT appearances after stroke are subtle and may be easily missed by the inexperienced. Parenchymal changes reported on CT imaging in the acute phase of stroke include mild sulcal effacement, blurring of the internal capsule, loss of the insular "ribbon" and of grey / white matter discrimination at the boundary of cortex and subcortex.[91,92] Following ischaemic stroke in middle cerebral artery territory, the "Dense MCA" sign occurs with a reported frequency of 35-50%.[93] The hyperintensity of the proximal part of the middle cerebral artery is thought to represent lodged embolus or thrombus in this vessel. Similar appearances may be seen in the posterior fossa following basilar artery occlusion. Although a useful indicator of ischaemic middle cerebral artery stroke or basilar thrombosis in patients with no parenchymal change on CT, non-occlusive chronic calcification of atheromatous deposits in the middle cerebral or basilar artery can give rise to similar appearances.[93]

1.09 Basic principles of magnetic resonance imaging

Magnetic resonance imaging (MRI) is a technique which provides high quality images of structures within the human body. It is based upon the principles of nuclear magnetic resonance, and was developed from techniques used by scientists to obtain information about the physical and chemical properties of molecules. The first successful experiments with nuclear magnetic resonance

were performed in 1946 by Bloch and Purcell. Their work led to the development of NMR spectroscopy as a powerful analytical tool in chemistry and biology. The potential of NMR to obtain images of the intact human body was recognised approximately thirty years later.

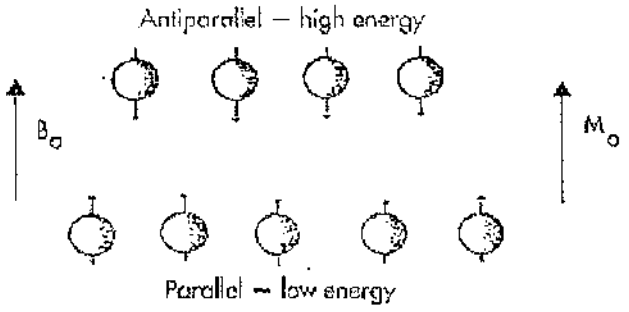
MRI was initially able only to provide tomographic images; however as the technique was further developed, more complex functional and volumetric studies became possible and the clinical utility of MRI became apparent. The technique has a wide range of clinical applications, and is increasingly used by clinicians from many specialities.

Magnetic resonance physics

The physical principles which underlie magnetic resonance imaging are complex. Images are generated by detection of signals emanating from the hydrogen nuclei which abound within the human body. The nucleus of a hydrogen atom comprises a single proton. This proton possesses a fundamental property of nature called spin, which causes it to behave like a very small magnet with a north and a south pole. When the proton is placed in an external magnetic field, it aligns itself along the field in one of two possible configurations, which possess differing energy states (figure 1.1). When the poles of the protons are aligned north-south south-north (N-S S-N) relative to the magnetic field, the energy state is low, and when the configuration is N-N S-S the energy state is high.

Figure 1.1 – Proton alignment in external magnetic field

B_o represents external magnetic field. M_o represents net magnetisation vector.



A proton in a magnetic field can move from low to high energy states by absorbing a photon. In order that this occurs, the photon must possess exactly the same amount of energy as the energy difference between the two states. The energy of a photon (E) can be calculated by the equation

$$E = h\nu$$

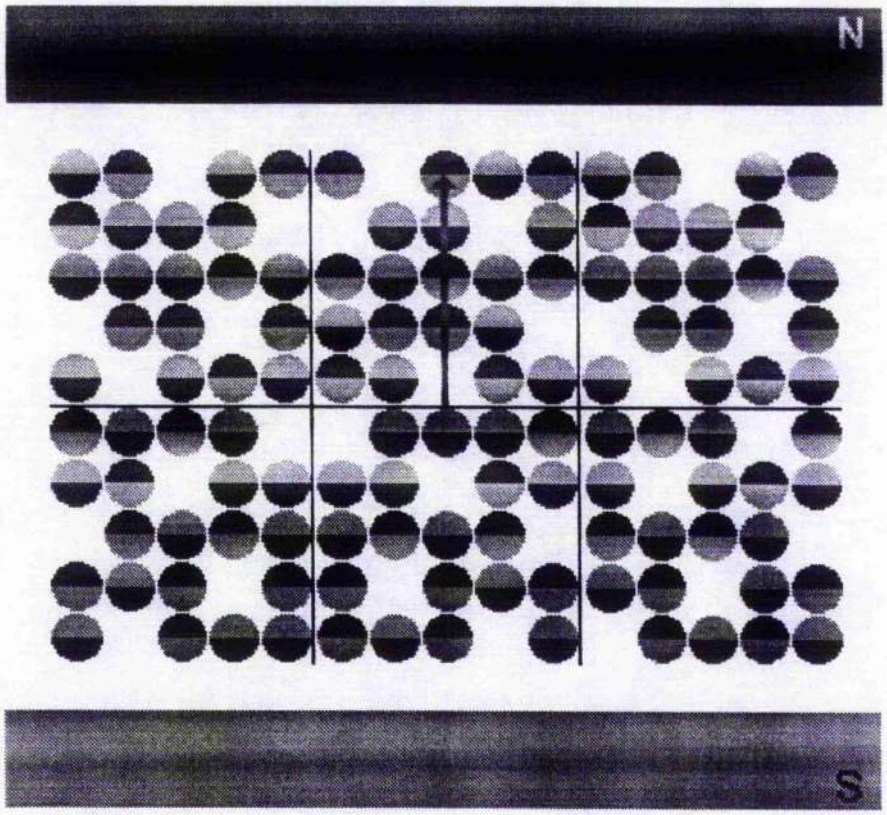
where ν is the frequency of the photon and h is Plank's constant ($h = 6.624 \times 10^{-34}$). In MRI, ν is called the resonance frequency or the Larmor frequency and lies within the radiofrequency range (15-80 MHz).

On a macroscopic scale, the sum of all of the magnetic fields generated by protons within an externally applied magnetic field can be considered. This net magnetic vector is proportional to (N^+-N^-) where N^+ represents the total number of protons in a high energy configuration and N^- represents those in a low energy configuration (figure 1.2).

At equilibrium, the position of the net magnetisation vector lies along the direction of the applied magnetic field and is called the equilibrium magnetisation M_0. It is possible to change the net magnetisation vector by exposing the nuclear spin system to energy of a frequency equal to the energy difference between the spin states. If enough energy is put into the system, it is possible to saturate it and maximise the net magnetisation vector. Once the exposure to external energy has ceased, the system returns to equilibrium. The time constant that governs the return to equilibrium is called the spin lattice relaxation time (T_1). The equation governing this behaviour as a function of the time (t) after displacement is

Figure 1.2 - Net magnetisation vector

Each circle represents one proton aligned in the magnetic field in one of the two possible configurations. The net magnetisation vector for one of the six blocks of protons shown is represented by an arrow.



$$M_z = M_0(1-e^{-t/T1})$$

where M_z represents the degree of displacement from the equilibrium position (figure 1.3).

In addition to displacement along the axis of the externally-applied magnetic field, the protons may be displaced in the plane perpendicular to the magnetic field. This is called transverse magnetisation. As with displacement along the axis of the magnetic field, the degree of displacement rapidly returns to the equilibrium value, giving off radiofrequency energy as it does so. The time constant which describes this behaviour is called the spin-spin relaxation time, T_2. The spin-spin relaxation time is always less than the spin-lattice relaxation time. It is calculated using the formula

$$M_{XY}=M_{XYO}e^{-t/T2}$$

Where M_{xy} is the magnitude of the displacement in the plane perpendicular to the magnetic field (figure 1.4).

In summary, the spin-spin relaxation time (T_2) is the time taken to reduce the transverse magnetisation by a factor of e. The spin lattice (T_1) relaxation time is the time taken to reduce the longitudinal magnetisation by a factor of e. The energy released by the protons as they return to the equilibrium position is recorded as an electrical signal which is subsequently processed and an image is generated.

By altering the parameters of the magnetic field gradients and radiofrequency pulses, images can be preferentially sensitised to particular structures and

Figure 1.3 – Relationship of spin lattice relaxation time (T1) to net magnetisation vector.

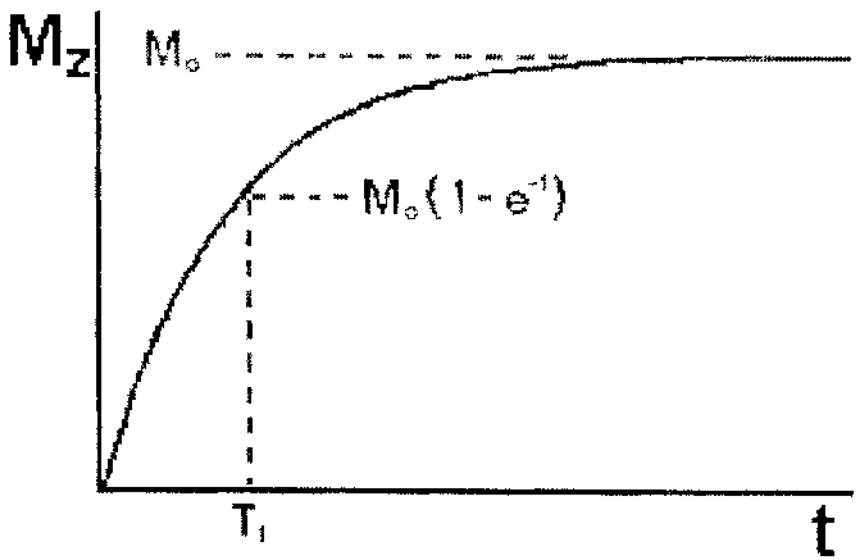
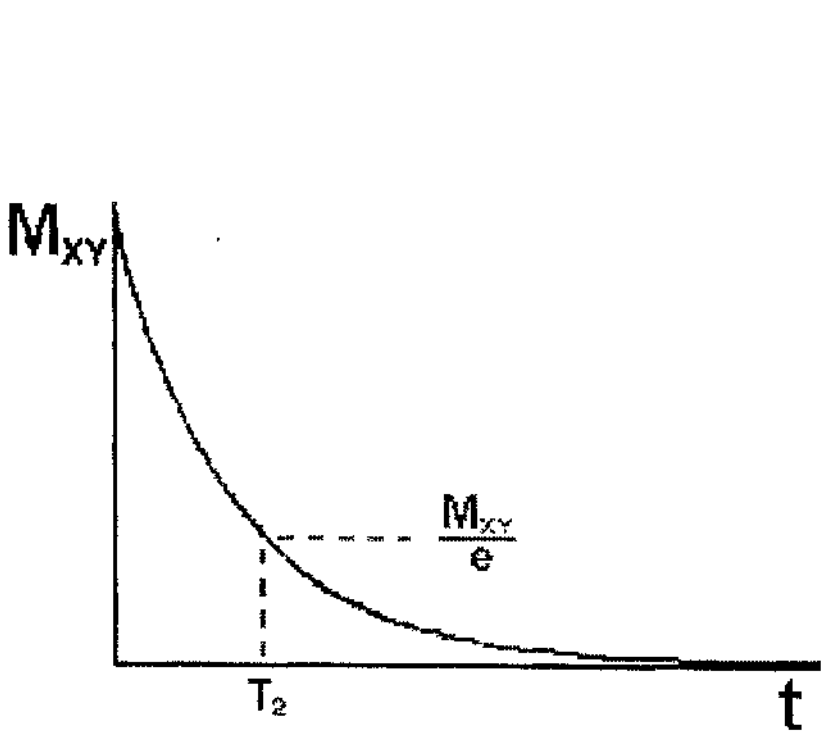


Figure 1.4 – Relation of spin-spin relaxation time (T2) to transverse magnetisation.



processes within the body. More detailed discussion of the physical principles of magnetic resonance is outwith the scope of this introduction.

Magnetic resonance imaging of ischaemic stroke

In the context of acute cerebral ischaemia, magnetic resonance imaging holds a number of advantages over other radiological modalities and is rapidly becoming the investigation of choice in patients with stroke. The main goals of neuroimaging in acute stroke have been

- To confirm a vascular cause of neurologic dysfunction.
- To provide radiological evidence of tissue ischaemia and define the vascular territories involved.
- To exclude haemorrhage or haematoma in anticipation of thrombolytic treatment.

Using techniques discussed later, MRI is capable of achieving these goals at an earlier stage in the natural history of a stroke event than other imaging modalities. Lesions in areas poorly - seen using CT or SPECT are more easily visualised on MRI due to greater anatomical detail and reduced artefact. The ability to diagnose and classify stroke early in the course of the illness allows the provision of early and appropriate thrombolytic therapy, may assist in the development and assessment of neuroprotective agents and will assume an increasingly important role as novel therapies for acute stroke enter clinical practice.

Although magnetic resonance imaging has a number of distinct advantages over other techniques, certain practical considerations limit its applicability. As

the generation of an image requires the application of strong magnetic gradients, metallic implants such as permanent pacemakers or foreign bodies represent an absolute contraindication to magnetic resonance imaging. As most magnetic resonance imaging examinations involve placing the patient in a confined and noisy environment, patients with claustrophobia or anxiety disorders are frequently unable to tolerate the scanning process. Many magnetic resonance sequences are exquisitely sensitive to patient movement and require adequate patient co-operation for the generation of clinically-useful images. Acutely-ill patients such as those with ischaemic stroke may be too restless or too agitated to undergo magnetic resonance scanning. Approximately one quarter to one third of all stroke patients referred for MR investigation are unable to be scanned for these reasons.

Detection of cerebral ischaemia by magnetic resonance depends upon the evaluation of four features.

- Vascular flow abnormalities.
- Mass effect.
- Parenchymal signal characteristics.
- Parenchymal enhancement by contrast material.

Assessment of vascular flow abnormalities using magnetic resonance

Cerebral ischaemia develops when cerebral blood flow drops from its normal level of approximately 55 ml/100g/min to 20-25 ml/100g/min. Normal flow through cerebral arteries is seen on MR images as an area of greatly reduced signal intensity within the lumen of the artery (the flow void phenomenon).

When flow through an artery slows or the artery is occluded, this flow void disappears. On T_1 weighted images the flow void is replaced by an area of iso- or hyperintense signal with respect to surrounding brain parenchyma Vascular flow abnormalities can be more easily seen when contrast media are administered. The arterial enhancement seen in areas of compromised blood flow is likely to be a manifestation of low velocity flow associated with vascular stenosis, occlusion or mass effect from cerebral swelling.

A more specific means of imaging the cerebral vasculature is magnetic resonance angiography. This technique provides detailed images of the arteries and veins without the need for the introduction of contrast agents. There are two general types of MRA

- Time of flight angiography.
- Phase contrast angiography.

Time of flight angiography uses slice selective 90 degree and 180 degree pulses. These pulses cause proton spin in opposing directions. In the absence of flow, no signal is seen as no protons experience both the 90 and 180-degree pulses. In the presence of flow, blood from the 90-degree plane flows into the 180-degree plane and an echo is produced from which an image is generated.

Phase contrast angiography is more complex. This method uses alternating bipolar pulses to impart spin first in one direction then the other. Stationary protons experience no net effect from this process as the opposite spins negate each other. Moving protons (i.e. those within flowing blood) experience

an additive effect from sequential pulses and give off a radiofrequency signal which is converted into an image.

Parenchymal signal changes

The remaining three factors which affect the detection of cerebral ischaemia by magnetic resonance are biological processes which take longer to develop and may not be apparent using conventional MR in the hyperacute phase of stroke. Parenchymal signal change and mass effect arise as a result of degradation of the blood - brain barrier, changes in tissue water content and loss of autoregulation.

Critically-ischaemic cerebral tissue experiences metabolic and cell-membrane dysfunction within the first few hours of the onset of ischaemia. This results in the accumulation of intracellular water, sodium, lactate and calcium with subsequent swelling of the affected area - a phenomenon termed cytotoxic oedema. This process is not well seen using conventional T2 weighted techniques and may resolve if normal perfusion is restored. Should ischaemia continue for six hours or more, a predominantly extracellular accumulation of water occurs which has been attributed to shift of macromolecules and water into the interstitium following breakdown of the blood brain barrier. This process, termed vasogenic oedema, may be visualised on conventional T_2 MRI sequences as an area of high signal. The radiologic change may not be visible until eight hours post onset of ischaemia.

It is clear that conventional T_1, T_2 and proton density images cannot be relied upon to demonstrate cerebral lesions until at least six hours after the ischaemic insult. Magnetic resonance angiography reveals occlusive lesions in large vessels acutely; however this technique is technically demanding, yields no information on brain parenchyma and the long acquisition time required to obtain a satisfactory image may lead to substantial movement artefact when used in the investigation of patients in the hyperacute phase of stroke. The ability to visualise and quantify early ischaemic cerebral injury is essential if design of clinical trials of neuroprotective therapies is to be optimised. Looking further into the future, early and accurate diagnosis of cerebral ischaemia will permit rational and judicious use of neuroprotective and thrombolytic treatment.

One of the theories underlying the development of neuroprotective agents is the concept of the ischaemic penumbra. This is an area of abnormally low perfusion thought to surround a central core of acutely-infarcted cerebral tissue. It is postulated that although this area is at risk of imminent infarction, restoration of blood flow could result in reversal of the ischaemic change and restoration of normal function. The ability to visualise this penumbra would permit accurate, early diagnosis, provide prognostic information early in the course of the illness and facilitate a rational decision regarding treatment in the hyperacute phase. As mentioned above, acutely-ischaemic brain cells swell and accumulate water due to membrane dysfunction. Regions affected by this process are not seen using conventional MR sequences or CT. Diffusion weighted magnetic resonance imaging is a relatively new technique

that detects the altered movement of water molecules through acutely-ischaemic brain tissue and hence allows localisation and quantification of hyperacute ischaemic lesions

Diffusion refers to the movement of molecules from areas of high concentration to areas of low concentration. This process occurs due to the random Brownian motion of molecules in solution. The rate of diffusion is dependent on the kinetic energy of the molecules and so is affected by temperature. The movement of water molecules through the brain is not truly random due to the presence of cell membranes and so the term "apparent" diffusion is used. The apparent diffusion of water molecules through brain parenchyma is detected with the addition of two strong gradient pulses to an otherwise standard pulse sequence. The first dephases the proton spins and the second completely rephases those protons which have not moved in position. When movement of protons occurs in the time between the two pulses, the spins are not perfectly in phase and signal attenuation results. The degree of signal attenuation depends upon both the degree of proton displacement and the strength of the diffusion weighting.

Although diffusion-weighted imaging has been used in the investigation of many human diseases, the technique has had greatest impact in the imaging of stroke. Prior to the development of diffusion imaging, identification, localisation and quantification of ischaemic cerebral damage using non-contrast magnetic resonance techniques was only reliable at least six hours after onset of symptoms. Using diffusion techniques, areas of infarction can

be visualised within minutes of onset of experimental stroke in cats,[94] and lesions as small as 4mm in diameter have been visualised within 100 minutes post ictus in humans.[95] On diffusion-weighted images, regions of ischaemic cerebral damage are visualised as areas of abnormally high signal intensity. The increase in signal represents a region of reduced net diffusion, or low apparent diffusion coefficient. The abnormalities observed on diffusion-weighted images usually disappear over 5-14 days as the pathophysiological processes to which the technique is sensitive resolve. The use of this technique has recently been validated as a tool for the early investigation of acute stroke in humans. The intensity of signal (hence the size of decrease in apparent diffusion coefficient) would appear to correlate directly with the degree of cerebral ischaemia and inversely with the likelihood of reversal of the radiologic change if the ischaemic area is reperfused. The clinical significance of this observation is yet to be established.

Caution is required in the interpretation of these images as other factors may cause hyperintense lesions on a diffusion-weighted image. The most commonly encountered of these is diffusion anisotropy, which is due to unilateral diffusion of water molecules along the "path of least resistance" perpendicular to the white matter tracts. Early studies with DWI in patients with stroke used long image acquisition times which lead to some movement artefact in a substantial proportion of images. The development of faster diffusion weighted sequences (using, for example, echo-planar imaging techniques which enable very rapid image acquisition) has gone some way towards obviating this problem.

Assessment of cerebral perfusion using magnetic resonance

In addition to the visualisation and quantification of cerebral ischaemia, cerebral perfusion may also be assessed using recently developed techniques. Perfusion studies may be performed by "tagging" inflowing blood using RF pulses or by injecting a bolus of contrast medium and following its progress through the cerebral circulation. The former method has the advantage of being non-invasive. Blood vessels leading into the slice to be studied are subjected to alternating radiofrequency pulses which, when summated, result in no net radiofrequency signal. Blood flowing into the designated slice is met with a different echo-planar radiofrequency pulse which alters the spin of the protons within it, leading to the generation of a radiofrequency signal. The intensity of the signal is proportional to the amount of blood flow and as a result a map of cerebral perfusion can be constructed. This technique is known as Echo Planar Imaging Signal Targeting with Alternating Radiofrequency ("EPISTAR").

Dynamic contrast-enhanced perfusion imaging involves tracking a bolus of intravenous contrast agent as it passes through the cerebral circulation. Rapid sequential imaging sequences are used to track the progress of a bolus of contrast agent through the cerebral circulation. The contrast material causes loss of signal intensity, which can be plotted over time. This data can be analysed to calculate cerebral blood volume, mean transit time (MTT) of the contrast agent and hence relative cerebral blood flow, which is a function of these two parameters. True quantitative analysis of blood flow is possible, as

the amount by which the contrast agent alters signal intensity is directly proportional to the blood volume of the area being studied.

The ability to quantify cerebral blood flow using magnetic resonance has potentially useful clinical applications in the context of acute cerebral ischaemia. The technique is sensitive to relatively minor degrees of hypoperfusion and does not rely upon the development of cellular ischaemic dysfunction. It has been postulated that the use of perfusion studies in conjunction with diffusion weighted and conventional MR sequences will permit a comprehensive assessment of the cerebrovascular status of the patient with acute stroke. Conventional T2-weighted images will exclude haemorrhage and may reveal an ischaemic lesion between 6 and 8 hours old. Hyperacute diffusion weighted and perfusion sequences may confirm clinical diagnosis of ischaemic stroke in patients with clinical signs but normal T2 scans. These novel techniques may also provide some prognostic information and consequently permit rational decisions to be made concerning neuroprotective and thrombolytic treatment at the earliest possible stage of the event. The ability to exclude haemorrhage, confirm ischaemia and possibly predict potential infarct progression using only one imaging modality represents a significant advance in the investigation of acute stroke. This should be of great benefit in the development and eventual clinical implementation of neuroprotective treatment.

1.10 Thrombolytic therapy for acute ischaemic stroke

In the United States thrombolysis is now established management for selected patients with acute ischaemic stroke presenting within 3 hours of onset of symptoms. As yet thrombolysis is unlicensed in Europe but is undergoing further clinical evaluation and assessment.

Thromboiysis has been used for a number of years in the management for patients with myocardial infarction and its efficacy has been demonstrated by a number of large randomised, placebo controlled trials. Thrombolysis leads to reperfusion of vessels occluded by coronary atherothrombosis which in turn reduces mortality and preserves left ventricular function.

Most strokes are atherothrombotic or embolic (85%) while the remainder are haemorrhagic. It was hoped that the same 'open artery hypothesis' would hold true for patients with ischaemic or non-haemorrhagic stroke i.e. that early reperfusion would improve both mortality and functional outcome. In patients with stroke it appears that a central core of densely ischaemic tissue is surrounded by a 'penumbral' zone of potentially salvageable tissue which is amenable to either neuroprotective or reperfusion therapy. The potential risks of thrombolysis are predictable from what we already know of the pathophysiology of stroke. Reperfusion injury may occur when toxic free radicals and inflammatory cells enter an ischaemic area following spontaneous reperfusion and haemorrhagic transformation is commonly seen in patients with large infarcts. Certain groups of patients could be thought of as at higher risk of secondary intracerebral haemorrhage as a result of

thrombolysis. Elderly patients already have a higher risk of spontaneous intracerebral haemorrhage, as have hypertensives and patients with embolic rather than simple occlusive atherothrombotic strokes. Delayed thrombolysis may simply lead to reperfusion of already irreversibly infarcted tissue and increase the risk of haemorrhage. When trials of thrombolytics were devised these issues were considered and are reflected in the design, entry and exclusion criteria of the studies. All studies incorporated a variable 'time window' outwith which patients were ineligible. all patients underwent CT scanning to exclude intracerebral haemorrhage as the primary pathology and some studies excluded large infarcts and hypertensive patients. The analysis of treatment efficacy is crucial in the design and interpretation of any trial of stroke therapy. Most studies incorporated a validated functional outcome score, for example the Barthel score.[96] These scores describe the patients' ability to perform activities of daily living. More detailed assessments of neurological function were also made using, for example, the Scandinavian Stroke Scale.[97]

Streptokinase studies

The MAST-E (Multicentre Acute Stroke Study- Europe)[98] and ASK (Australian Stroke Study)[99] treated patients with streptokinase (1.5 M units over 1 hour) or placebo within 6 and 4 hours respectively. In both studies there was an increase in early mortality sufficient to result in the safety committees abandoning both studies prior to completion. This correlated with a high incidence of complicating intracerebral haemorrhage. In the MAST-E study

36% (*n=156*) patients treated with streptokinase *Vs* 3% (*n= 154*) placebo patients suffered a fatal haemorrhagic transformation of infarct.

The MAST-I (MAST-Italy) study evaluated treatment with SK and aspirin compared with SK or aspirin alone or placebo.[100] Again the results suggested an increase in early mortality which was more marked in patients receiving both SK and aspirin, while aspirin alone appeared to be safe but of no clear benefit.[101]

Meta-analysis of all streptokinase results failed to reveal factors which predisposed to early mortality. Streptokinase significantly increased early mortality while there may have been a trend towards improved outcome in survivors. To date, no randomised controlled trial has supported the use of intravenous streptokinase as therapy for acute ischaemic stroke. Further studies of streptokinase using different patient selection criteria, lower doses of thrombolytic and prohibition of antiplatelet or anticoagulant co-administration have been proposed however at present no such trial is underway. Although there remains some debate, it is unlikely that streptokinase will have any role in the management of acute ischaemic stroke.

Recombinant tissue plasminogen activator studies

Four major trials have evaluated the role of recombinant tissue plasminogen activator (rt-PA) in patients with acute stroke. There are theoretical reasons why rt-PA may be more effective: first rt-PA is known to be more 'clot specific'

that is it causes less generalised activation of plasminogen, instead having a more selective action at the site of the clot itself; this may make haemorrhagic complications less likely. Second rt-PA is less antigenic than streptokinase and is not associated with a fall in blood pressure during the infusion. This may be of significance as reducing blood pressure immediately after ischaemic stroke has been associated with worse outcome.[47] Finally, angiographic studies in patients with acute myocardial infarction suggest rt-PA is more effective than streptokinase in reperfusing occluded coronary vessels. The ECASS 1 (European Co-operative Acute Stroke Study)[101] the NINDS (National Institute of Neurological Diseases and Stroke rt-PA Stroke Study)[102] and ECASS 2[103] studies evaluated rt-PA at doses of 1.1 mg/kg, 0.9 mg/kg and 0.9 mg/kg respectively. Patients were treated within 6 hours in the ECASS studies and 3 hours in the NINDS study. The Alteplase Thrombolysis for Acute Noninterventional Therapy in Ischaemic Stroke (ATLANTIS) study used a dose of 0.9 mg/kg administered within an initial time window of 6 hours. The time window was altered twice during the study. Although the results of the ATLANTIS study are not yet published, the trial methodology and preliminary data are briefly presented below.

ECASS 1

The higher dose of rt-PA used in the ECASS 1 study is equivalent to that used in the treatment of patients with myocardial infarction. Dose ranging studies have shown an increasing incidence of complicating haematoma formation in patients receiving doses greater than 0.85 mg/kg, and this could have contributed to the increased incidence of haemorrhages seen in the ECASS 1

study. The results of ECASS 1 are also notable for the large proportion of patients excluded from the target population analysis (109 out of 620 randomised). The protocol intended to exclude patients with greater than one third MCA territory stroke on CT, i.e. those with large infarcts. Sixty-six such patients were randomised, constituting the largest group of protocol violators. Survival in protocol violators was significantly worse than for those meeting the entry criteria. While analysis of the target population results suggested an improvement in functional outcome at 3 months (p=0.03) the more rigorous intention to treat analysis was negative. Mortality was non-significantly higher in the rt-PA group in both ITT and TP analysis. In summary the ECASS study did not show enough benefit to justify thrombolysis with rt-PA up to 6 hours after stroke onset although an improvement in functional outcome (Barthel and Rankin score) was seen in the target population analysis of the rt-PA receiving patients.

NINDS

The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study assessed a lower dose of rt-PA (0.9mg/kg) given within three hours of onset of CT confirmed ischaemic stroke. The results suggest an improvement in functional outcome at 3 months, with those in the treated group 30% more likely to have negligible disability at this timepoint. There was no difference in mortality in the actively treated patients although there was an increase in symptomatic intracerebral haemorrhage 7% *Vs* 1% (rt-PA *Vs* plac). Mortality ascribed to intracerebral haemorrhage was 3% Vs 0.3%.

ECASS 2

The aim of the ECASS 2 study was to investigate the safety and efficacy of rt-PA given in a dose of 0 9 mg/kg within 6 hours of onset of ischaemic stroke. Strenuous efforts to improve the quality of CT interpretation were made, and significantly fewer intracranial haemorrhages were seen in comparison with the original ECASS study. The primary outcome measure was the proportion of patients reaching modified Rankin[104] Scores of 0 or 1 (i.e. little or no disability) at three months. The study failed to demonstrate a statistically significant difference between treated and placebo groups; however when the outcome measure was re-dichotomised to define good outcome as mRS of 0-2, a beneficial effect of rt-PA was seen. The authors concluded that rt-PA treatment leads to a clinically relevant improvement in outcome without increased morbidity and mortality despite increased symptomatic haemorrhage, however the failure of the trial to demonstrate efficacy using pre-determined endpoints has led to the widespread interpretation of the trial as neutral. Incorporation of the ECASS 2 data into a meta-analysis of trials of rt-PA in acute stroke reveals a favourable odds ratio of 0.67 (95% CI 0.56 - 0.80) with respect to death and disability.

ATLANTIS

The ATLANTIS study was designed to assess the safety and efficacy of intravenous rt-PA 0.9 mg/kg within 6 hours of ischaemic stroke. Two years after recruitment commenced the time window was changed to 0-5 hours due to adverse interim safety analysis in the 5-6 hour group. After a further three years the time window was further modified (3-5 hours) due to the results of

the NINDS study. With the exception of the time window, entry criteria were very similar to those employed by the NINDS investigators. The ATLANTIS study was terminated prematurely in July 1998 following further interim safety analysis which concluded that "treatment was unlikely to prove beneficial". Although full data have not yet been published, it is understood that the use of intravenous rt-PA beyond three hours after stroke onset is not supported by the ATLANTIS study.

Conclusions

The NINDS study was the first to demonstrate that early intervention can improve the outcome for patients with acute ischaemic stroke. The ECASS 2 study failed to reproduce the convincingly positive results of the NINDS trial; however meta-analysis suggests a beneficial effect of intervention with rt-PA. Thrombolysis with intravenous rt-PA in patients with acute ischaemic stroke can be justified when used judiciously within an experienced centre. Although it has been argued that the potential benefit of streptokinase has yet to be fully investigated,[105] on the basis of current evidence its use cannot be justified in the context of acute ischaemic stroke.

1.11 Rationale for the studies performed

The theme of the studies presented in the following chapters is that outcome following stroke may be influenced or predicted. The work presented addresses several different of facets of stroke management including acute therapy (chapters 3 and 7), brain imaging (chapters 5 and 6) and secondary preventative strategies (chapters 2 and 4). The broad range of factors which

may influence prognosis after stroke has necessitated a variety of investigative approaches both prospective and retrospective.

Chapter two reports a study designed to investigate the effect of intervention with the angiotensin converting enzyme inhibitor perindopril upon blood pressure, cerebral perfusion and renal perfusion in hypertensive stroke patients with carotid artery disease. There is little published evidence to guide clinicians treating such patients: failure to control blood pressure is associated with the risk of recurrent stroke and other morbidity however intervention to lower blood pressure carries the theoretical risk of cerebral hypoperfusion within the territory of a stenosed carotid artery. The angiotensin converting enzyme inhibitor perindopril is known to reduce blood pressure without adversely affecting cerebral perfusion in the context of acute ischaemic stroke; I hypothesised that a similar relationship would be observed following treatment of non acute stroke patients with carotid artery disease. This study is intended to provide a little evidence for clinicians dealing with secondary prevention of stroke in a difficult patient population; further research into the mechanistic basis of the observations described is required.

In chapter three a pilot study of rigorous glycaemic control following ischaemic stroke is examined. Recent evidence has suggested that hyperglycaemia (blood glucose > 8 mmol/L) at the time of ischaemic stroke is associated with poor outcome. A large definitive study to investigate the potential benefit of intervention with insulin in hyperglycaemic acute stroke patients is required; the study presented sought to investigate the safety and feasibility of one

method of glycaemic control in this population. This "proof of concept" study is necessary before a larger trial, powered to provide evidence of treatment efficacy, can be performed.

Chapter four reports a retrospective study designed to further investigate the relationship between lipid profile and outcome after stroke. There has been much debate over recent data which suggested that low serum cholesterol concentration at the time of stroke is associated with poor prognosis. Although triglyceride concentration is an established risk factor for the development of vascular disease, the prognostic significance of triglyceride concentration following stroke has been largely unexplored. The findings of the study reported in chapter four extend our knowledge of the prognostic significance of lipid parameters following stroke however the mechanistic basis for the observations remains elusive and further study will be required before any therapeutic implications can be derived.

The study reported in chapter five addresses the prognostic implications of visible infarction on CT scan following lacunar stroke. Prognosis following lacunar stroke is variable and few reliable indicators of eventual outcome have been identified in this group of patients. Previous studies have examined the effect of infarct size on outcome and the prognostic significance of early changes on CT scan following ischaemic stroke; no published study to date has examined a large cohort of patients with small subcortical cerebral lesions, correcting for other factors known to influence outcome. Early acquisition of prognostic information may facilitate provision of appropriate

rehabilitation and enable more efficient use of scarce resources. The aim of the work presented is to investigate the potential prognostic significance of an investigation that is routinely acquired and easily assessed.

With the advent of acute therapies for acute ischaemic stroke, a greater need for reliable early brain imaging has evolved. As discussed above, interpretation of current conventional brain imaging modalities early after ischaemic stroke is unreliable. The ability of diffusion-weighted magnetic resonance imaging to detect acute ischaemic stroke has been established and the technique is being increasingly applied in clinical practice, however the implementation of diffusion-weighted magnetic resonance imaging has been limited by the relatively high technical specifications required. Chapter six describes the development and implementation of a new diffusion-weighted magnetic resonance sequence which requires significantly lower technical capabilities and can hence be employed by centres hitherto unable to use the technique. The feasibility and clinical utility of the new sequence in the investigation of patients with acute ischaemic stroke are critically assessed, and areas of further potential for development are identified. Although the work presented does not represent an entirely new development in magnetic resonance imaging, it is hoped that the sequence described will extend the range of imaging modalities available to centres with less powerful magnetic resonance imaging machines.

Chapter seven reports the preliminary experience of thrombolytic therapy in a British stroke centre. As intravenous thrombolytic therapy for ischaemic stroke

is not widely used in the UK, little evidence of the safety of the treatment within UK stroke units exists. Significant differences in the provision of acute stroke care between the United States of America and the United Kingdom make interpretation of similar American studies difficult. Despite the relatively small numbers of patients treated it is hoped that this report will contribute to the ongoing debate over the implementation of thrombolytic therapy in British clinical practice.

Chapter Two

The effect of perindopril on cerebral and renal perfusion in stroke patients with carotid disease

2.01 Introduction

Control of high blood pressure is the cornerstone of primary stroke prevention. A reduction in mean arterial blood pressure of 5 mmHg reduces population risk of stroke by approximately 30%.[30] The effect of blood pressure control on secondary prevention of stroke is less well defined, however the benefit is likely to be greatest in stroke patients with underlying carotid or intracranial arterial disease. Although little prospective evidence on the effect of blood pressure control on secondary stroke prevention is available at present, a large multi-centre randomised placebo-controlled study (PROGRESS) designed to clarify the relationship is underway and is due to report in 2001. The PROGRESS study has enrolled 6038 patients with cerebrovascular disease and normotension or mild to moderate hypertension, and is examining the effect of the angiotensin converting enzyme inhibitor perindopril alone or in combination with a thiazide diuretic on secondary incidence of stroke. Perindopril is an ACE inhibitor with a gradual onset of action and a relatively long half-life allowing once daily dosing. It is less likely to cause first-dose hypotension than captopril or enalapril.[106]

Little evidence exists to guide the choice of antihypertensive agent, or the timing of its introduction following the cerebrovascular event. In the first few days after stroke, cerebral autoregulatory mechanisms are deranged, hence blood pressure fluctuations may lead to significant changes in cerebral

perfusion. Some conventional antihypertensive medications may lower cerebral blood flow and worsen outcome after acute ischaemic stroke probably as a result of reduced cerebral perfusion within and adjacent to the affected area.[107] In acute ischaemic stroke patients, clinical trials involving early administration of agents which may lower blood pressure such as nimodipine and lifarizine have shown a correlation between blood pressure reduction and poor clinical outcome.[47,48] Conversely, more recent studies have suggested that no clinically-significant change in cerebral perfusion occurs following administration of angiotensin converting enzyme (ACE) inhibitors to patients early after ischaemic stroke.[51,108] This is thought to be due to increased vessel wall compliance and dilatation of the extracranial vessels.

Poor cerebral perfusion is associated with a greater risk of stroke in patients with carotid disease.[109] Since patients with recent stroke who may have unrecognised carotid disease are already treated with antihypertensive drugs and as they will form an unidentified sub-group of the PROGRESS trial, it is desirable to discover the effects of such treatment on cerebral perfusion. As ACE inhibitors are also known to impair renal function in patients with critical renal artery stenosis and the prevalence of clinically-significant renal artery disease in patients with cerebrovascular disease is unknown, we sought to investigate effect of ACE inhibitors on cerebral perfusion and glomerular filtration in the sub-group of hypertensive stroke patients with moderate to severe carotid stenosis or carotid occlusion.

2.02 Methods

We performed a randomised, double-blind, placebo controlled study of oral perindopril (4 mg daily for 14 days) in patients with a CT or MRI confirmed diagnosis of ischaemic stroke, mild to moderate hypertension (mean arterial blood pressure greater than 100 mmHg) and carotid disease ranging from moderate stenosis to occlusion as assessed by Doppler ultrasonography. Patients whose index stroke had occurred between two weeks and six months prior to randomisation were considered eligible. Three consecutive blood pressure readings were required to fall within the inclusion range over a period of not less than 24 hours prior to randomisation. Patients with potentially operable carotid disease were excluded from the study in order not to delay surgery, as were patients with pre-existing moderate to severe renal impairment (serum creatinine > 200 μmol/lit). Any pre-existing antihypertensive therapy was continued for the duration of the study, as shown in table 2.1.

Ethical approval was obtained from the West Ethical Committee, and patients gave written informed consent to participate. Clinical and neurological assessment using the NIH stroke scale[110] was made prior to study entry and repeated on day 14. Blood pressure was measured semi-automatically using Marquette oscillometric equipment (Marquette Electronics, Wisconsin) pre treatment in triplicate and then hourly in triplicate for the first 8 hours after dosing. Blood pressure was repeated in triplicate at 24 hours and at two weeks.

Total carotid blood flow was calculated from bilateral internal carotid artery insonation (Acuson 128, 5 MHz probe, Acuson, California). Arterial flow was calculated as:

$$[\pi \times (\text{diameter})^2 \times \text{mean velocity}] / 4$$

Details of Doppler methods employed have been published previously.[111] Middle cerebral artery velocity and resistance index were measured by transcranial Doppler (Nicolet EME TC2000, 2 MHz probe, Nicolet, Warwick, UK). Doppler recordings were taken pre-treatment, at 2.5, 5.5, 7.5 and 24 hours post dose and at 2 weeks. Routine safety biochemistry and haematology were collected at entry and at the conclusion of the study. Glomerular filtration rate was measured using chromium (^{51}Cr) radiolabelled ethylene diamine tetraacetic acid (EDTA) prior to dosing and at two weeks. Regional cerebral perfusion was measured using ^{99m}Tc hexamethyl propylene amine oxide single photon emission computed tomography (HMPAO SPECT) pre-dose and at the estimated time of peak drug effect (approximately 6 hours) following first dose of perindopril or placebo. Quantification of cerebral blood flow was obtained using a technique described by Matsuda[112,113] which involves dynamically imaging the bolus injection of ^{99m}Tc HMPAO, and using this as a reference level.

SPECT was undertaken in the department of nuclear medicine of the Western Infirmary using a Picker Prism 2000XP large field of view double-headed gamma camera. On each occasion 500 MBq ^{99m}Tc HMPAO dissolved in 5 millilitres of 0.9% saline was injected as a bolus over one second through an 18-gauge

intravenous catheter, followed by a further flush bolus of 10 millilitres of 0.9% saline. An anterior image of 200x1s frames was then acquired. The patient's head and as much as possible of their heart were included in the field of view. SPECT imaging was undertaken 15 minutes later using a circular orbit and acquiring 60 angles around 360° with 20s per angle. Analysis of the SPECT data was undertaken using a combination of Picker supplied software and some specialised software written in house. The data were reconstructed using a back projection algorithm with a Butterworth filter of order 3.14. Attenuation correction was undertaken using the Chang algorithm. A brain perfusion index (BPI) was calculated from the first pass data as described by Matsuda.[112,113] The unaffected hemisphere was used as the reference region. The reconstructed data were reorientated to transaxial oblique slices (8mm thick) parallel to the orbito-meatal line. Image registration was undertaken, registering the two sets of SPECT data for each of the patients.

An elliptical region of interest was manually fitted to the outer edge of each transaxial oblique slice for each of the sets of data and a set of templates constructed as previously described.[114] Regional cerebral blood flow in ml/100g/min was then calculated in each of the segments of the template using the previously obtained BPI. The difference in regional cerebral blood flow can then be calculated for each of the segments.

Measurement of glomerular filtration rate was achieved using the standard single injection method. A solution of ^{51}Cr-EDTA with a total activity of 1.6 MBq was administered as a bolus injection. Blood samples were drawn at baseline (immediately prior to the injection) and at 2, 3 and 4 hours following the injection. The plasma activity of each of these samples was measured and the rate of decline of plasma activity was used to estimate the glomerular filtration rate.

Results were analysed by repeated measures ANOVA and ANCOVA with the use of Statistica for Windows version 5.1 (Statsoft inc. OK) and Arcus Quickstat Biomedical version 1.2 (Research Solutions, UK). With a sample size of 24 patients we expected to detect a difference in cerebral perfusion (as assessed by carotid Doppler) of 16% with 80% power.

2.03 Results

Tolerance and Safety

A total of 24 patients were recruited into the study. Patients were well matched with regard to age, sex, baseline blood pressure and stroke severity as assessed by NIH score. The clinical details, brain imaging and carotid Doppler findings of patients at entry into the study are shown in Table 2.1.

Patient one in the treated group was withdrawn on day 3 of the study following an episode of acute renal failure which necessitated temporary haemodialysis.

Serum creatinine at baseline had been 170 μmol/L, serum potassium 4.9 mmol/L. Routine safety bloods performed on day 3 revealed rises in both of these parameters to 240 μmol/L and 9.0 mmol/L respectively. Temporary haemodialysis was required to reverse the hyperkalaemia, and biochemical parameters returned to pre-morbid levels within two days. The patient was well at the conclusion of the study; he was normokalaemic with serum creatinine levels consistently between 160 and 175 μmol/L without renal replacement therapy. No other adverse events were encountered. With the exception of the subject mentioned, no significant change in safety bloods was seen. Mean NIH scores improved in both groups over the duration of the study. No difference in improvement between the groups was observed.

Blood Pressure

A significant fall in systolic (p=0.028), mean arterial (p=0.017) and diastolic (p=0.04) blood pressure was observed in perindopril-treated patients compared with the placebo group. Figures 2.1, 2.2 and 2.3 show change in blood pressure expressed as percentage change from baseline for each group. Figure 2.4 shows the absolute values of mean arterial blood pressure at each timepoint. At baseline, blood pressure was 161±17.6 / 86±7 in the perindopril group and 164±17.5 / 85±8.6 in the placebo group. After two weeks' treatment blood pressure was 143±22.6 / 77±13.4 in the perindopril group and 163±16.1 / 86±8 in the placebo group, i.e. a placebo-corrected fall of 17 / 10 mmHg. No associated change in heart rate was seen in either group (figure 2.5).

Internal Carotid Artery Flow

No significant difference in total internal carotid artery flow was seen in the treated group compared to the placebo group (p=0.37). Figure 2.6 shows the percentage change in total internal carotid artery flow at each timepoint for treated and placebo groups. A non-significant trend towards an increase in total internal carotid artery flow was observed on the first dosing day. In the treated group, the 95% confidence interval for percentage change in ICA flow from baseline at 5.5 hours post dose ranged from –3.8% to +41.4%. The equivalent confidence interval in the placebo group ranged from –14.6% to +21%. In treated patients with asymmetrical haemodynamically-significant carotid artery lesions, no significant difference in relative flow through each artery was observed. Absolute values for blood flow through affected and unaffected arteries from such patients in the treated group are given in figures 2.7, 2.8 and 2.9.

Middle Cerebral Artery Velocity and Resistance Index

Successful insonation of both middle cerebral arteries was achieved in seven of the treated patients and eight of the placebo patients. No significant difference in change in middle cerebral artery velocity over time was seen in the treated group compared to the placebo group (p=0.46). These data are shown graphically in figure 2.10. The middle cerebral artery resistance index is a measure of arterial tone and distensibility. No significant difference (p=0.07) in the change of this parameter was observed between the two groups (figure 2.11).

Glomerular Filtration Rate

Figure 2.12 shows the mean glomerular filtration rate (± 2SE) for each group at baseline and at two weeks. No significant change was seen within or between groups (p=0.6). Repeat GFR estimation was not performed on the patient who developed acute renal failure.

SPECT Measurements

22 of 24 patients completed the SPECT protocol. One patient in the treated group (patient 11) failed to complete the SPECT protocol due to claustrophobia; in one placebo recipient (patient 12) the SPECT data images were unsuitable for analysis. An example of one of the SPECT studies obtained, including the template used for analysis, is given in figure 2.13. Analysis of both whole hemisphere and focal brain perfusion indices was undertaken. Figure 2.14 shows the percentage change in whole hemisphere brain perfusion index (BPI) from baseline for both the affected and unaffected hemispheres in both treated and untreated groups. No significant deviation from baseline was observed in either treated or untreated groups (p=0.43). No significant difference between placebo and active groups was observed. Figure 2.15 shows the percentage change in BPI within the template zones which contained or were immediately adjacent to the ischaemic lesion. No significant difference in percentage change in peri-infarct perfusion was seen either between or within groups following perindopril administration (p=0.27).

2.04 Discussion

Perindopril lowered blood pressure without adversely affecting global or regional cerebral perfusion in hypertensive stroke patients with moderately stenosed to occluded carotid arteries. Of twelve patients treated with perindopril, one patient developed acute renal failure with required temporary haemodialysis. The study was not designed to demonstrate any long-term effect on neurological outcome, however no drug-associated neurological deterioration was seen.

In the execution of this study I have adopted some of the methodology of an earlier study[108] designed to investigate the effect of perindopril upon cerebral perfusion early after stroke. The technique used by both studies to assess internal carotid artery blood flow assumes that the lumen of the vessel being studied is cylindrical, i.e. that the cross-sectional area of the vessel can be calculated using its diameter. If the atheromatous lesions in the arteries of the patients studied cause a non-concentric reduction in cross-sectional area of the vessel, errors may be introduced in the calculation of internal carotid artery flow. Although this may lead to inaccuracies of quantitative ICA blood flow measurement, no significant change over time in arterial diameter was seen within ($p=0.7$) or between ($p=0.31$) groups; hence the comparison of the magnitude of change in flow between groups remains valid.

The potential source of inaccuracy may influence the power calculation of the study. The calculation was based upon variability data acquired during earlier studies of patients with normal carotid arteries, and suggested that with a sample size of 24 patients, a 16% difference in internal carotid artery flow could be detected with 80% power. Examination of the variability of the SPECT data acquired during this study allows a further power calculation for future studies. Using variability data from the SPECT scans performed during this study, it has been calculated that a sample size of 24 patients will allow detection of a 6% difference in hemispheric cerebral perfusion (as assessed by SPECT) with 80% power.

Although a previous study has examined the effect of perindopril on global cerebral perfusion early after ischaemic stroke in patients with normal carotid arteries,[108] this study is the first to investigate the effects of perindopril in stroke patients with carotid arterial disease. Control of hypertension in this group of patients is associated with the theoretical risk of reduction in cerebral perfusion distal to the site of a stenotic lesion. Patients with severe carotid arterial disease may have an increased prevalence of atheromatous disease elsewhere. The use of angiotensin converting enzyme inhibitors in patients with renal artery stenosis may lead to adverse consequences, and renal function should be closely monitored following introduction of ACE inhibitor therapy. Our data suggests that perindopril will reduce blood pressure without reduction in global or focal cerebral

perfusion as assessed by Doppler and SPECT respectively, however the mechanistic basis of this observation remains unclear

In rats, angiotensin II receptors within large cerebral arteries are involved in cerebral autoregulation following a rise in blood pressure, and inhibition of ACE resets cerebral autoregulation at a lower level.[115] Studies of the effect of ACE inhibition in healthy volunteers have shown reduced in blood pressure with increased cerebral blood flow (as assessed by transcranial Doppler). The middle cerebral artery flow velocity in the volunteers did not change, however the resistance index increased, suggesting vasoconstriction in the cerebral arterioles.[116] In hypertensive patients without a history of stroke, the ACE inhibitor captopril has been shown to increase cerebral perfusion (measured with Xe^{133} SPECT) while lowering blood pressure. An inverse correlation between magnitude of blood pressure fall and mean cerebral blood flow was observed.[117]

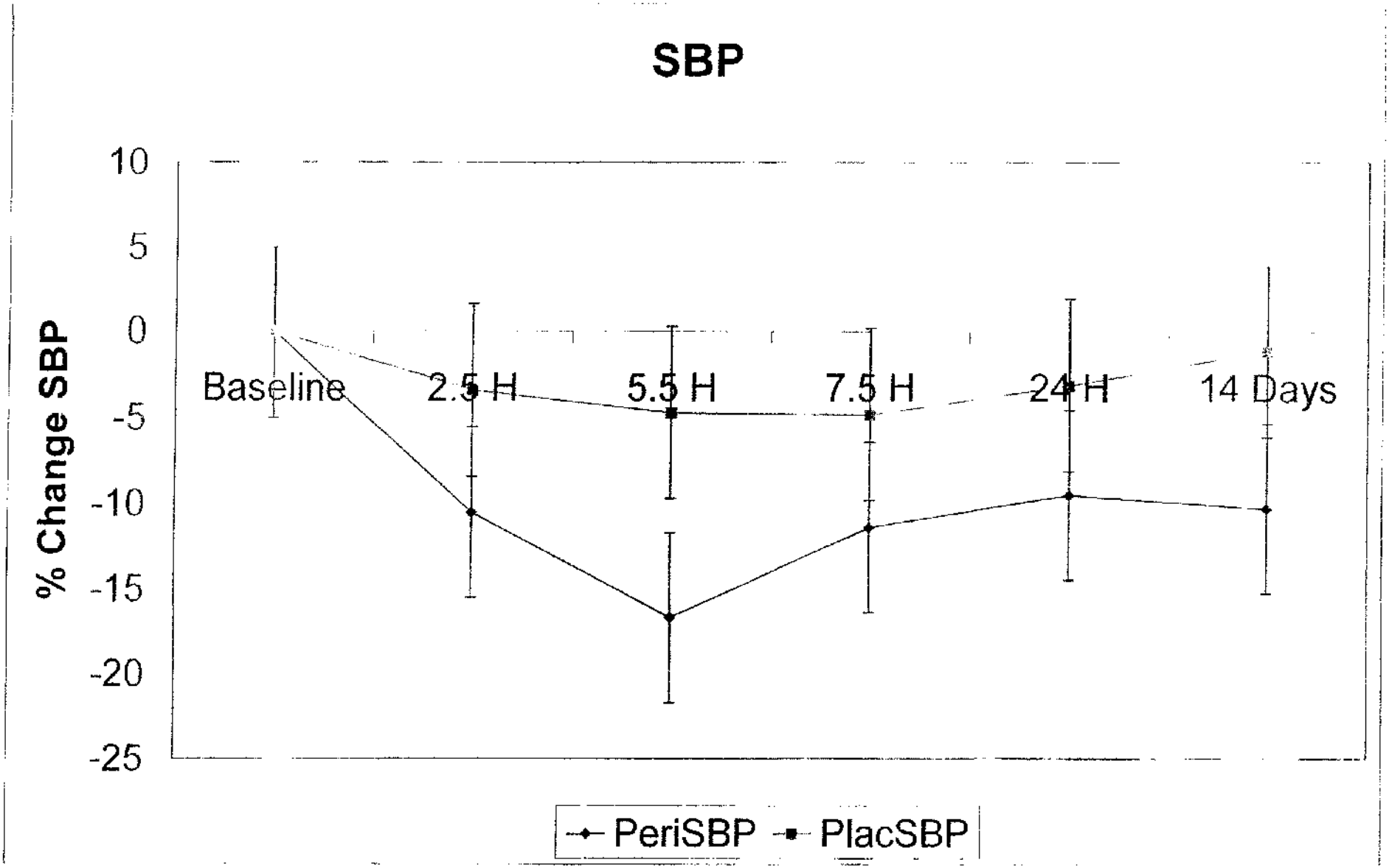
The deleterious effect of ACE inhibition upon glomerular filtration rate in patients with renal artery stenosis is well recognised.[118] Atherosclerosis is a generalised disease and the coexistence of haemodynamically-significant atheroma in the renal, carotid, and lower limb vessels has been documented.[119] In a large case-control study of hypertensive patients, carotid artery ultrasound revealed the prevalence of significant atheroma in 83% of patients with known renovascular hypertension and in 43% of patients with essential hypertension.[120] An overall trend for patients with increasingly severe renal artery disease to have

increasingly severe degrees of carotid disease has also been reported. This correlation lead the authors to conclude that non-invasive screening of carotid arteries should be undertaken in patients with severe renal artery stenosis.[121] The prevalence of severe renal artery stenosis among stroke patients with carotid arterial disease is as yet undefined.

2.05 Conclusion

We conclude that in patients without clinically-significant renal artery stenosis, the angiotensin converting enzyme inhibitor perindopril safely reduces blood pressure without adversely affecting global or focal cerebral perfusion. As renal artery stenosis is known to be associated with haemodynamically significant carotid disease and may be reliably detected using non-invasive techniques,[122] screening of this patient population for the presence of renal artery stenosis should be considered.

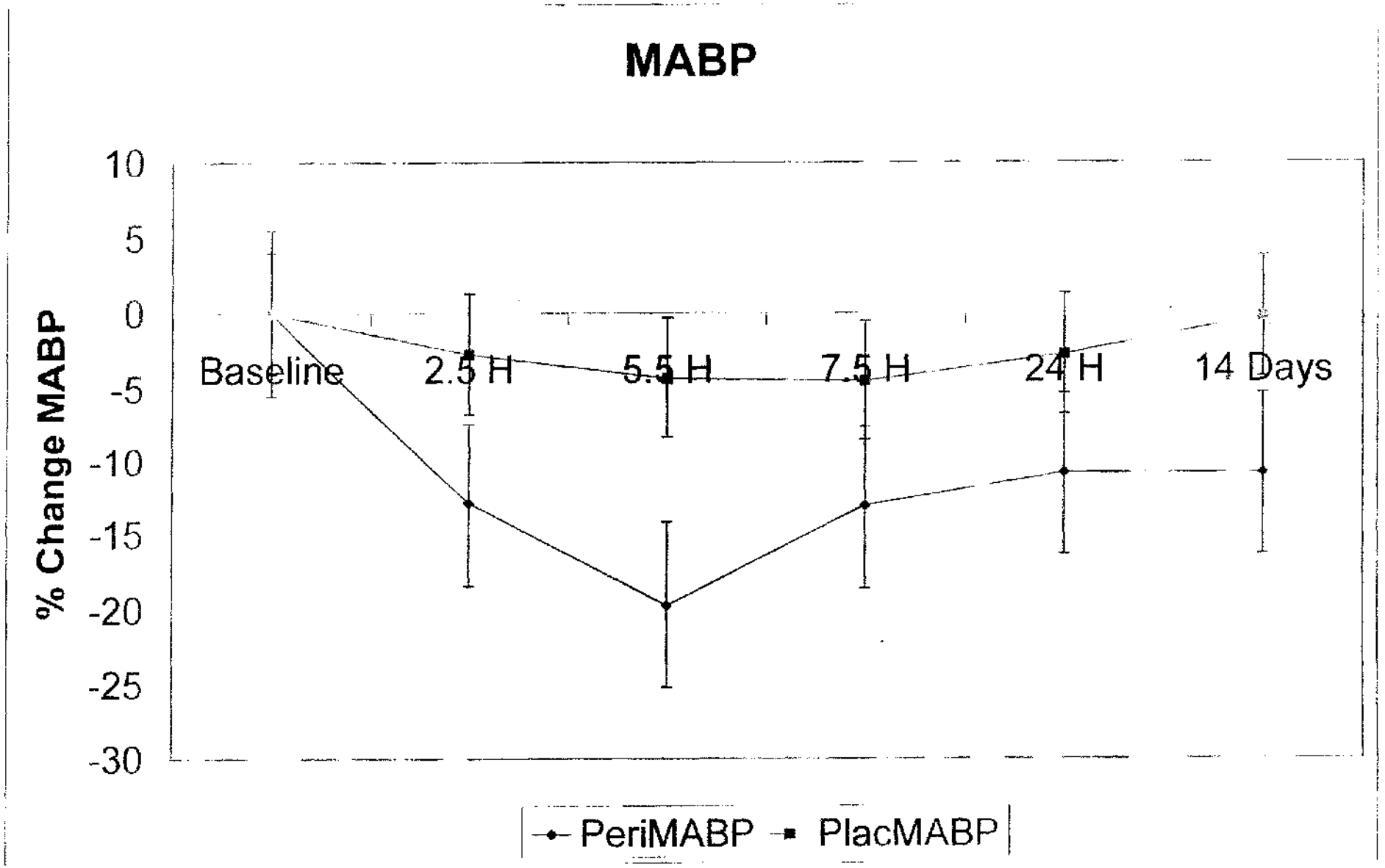
Figure 2.1 Change in systolic BP



Error bars represent +/- 2SE

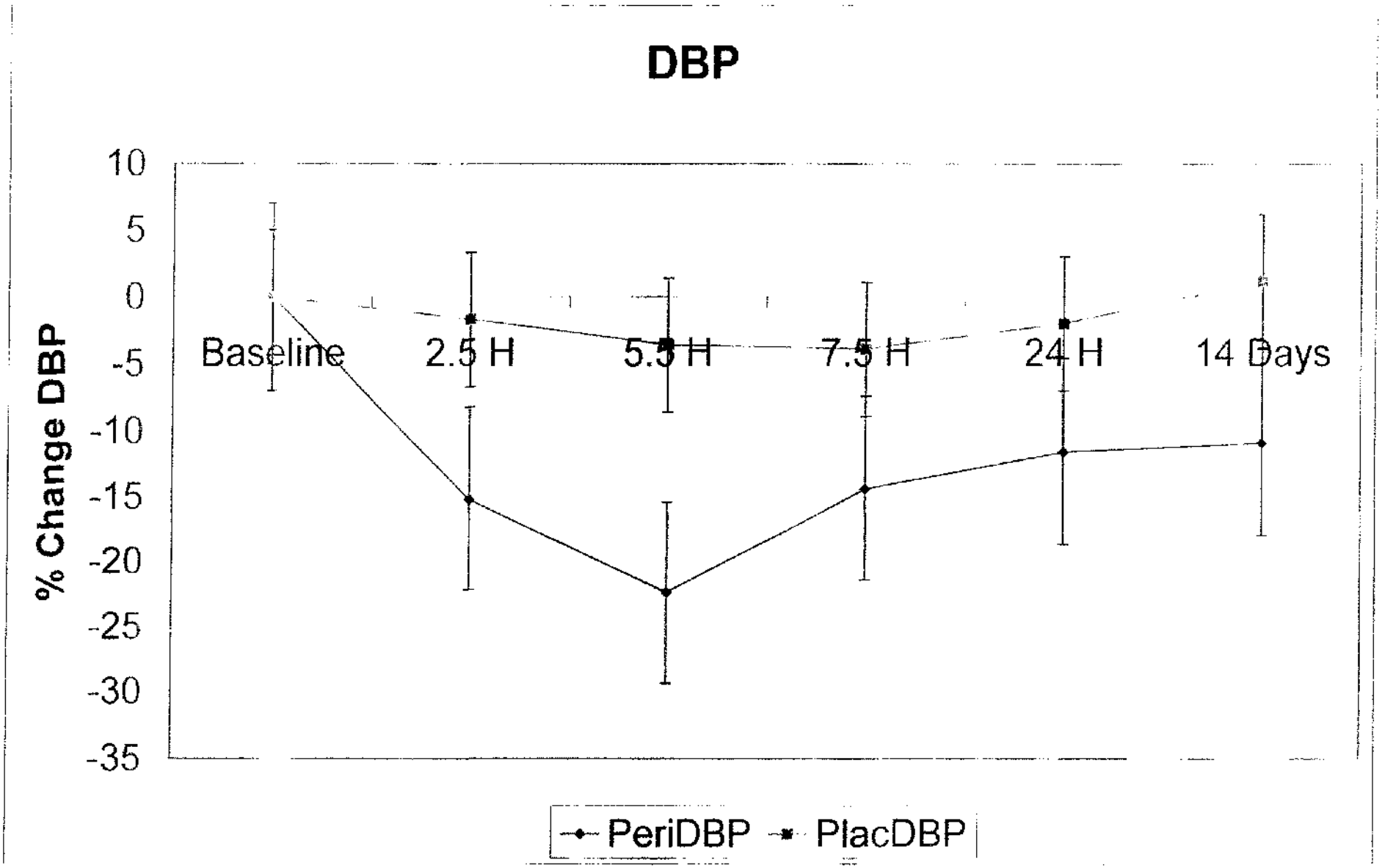
Figure 2.2

Change in MABP



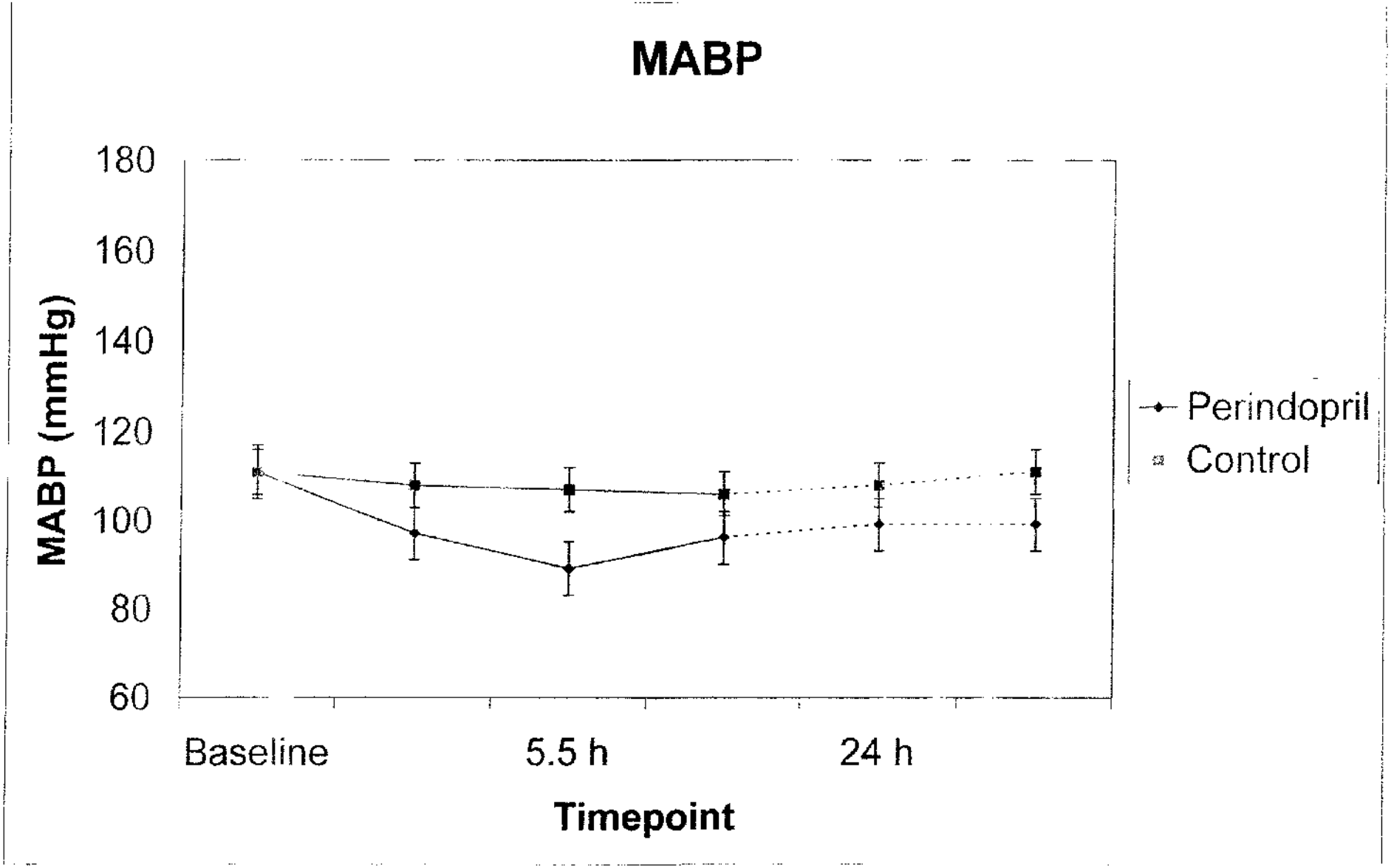
Error bars represent +/- 2SE

Figure 2.3 Change in diastolic BP



Error bars represent +/- 2SE

Figure 2.4 MABP values



Error bars represent +/-2SE

Figure 2.5 Change in heart rate

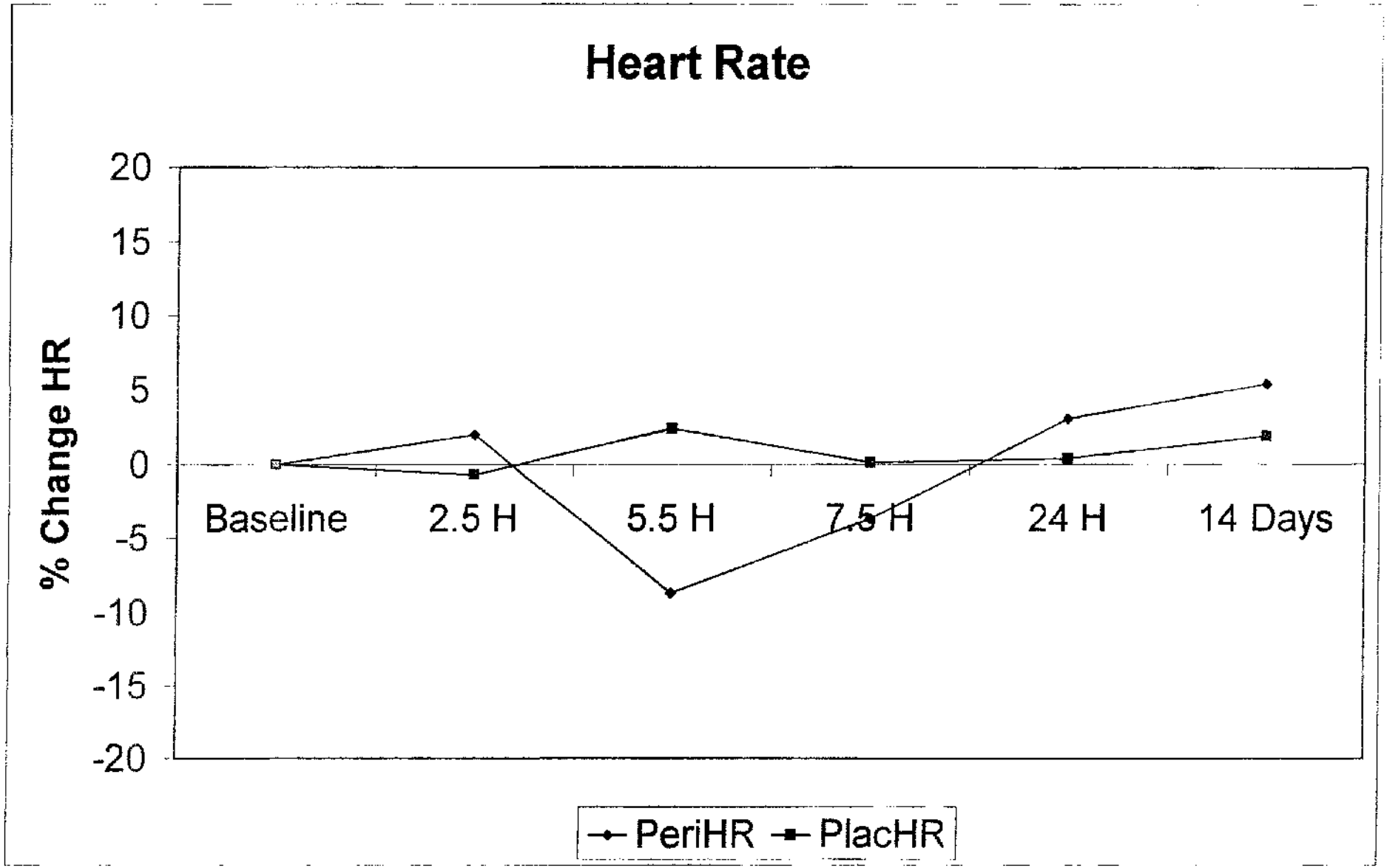
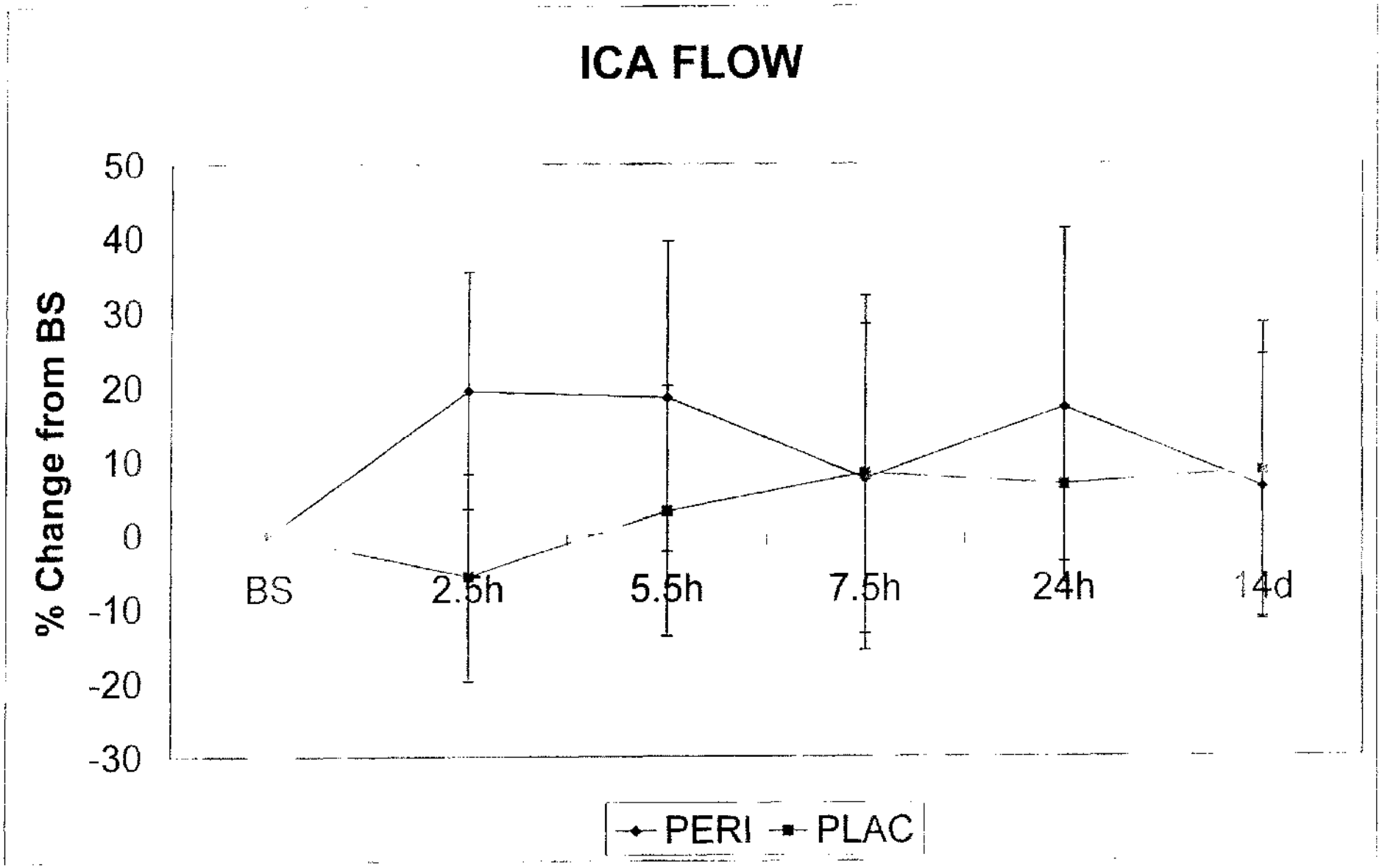


Figure 2.6

Change in ICA flow



Error bars represent +/-2SE

Figure 2.7 ICA flow - Treated group patient 1

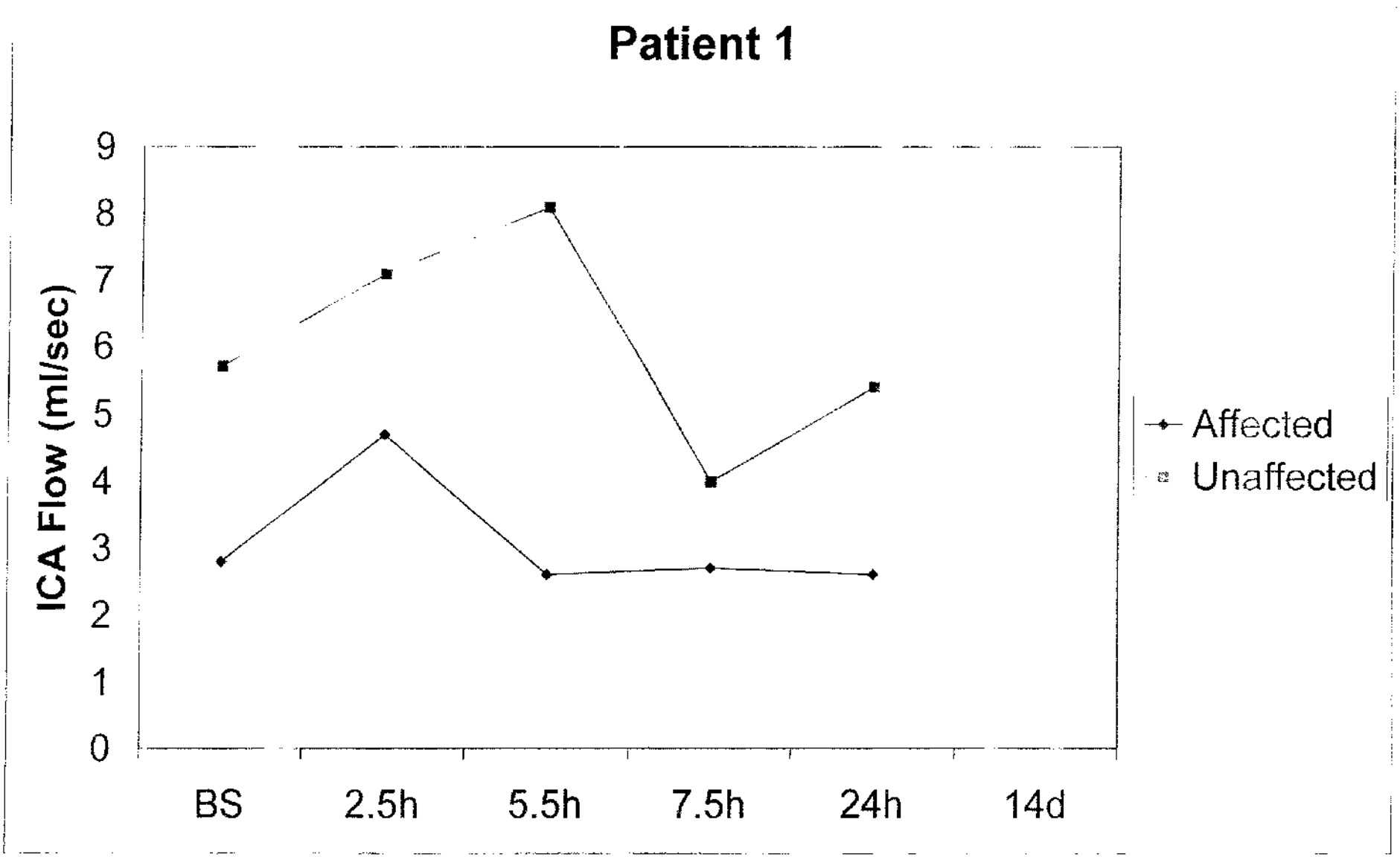


Figure 2.8 ICA flow - Treated group patient 2

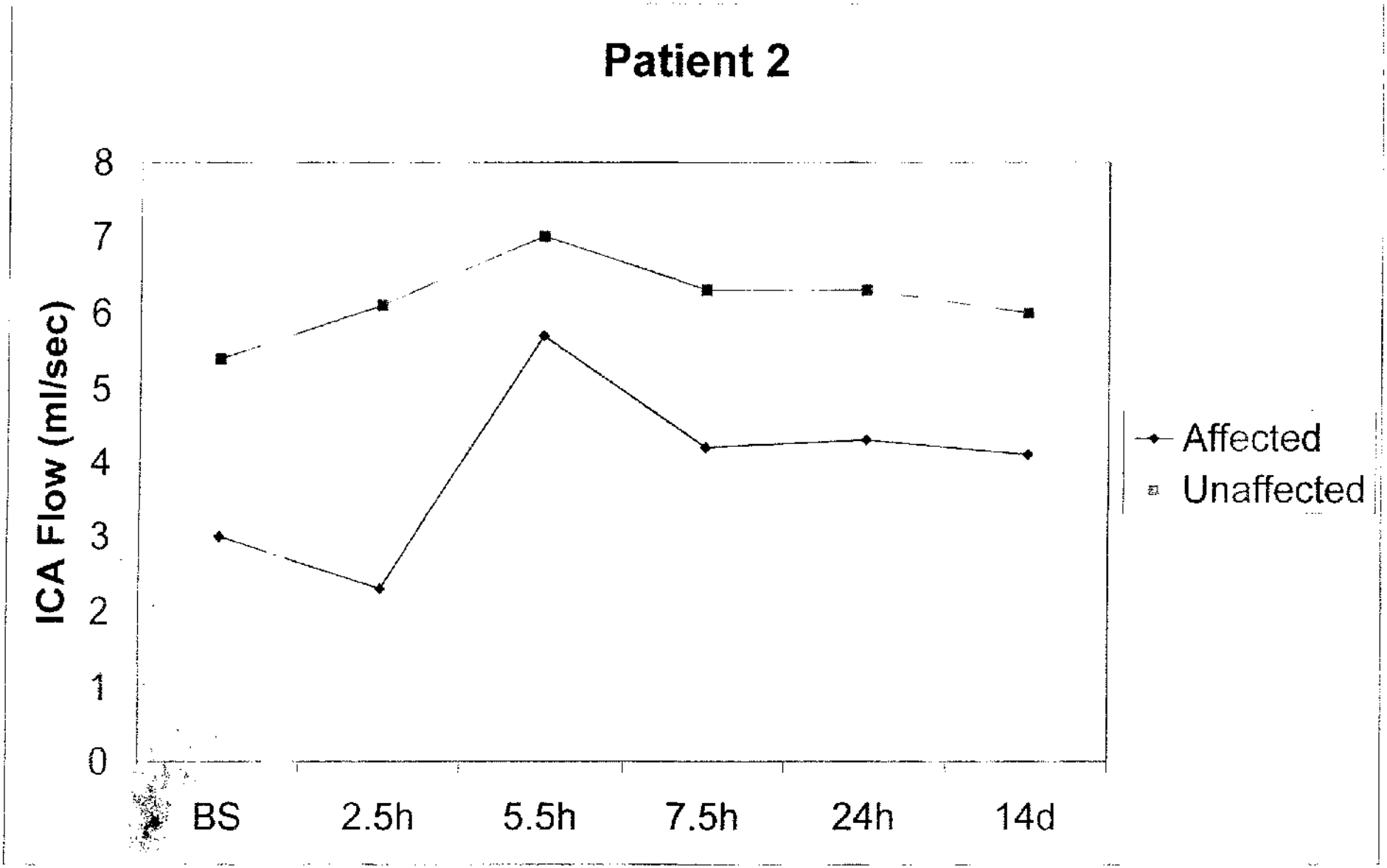


Figure 2.9 ICA flow - treated group patient 8

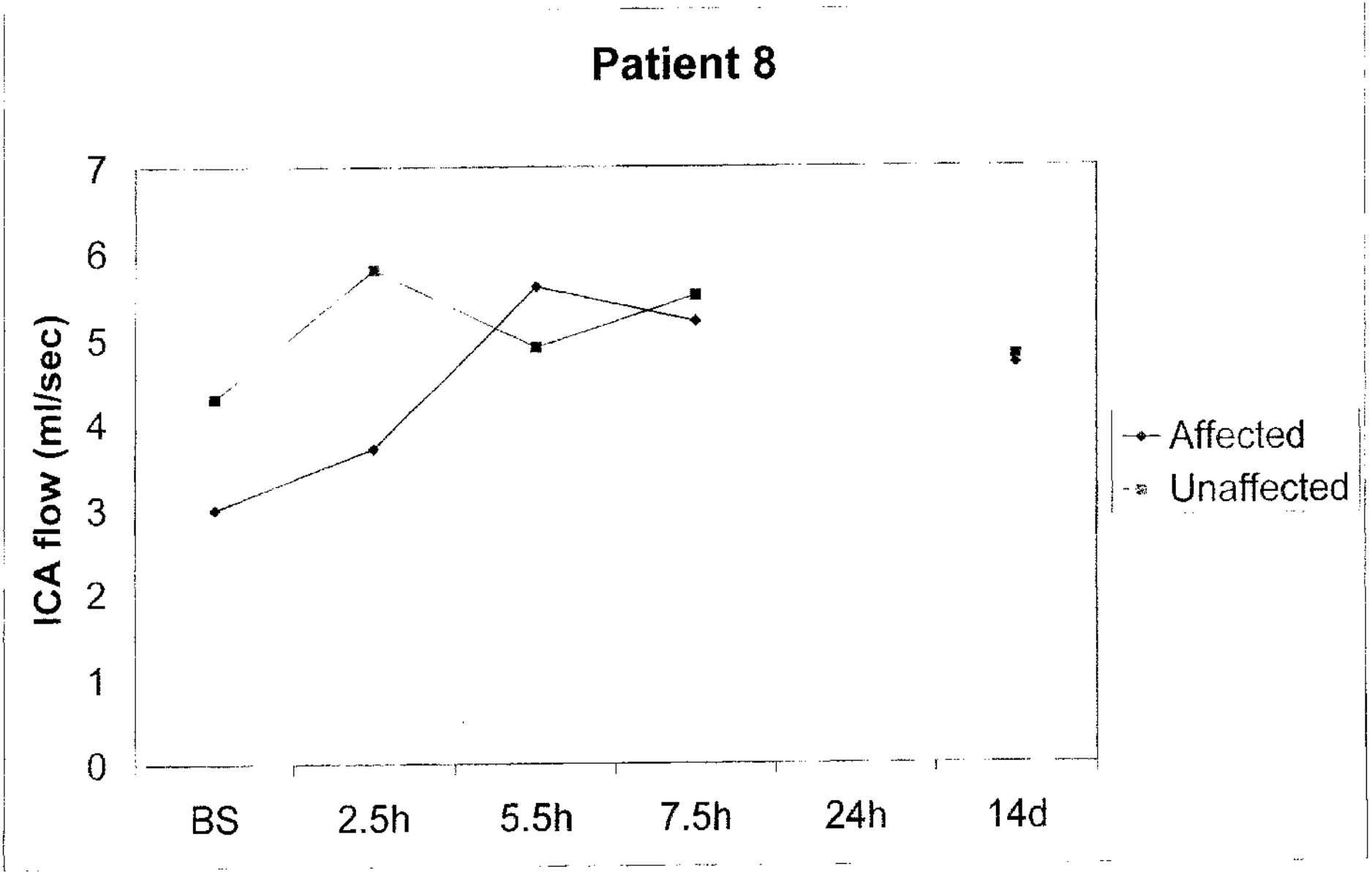
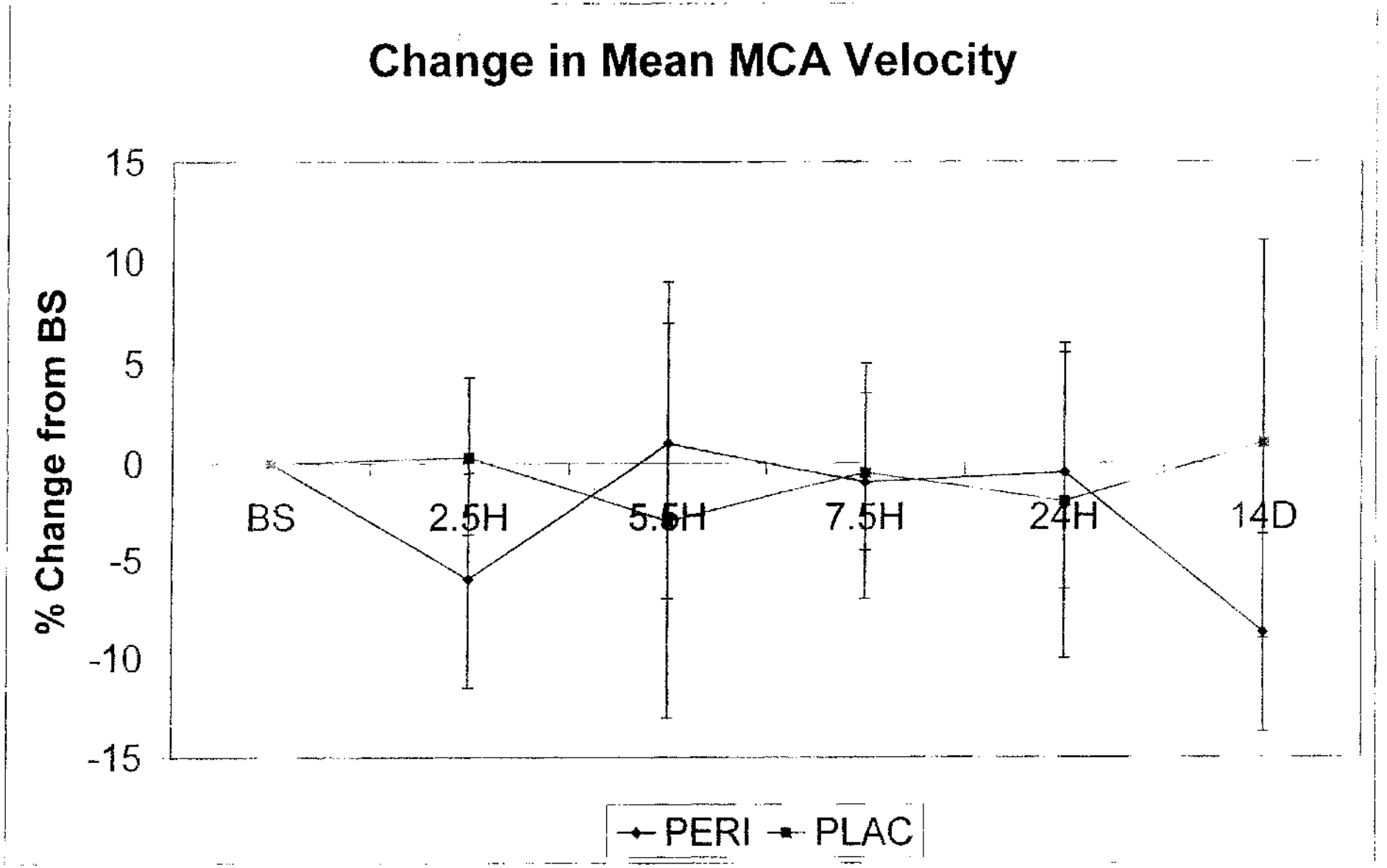
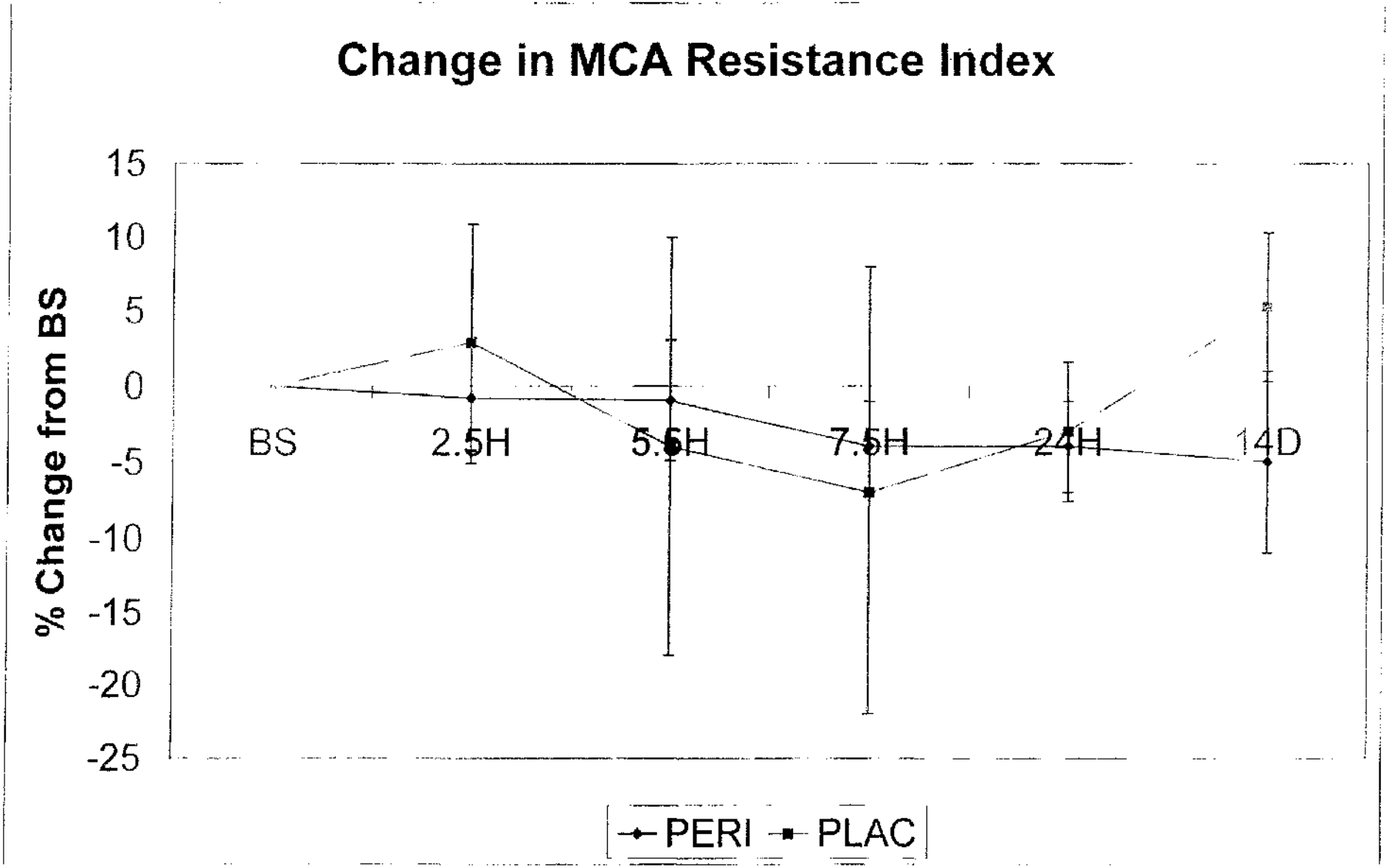


Figure 2.10 Change in MCA velocity



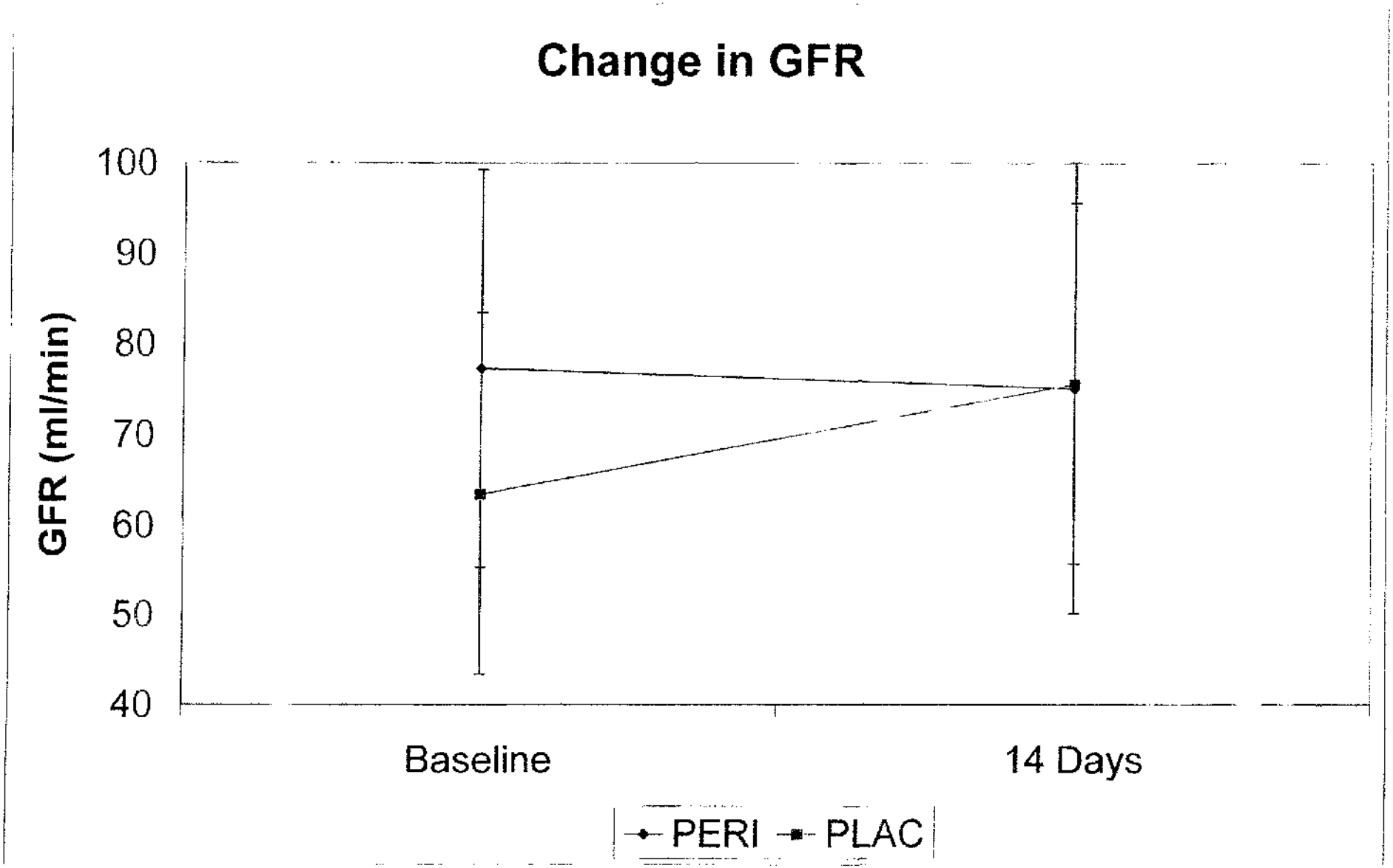
Error bars represent +/-2SE

Figure 2.11 Change in MCA resistance index



Error bars represent +/-2SE

Figure 2.12 Change in GFR



Error bars represent +/-2SE

Figure 2.13 Example of a SPECT scan with template used for analysis of focal cerebral perfusion

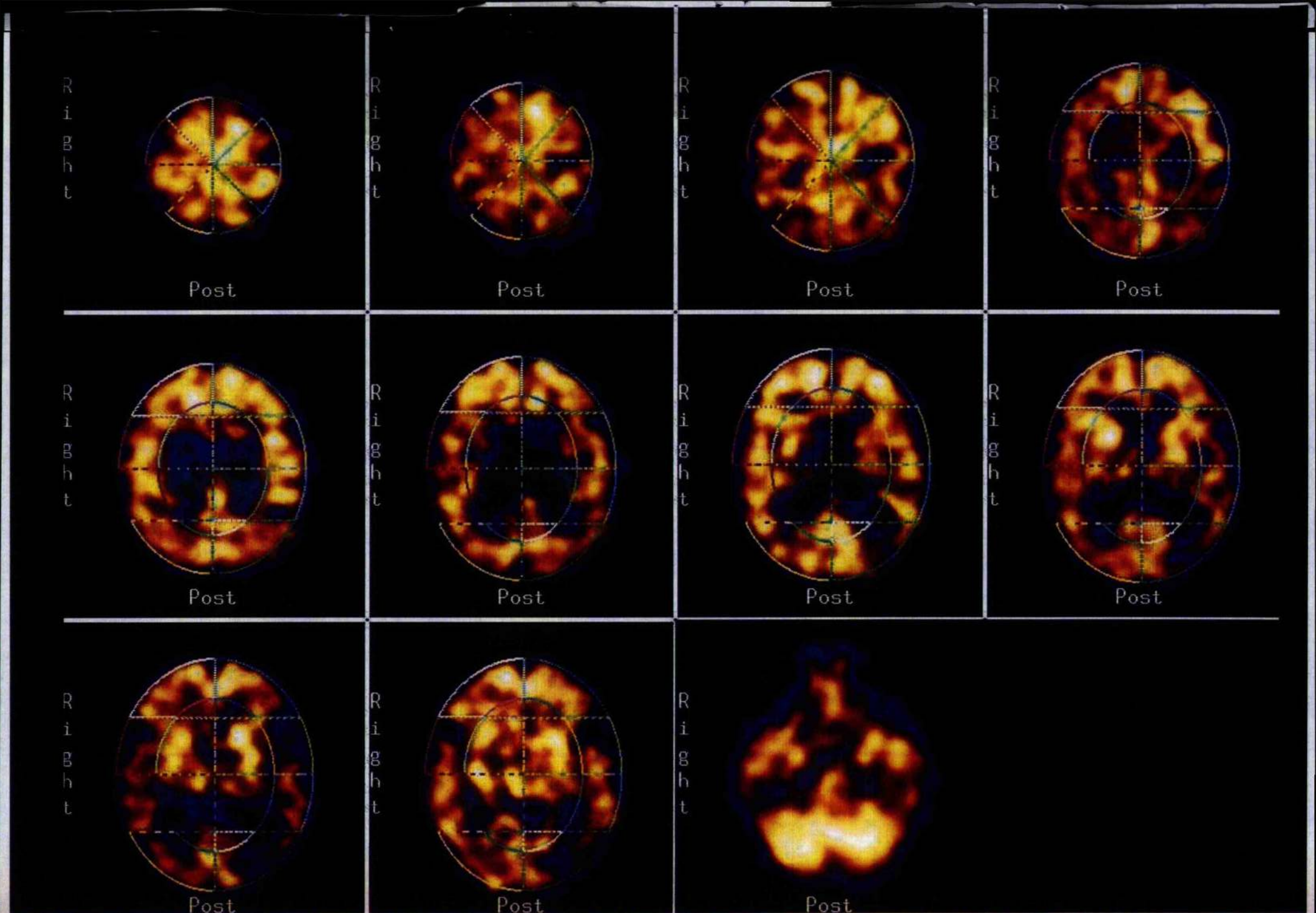

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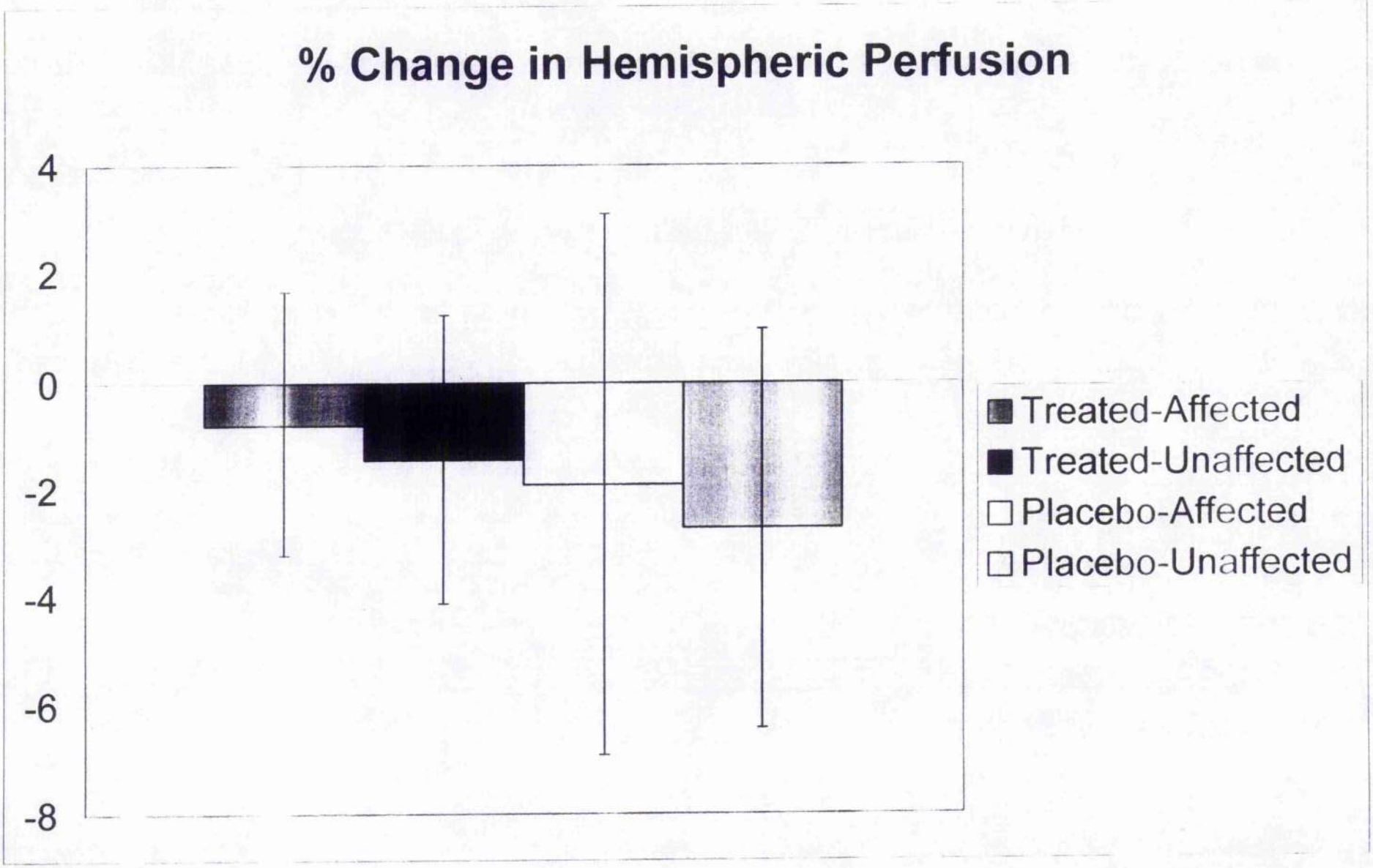
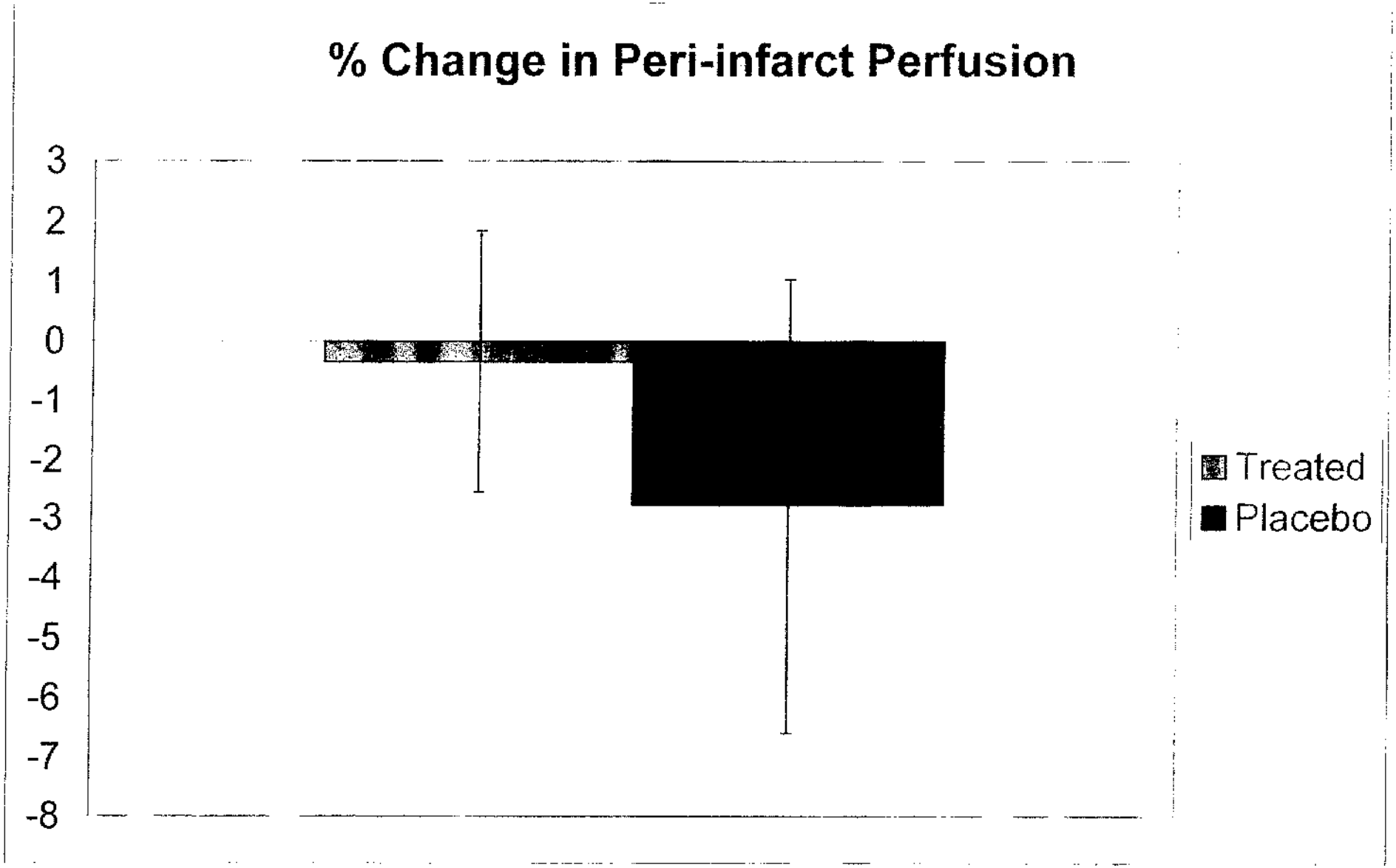
Figure 2.14 Change in hemispheric perfusion



Error bars represent +/-2SE

Figure 2.15 Change in focal perfusion



Error bars represent +/-2SE

Table 2.1a **Treated Group Demographics**

Pat No	Age	Sex	Prior HBP	BP therapy	Days since CVA	CT/MRI	Clinical	Doppler	Initial NIH	Interval NIH	Initial MABP
1	77	M	N	None	15	L Cortical	L PACS	L Severe	7	4	102
2	78	M	N	None	16	L Subcort	L LACS	L Mod	3	1	105
3	70	M	Y	Bisop.	22	L Subcort	L LACS	Bilat Sev	5	4	104
4	70	M	Y	Atenolol	20	R Cortical	R PACS	R Occ	6	5	125
5	79	M	Y	BDF	25	L Cortical R Subcort	L LACS	Bilat Sev	7	7	106
6	83	F	Y	None	44	R Brainst	POCS	L Severe R Occ	3	3	110
7	68	F	Y	BDF	50	R Cortical	R PACS	R Occ	6	6	105
8	64	M	Y	BDF	62	L Subcort	L LACS	L Mod	3	3	124
9	70	M	N	None	17	L Cortical L Subcort	L PACS	L Occ R Severe	3	0	112
10	68	F	Y	None	14	R Pontine	POCS	Bilat Sev	4	1	120
11	60	F	Y	None	25	L Cortical	L PACS	L Occ	5	3	110
12	59	M	Y	None	15	R Subcort	R LACS	R Mod	2	1	113
Mean (SD)	70.5 (7)				27 (16)				4.5 (1.7)	3.2 (2.2)	111 (8)

Table 2.1b **Placebo Group Demographics**

Pat No	Age	Sex	Prior HBP	BP therapy	Days since CVA	CT/MRI	Clinical	Doppler	Initial NIH	Interval NIH	Initial MABP
1	57	M	N	None	15	R Subcort	R LACS	L Mod	2	1	118
2	72	F	N	None	62	R Pontine	POCS	R Occ L Sev	5	3	116
3	78	M	Y	BDF	19	L Cortical	L PACS	R Occ	7	7	121
4	71	M	N	None	22	L Medulla	POCS	R Mod	3	3	102
5	57	M	Y	BDF	16	L Cortical	L PACS	R Sev	8	8	111
6	74	F	Y	Atenolol	27	R Subcort	R LACS	L Mod	2	0	120
7	87	F	Y	None	34	L Cerebel	POCS	R Occ L Sev	1	0	101
8	86	F	N	None	17	R Subcort	R LACS	R Occ	5	4	118
9	64	M	Y	Bisop	27	R Subcort L Cortical	R LACS	R Mod L Occ	4	2	119
10	62	F	N	None	34	R Subcort	R PACS	L Occ R Mod	5	2	101
11	65	M	N	None	14	L Cortical	L TACS	L Occ	11	9	104
12	71	M	Y	BDF	28	L Subcort	L LACS	L Mod	4	2	111
Mean (SD)	70.3 (10)				26 (13)				4.7 (2.8)	3.4 (3)	112 (8)

Chapter Three

A randomised, pilot study to investigate the feasibility of glucose lowering with insulin in hyperglycaemic acute ischaemic stroke patients

3.01 Introduction

Specific therapeutic intervention can improve outcome in patients with acute ischaemic stroke. Early thrombolysis is associated with a reduction in mortality and an improved functional outcome.[102] Other forms of treatment (for example neuroprotective agents) are currently undergoing therapeutic trials. Attention has recently been focused upon the significance of elevated levels of blood glucose in patients presenting with either myocardial infarction (MI) or stroke. Tight glycaemic control immediately following MI in diabetic patients improves outcome at one year;[123] however similar prospective evidence to guide clinicians in their management of hyperglycaemic acute stroke patients does not exist.

Preclinical evidence suggests that hyperglycaemia predisposes to poor outcome.[124-128] In experimental models of stroke, hyperglycaemia augments ischaemic brain injury following cerebral ischaemia, possibly as a result of lactate accumulation. A more detailed discussion of the preclinical work in this field can be found in chapter one. In humans with acute stroke, a correlation between high levels of blood glucose and poor outcome has been reported in several series.[129,130] It was initially suggested that the relationship was an epiphenomenon and that blood sugar is elevated in patients with more severe strokes (as part of an exaggerated stress response); however, studies of biochemical markers of stress failed to substantiate this.[131,132] Two large-scale studies have further clarified the nature of the relationship between acute ischaemic stroke, hyperglycaemia and the stress response.[70,133] Both studies

demonstrated an adverse effect of hyperglycaemia following acute stroke, after correcting for stroke severity and other factors known to predict outcome.

Evidence accrued from these studies suggests plasma glucose level greater than 8 mmol/L is an independent predictor of poor prognosis. The effect of hyperglycaemia on mortality was considerable; the estimated relative hazard of 1.87 calculated in one study[70] was greater than for haemorrhagic versus ischaemic stroke, and comparable to the excess risk conferred by adding twenty years to a patient's age. A randomised trial is warranted to examine the potential benefit of intervention with insulin to lower blood glucose after stroke, and to define more accurately the potential risks of iatrogenic hypoglycaemia and disturbance of fluid balance. As discussed in chapter one, insulin may exert a beneficial effect through a number of local and systemic mechanisms. Through lowering blood glucose, insulin reduces availability of substrate for anaerobic glycolysis and hence may reduce lactate accumulation within ischaemic human brain tissue. In addition, a neuroprotective effect of insulin acting at cerebral insulin-like growth factor receptors and independent of its hypoglycaemic action has been postulated. Systemic effects of insulin, such as correction of disturbed lipoprotein patterns, effects on platelet function and on plasminogen activator inhibitor levels may also contribute to any beneficial effect. We performed a pilot study to examine the feasibility of a large-scale clinical trial comparing stroke outcomes in hyperglycaemic patients presenting within 24 hours of stroke randomised to either strict or routine glycaemic control.

3.02 Methods

The Acute Stroke Unit of the Western Infirmary in Glasgow serves a catchment population of 220 000. All patients who present within 72 hours of onset of acute neurological deficit with no known alternative to a vascular cause are admitted irrespective of age or severity of neurological deficit. All clinical data and results of investigations are gathered prospectively. The aim is to complete all investigations, including computed tomography or magnetic resonance imaging where appropriate, within 72 hours. Patients admitted to our unit within 24 hours of onset of CT-confirmed ischaemic stroke, with venous blood glucose greater than 8 mmol/L and Glasgow Coma Scale greater than 8 were considered eligible for the study. Patients in whom insulin administration was considered essential (known insulin-requiring diabetes mellitus or severe metabolic derangement) and patients with clinical evidence of infection or congestive cardiac failure were excluded. After informed consent (or informed assent of relatives in patients with language disorder) was obtained, patients were randomised to either standard management or rigorous control of hyperglycaemia. The aim of the latter approach was to maintain blood sugar between 5 and 8 mmol/L. Those patients randomised to the active treatment arm received an infusion of insulin (Human Actrapid, Novo Nordisk) with dosage adjustment according to a sliding scale (Table 3.1) together with an infusion of crystalloid (either 5% dextrose or 0.9% saline as dictated by blood glucose concentration). Insulin was given continuously via an infusion pump attached to an intravenous cannula and the

dose titrated according to the most recent blood sugar reading. Blood sugar monitoring was carried out every 2 hours in accordance with guidelines issued by our local Drug and Therapeutics Committee. The rate of intravenous crystalloid infusion was dependent upon cardiac status and fluid balance. The infusion of insulin by sliding scale and appropriate crystalloid continued for 48 hours. On cessation of the infusion, any pre-existing oral hypoglycaemic therapy was re-instituted. Patients in the control group received standard management of their blood sugar, which comprised the continuation of any pre-existing oral hypoglycaemic therapy if swallowing was satisfactory, and blood glucose monitoring with the same frequency as the insulin group. Patients in the control group received intravenous infusion of crystalloid (0.9% saline ± 5% dextrose) for the first 48 hours of the study. The rate of the infusion was determined by the fluid balance and clinical state of the patient. Standard hospital diet was provided to patients in both groups if swallowing ability was satisfactory. Tube feeding was not instituted in any patient during the infusion period. All pre-existing oral hypoglycaemic therapy was discontinued in the insulin recipients; the antihypertensive medication of every patient recruited was discontinued in accordance with our protocol. Ethical approval from the West ethics committee was obtained prior to recruitment.

The blood glucose concentrations and infusions were monitored and rates of insulin delivery adjusted by ward nursing staff; protocols were provided to guide insulin dose titration. Blood glucose was monitored at the bedside using standard

hexokinase assay (Bayer Glucometer strips: Bayer Diagnostics, Leverkusen, Germany). Readings were taken at two-hourly intervals for 48 hours after commencement of infusion. Supine blood pressure was measured at four-hourly intervals during the infusion period using Marquette oscillometric equipment (Marquette Electronics, Wisconsin). Monitoring protocols were identical for both the treated and control arms of the study.

As the protocol required titration of insulin dose against regular blood glucose measurements, it was not practicable to conduct the study double or single blind, and an open study design was adopted. Patients underwent laboratory, clinical and neurological assessments (including NIH Stroke Scale) at baseline and clinical assessments were repeated one month later.

This was a pilot study designed to establish the safety of insulin treatment in patients with moderate hyperglycaemia following acute ischaemic stroke. Mortality figures at one month were collected and used as the principal outcome measure of the study: as other clinical and radiological assessments may have been prone to observer bias they were used as secondary endpoints only. HbA1c was collected at baseline and values less than 5% were considered as indicative of pre-stroke normoglycaemia. The secondary endpoints were NIH scores at baseline and at one month, and safety laboratory measurements, including assessment of the frequency and severity of iatrogenic hypoglycaemia.

3.03 Results

Demographics

25 patients were recruited, of whom 13 were treated with insulin and 12 received conventional treatment. The groups were well matched with regard to age, sex, elapsed time between ictus and commencement of infusion, initial plasma glucose concentration, diabetic status and initial stroke severity (as assessed by NIH score). Patient demographics are shown in Table 3.2.

Blood Glucose

The capillary glucose concentration of patients within the treated group fell to lower values within 4 hours of commencement of infusion and a significant difference in capillary blood glucose between the two groups was apparent during the early stages of the infusion of insulin (Figure 3.1). Repeated measures analysis of variance revealed highly significant group ($p=0.001$) and time ($p=0.009$) differences. No significant group / time interaction was seen. A smaller proportion of insulin recipients strayed outwith the desired range of capillary glucose concentrations (5.0-7.9 mmol/lit) during monitoring (Figures 3.2 and 3.3). A significant reduction in mean area under glucose / time curve was seen in the insulin treated group (figure 3.4).

Insulin dose and Infusate Volume

Mean total insulin dose per patient was 80.8 units in 43 hours (SD 12). A histogram of insulin infusion rate at each timepoint is given in figure 3.5. Patients in the treated arm of the study received relatively more 5% dextrose and less 0.9% saline than patients in the control group, in accordance with the sliding scale protocol. The mean total volume of crystalloid infused in each group is given in figure 3.6.

NIH Score

NIH scores were measured at baseline and at one month. Mean baseline NIH score in the treated group was 7.6 (range 2-28). The equivalent figure in the control group was 8.3 (range 4-17). At one month, NIH score was unchanged or improved in all 24 survivors. Mean NIH score in the treated group survivors at one month was 3.75 (range 0-17), and 6 (range 2-13) in the control group. The change in mean NIH scores of the survivors is shown in figure 3.7. Due to the small numbers involved, further analysis of the NIH data was not undertaken.

Blood Pressure

Supine blood pressure was measured at regular intervals during the infusion period using Marquette oscillometric equipment. No significant differences in systolic or diastolic blood pressure were observed between the groups on admission or for the duration of the infusion. These data are shown in figure 3.8.

Adverse events

No significant adverse events were observed in either group during the infusion period. One symptomatic episode, possibly attributable to iatrogenic hypoglycaemia occurred in one of the insulin recipients. This comprised subjective sensations of hunger and anxiety, and was associated with a capillary blood glucose reading of 4.0 mmol/lit (reference range 2.8-6.0 mmol/lit). The symptoms resolved promptly following oral dextrose administration, and capillary blood glucose had risen to 5.2 mmol/lit when retested two hours later.

Mortality

One death occurred in the treated group. The patient succumbed to hypostatic bronchopneumonia three weeks after admission with extensive hemispheric cerebral infarction. There were no deaths in the untreated group.

3.04 Discussion

We have demonstrated that attenuation of hyperglycaemia and maintenance of blood glucose within a pre-defined range following acute ischaemic stroke is safely achievable using intravenous administration of rapid-acting insulin by sliding scale. The study protocol excluded comatose patients and hence allowed assessment of the incidence of symptomatic episodes of hypoglycaemia. In common with other studies of insulin administration to hyperglycaemic patients [70 134] we found these to be rare and relatively mild. One patient in the

treated group had symptoms consistent with hypoglycaemia, however glucose concentration measured during the symptoms was at the lower end of the normal reference range. No blood glucose readings below the lower end of the reference range were recorded in either group during the monitoring period. As can be seen from the NIH data presented, recruits into the study tended to have mild stroke related symptoms. The mean baseline NIH scores of patients in treatment and control groups were 7.6 and 8.3 respectively. In an unselected cohort of 200 consecutive patients presenting to our unit, the equivalent figure was 9.7. Although retrospective evidence suggests that the detrimental effect of hyperglycaemia is greatest in patients with small subcortical cerebral infarction, the adverse prognostic effect of hyperglycaemia was seen across a broad range of stroke severity.[70] Our study sought to investigate the feasibility of this method of blood glucose control when applied to a busy clinical stroke unit. The design of the study was simple and pragmatic in order to minimise the burden to nursing staff. All patients receiving intravenous insulin are monitored with two-hourly fingerprick blood glucose estimation in accordance with local protocols, and no significant problems with data collection were encountered during this study. Insulin infusions were prepared daily by either nursing or medical staff; this task caused no significant inconvenience for either group.

A large scale prospective study to investigate the potential benefit of intervention with insulin in hyperglycaemic humans following acute stroke is warranted, and our study has demonstrated the feasibility of one approach to metabolic

intervention after stroke. Relatively little published work on the effect of insulin on human ischaemic brain tissue exists therefore a pragmatic approach must be taken in the design of such a trial. Due to the paucity of evidence, a number of issues arise; these are discussed below.

1. Hyperglycaemic threshold above which randomisation should be considered. The recent observational study discussed earlier[79] reported a poor outcome in stroke patients whose level of blood glucose exceeded 8.0 mmol/lit on presentation. Further analysis of the data used in that study reveals a relationship between poor outcome and elevated blood glucose at relatively lower levels of glucose concentration. The relative hazard of poor outcome is as great at a threshold of 7.5 mmol/lit (1.7964) as it is at 8.0 mmol/lit - it may be that a threshold of 7.5 mmol/lit for both recruitment and upper end of "target" range should be considered in the light of this information.

2. Time window within which patients can be recruited. As the potential benefit of insulin in hyperglycaemic patients with stroke may arise from any of a number of diverse effects both local and systemic, it is difficult to argue a cogent case for any particular time window. There is evidence to suggest that any direct neuroprotective effect of insulin mediated by central IGF-1 receptors and independent of hypoglycaemic action may occur exclusively during the hyperacute phase of stroke.[84] It is also unlikely that neuronal death associated with excess accumulation of lactate following hyperglycaemic ischaemic stroke will be affected by insulin-mediated euglycaemia outwith the

hyperacute phase. Conversely, both positron emission tomography (PET) and magnetic resonance spectroscopy studies have suggested that cellular activity within the ischaemic penumbra continues in some cases for greater than 24 hours after ictus [35]. In addition, evidence from other studies of intervention with insulin following acute vascular occlusion[123] suggests that subacute intervention with insulin may be beneficial, although the underlying mechanisms remain elusive. Analysis of data from Glasgow[70] suggests that elevated levels of blood sugar measured at 24 hours post ictus continue to confer a relatively poor prognosis. The DIGAMI study of insulin infusion following myocardial infarction in diabetic patients used a 24 hour time window. The investigators reported a significant improvement in both short and long-term mortality within the treated group and concluded that intravenous insulin therapy in hyperglycaemic diabetic patients with acute myocardial infarction was beneficial if initiated within 24 hours of onset. In addition, the prevailing erroneous perception of stroke as a non-urgent condition frequently leads to delay in presentation to hospital in the U.K. A broader time window should greatly facilitate recruitment of patients. Subgroup analysis of those patients treated within a shorter time window may permit detection of any hyperacute neuroprotective effect upon the ischaemic penumbra.

3. The duration of insulin infusion. As a substantial proportion of the patients recruited into the study will not be diabetic and will have no need of

hypoglycaemic therapy in the longer term, the minimum duration of insulin therapy must be defined. A period of 48 hours was used for the pilot study and was found to be well tolerated, with no clinically significant hypoglycaemic episodes; however it was observed that a substantial proportion of those non-diabetic individuals randomised to receive insulin maintained euglycaemic levels with minimal supplemental insulin towards the end of the prescribed dosing period. It may be that cessation of the infusion could be considered before 48 hours have elapsed should it become apparent that the supplemental insulin is no longer required to maintain euglycaemia. A further issue is the maintenance of long term insulin therapy. The DIGAMI investigators observed a continuing survival advantage of long-term subcutaneous insulin in diabetic patients following acute myocardial infarction. Although the observed benefit may have been attributable to cessation of potentially cardiotoxic sulphonylurea therapy, it has been postulated that the systemic effects of insulin upon platelet function and lipid metabolism may be protective against further vascular events. In order to detect any benefit provided by insulin therapy in the longer term following stroke, non insulin dependent diabetic individuals recruited into the study could be converted to subcutaneous insulin for the remainder of the trial in a fashion analogous to the DIGAMI study.[123] Conversion to subcutaneous insulin for one year after the index event would enable detection of any sustained benefit of ongoing insulin administration however this approach may lead to increased withdrawals from the study.

4. The primary endpoint The pilot study used mortality alone as a primary endpoint, with neurological deficit (NIH score) as a secondary endpoint. Mortality alone was used as a primary endpoint for both statistical and logistical reasons. In Scotland, record-linkage of clinical information allows rapid and accurate collection of mortality data with a minimum of effort on the part of the investigators. The initial retrospective study[70] suggested a significant and early divergence of survival curves between hyperglycaemic and euglycaemic stroke patients. In addition, as the trial design was unblinded, the use of a primary endpoint that was not open to bias represented a logical choice. It may be that more functional measures of outcome (such as Barthel or Rankin scores) should be used and combined with mortality to permit dichotomised primary endpoints (such as dead or severely disabled versus alive and at home) in a larger, definitive study. The use of more widely-recognised outcome measures is to be encouraged, however this approach may leave the study open to criticisms of bias as the design is currently unblinded and implementation of a blinded study of insulin infusion may prove difficult.

5. The duration of follow-up. As the initial retrospective study[70] indicated that the majority of excess early mortality in hyperglycaemic stroke patients occurred early, the follow-up period of the pilot study was limited to one month. It is anticipated that the proposed multi-centre protocol would employ a longer

period of follow-up as a larger, multi-centre trial may be in a position to detect ongoing long term benefit of intervention with insulin particularly if the duration of insulin therapy is increased as mentioned above.

6. The method of insulin administration. For logistical reasons a sliding scale of insulin infusion was used in the pilot study. Local nursing staff were experienced in using this method of glycaemic control and during the pilot study no significant difficulties with infusion preparation or administration arose. The sliding scale insulin regime involves regular monitoring of capillary blood glucose (12 times daily) with subsequent titration of "Actrapid" insulin dose as delivered by a syringe driver. The dose of insulin to be administered varies directly with the glucose measurement and is prescribed in advance by medical staff. A concurrent dextrose infusion is provided at variable rate as dictated by the patient's cardiac status, metabolic status and fluid balance. An alternative regime is also commonly used in Europe and North America. The "GKI" regime uses infusion of a solution containing glucose, potassium and insulin at variable rate to maintain euglycaemia. It is relatively simple to administer and obviates any risk of infusion pump malfunction causing iatrogenic hypoglycaemia. It is somewhat less flexible than the sliding scale regime as glucose and insulin infusions cannot be varied independently of each other. As with sliding scale insulin administration, two-hourly monitoring of blood sugar is routinely used. A recent prospective study has recently reported a similar physiological but attenuated glucose response to acute

stroke using the GKI regime.[133] Some problems are faced when using either method. Both sliding scale and GKI regimes are reactive rather than proactive as they respond to fluctuations in blood sugar only after they have occurred. Maintenance of euglycaemia can prove difficult in patients who are eating and drinking normally, as such patients tend to develop post-prandial hyperglycaemia before the insulin infusion rate is increased. While both sliding scale and GKI regimes have attracted criticism in the literature,[136,137] no clearly superior alternative has yet been reported.

3.05 Conclusion

The relationship between hyperglycaemia and outcome following cerebral ischaemia is not yet fully understood. There is reasonable evidence to suggest that hyperglycaemia immediately following cerebral ischaemia is not entirely attributable to a stress response and that it predicts a relatively poor prognosis. Although there is a paucity of prospective data at present, current evidence suggests that intervention to reduce excessively high levels of blood sugar while avoiding hypoglycaemia is warranted, and that this can safely and effectively be achieved using intravenous administration of insulin by sliding scale. Larger trials are required to clarify optimal metabolic management following ischaemic stroke.

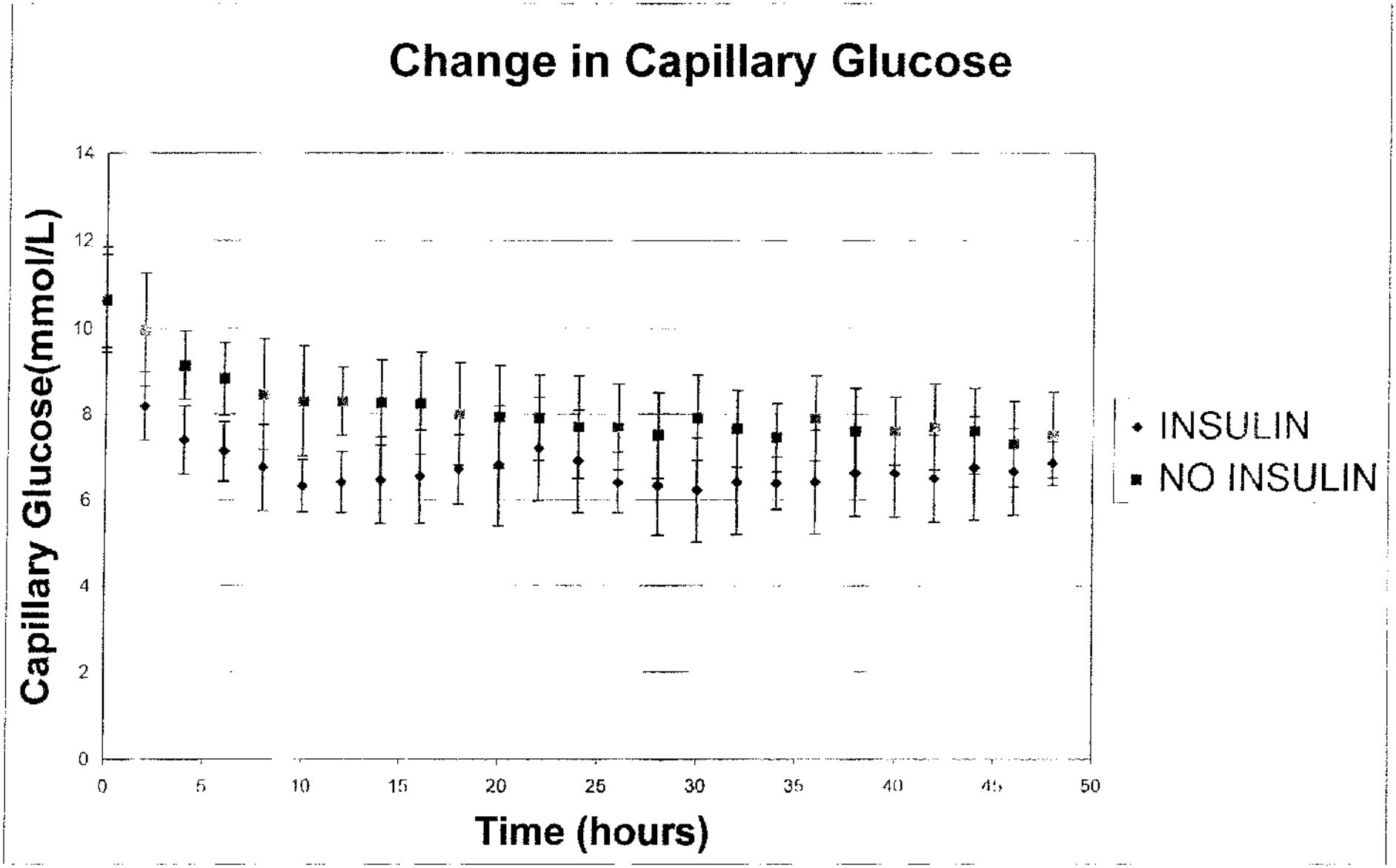
Table 3.1 Sliding Scale

Blood Glucose (mmol/lit)	Infusate	Dose of Intravenous Actrapid Insulin (Units/h)
0-6	5% Dextrose	0
6.1-8	5% Dextrose	2
8.1-11	5% Dextrose	3
11.1-15	0.9% Saline	4
15.1-20	0.9% Saline	5
>20	0.9% Saline	6

Table 3.2 Patient demographics

	Insulin group (n=13)	**Control group (n=12)**
Age (± 2SE)	73.3 (6.9)	76.7 (5.5)
NIH (± 2SE)	7.6 (4)	8.33 (2.5)
Initial blood glucose(± 2SE)	10.7 (0.9)	10.6 (0.8)
Known NIDDM	7	6
Initial HbA1c	6.2 (0.94)	5.6 (1.04)
Number of males	5	5
Initial SBP (± 2SE)	159 (10)	166 (12)
Initial DBP (± 2SE)	87 (5)	87 (8)
HbA1c > 5%	8	5

Figure 3.1 Capillary glucose concentration



Error bars represent +/-2SE

Figure 3.2 Proportion with glucose >8 mmol/L

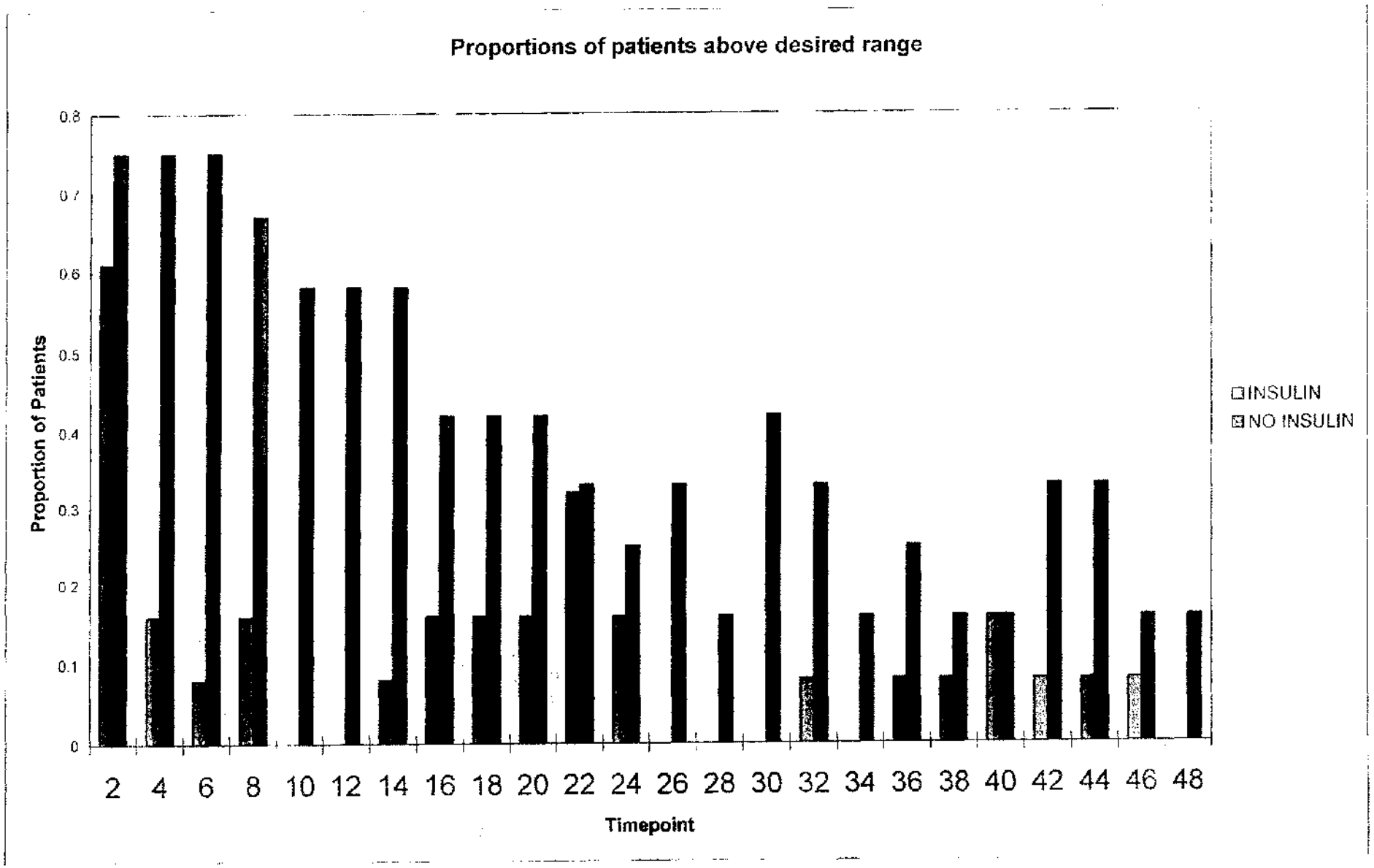


Figure 3.3 Proportion with glucose <5 mmol/L

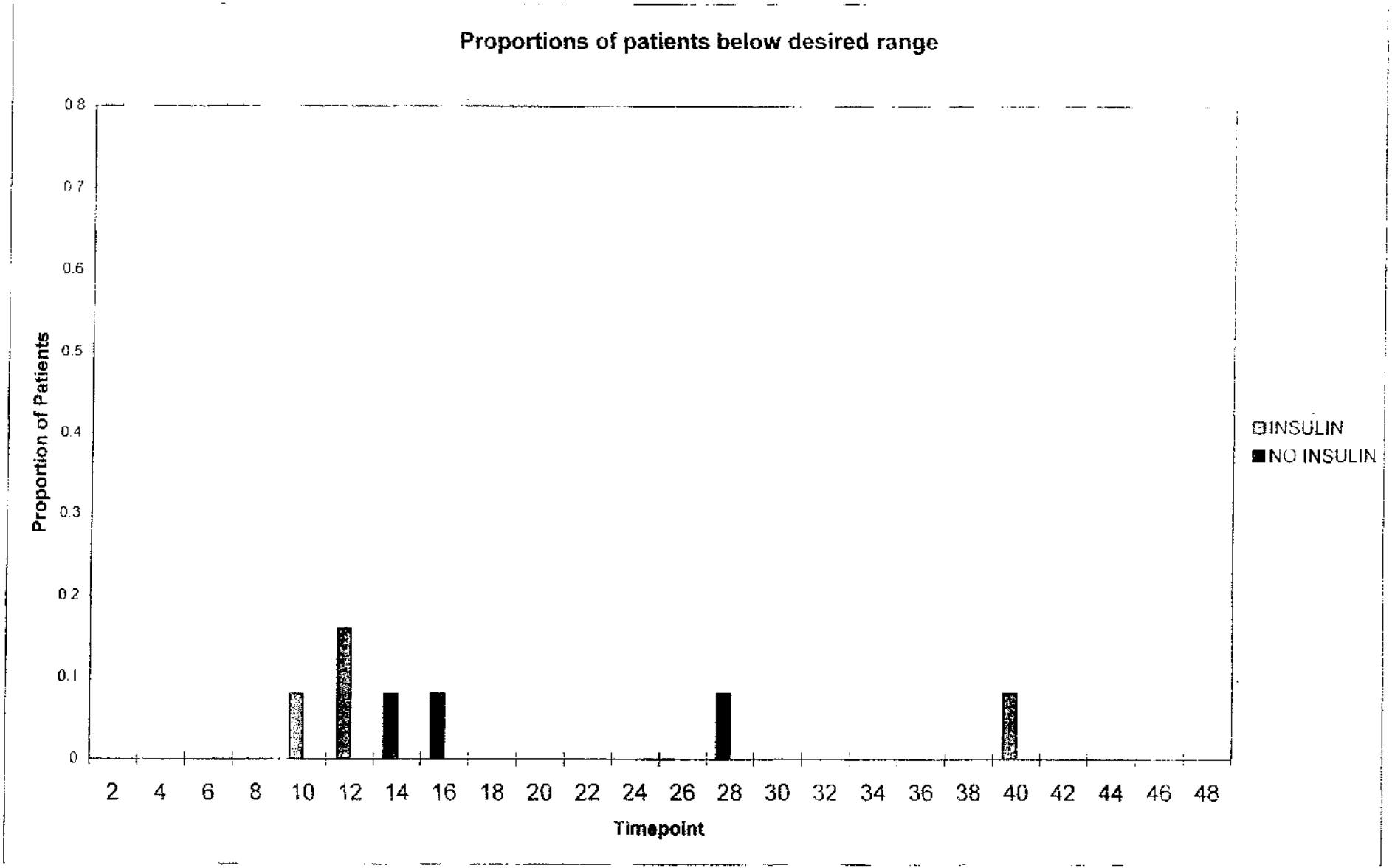
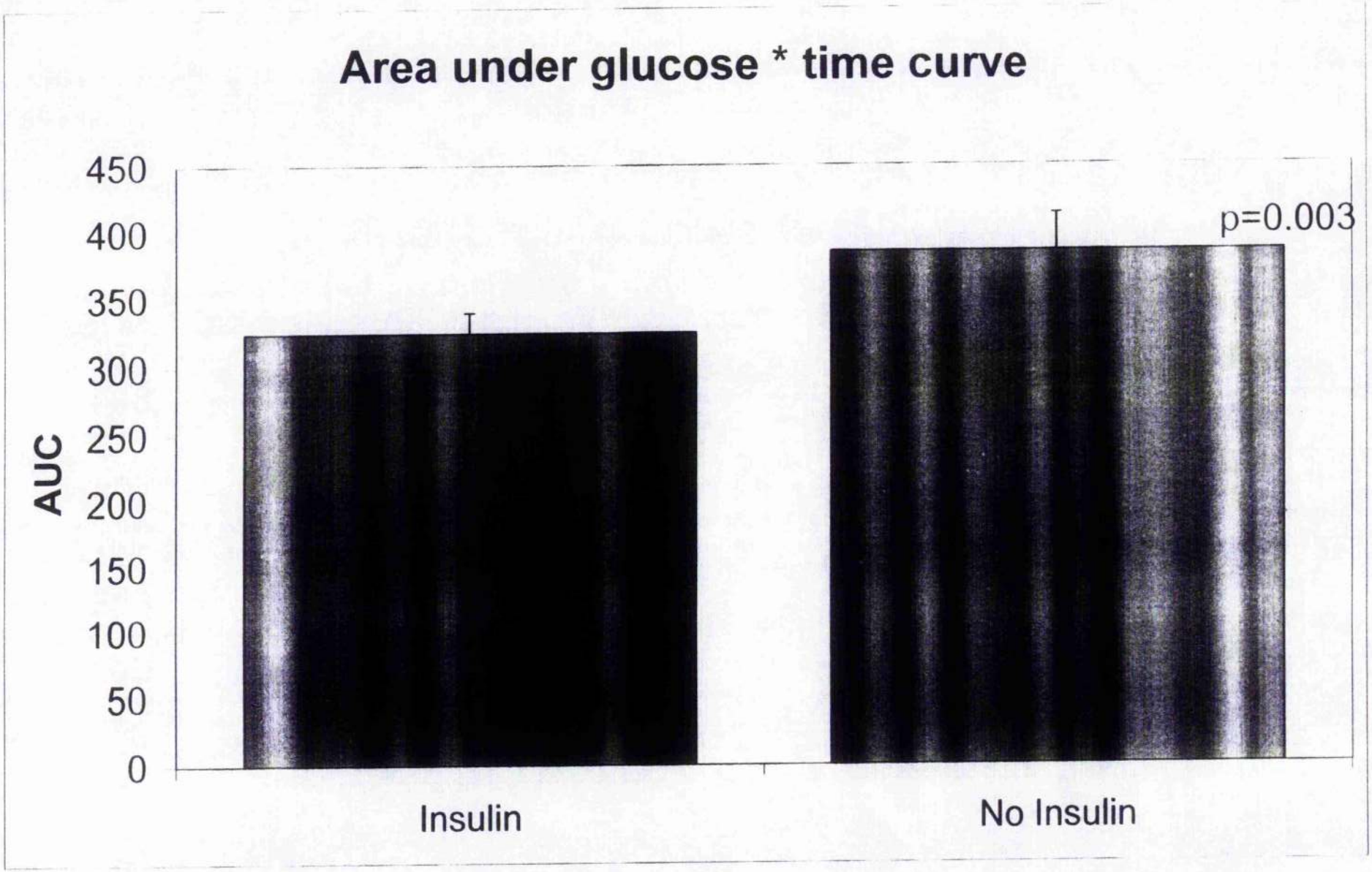


Figure 3.4

Area under glucose / time curve



Error bars represent +/-2SE

Figure 3.5 Mean insulin dose at each timepoint

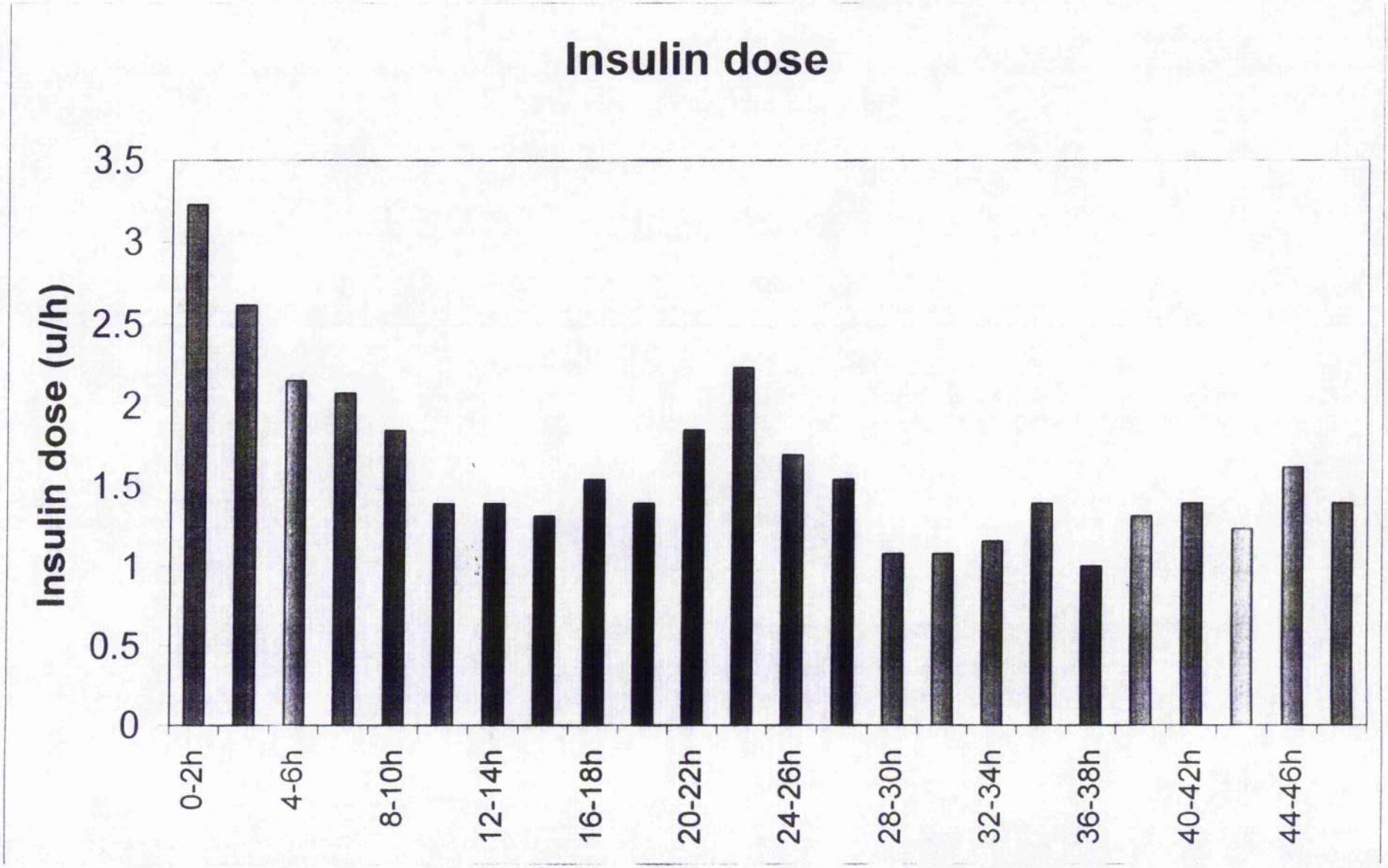


Figure 3.6

Infusate volume

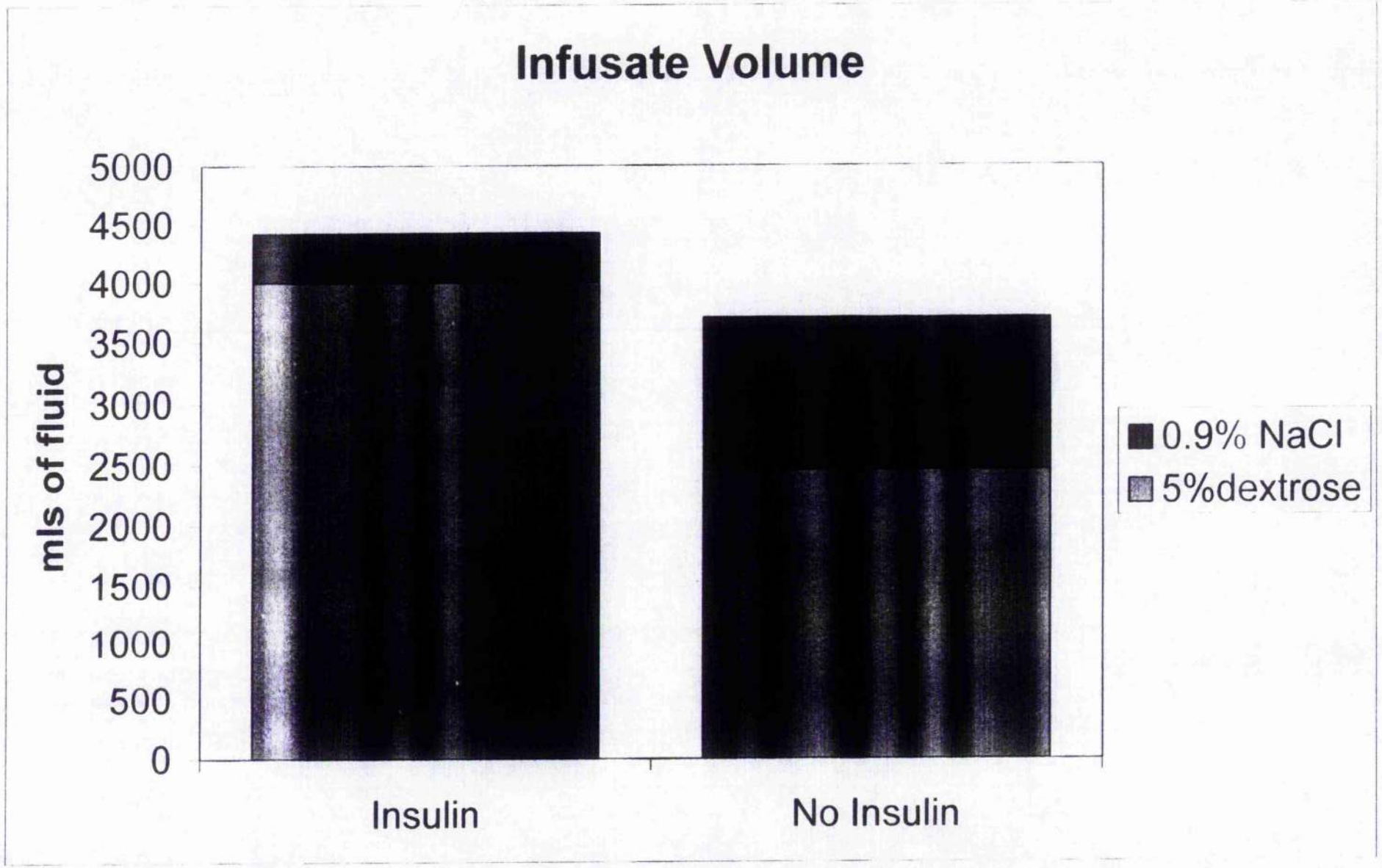
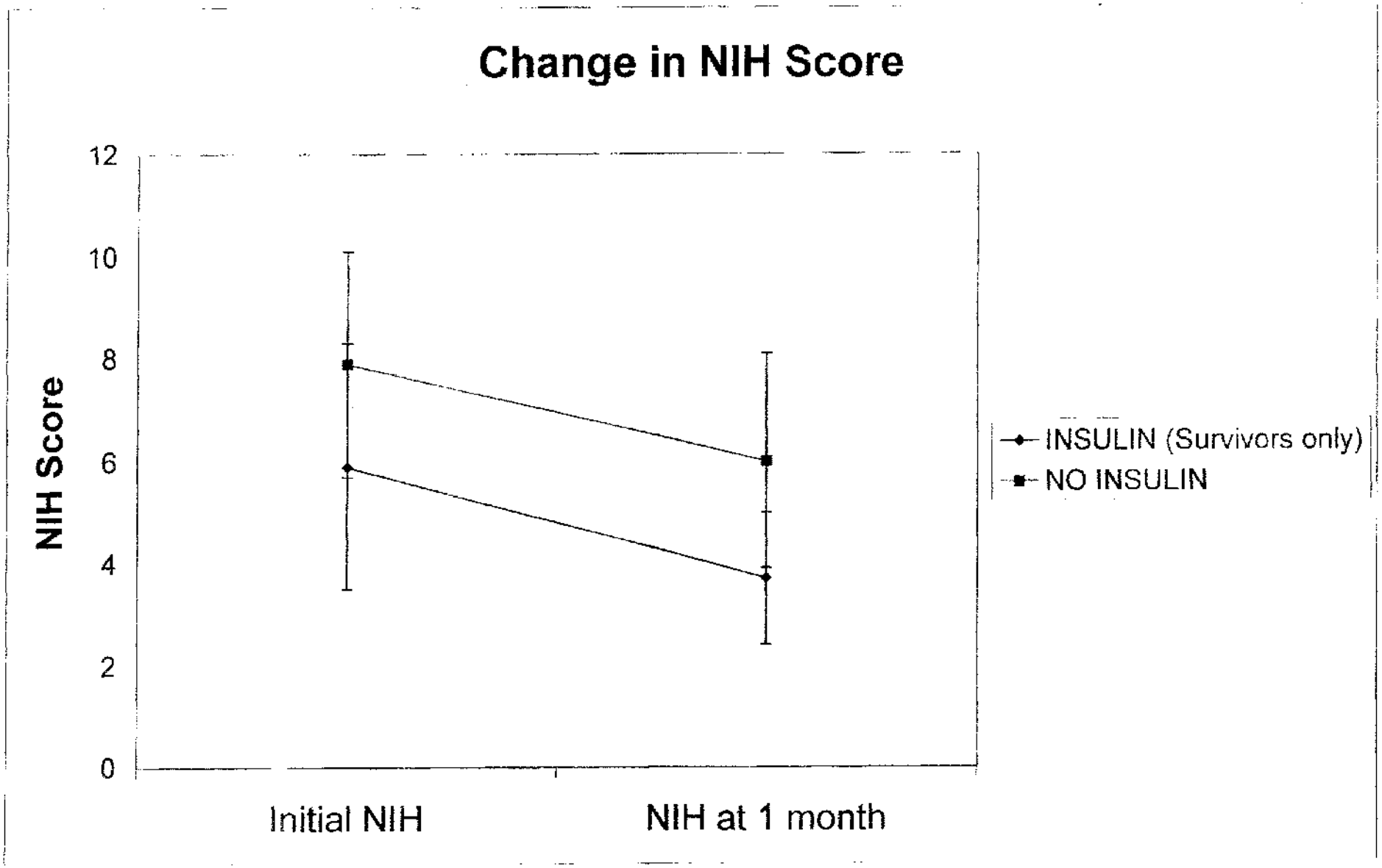
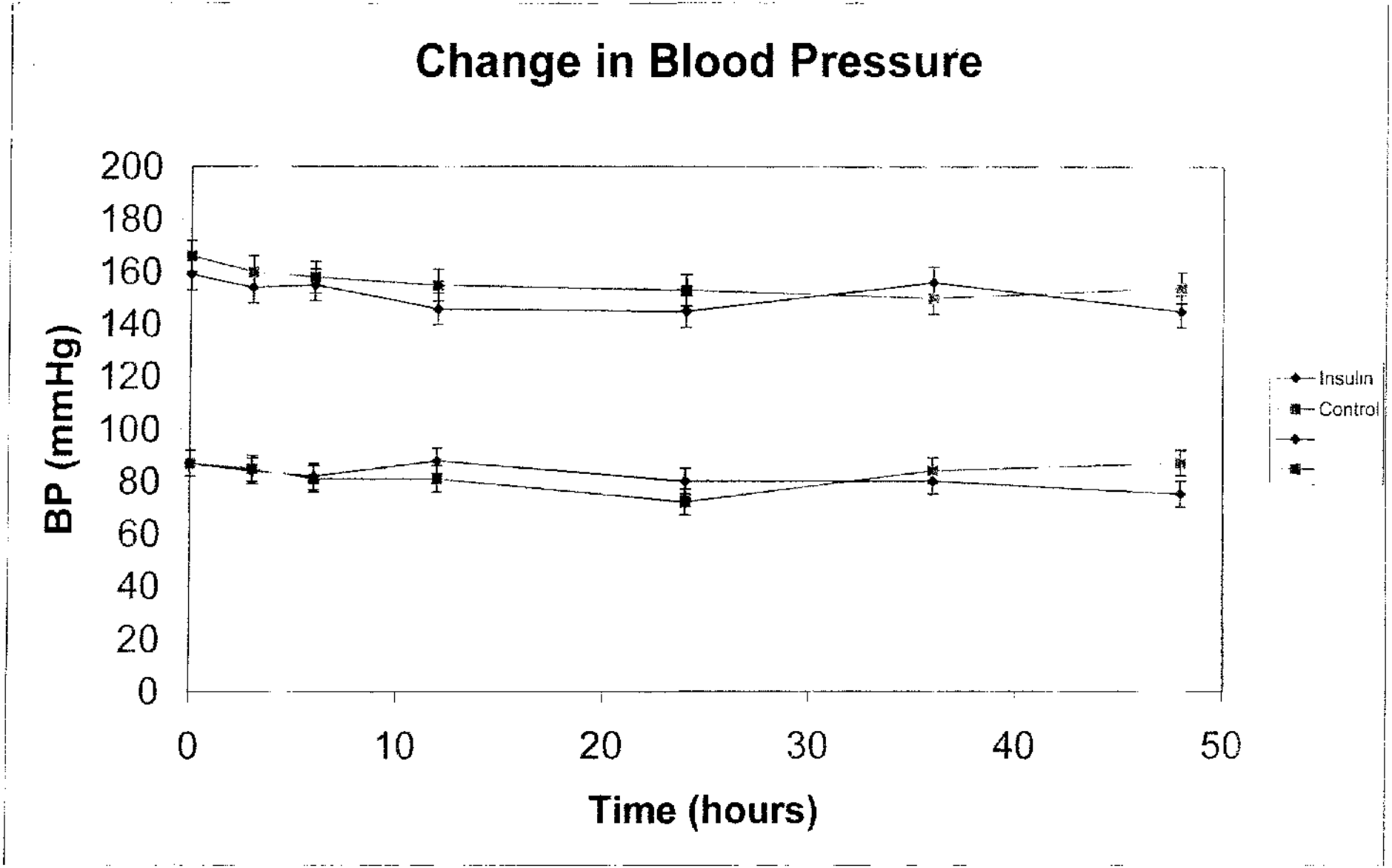


Figure 3.7 Change in NIH score



Error bars represent +/-2SE

Figure 3.8 Change in blood pressure



Error bars represent +/-2SE

Chapter Four

Low triglyceride, not low cholesterol concentration, independently predicts poor outcome following acute stroke

4.01 Introduction

Recent data from our group has shown an unexpected and 'counter-intuitive' association between poor stroke outcome and lower serum cholesterol.[138] This finding has been corroborated by others.[139] The biological plausibility for this relationship is uncertain, however It has been speculated that lower serum cholesterol in relatively elderly patients may simply reflect poor nutritional status, which could predispose to a poor outcome after stroke.[138] A second possibility is that high cholesterol post stroke may be protective through increasing gamma-GT;[140] this enzyme has a role in amino acid uptake and transport and thereby could reduce the neurotoxic effects of amino acids. Alternatively, the relationship between cholesterol and stroke may simply rely on (or be explained by) the immediate post stroke effects of inflammation on cholesterol concentrations.[141] Whatever the mechanism, it would seem sensible to caution on lipid lowering therapy for prevention of stroke until better prospective data are available.

Cholesterol, however, is not the only lipid parameter measured in relation to assessment of vascular risk. Triglyceride concentration has also been linked to coronary heart disease and stroke in prospective studies, and recent observations have tended to support the notion that there may be a clinically relevant interaction between cholesterol and triglycerides in the risk of coronary heart disease.[142,143] In addition there exists a strong association between triglyceride and insulin resistance. Indeed, hypertriglyceridaemic individuals are at increased risk of Type 2 diabetes mellitus, consistent with the key role of insulin resistance in both conditions.[144]

Despite such associations, there are currently no data on the relationship between triglyceride and stroke outcome. Such information may yield further mechanistic information on the relationship between lipids and outcome following stroke. For example, if poor outcome following stroke was unrelated to low triglyceride, then this would imply that the cholesterol link is specific, implicating an association of low LDL-cholesterol (± low HDL-cholesterol) concentrations to stroke outcome. If, on the other hand, low triglyceride were a strong predictor of stroke, then this would point towards an association also of triglyceride-rich lipoproteins with stroke outcome.

Alternatively, since hyperglycaemia is common following acute stroke and predicts a poor outcome,[70] there may be an underlying susceptibility to insulin resistance in such individuals.[145] In this scenario, we may anticipate an association of high triglyceride with poor stroke outcome, because as noted above hypertriglyceridaemia is a common feature of insulin resistance.

The aim of this long-term follow-up study therefore, was to explore the association between triglyceride and outcome following stroke. If an association was present, we wished also to examine whether this association was stronger than that of cholesterol with outcome following stroke.

4.02 Subjects and Methods

The Acute Stroke Unit of the Western Infirmary in Glasgow serves a catchment population of 220,000. All patients who present within 72 hours of onset of acute neurological deficit with no known alternative to a vascular cause are admitted irrespective of age or severity of the neurological deficit. Approximately 800 patients

are admitted each year. All patients have their stroke subtype categorised using the Oxfordshire Community Stroke Project clinical classification, and brain imaging using either CT or MRI is performed within 72 hours of admission on all patients with a clinical diagnosis of stroke. Clinical, radiological and biochemical data from each patient are reviewed and verified by a neurologist, radiologist and stroke physicians before being recorded prospectively on a computer database.

Biochemical data (including random glucose concentration and lipid profile) are obtained routinely from all patients on the day of admission. Cholesterol concentration was measured using a standard cholesterol oxidase antipyrine (CHOD-PAP) assay (Beckman diagnostics, Fullerton, Ca.). Triglyceride concentration was measured using a glyceryl phosphate (GPO) enzymatic technique. Both assays were performed using a Beckman CX4 analyser (Beckman diagnostics, Fullerton, Ca.). Fasting samples for glucose and lipid estimation are also drawn early the following morning, along with a standard battery of haematological and biochemical tests. In this study we used the random glucose concentration if it was taken; otherwise we used the fasting sample. Glucose concentration was recorded both as a continuous variable and as a binary variable (≤8mmol/l, normoglycaemic; >8mmol/l, hyperglycaemic). The upper limit of normal for a fasting plasma glucose concentration is 6.5mmol/l. As not all glucose measurements in our study were fasting, 8 mmol/l was taken as the cut point for hyperglycaemia. Other potential prognostic variables considered were age, stroke type (ischaemic or haemorrhagic), admission blood pressure (systolic and diastolic), smoking status, presence of atrial fibrillation, time to resolution of symptoms (≤72 hours or >72 hours) and Oxford Community Stroke Project Category. We examined the effect of known

surrogate biochemical markers of insulin resistance (serum urate concentration, serum triglyceride concentration, gamma GT concentration and serum cholesterol level) on outcome by constructing a proportional hazards regression model and incorporating them in the analysis, after adjusting for known prognostic factors. Patients in the study presented to our acute stroke unit between June 1990 and September 1997. Those with previously diagnosed diabetes or a final diagnosis other than acute stroke were excluded from the analysis.

Survival and placement follow-up were by record linkage to the Scottish deaths register and to a national database of hospital discharge records. The method of record linkage was validated in an epidemiological study of hypertension, and has been used for monitoring end points and adverse events in a large clinical trial.[146,147] Record linkage provides reliable patient follow up; however, admissions to private hospitals or to institutions outside Scotland are not detected.

4.03 Statistical analysis

Our main analysis used a stepwise Cox proportional hazards regression model[148] to estimate the effect of markers of insulin resistance (serum urate concentration, serum triglyceride concentration, gamma GT concentration and serum cholesterol level) on survival following stroke. A separate baseline survival function was fitted for each of five strata of patients defined by OCSP category and stroke type (primary intracerebral haemorrhage, total anterior circulation infarction, partial anterior circulation infarction, posterior circulation infarction, and lacunar infarction). This was done since these variables would not satisfy the proportional hazards assumption.

The effects of serum urate, triglyceride, gamma GT and cholesterol concentrations, were determined after correcting for other significant prognostic variables (selected from age, time to resolution of symptoms, smoking status, systolic blood pressure, diastolic blood pressure, mean arterial pressure, presence of atrial fibrillation and hyperglycaemia). The assumption of proportional hazards was checked for all variables included in the model. A quadratic term for the blood pressure measurements was permitted in the proportional hazards modelling.

4.04 Results

We studied 1312 non-diabetic patients with computed tomography confirmed acute stroke and biochemistry data available. The cohort used included those patients previously studied.[138] Blood samples were drawn in a fasting state within 24 hours of admission to our unit. The mean follow up time in survivors was 1196 days (range, 94 to 2764 days). Table 4.1 gives the distribution of the clinical and biochemical features studied.

With the exceptions of serum triglyceride level and the plasma glucose level (continuous variable), all variables satisfied the assumption of proportional hazards. In the case of plasma glucose level this was resolved by using the binary representation of the variable with 8 mmol/l as the cut-off point for hyperglycaemia. Categorisation of the triglyceride measurement into quartiles satisfied the proportional hazards assumption and this representation of the variable was therefore used. Table 4.2 gives the results of the proportional hazards modelling.

Increased age, presence of AF, hyperglycaemia, and lower triglyceride level independently predicted (all $p<0.001$) higher mortality. The early resolution of symptoms unsurprisingly was an independent predictor of lower mortality (Table 4.2). Although serum cholesterol level predicted outcome after adjusting for other prognostic factors, it did not remain significant when triglyceride level entered the model. Urate level and current smoking status did not independently predict mortality. Elevated systolic blood pressure, diastolic blood pressure and mean arterial pressure were not significant linear or quadratic predictors of higher mortality. Table 4.3 shows examples of the predicted 6 month survival for patients with different stroke types, serum triglyceride concentrations, and age. Figure 4.1 shows survival curves, estimated from the proportional hazards model, for the four quartiles of triglyceride level in a typical patient. Even at 6 months, there was approximately a 20% better survival in patients in the top quartile of triglyceride concentrations relative to those in the lowest quartile.

4.05 Discussion

Our data suggest that the concentration of triglyceride independently influences survival of patients with acute stroke. More importantly, triglyceride concentration was more strongly predictive of outcome than cholesterol and in fact, cholesterol concentration was not a significant independent predictor once triglyceride was entered into the model. We also examined the effect of Gamma-GT on outcome, but this parameter had no relationship to outcome.

What is the potential explanation of our results, and do they provide any further insights into potential biological mechanisms for these associations? First, the association of low triglyceride with outcome following acute stroke argues perhaps more strongly against an confounding effect of inflammation in this relationship. Butterworth et al[141] demonstrated that triglyceride concentration does not change in the first week after an ischaemic stroke; in contrast, they noted that cholesterol (and LDL-cholesterol) does fall following stroke and significantly so by 1 week. Second, since at a total fasting plasma triglyceride concentration of 1.3 mmol/L (the median concentration in our cohort), the majority of measured triglyceride circulates in the form of very low density lipoprotein particles[149] (which also carries cholesterol), our results would be more suggestive of an association of low VLDL (rather than or in addition to LDL) concentration with poor stroke outcome. In other words, it is the strong association of triglyceride in the form of VLDL with stroke outcome that accounts for the previously noted relationship of low cholesterol with outcome following stroke. This suggestion, however, requires direct examination in future studies. Third, the association of low triglyceride with poor outcome post stroke is consistent with a nutritional deficiency element. A significant fall in triglyceride can be effected with energy restriction, and independently of changes in weight.[150] In addition, there are abundant data to show that energy restriction reduces triglyceride much more than cholesterol,[150-152] indicating that triglyceride is a better indicator of poor nutritional status.

With respect to this nutritional argument, it is of interest to note the recent observation by Gillman et al[153] of an inverse association of dietary fat with

development of ischaemic stroke in men. Could low dietary fat intake (or status) also influence long term outcome following acute stroke?

Triglyceride also correlates strongly to body mass index and waist circumference, and again in both sexes does so better than cholesterol.[154] Unfortunately information on weight and waist circumferences was not collected in this study but clearly such parameters require investigation as predictors of outcome following stroke in future studies.

4.06 Conclusion

In conclusion, low fasting plasma triglyceride is a strong and independent predictor of poor outcome following stroke, whereas cholesterol is not. Clearly, future studies are needed to examine in greater detail the mechanisms explaining the association of lipids (and lipoproteins) with outcome following stroke.

Table 4.1 **Clinical and biochemical factors on presentation (n=1312)**

Median age	70
Male sex	646 (49)
Smoker	626 (48)
Median systolic blood pressure (mm Hg)	159
Median diastolic blood pressure (mm Hg)	90
Median mean arterial pressure (mm Hg)	113
Atrial fibrillation	114 (9)
Ischaemic stroke	1199 (91)
Oxford classification	
Total anterior circulation infarct	224 (17)
Partial anterior circulation infarct	441 (34)
Posterior circulation infarct	133 (10)
Lacunar infarct	386 (29)
Other	15 (1)
Hemorrhagic stroke	113 (9)
Symptoms resolved within 72 hours	170 (13)
Median plasma glucose (mmol / L)	6.1
Hyperglycemia	216 (16)
Median serum cholesterol (mmol / L)	5.8
Median serum triglyceride (mmol / L)	1.3
Median serum urate (mmol / L)	0.32
Median serum γ-GT (U / L)	29

Figures shown are number (percentage) of patients unless otherwise stated.

Table 4.2 **Proportional hazards modeling of mortality**

	Relative hazard	95% confidence interval	p-value
Increasing age (per decade)	1.56	(1.45 , 1.72)	< 0.0001
Increasing triglyceride level (per quartile)	0.84	(0.77 , 0.92)	< 0.0001
Hyperglycemia	1.47	(1.18 , 1.83)	0.001
Symptoms resolved within 72hrs	0.59	(0.41 , 0.85)	0.004
Atrial fibrillation	1.37	(1.08 , 1.75)	0.009

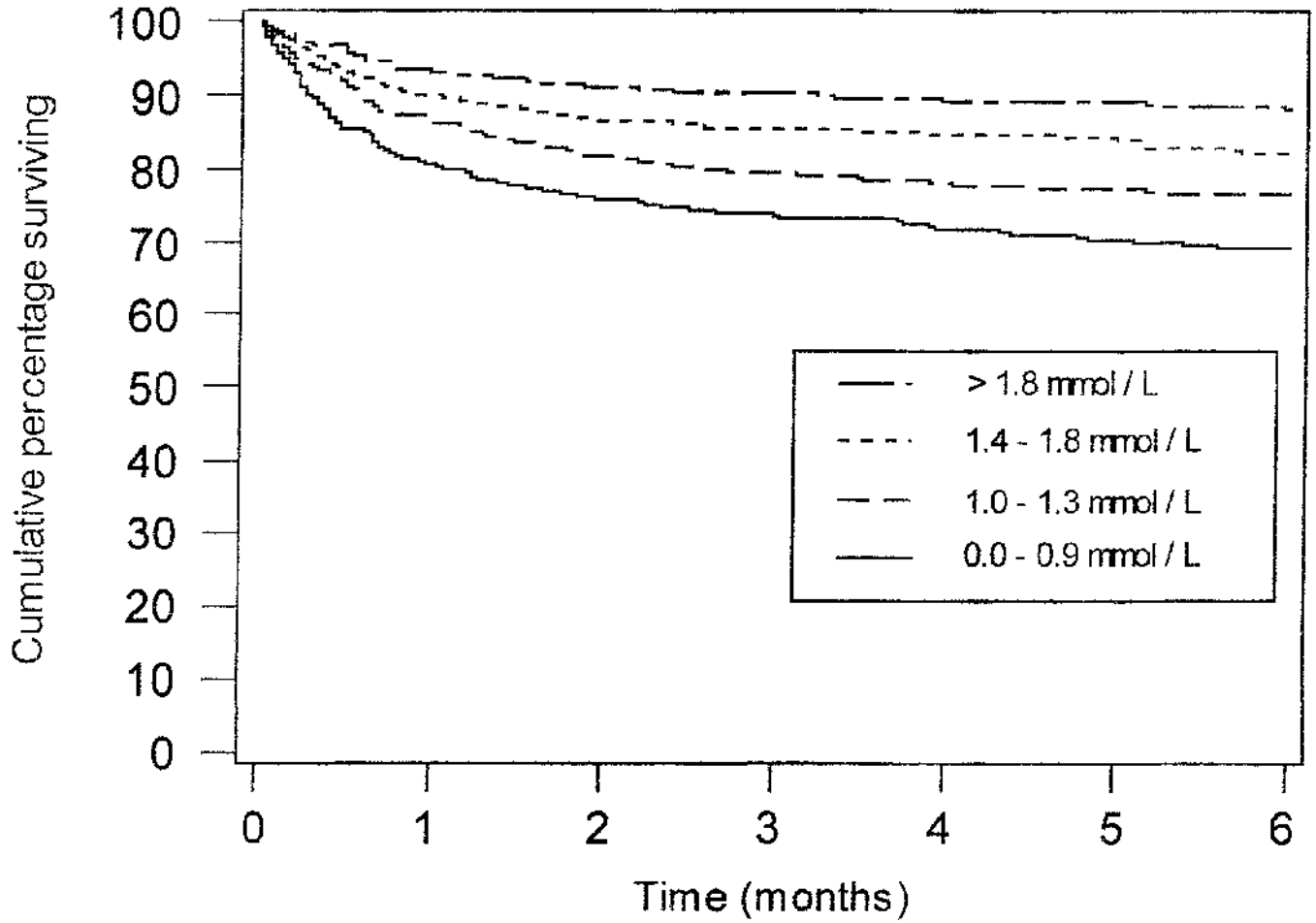
Table 4.3 **Survival rates estimated from proportional hazards model**

Oxford classification	Age	Triglyceride level	6-month survival rate (%)
Total anterior circulation infarct	50 years	0.8 mmol / L	80.7
Total anterior circulation infarct	50 years	2.5 mmol / L	88.1
Total anterior circulation infarct	75 years	0.8 mmol / L	51.1
Total anterior circulation infarct	75 years	2.5 mmol / L	67.1
Lacunar infarct	50 years	0.8 mmol / L	95.4
Lacunar infarct	50 years	2.5 mmol / L	97.2
Lacunar infarct	75 years	0.8 mmol / L	86.3
Lacunar infarct	75 years	2.5 mmol / L	91.6

Survival rates are calculated for patients with the above characteristics and without AF, early resolution of symptoms or elevated plasma glucose level. 0.8 mmol / L and 2.5 mmol / L are the median serum triglyceride concentrations within the lower and upper triglyceride quartiles respectively.

Figure 4.1 Triglyceride level and survival after stroke

Kaplan-Meier survival curves for the four quartiles of serum triglyceride level.


Cumulative percentage surviving
100
90
80
70
60
50
40
30
20
10
0
0
1
2
3
4
5
6
Time (months)
> 1.8 mmol / L
1.4 - 1.8 mmol / L
1.0 - 1.3 mmol / L
0.0 - 0.9 mmol / L

Chapter Five

The prognostic significance of visible infarction on computed tomography following lacunar stroke: Results of a long-term follow-up study

5.01 Introduction

Lacunar strokes are characterised by a number of well-recognised clinical features,[155] explained pathologically by small (0.2-15mm^3), deep infarcts caused by hypertension-induced changes in small perforating arteries. Lacunar syndromes are characterised by lack of impairment of higher cerebral functions such as praxis, language and gnosis. Symptoms are usually well-localised, the most common clinical manifestation being pure motor stroke. Other recognised lacunar syndromes comprise pure sensory stroke, sensorimotor stroke, dysarthria-clumsy hand syndrome and ataxic hemiparesis. As defined by the Oxford Community Stroke Project,[28] lacunar symptoms account for approximately 25% of all presentations with ischaemic stroke. Substantial variability in functional outcome, and relatively few factors predictive of death or degree of recovery have been observed in this group.[156] The recognised prognostic indicators following lacunar stroke are age, degree of neurological dysfunction, blood glucose concentration at presentation and functional disability one week after the index stroke.[156] Although computed tomography (CT) of patients with a clinical diagnosis of lacunar stroke performed within the first ten days shows evidence of cerebral infarction in 50-60%,[157] the prognostic significance of a visible ischaemic lesion on CT is unclear. A number of recent studies have addressed the prognostic value of CT appearances following cerebral infarction of any size or location[158-160] however only one[158] analysed the

significance of an appropriately-placed visible ischaemic lesion on CT (as opposed to no visible lesion) in a small (n=258) subgroup of patients with lacunar symptoms, using a multiple logistic regression model to adjust for confounding baseline variables. We used prospectively-acquired data from a larger cohort to compare the outcome of lacunar stroke patients in whom CT scanning had revealed an appropriate ischaemic lesion with those lacunar stroke patients in whom no such lesion had been identified.

5.02 Subjects and Methods

The Acute Stroke Unit of the Western Infirmary in Glasgow serves a catchment population of 220,000. All patients who present within 72 hours of onset of acute neurological deficit with no known alternative to a vascular cause are admitted irrespective of age or severity of neurological deficit. Approximately 800 patients are admitted each year. All patients are classified using the Oxford Community Stroke Project[28] classification, and brain imaging using either CT or MRI is performed within 72 hours of admission on all patients with a clinical diagnosis of stroke. Prior to July 1994, a Phillips Tomoscan 310 CT scanner was used to perform computed tomography on stroke patients; subsequently a Phillips Tomoscan SR7000 has been used. A standard protocol has been consistently employed, taking 5mm slices through the brainstem and 10mm slices through the cerebral hemispheres. Clinical and radiological data from each patient are reviewed and verified by a neurologist, radiologist and stroke physicians before being recorded prospectively on a computer database.

Using the database, we identified 633 patients who presented with symptoms consistent with lacunar stroke between June 1990 and February 1998. Of these patients, 114 were either imaged with CT at greater than 30 days following onset of symptoms, or imaged with magnetic resonance; these patients were excluded from the analysis. Forty-one patients with intracranial haemorrhage or non-cerebrovascular lesions on CT were also excluded. Due to the insensitivity of x-ray computed tomography in the detection of early cerebral ischaemia, the 57 patients in whom CT scans had been performed within 12 hours of onset of symptoms were not included in the analysis. A further 17 patients were excluded due to incomplete follow-up data.

The remaining 404 patients were divided into two groups, depending on the appearance of the CT scan. Patients with a low-attenuation area on the CT scan consistent with an ischaemic lesion in an appropriate region of brain to explain the presenting symptoms were classified as "CT positive". Patients with either a normal CT scan of brain or a scan which showed a lesion in an area inconsistent with the presenting symptoms were classified as "CT negative".

A series of known or suspected prognostic factors were recorded for each patient: blood pressure (systolic, diastolic, and mean arterial), age, smoking, plasma glucose level, serum cholesterol level and serum triglyceride level. Delay from stroke onset to scanning was also noted.

Patients were followed up for survival and subsequent hospital admissions using record linkage[147] to the national registers of deaths and hospital discharge records (SMR1). The technique of record linkage has been validated as a method of endpoint detection in a large clinical trial.[151] Record linkage is a reliable method: false positive and false negative linkage rates are approximately 1%.[147] The method does not detect admissions to private hospitals or episodes of care in institutions outside Scotland.

We considered three outcome measures: survival time, outcome at 6 months after the stroke and total hospital length of stay for the stroke admission. Six month outcome was categorised as good (alive at home) or poor (alive in care or dead).

5.03 Statistical Methods

Survival times in patients with and without visible infarction on CT were compared using a log-rank test. A proportional hazards model[148] was then used to assess the effect of visible infarction on survival after correcting for other clinical and biochemical factors known or suspected to influence prognosis.

A comparison of six-month outcome with respect to presence or absence of infarction on CT was performed using a chi-squared test. We then used stepwise logistic regression to adjust for significant prognostic factors before assessing the effect of CT findings on six-month outcome.

A Mann-Whitney test was used to compare median total length of hospital stay in patients with and without visible infarction on CT.

5.04 Results

We identified 404 patients who had been admitted to the Acute Stroke Unit of the Western Infirmary with a clinical diagnosis of lacunar stroke and who met the other criteria detailed above. Of these, 142 (35.1%) were scanned between 12 and 24 hours of onset of symptoms, 183 (45.3%) were scanned between one and three days of onset, and 79 (19.6%) were scanned between three days and thirty days after onset. Only seven of these patients were scanned at an interval of greater than 10 days after stroke onset: this delay was due to the CT scanner being broken. The mean elapsed time between onset of symptoms and CT scan was 2.7 days. Table 5.1 describes the clinical, radiological and biochemical features of the patient group.

Figure 5.1 shows the Kaplan-Meier survival curves for CT positive and CT negative patients. There was no difference in survival between the two groups ($p=0.29$, log-rank test). After adjusting for other significant prognostic factors (age, $p<0.0001$; plasma glucose level, $p=0.03$) in a proportional hazards model, presence of visible infarction remained non-significant ($p=0.40$). Table 5.2 gives the results of the proportional hazards modelling.

After adjustment for the other significant factor (age, p=0.0001), there was no significant difference in six month outcome between CT positive and CT negative patients (p=0.61; see table 3). Median total length of hospital stay was not significantly different between the two groups (CT positive, 9 days; CT negative, 8 days; Mann-Whitney test, p=0.29).

5.05 Discussion

Our data suggest that following lacunar stroke, the presence of visible infarction on CT scan performed between twelve hours and thirty days of ictus is not predictive of outcome or duration of hospital stay. This conclusion is consistent with other published work[159,160] however unlike these studies, our findings are based upon observations from a large cohort of patients with correction for other factors known to predict outcome and long-term follow-up. A further study of the prognostic significance of computed tomography findings following stroke in which a subgroup of patients with lacunar stroke was examined reached different conclusions.[158]

In this previous study, a more complex multiple logistic regression model was used to investigate the prognostic effect of visible infarction in 258 patients with lacunar stroke, and the authors reported that visible infarction was an adverse prognostic indicator of borderline significance. Delay from onset of symptoms to CT scan was a significant prognostic factor. This was not the case in our study, since all patients were scanned according to our Acute Stroke Unit protocol and

severity of deficit did not substantially influence the delay in scanning. Addition of the scan delay variable to any of the analyses presented here did not alter any of the results.

Other methodological differences between our study and that of Wardlaw et al[156] are less likely to account for the difference in results. We used a less complex model with fewer prognostic variables, although in the subgroup of patients with lacunar symptoms parameters used in the previous study's model such as diminution in conscious level and language disorder would not, by definition, have been of relevance. Our study used one radiologist to interpret the CT scans, unblinded to clinical data. The previous study used several similarly unblinded observers, however inter-observer variability in the interpretation of CT brain scans has been shown to be satisfactory.[162-165]

A further possible explanation for the disparity in results between the two studies lies in the assessment of the CT images. An association between infarct volume on CT scan and functional outcome in patients with lacunar and other stroke subtypes has been reported;[165] it is possible that the cruder dichotomisation of scan appearance into "present" or "absent" in both studies may have masked an underlying concordance in data.

5.06 Conclusion

We conclude that in our cohort of patients, having corrected for other prognostic variables, the presence of visible infarction on CT brain scan performed between twelve hours and thirty days of onset of lacunar symptoms is not predictive of duration of hospital stay or of longer-term outcome.

Table 5.1 **Characteristics of patients studied**

	n=404
Male sex	201 (49.8)
Infarct visible on CT scan	170 (42.0)
Current smoker	163 (41.7)
Delay (days from stroke onset to CT scan)	2 (1 to 3)
Age	71 (61 to 79)
Blood pressure	
Systolic	160 (142 to 185)
Diastolic	90 (80 to 100)
Mean arterial	113 (102 to 127)
Plasma glucose level (mmol/lit)	6.0 (5.1 to 7.3)
Serum cholesterol level (mmol/lit)	5.7 (4.9 to 6.6)
Serum triglyceride level (mmol/lit)	1.4 (1.0 to 2.0)

For the variables sex, visible infarction and current smoker, data are presented as number of patients (percentage of patients). The remaining variables are described as median (interquartile range).

Table 5.2 **Results of proportional hazards modelling of survival**

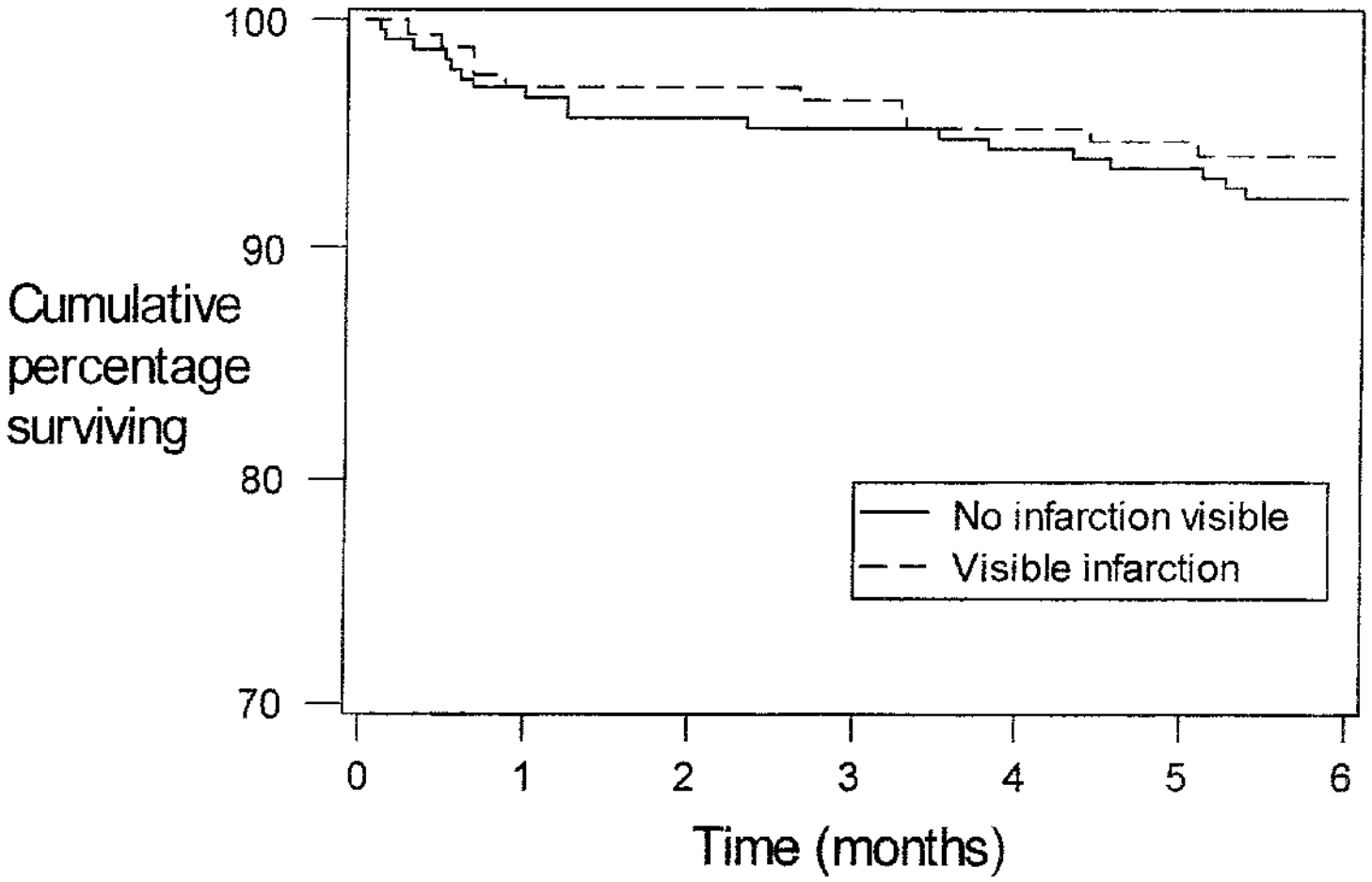
Variable	Relative hazard	95% confidence interval	p-value
Age (per decade)	1.67	1.37 to 2.02	<0.0001
Plasma glucose level (per mmol/l)	1.08	1.01 to 1.16	0.03
Infarction visible on CT	0.84	0.54 to 1.28	0.40

Table 5.3 **Six-month outcome: results of logistic regression analysis**

Variable	Odds ratio	95% confidence interval	p-value
Age (per decade)	0.52	0.39 to 0.71	0.0001
Infarction visible on CT	1.19	0.63 to 2.23	0.61

Odds ratios refer to the odds of good (alive, at home) versus poor (alive in care or dead) outcome.

Figure 5.1 Kaplan-Meier survival curves for CT positive and CT negative patients



Chapter Six

Diagnostic diffusion-weighted magnetic resonance imaging of acute ischaemic stroke at 1.0 Tesla

6.01 Introduction

Magnetic resonance imaging has long been used in the investigation of structural pathology of the brain. More recently, it has been used to investigate pathophysiological processes such as acute cerebral ischaemia. With the advent of thrombolytic treatment for ischaemic stroke, the ability to image ischaemic brain tissue within the therapeutic time window of 180 minutes has become clinically advantageous.[167] The accurate localisation and quantification of ischaemic damage in the hyperacute phase of stroke may also permit more rational assessment of the effects of neuroprotective therapies. Conventional magnetic resonance sequences are unable reliably to identify ischaemic brain tissue within 6 hours of onset of acute ischaemic stroke.[168] Diffusion-weighted magnetic resonance imaging (DW-MRI) is highly sensitive to changes in the diffusion of water molecules through tissue, such as those which occur in acutely-ischaemic brain. As discussed in chapter one, the signal intensity of a tissue being imaged with magnetic resonance depends upon proton density, T1, T2 and T2* relaxation processes of the spins within each imaging element or voxel. The random diffusion of protons within the tissue being studied will lead to random shifts in spin orientation and consequent loss of signal. The rate and direction of proton diffusion are affected by molecular and macromolecular barriers to diffusion, hence water molecules within tissue travel approximately half of the distance travelled by free water molecules in the same time interval. The potential clinical application of diffusion-weighted imaging was realised when abnormal DW-MRI sequences in association with normal T1 and T2 weighted

MRI studies were observed in the cat middle cerebral artery occlusion model within minutes of onset of ischaemia.[169-175] In experimental models, reduction in cerebral blood flow to 15-20 ml/100g/minute is associated with a profound fall in the diffusibility of water through the ischaemic tissue, termed the apparent diffusion coefficient (ADC).[172] This fall in ADC arises as a result of influx of water molecules through the membranes of ischaemic cells. Once trapped within ischaemic cells, the diffusion of water molecules is slowed and restricted. Clinical studies[176-179] have confirmed the ability of this technique to identify early ischaemic cerebral damage in stroke patients before any abnormalities are seen with conventional imaging techniques such as x-ray computed tomography or T2-weighted magnetic resonance scanning.

As DW-MRI is sensitive to the Brownian motion of water molecules within brain tissue, the technique is highly susceptible to macroscopic patient movement artefact. Severe image distortion may arise as a result of voluntary or involuntary patient head movement, breathing motion, and brain and CSF pulsation as a result of cardiac systole. Large-scale movement of the tissue under examination causes severe "ghosting" artefact in the image which often renders it difficult to interpret. The two main methods of compensation for patient movement are the use of navigator echoes[180-184] to correct for movement, or rapid image acquisition with echo-planar techniques. Navigator echoes involve the addition of an extra spin echo to the sequence, which enables motion-induced phase variations to be identified and corrected. Cardiac gating is also used to reduce image distortion

arising from brain pulsation further. Although successful, the use of navigator echoes requires extensive image post-processing which significantly prolongs overall scan time and hence detracts from the usefulness of the technique in the context of acute stroke. With improved hardware, distortion due to patient movement can be reduced by obtaining the images in less than one second using Echo-Planar Imaging (EPI).[185-187] This technique uses rapid gradient pulses to produce an image within 100 ms. Although EPI allows rapid image acquisition, image quality is often compromised by distortion artefacts that occur at boundaries of regions with differing magnetic resonance characteristics, such as air-tissue interfaces. As a result of this distortion, areas of brain adjacent to the sinuses are often very poorly visualised during echo-planar imaging. Only with machines at field strengths of at least 1.5 T have the manufacturers made concerted efforts to resolve these distortions. Hence to date the implementation of DW-MRI has been hindered and restricted to major centres and optimised research sites. Therefore, in a centre with a 1.0 T magnet an alternative approach to EPI is required. To attempt to overcome this difficulty we reverted to a compromise between acquisition time and image quality, the use of a time-reversed steady state sequence (given the acronym "PSIF" as it is a reversed form of the fast imaging with steady-state precession or "FISP" sequence) which had previously been used at high (1.5 T) magnetic field strength.[188] We developed the sequence further and optimised it for use on our standard 1.0 Tesla scanner. The partial flip angle and low repetition time used by the PSIF sequence enables the stimulated echo (or "left-over") component of the signal to

be detected and used to generate an image.[189] The application of a diffusion gradient during each repetition allows a heavily diffusion-weighted stimulated echo to be measured. A graphical representation of the pulse sequence timing diagram is shown in figure 6.1. We sought to investigate the clinical utility of such a sequence at 1.0 T in the investigation of patients with ischaemic stroke.

6.02 Methods

Patients

The Acute Stroke Unit of the Western Infirmary in Glasgow serves a catchment population of 220,000. All patients who present within 72 hours of onset of acute neurological deficit with no known alternative to a vascular cause are admitted, irrespective of age or of severity of neurological deficit. Patients are classified using the Oxford Community Stroke Project[28] classification. Between June and September 1998, consecutive patients presenting to the Acute Stroke Unit with a provisional diagnosis of stroke within 8 hours of onset of neurological deficit and with no contra-indications to MR scanning underwent both conventional and diffusion-weighted magnetic resonance imaging. A full explanation of the procedure was given to each patient and verbal consent was obtained prior to imaging.

Imaging Protocol

A Siemens 1.0 T Impact Expert Scanner was used with a PSIF based diffusion weighted sequence programmed using the Pargen Sequence Editor.

Parameters were:

Repetition Time 29 ms Echo Time 9 ms Flip Angle 50°

Field of View 240 mm Slice Thickness 6 mm.

A strong diffusion gradient of 16.5 mTm^{-1} was applied for 7 ms. A weak gradient of 3 mTm^{-1} was also used.

Five acquisitions were taken to ensure good signal-to-noise in the image and to minimise any remaining artefact.

Diffusion weighted imaging was performed by taking transverse sections through the head to determine the possible location of an acute stroke. On definite depiction of the stroke area, strong diffusion weighted imaging was repeated in the other two gradient directions.

Baseline images using a very weak diffusion weighting setting were also acquired for comparative purposes. All images were reviewed at the time of acquisition by a consultant radiologist. The addition of the DW sequence to the conventional series of sequences prolonged overall scan time by approximately ten minutes.

6.03 Results

Sequence optimisation

A standard water phantom was used to optimise the DW-PSIF sequence. Optimisation was achieved by increasing the diffusion gradient amplitude and

limiting the repetition time. If the repetition time is increased then the scan time will increase, and as a result the images will be more susceptible to patient movement artefact. If the diffusion gradient is increased then the images will also be more susceptible to patient movement artefact as the scan will become more sensitive to tiny movements.

The signal (S) measured in the presence of the diffusion gradient can be described as

$$S = S_o\, e(-bD^*)$$

Where S_o is the signal intensity in the absence of a diffusion gradient, D^* is the apparent diffusion coefficient of the tissue and b is a measure of the diffusion sensitivity.

The diffusion sensitivity, b, is proportional to the square of the gradient strength, although it is also dependent on the type of diffusion sequence used.
When the natural log of (S/S_o) is plotted as a function of the b value, the slope of the resultant line is –bD. Determination of the b value of a diffusion-weighted PSIF sequence is mathematically complex, as the stimulated echo component of the signal is used to generate the image. The natural log of S/So was plotted against the square of the gradient strength, and the gradient of (b/k)D was found to be –0.00423 (figure 6.2) for the original sequence, and –0.0078 (figure 6.3) for the optimised sequence. (In this calculation, k represents the constant necessary

to adjust for the use of a stimulated echo sequence.) The optimisation process had therefore increased the sensitivity of the sequence by a factor of 1.84.

Using a standard echo-planar diffusion-weighted sequence with a known b value of 1000 and a standard water phantom, a calibration experiment was performed. The natural log of S/So using the echo-planar sequence was –2.26; the optimised PSIF sequence yielded a value of –2.3 when the same phantom was used. The diffusion sensitivity of the optimised PSIF sequence was therefore calculated to be 1025 s/mm^2. The relative gradients of the original and optimised sequences are shown in Figure 6.4.

Clinical application

10 patients (8 male, 2 female) were scanned. Median age was 66.5 years (range 50-83 years), median NIH score on presentation was 5 (range 1-16). 2 patients had symptoms and signs consistent with total anterior circulation stroke, 3 had partial anterior circulation stroke, 2 patient with lacunar symptoms and 2 patients had posterior circulation stroke. One patient with transient global amnesia was scanned at 6 hours after onset of symptoms.

All patients were scanned within 8 hours of onset of neurological deficit. Median elapsed time between onset and scan was 5 hours 55 minutes (range 2 hours 35 minutes to 7 hours 10 minutes). Clinical and radiological findings are summarised in table 6.1. Evidence of cerebral ischaemia was seen as an area of

signal hyperintensity in the DW-MRI sequences of all patients scanned. In 5 patients, corresponding T2-weighted MRI sequences revealed no overt abnormality. In these cases, the abnormal diffusion-weighted image provided strong radiological evidence to support a diagnosis of ischaemic stroke. In two of these five cases (patients 6 and 10) a differential diagnosis of migraine had been considered as a cause for their symptoms (right hemiparesis and right-sided visual disturbance respectively). The abnormal diffusion-weighted images allowed exclusion of this possibility and influenced early management. Figure 6.5 shows the images acquired from patient 10.

In cases 5 and 8 (figure 6.6), evidence of previous cerebral infarction was apparent on T2-weighted MRI sequences. The images from patient 8 are shown in figure 6.6. As diffusion-weighted images are sensitive only to recent ischaemic lesions, the DWI sequence in these patients allowed differentiation of old lesions from new areas of cerebral ischaemia. The cause of a further episode of focal neurological disturbance in patients with previous stroke is often elusive. Such patients are at increased risk of further cerebrovascular events, however they may also be prone to seizure activity as a result of previous cerebral injury. In order to provide appropriate therapy, it is important to be able to distinguish these two causes. Diffusion-weighted imaging allowed positive identification of a new cerebral ischaemic lesion in these patients, and management was influenced accordingly.

In patient 1, tiny parietal lesions were seen on the diffusion-weighted images, which did not register on the T2-weighted sequence. The spatial resolution of the PSIF sequence enabled these lesions to be visualised; it is notable that the coarser matrix used by many echo-planar diffusion weighted sequences would not reliably detect such lesions. Figure 6.7a shows the diffusion-weighted PSIF image, figure 6.7b shows the same image when displayed using a coarser matrix, typical of EPI images from a 1.5T scanner.

Repeat imaging at an interval of one month was performed in three patients. In each case established cerebral infarction was seen in the area identified by the DW sequence. Early and interval images from patient 7 are given in figure 6.8. A lesion in the subcortical area of the left parietal lobe was demonstrated on the early diffusion-weighted image; interval imaging with CT confirmed established cerebral infarction involving this area.

6.04 Discussion

Diffusion-weighted MRI has the potential to be widely applied in both clinical and research environments. Recent trials of thrombolytic therapy for acute ischaemic stroke have emphasised the difficulties in the interpretation of CT imaging acutely following stroke.[101,102] Diffusion-weighted magnetic resonance imaging will enable early, accurate diagnosis, quantification and topographical localisation of early ischaemic stroke. It has been proposed that, in combination with perfusion imaging, diffusion-weighted MRI may represent a more sensitive

tool for identifying patients most likely to respond to thrombolytic therapy,[190] and one early study appears to support that hypothesis.[166]

Diffusion-weighted MRI also has potential application in the development of novel neuroprotective therapies. As the clinical course of stroke is highly variable, efficacy studies using clinical endpoints require large numbers of recruits to demonstrate an effect. As neuroprotective therapy should reduce the size of the eventual damage to the brain, the ability to assess changes in infarct volume over time using both DW-MRI and T2-weighted imaging may become a rational useful surrogate endpoint in the evaluation of new drugs. As the pathophysiology of white matter ischaemia differs from that of cortical ischaemia, it may be that pharmacologically different agents will be required to treat ischaemic stroke, depending upon the area of brain affected. Accurate, early localisation of cerebral ischaemia will be necessary to enable administration of the appropriate drug.

The high field-strength scanners required for diffusion-weighted imaging are not widely available in the UK centres where patients with stroke are routinely managed. Our aim in this study was to develop and validate a diffusion-weighted MRI sequence which operates using a conventional MRI scanner and hence has the potential to expand the availability of diffusion-weighted imaging. Due to the pragmatic nature of the study, we did not attempt to quantify restricted diffusion by calculating apparent diffusion coefficient (ADC), nor did we generate ADC

maps. Although theoretically possible using our sequence,[191] ADC map generation would have significantly increased the duration of the scan.

6.05 Conclusion

In conclusion, we have found that clinically useful DW-MRI imaging of acute ischaemic stroke is possible using a 1.0 T scanner without EPI. Although analysis of ADC values has been shown to provide additional information particularly on the age of the lesion,[192,193] we have found that omission of ADC calculation does not significantly detract from the clinical utility of the sequence. Recent literature has confirmed the robust and reliable nature of diffusion-weighted MRI in the characterisation of early cerebral ischaemia.[166,194] A strong case for the use of magnetic resonance (including diffusion-weighted MR) as the principal imaging modality for initial investigation of the acute stroke syndrome is emerging, and on the basis of accumulating evidence more widespread implementation of diffusion MR has been proposed.[195]

The ability to perform DW-MRI on a 1.0 T scanner should enhance the applicability of the technique, and improve the ability of centres with conventional MRI scanners to investigate patients with acute ischaemic stroke.

Table 6.1 **Patient Characteristics**

Number	Age/Sex	Delay	OCSP	T2 MR	DW MR
1	51 M	5'45	L PACS	Scattered subcortical high signal	L parietal infarct
2	73 M	4'00	L POCS	Normal	L pontine infarct
3	58 M	6'00	TGA	Normal	L medial temporal high signal
4	81 F	6'10	L PACS	Atrophy. Nil focal	L parietal infarct
5	53 M	5'00	L LACS	Old R pontine infarct	New L subcortical infarct
6	66 M	6'30	L LACS	Normal	L subcortical infarct
7	67 M	2'35	L TACS	Normal	L parietal infarct
8	83 F	7'10	L TACS	Extensive infarction L cerebral hemisphere	New L subcortical infarct
9	82 M	6'15	L PACS	Diffuse small vessel disease. Nil focal	L frontoparietal infarct
10	50 M	4'45	L POCS	Normal	L occipital infarct

Figure 6.1 Pulse sequence timing diagram of the PSIF sequence.

This diagram illustrates the timing of introduction of the radiofrequency pulse (top line) and subsequent application of magnetic gradients in each of the three orthogonal planes (phase, read and slice).

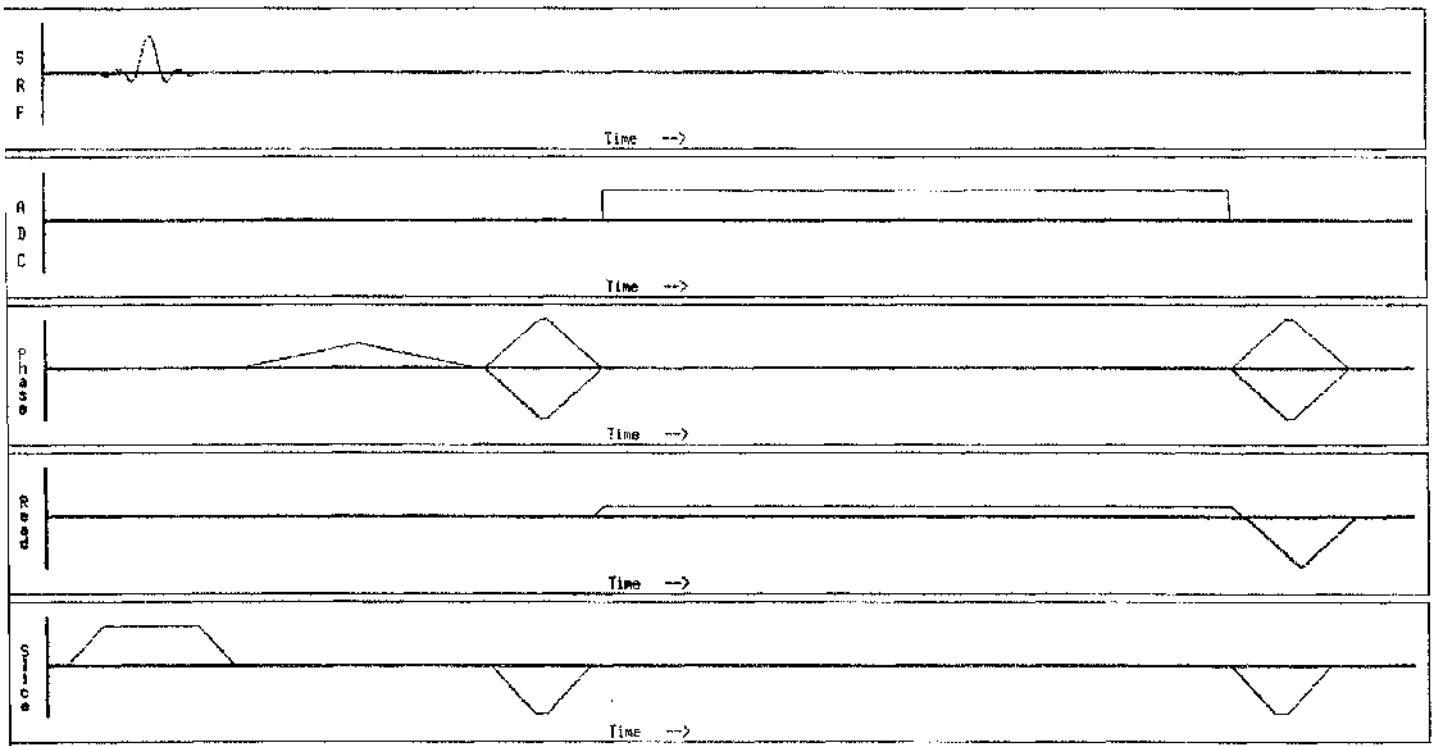


Figure 6.2 Calculation of b value of original sequence.

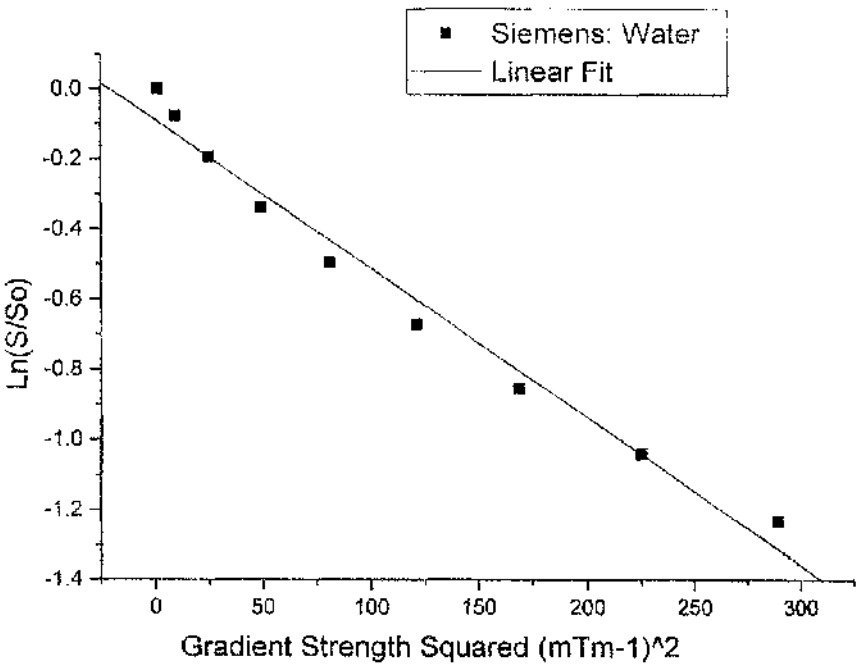


Figure 6.3 Calculation of b value of optimised sequence.

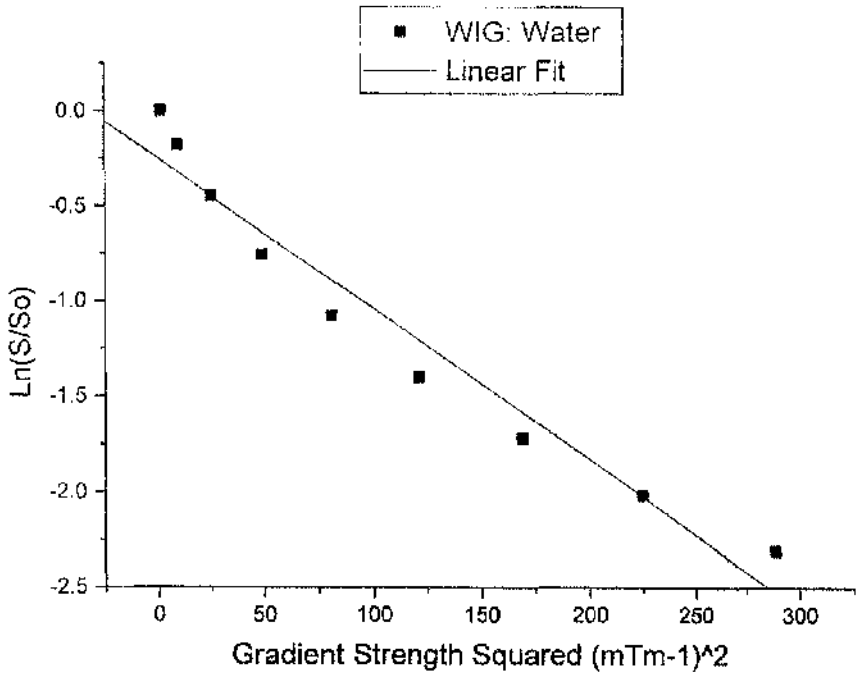
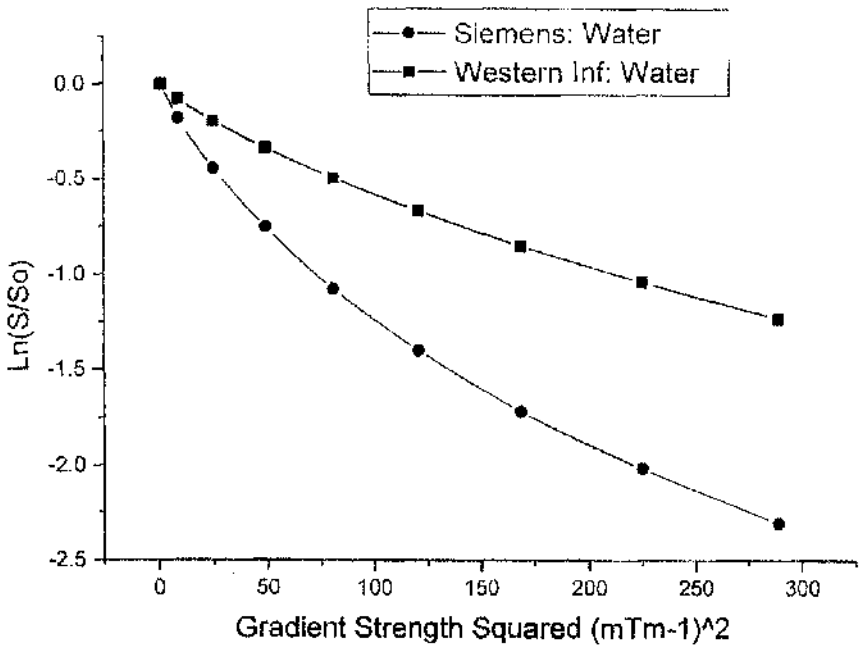


Figure 6.4 Comparison of gradients of original and optimised sequences.



Legends to Figures

Figure 6.5a T2 Weighted scan of 50 year-old man with right homonymous hemianopia of 7 hours' duration.

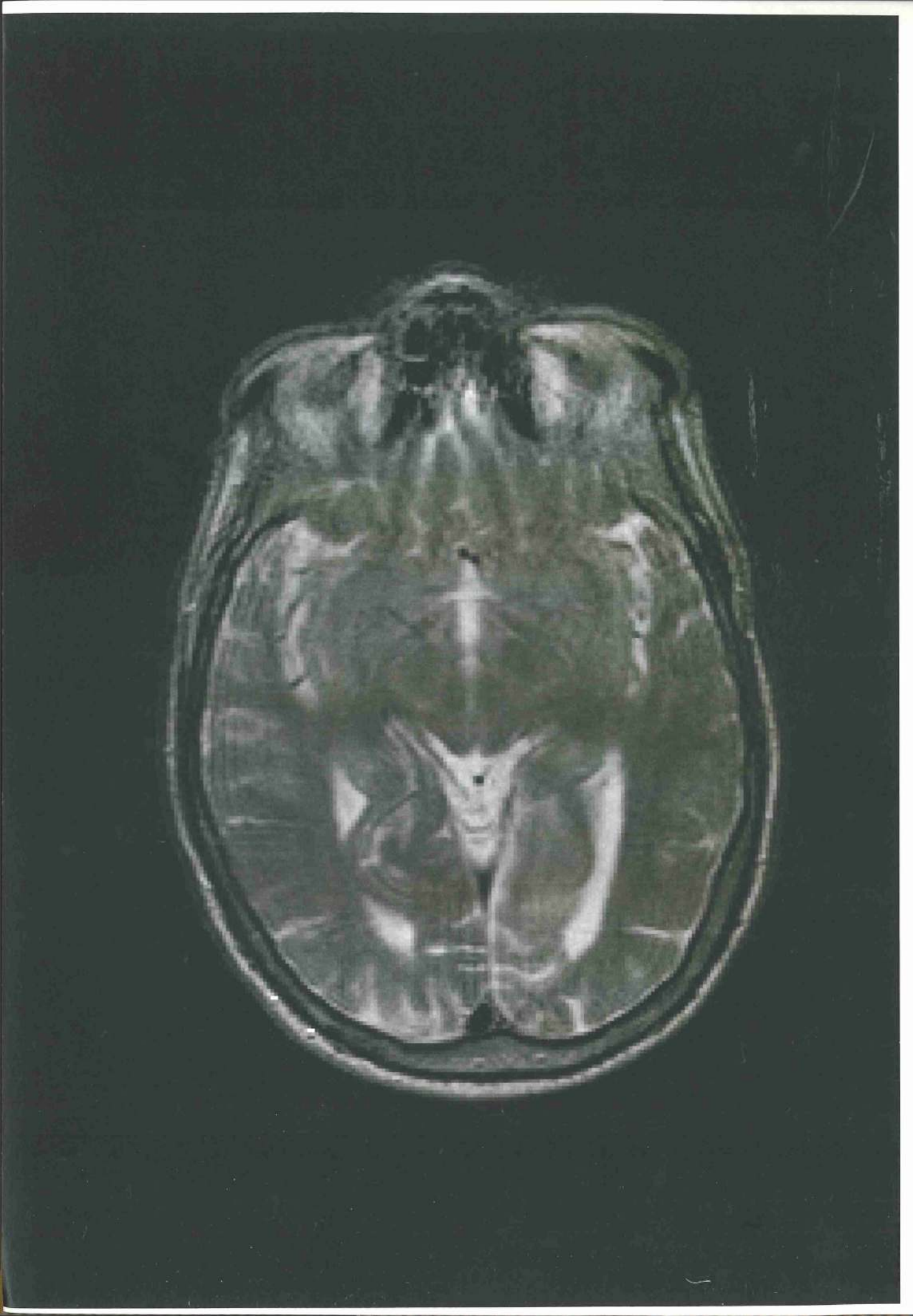


Figure 6.5b Diffusion weighted image of same patient taken several minutes earlier.

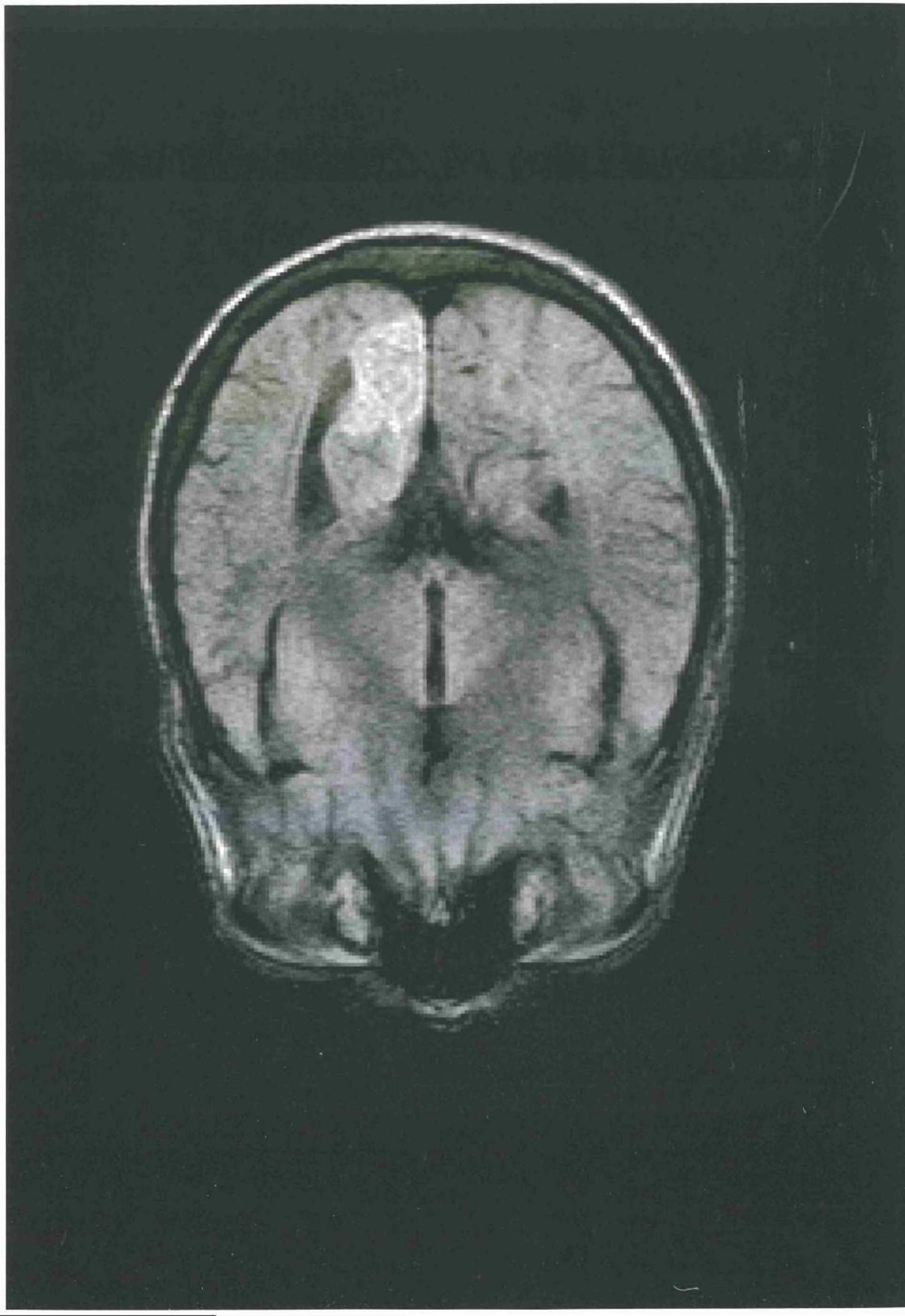


Figure 6.6a T2 Weighted scan of 83 year-old woman with sudden deterioration in longstanding right limb weakness.

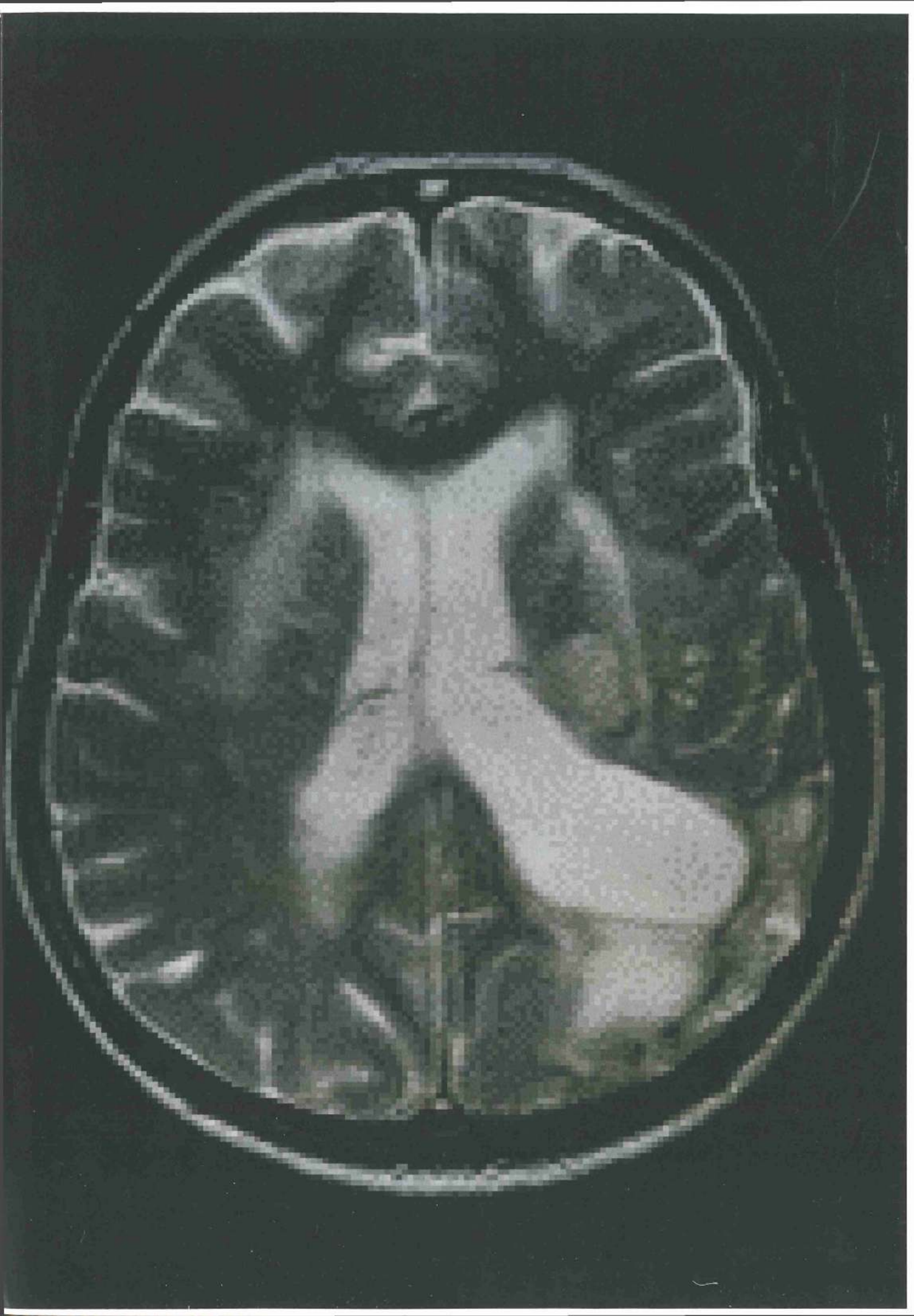
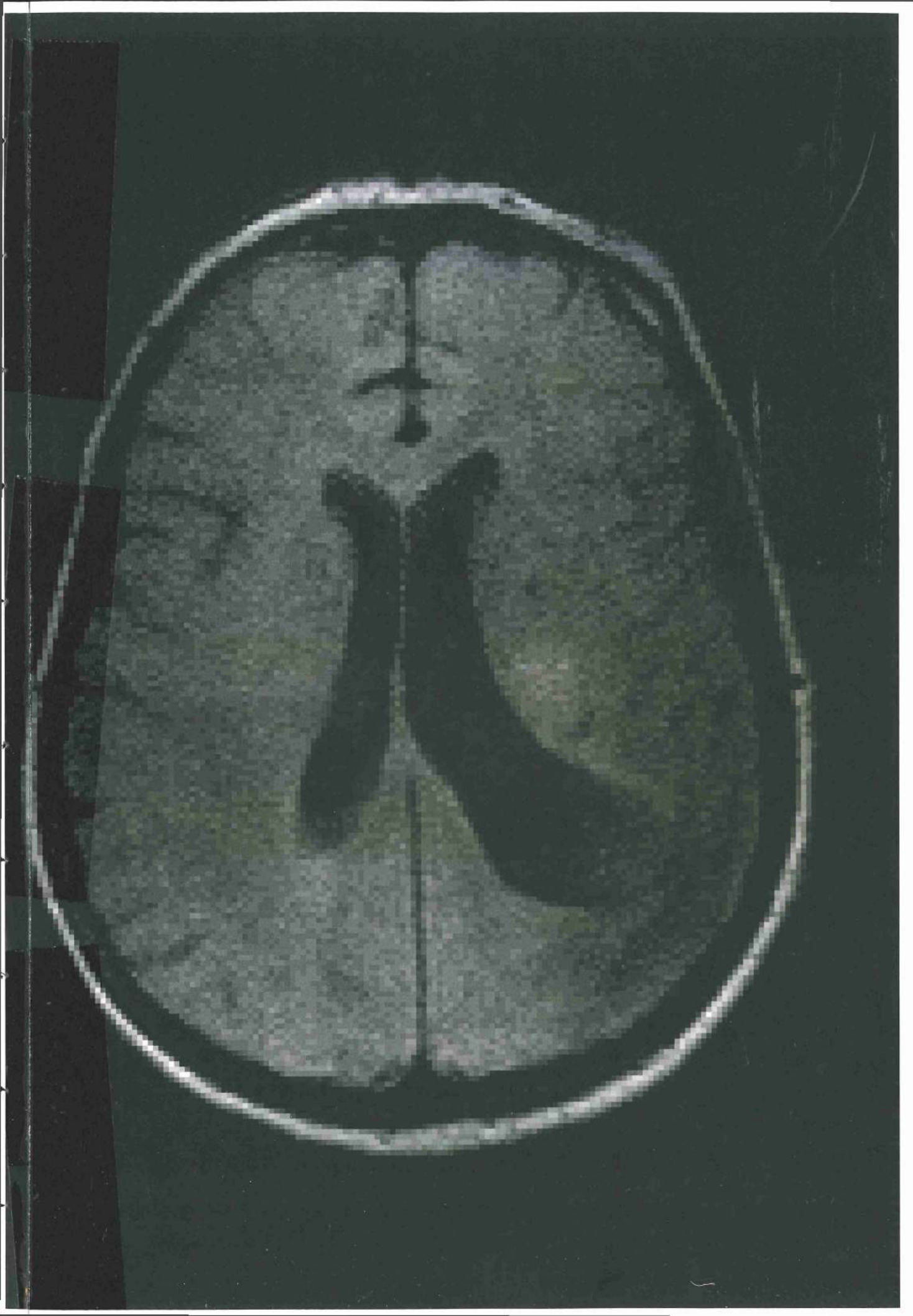
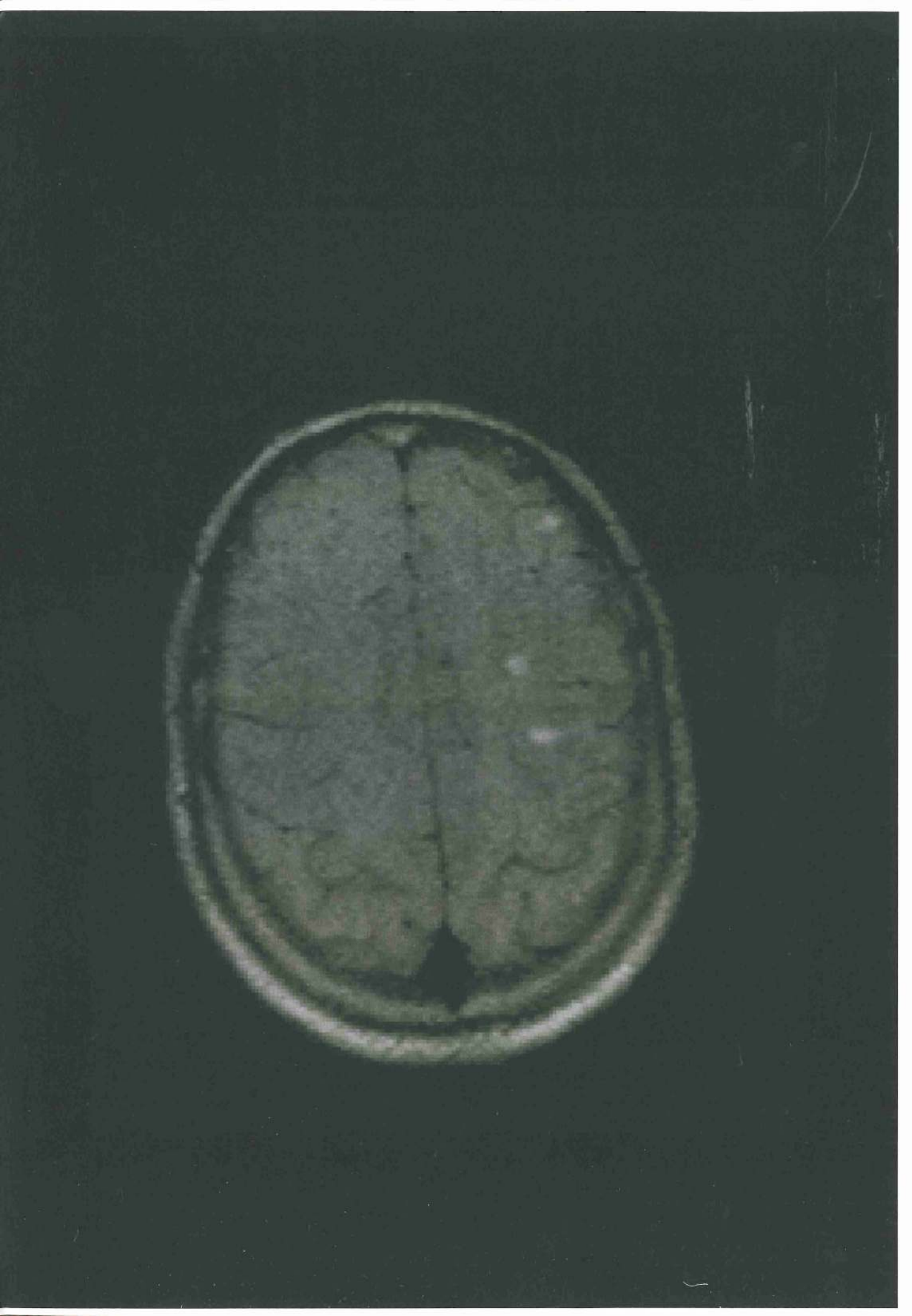


Figure 6.6b Diffusion-weighted scan of the same patient taken at the same time. The new left subcortical lesion is demonstrated.



Figures 6.7a and 6.7b Diffusion weighted images of a 51 year-old man with right hemiparesis and a language disorder, demonstrating tiny cortical and subcortical areas of abnormally high signal, which are not seen when a coarser (EPI) matrix is used.



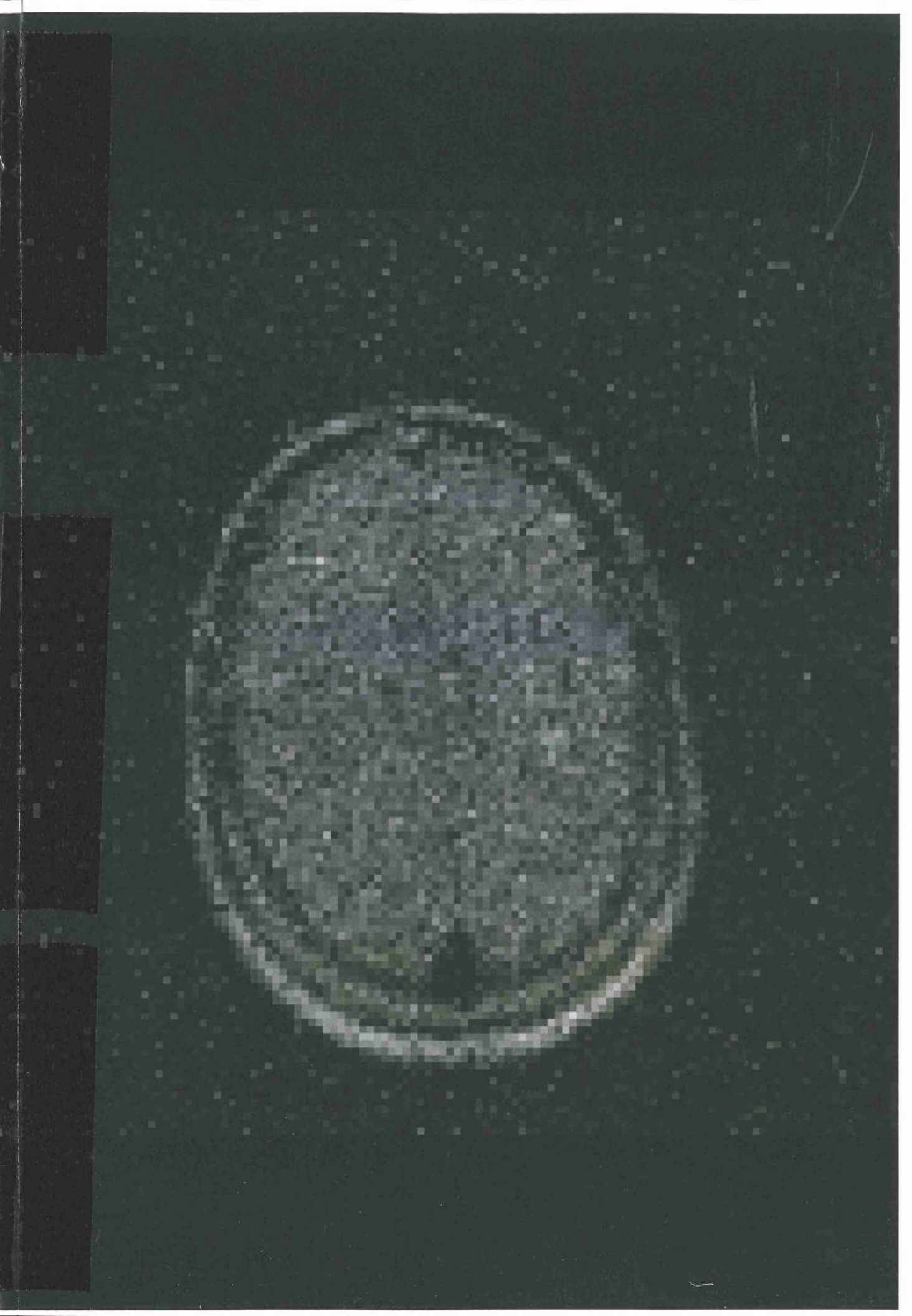
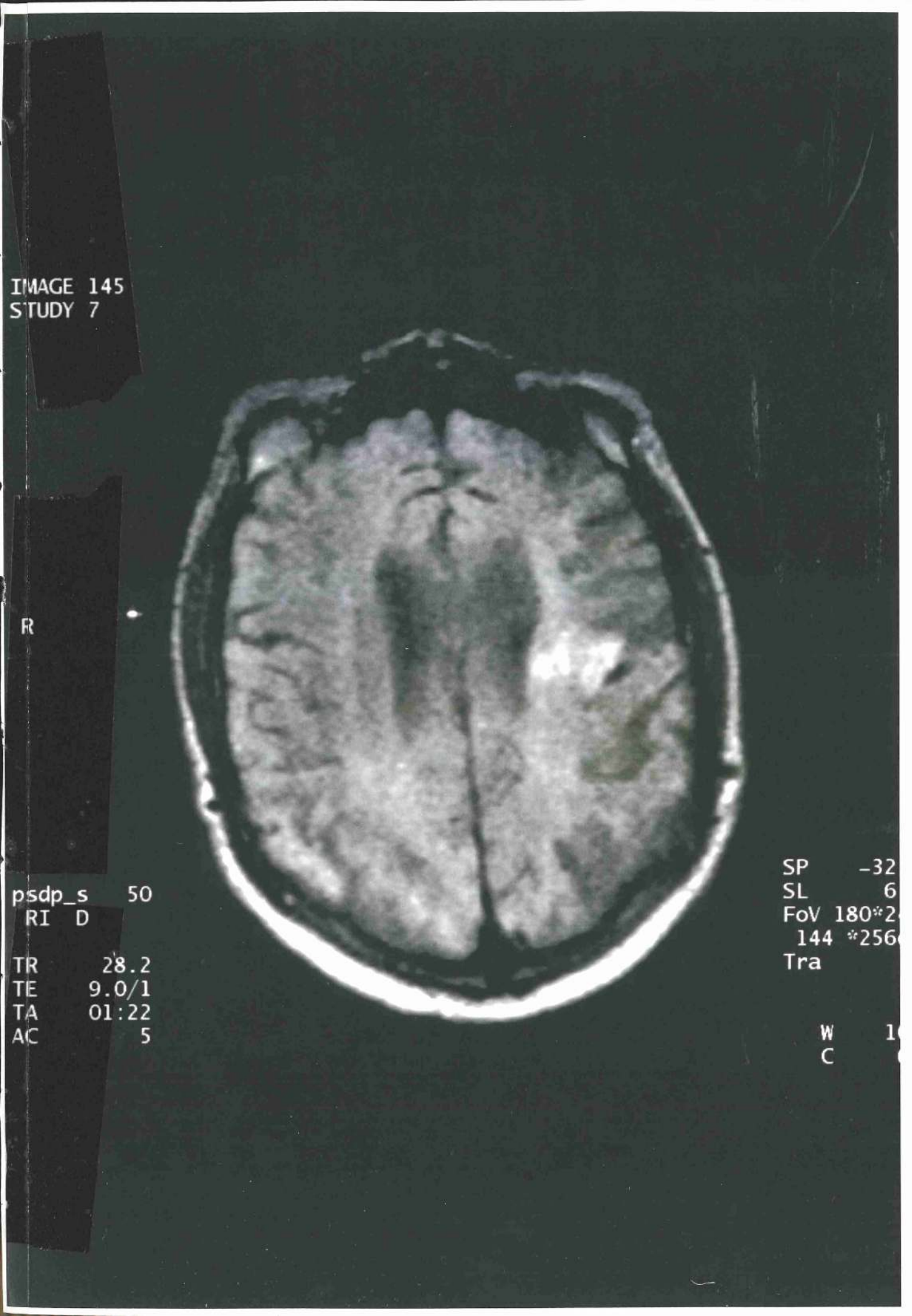


Figure 6.8a Diffusion-weighted scan of a 67 year old man with language disorder, right limb weakness and right visual field deficit.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STUDY 7
R
psdp_s 50
RI D
TR 28.2
TE 9.0/1
TA 01:22
AC 5
SP -32
SL 6
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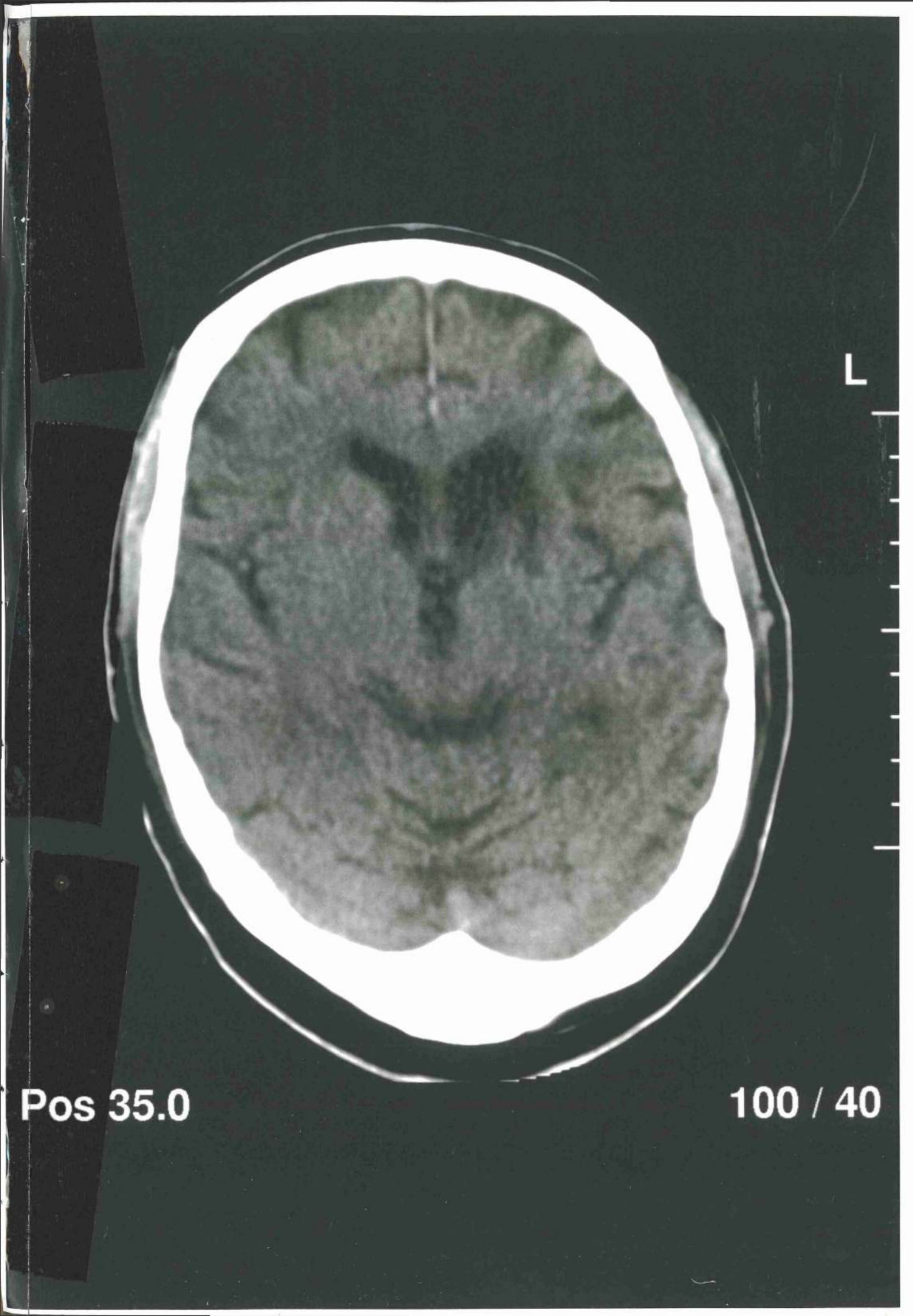
Figure 6.8b Interval CT scan of same patient showing established left hemispheric cerebral infarct.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Chapter Seven

Intravenous thrombolysis in acute ischaemic stroke: preliminary experience with rt-PA in a UK hospital

7.01 Introduction

The National Institute of Neurological Disorders and Stroke (NINDS) trial evaluated 624 patients with acute stroke. 0.9 mg/kg recombinant tissue plasminogen activator (rt-PA) or placebo was administered within 3 hours of onset of stroke symptoms. All patients underwent emergency CT scanning before administration of rt-PA and half the patients were treated within 90 minutes. Patients treated with rt-PA were 30% more likely to have minimal or no disability at three months than those given placebo. This improvement in outcome occurred despite a 10 fold increase in the rate of symptomatic intracerebral haemorrhage (6.4% vs 0.6%). Even with this higher rate of secondary haemorrhage mortality was similar in the two groups (17% in the thrombolysed group vs 21% in the placebo group).[102]

The European Cooperative Acute Stroke Study (ECASS) study randomised patients to a higher dose of 1.1 mg/kg rt-PA or placebo within a longer time window (6 hours of stroke). There was no improvement in functional outcome in the intention to treat analysis but reanalysis after exclusion of a large number of ineligible patients from the target population suggested a significant improvement in functional outcome in rt-PA receiving patients ($P = 0.035$).[101] A further ECASS trial, assessing the lower dose of rt-PA used in NINDS and a 6 hour time window failed to show significant benefit when pre-specified end points were examined; the trial has been interpreted as neutral.[103]

Earlier studies evaluating streptokinase (SK) therapy reported negative results with a high frequency of complicating cerebral haemorrhages and a resultant increase in early mortality in the treated groups.

The only clearly favourable trial of thrombolysis in acute ischaemic stroke was the NINDS study; however meta-analysis of the use of thrombolysis in this context has shown consistent benefit from rt-PA and no benefit from streptokinase.[196]

On the basis of the NINDS study results, rt-PA is licensed for the treatment of ischaemic stroke in the United States. It is currently used outwith license in Europe for the same indication. Concerns over the safety and practicability of this treatment when employed outwith US centres have been raised, due to differences in the provision of acute stroke services between Europe and North America.

7.02 Subjects and Methods

The acute stroke unit of the Western Infirmary in Glasgow serves an urban catchment population of 220,000. Patients with symptoms and signs consistent with a clinical diagnosis of stroke are admitted regardless of age or severity of neurological deficit, amounting to approximately 800 patients per year. Thrombolysis has been used selectively within the unit since 1996. We report a consecutive, prospectively evaluated series of patients with acute ischaemic stroke treated with rt-PA.

Between April 1996 and November 1998, rt-PA was administered on sixteen occasions to fifteen patients. Thrombolytic therapy was only given after discussion with a consultant stroke physician. Eligibility criteria were according to the entry criteria of the NINDS study and the American Heart Association guidelines,[102] i.e. administration of treatment within three hours of onset of symptoms following a normal CT scan. In general, patients were excluded if they were at risk of haemorrhage, (for example bleeding diathesis, active gastro-intestinal ulceration or recent surgery), if the focal neurological signs were attributable to metabolic disturbance or seizure activity, or if the deficit was rapidly resolving. Although the NINDS blood pressure criteria were applied,[102] intervention to reduce blood pressure in order to meet American Heart Association guidelines was not undertaken. Our patients are therefore likely to have a higher blood pressure than patients entering the NINDS trial.

Clinical and Imaging Protocol

Clinical and radiological data were prospectively recorded. Neurological and functional deficit were classified at admission according to the Oxford Community Stroke Project subtype[28] and NIH Stroke Scale.[102] Patients were reviewed at three months using the Rankin score and Barthel index. The reliability and reproducibility of these measures have been confirmed previously.[96,197]

Patients who were potentially eligible to receive thrombolytic therapy underwent emergency brain CT. Twenty four hour access to CT scanning was

available, and following discussion with the radiology department, potential suitability for treatment with rt-PA was classified as an indication for emergency CT brain scan. All scans were reported by a consultant or senior registrar in radiology at the time of scanning. If there were no signs of major cerebral infarction or haemorrhage 0.9 mg/kg intravenous rt-PA was administered following an informed discussion amongst senior medical staff, patients and other family in attendance. 10% of the total dose was given as a bolus and the remainder infused over 60 minutes.

Heparin was co-prescribed in one patient with severe carotid disease and a fluctuating neurological deficit; otherwise no patient received concomitant anticoagulant therapy. Antiplatelet therapy was disallowed for 24 hours after administration of rt-PA. All but one patient (who was scanned 7 days post thrombolysis) underwent follow up CT scanning between 48 and 72 hours, to evaluate the frequency of haemorrhagic transformation.

Patients received routine clinical care from the medical and nursing staff of the acute stroke unit. Death, new stroke, significant systemic bleeding and intracranial haemorrhage were monitored as primary adverse events. An intracerebral haemorrhage was considered to be symptomatic only if it was associated with a deterioration in neurological status.

7.03 Results

Patient demographics and clinical details are summarised in Table 7.1. The mean elapsed time from onset of deficit to start of thrombolytic infusion was 141 minutes (SD 42).

All patients received appropriate bolus and infusion doses of rt-PA. No symptomatic intracranial haemorrhages were reported. One clinically-silent haemorrhagic transformation of left middle cerebral artery infarction was detected on a follow-up scan three days after treatment.

Of nine patients with total anterior circulation stroke, three were independently living at home three months after presentation, three were alive and dependent and three had died. In total, five patients were independently living at home and a further two patients were living at home with support. Five patients were significantly disabled and dependent, either in hospital or in nursing home care. Outcome at three months as measured by NIH score and Barthel index is shown in figure 7.1 and 7.2, along with the NIH scores and Barthel indices of participants in the second part of the NINDS trial for comparison. Also shown for comparison are the reported results[198] of a German acute stroke unit.

Four patients died following treatment with rt-PA. Two deaths were attributed to massive cerebral infarction complicated by cerebral oedema, and two to hypostatic bronchopneumonia.

Patient 3 made a full recovery from a left lacunar syndrome following thrombolysis and underwent left carotid endarterectomy for a high-grade symptomatic carotid stenosis three days later. He developed a posterior circulation stroke within 10 days of carotid endarterectomy and was treated with a second infusion of rt-PA. Follow up CT scan revealed a new pontine infarction.

Patient 4 presented with a dominant hemisphere total anterior circulation infarction having undergone uncomplicated coronary angiography one week before. Initial CT showed no abnormality. Following administration of rt-PA, he developed an extensive groin haematoma over the site of the arterial puncture. No change in focal neurological signs occurred immediately following administration of rt-PA; however 30 hours following thrombolysis, conscious level deteriorated and signs consistent with brainstem compression were observed. Repeat CT scan revealed an extensive left hemispheric infarction with evidence of cerebral oedema and the patient died three days later.

Patient 6 developed left hemispheric total anterior circulation infarction whilst being treated in hospital for pneumonia. She was treated with rt-PA 100 minutes after onset of symptoms. No change in neurological status was observed immediately following dosing. Conscious level deteriorated the following day, and repeat CT scan performed 36 hours after onset of symptoms revealed massive left hemispheric infarction with significant mass

effect. Her conscious level deteriorated progressively and she died two weeks after onset of symptoms.

7.04 Discussion

Few patients referred to the acute stroke team are assessed and CT scanned by 180 minutes of onset of symptoms. The delays in presentation to hospital, CT scanning and reconstitution and administration of thrombolytic drug are summarised in figure 7.3. It is significant that three patients in this series were already in hospital at the time of stroke. Figure 7.4 illustrates the pattern of delay in presentation to hospital in a series of 200 consecutive patients referred to the Acute Stroke Unit between April and July 1998. In most cases delay is due to procrastination in contacting the primary care team or delay in GP referral to hospital. In our local area since March 1998, patients with symptoms consistent with stroke of less than three hours' duration have been afforded highest priority by the local ambulance service and are now brought directly to hospital as an emergency.

Failure in the early identification of stroke may also occur because of a lack of awareness on behalf of the patient or their relatives. Increased awareness of symptoms and the perception of stroke as a medical emergency may be achieved through educational campaigns directed at "at risk" populations. The accurate identification of a well-defined time of onset is a prerequisite for the safe use of thrombolytic therapy. Failure of the patient or clinician to identify

pre-existing subtle neurological symptoms which suddenly worsened may lead to inappropriate use of rt-PA.

A further consideration is the means through which medical attention is sought. Approximately 50% of admissions to our unit are referred by local general practitioners. In areas where the emergency services treat stroke with the same priority as acute myocardial infarction, it has been shown that admission to hospital is significantly more rapid. This often bypasses the primary care physician; instead the emergency paramedical services are the initial point of patient contact.[199]

Once within the hospital, the narrow therapeutic time window demands rapid and accurate assessment by medical staff. The presence of a dedicated "stroke team" with access to emergency 24 hour on site CT scanning are prerequisites for thrombolytic stroke therapy. It is therefore likely that this treatment could only be offered at specialist centres.

It is vital that the outcome in patients receiving unlicensed thrombolytic therapy be continuously audited, to ensure patients are not being exposed to unacceptable risk. Baseline median NIH scores in both the placebo and treated cohorts of the NINDS study were significantly lower (14) than those patients treated in Glasgow (19).[102] In other words our stroke population were more severely affected than those typically recruited to the NINDS study. The medical staff involved currently only consider thrombolytic therapy in those

patients with severe stroke, conscious of adversely affecting patients who might otherwise have a good prognosis. In contrast to the NINDS study, no treatment to lower blood pressure acutely following ischaemic stroke is undertaken in patients eligible to receive thrombolytic therapy. Patients with total anterior circulation syndromes have a poorer prognosis than many forms of cancer i.e. 95% death or dependency at 6 months.[197] Despite the relative severity of patients treated the outcome was satisfactory in comparison with that expected of untreated patients with similar degrees of disability at presentation. Figure 7.5 compares the three month functional status of thrombolysed patients with total anterior circulation infarction with interval outcome data from the untreated OCSP cohort.[28] A trend towards improved functional outcome in these severely-affected patients is apparent although formal analysis of efficacy is not possible given the small numbers involved.

7.05 Conclusions

rt-PA is currently licensed for the treatment of acute ischaemic stroke in the US. Limited approval for its use in the Europe is under consideration. This may depend on the interpretation of results of recently completed clinical trials, such as ECASS II[103] Although the ECASS II trial was interpreted as neutral, incorporation of its data into a meta-analysis of trials of rt-PA within 3 hours of onset of acute stroke reveals a favourable odds ratio of 0.67 (95% CI 0.56 - 0.80) with respect to death and disability.[200,201]

Although many clinical and logistical difficulties complicate its use, our patient outcome and complication rate are in keeping with those of the NINDS

investigators, while the small numbers involved preclude formal statistical analysis. We conclude that the judicious use of this treatment within a specialist unit appears safe, but requires careful continued audit. Substantive changes in the public and primary care providers' perception of stroke as a medical emergency will be required before this treatment is more widely applied.

Table 7.1 **Patient Characteristics**

Age/ Sex	OCSP	NIH Pre	NIH Day 3	CT Post	Rankin Month 3	Status Month 3
77 M	TACS	23	1	Small Infarct	1	Home
69 F	PACS	15	3	Small Infarct	1	Home
89 M	LACS	14	13	Subcort. Infarct	4	Hospital
89 M	POCS	23	18	Pontine Infarct	4	Hospital
64 M	TACS	19	--	Large Infarct	--	Died day 2
85 F	TACS	30	9	Small Infarct	2	Home
93 F	TACS	25	24	Large Infarct	--	Died day 10
72 F	TACS	21	8	Small Infarct	3	Home
59 M	PACS	8	0	Normal	1	Home
80 F	TACS	25	22	Small Infarct	4	Hospital
84 M	PACS	14	12	Large Infarct	--	Died week 6
60 M	POCS	5	1	Small Infarct	1	Home
70 M	LACS	6	5	Small Infarct	2	Home
63 M	TACS	19	23	Large Infarct	--	Died week 5
58 M	TACS	25	23	Large Infarct	3	Hospital
87 F	TACS	20	19	Silent Haem. Trans.	4	Nursing home

Figure 7.1 NIH scores at three months

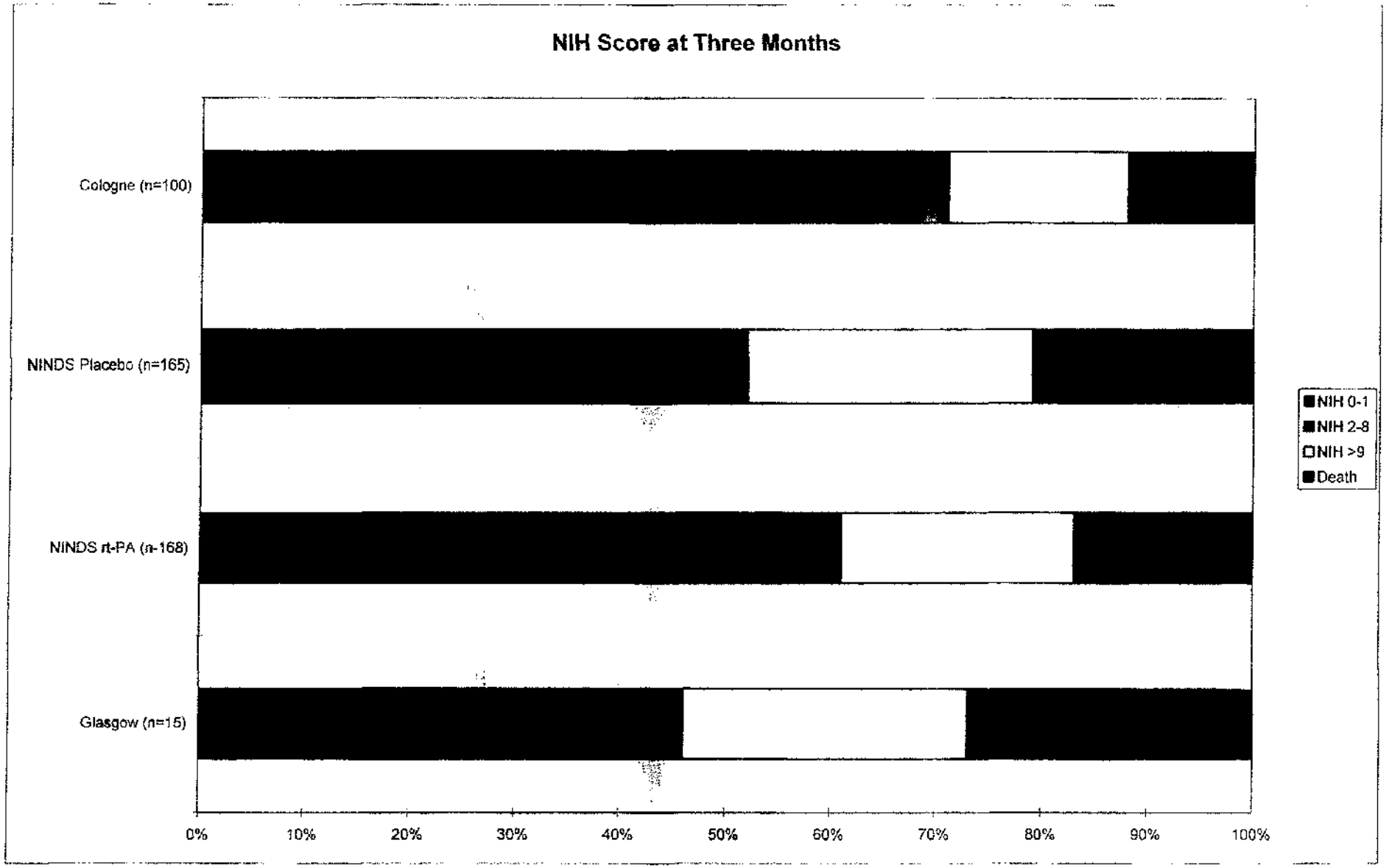


Figure 7.2 Barthel scores at three months

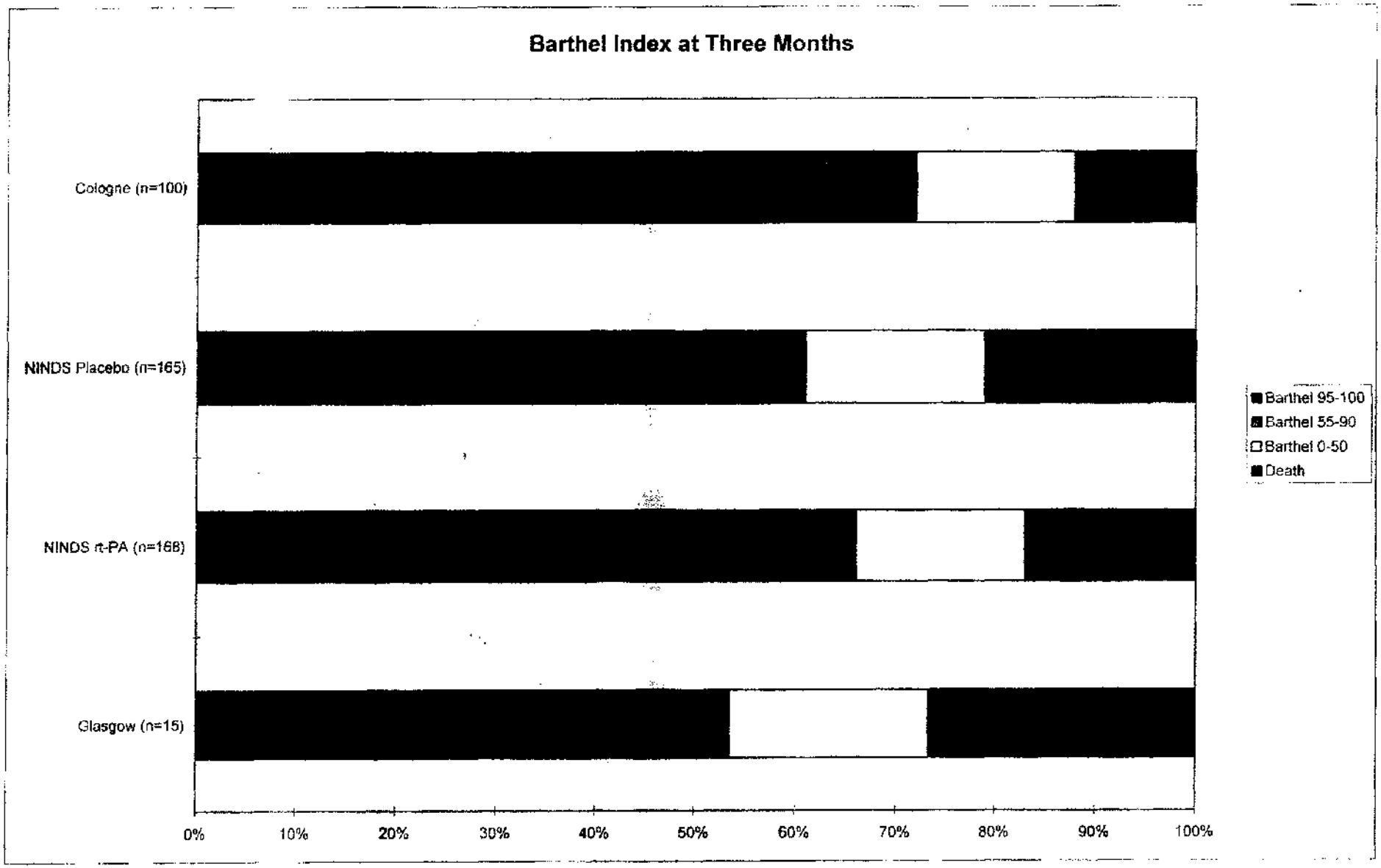


Figure 7.3 Factors delaying rt-PA administration

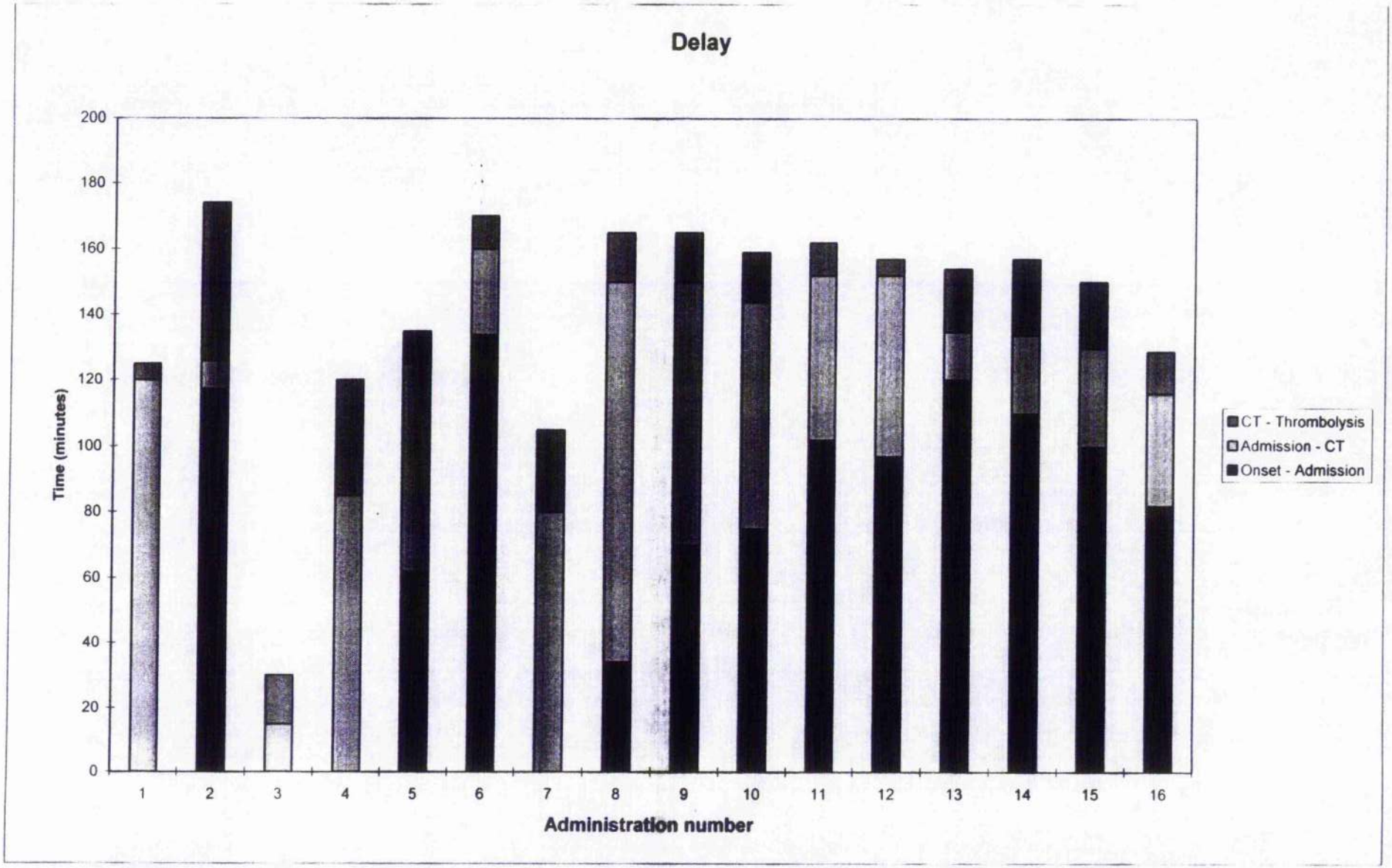


Figure 7.4 Delay in presentation of a cohort of 200 patients

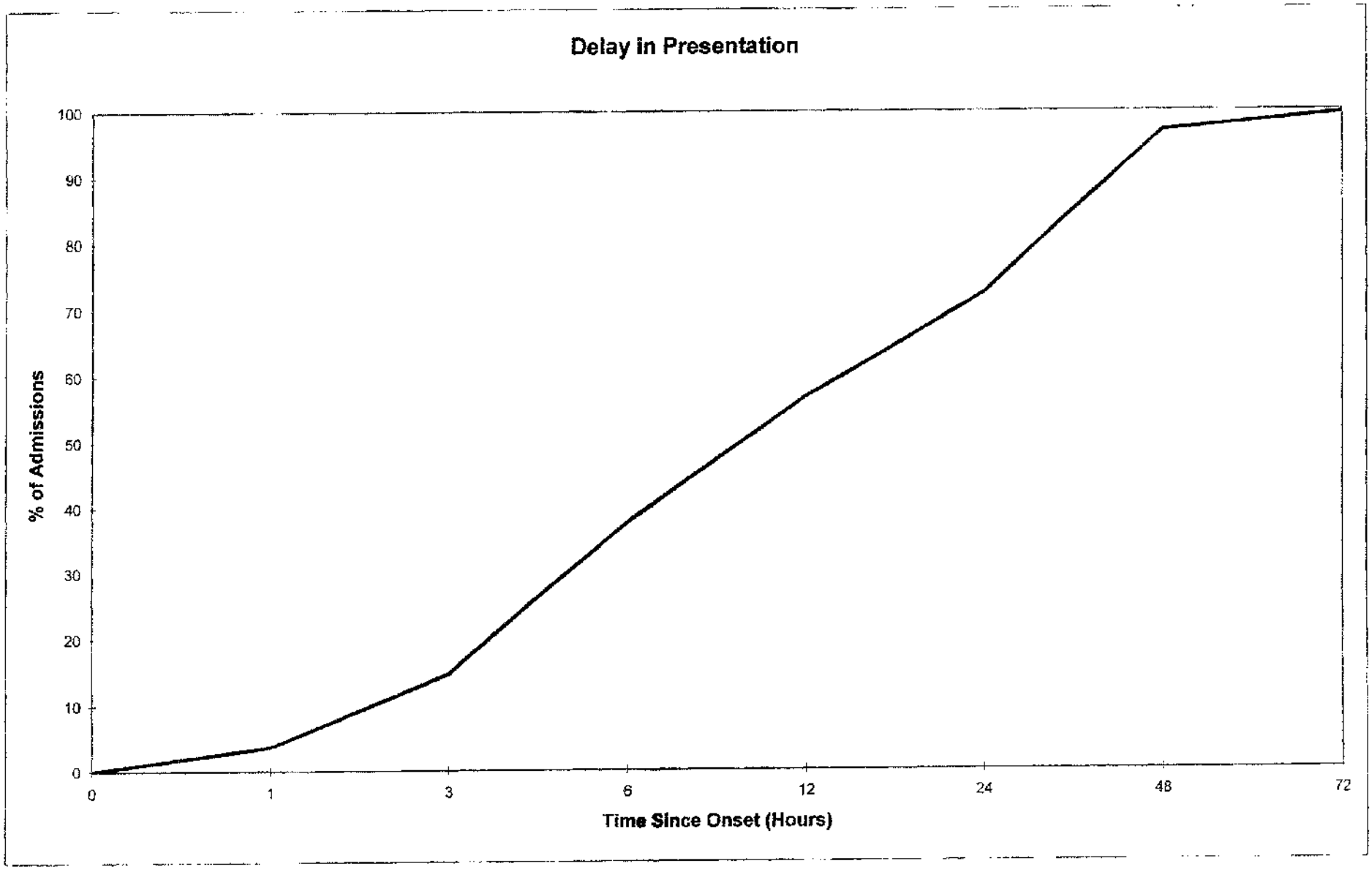
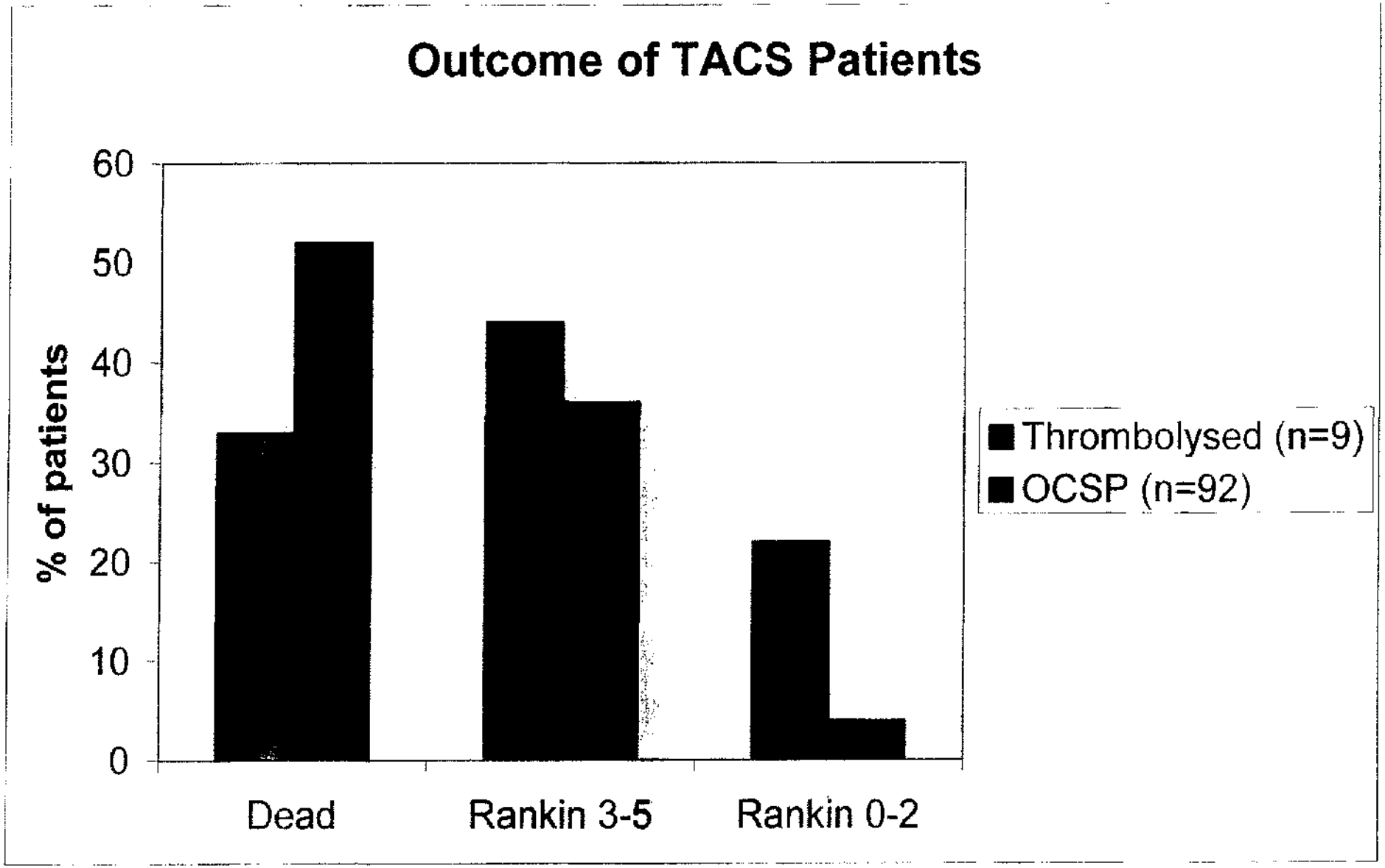


Figure 7.5 Outcome of patients with TACS



Chapter Eight

Discussion of results and future directions

8.01 Introduction

The underlying theme of the work presented in this thesis has been prognosis following stroke, and how it may be influenced or predicted. Three approaches to this theme have been adopted. I have sought to identify potential prognostic factors after stroke, such as serum triglyceride concentration and computed tomography appearances; to demonstrate in selected populations the safety and practicality of simple and potentially beneficial interventions such as control of blood glucose and blood pressure; and to implement novel techniques such as thrombolysis and diffusion-weighted magnetic resonance imaging of hyperacute stroke in the setting of a U.K. hospital. This chapter will provide a synopsis of the results of each study, together with a discussion of potential future research.

8.02 ACE Inhibition and Cerebral Perfusion

In chapter two the effect of the angiotensin converting enzyme inhibitor perindopril upon cerebral and renal perfusion in moderately-hypertensive stroke patients with extracranial carotid artery disease was investigated. I hypothesised that perindopril would reduce blood pressure without significantly affecting cerebral perfusion. Using transcranial Doppler ultrasound, carotid Doppler ultrasound, dynamic HMPAO bolus imaging and single photon emission computed tomography, global and focal cerebral perfusion were assessed. No significant change in global or peri-infarct cerebral perfusion was observed in the treated group. There was no drug-associated neurological deterioration, however an episode of acute renal

failure occurred in one of the treated patients; this was attributed to pre-existing bilateral renal artery stenosis.

There was a degree of heterogeneity in the age, size and location of the index cerebral infarction of the patients recruited into the study. The degree and extent of the carotid artery disease was also variable. Given the possible permutations of carotid artery disease severity and extent of cerebral infarction, recruitment of an entirely homogenous group of patients is impracticable however further studies may be required to fully elucidate the effect of ACE inhibition on cerebral perfusion in specific subtypes of cerebral infarction.

The cerebral perfusion data were consistent with those data acquired from a similar study which examined the effect of ACE inhibition upon cerebral perfusion early after ischaemic stroke.[168] The mechanistic basis of effect of ACE inhibition upon cerebral perfusion remains elusive. In rats, angiotensin II receptors are known to be involved in the regulation of cerebral perfusion[115] and it is likely that a similar mechanism is implicated in humans. The study was not designed to examine the effect of blood pressure reduction with ACE inhibitors upon long-term outcome, and the cohort of patients studies had relatively mild degree of neurological impairment relative to an unselected cohort of stroke survivors, hence no conclusions concerning longer-term outcome can be drawn. A large-scale study is currently underway to investigate the potential effect of angiotensin converting enzyme inhibition on secondary incidence of stroke. Although the cerebral perfusion data are

consistent with our original hypothesis, the episode of reversible acute renal failure observed in one treated patient raises some concern. It is known that patients with severe renal artery disease are significantly more likely to have extracranial carotid atheroma,[119] however the prevalence of significant renal artery disease among patients with extracranial carotid artery atheroma is unknown. We conclude that intervention with ACE inhibitors will lower blood pressure in this population without adversely affecting cerebral perfusion, but that simple, validated non-invasive screening for the presence of renal artery stenosis (such as scintigraphy) should be considered prior to introduction of therapy.

8.03 Control of Hyperglycaemia after Ischaemic Stroke

Chapter three reported the results of a pilot study designed to investigate the safety and feasibility of rigorous glycaemic control early after ischaemic stroke in patients with moderate hyperglycaemia. The deleterious effect of hyperglycaemia following experimental stroke has been extensively studied,[71-73,124-128] and clinical evidence to support a similar adverse effect in stroke patients has been published.[70,129,130] In animal models, intervention with insulin following hyperglycaemic stroke reduces infarct volume and improves functional outcome.[84] Twenty-four hyperglycaemic patients with acute stroke were randomised to receive either standard management of blood sugar or rigorous control using insulin infusion titrated in accordance with a sliding scale. The ability of the sliding-scale insulin regime to maintain blood glucose concentrations in the pre-specified range was demonstrated. Symptoms consistent with hypoglycaemia were observed one occasion; these were

minor and reversed rapidly following administration of oral dextrose. The purpose of the study was to investigate the feasibility and safety of one method of glycaemic control following acute ischaemic stroke: no conclusions concerning efficacy can be drawn as the study was not powered to demonstrate an effect upon outcome. Data acquired during this study may however be useful in the preparation of power calculations for future studies. I conclude that a larger trial is now warranted to investigate this. The pilot study has demonstrated the feasibility of the protocol used, however alternative means of glycaemic control such as the glucose-potassium-insulin ("GKI") regime have also been employed in the context of hyperglycaemic ischaemic stroke patients.[134] The GKI regime is simple and easy to administer, but is less flexible as the relative rates of insulin and dextrose infusion cannot be varied independently of each other. A further study to compare the relative merits of these two approaches may permit the design of a definitive large-scale study to be optimised.

8.04 The Prognostic Significance of Triglyceride Concentration

Chapter four reports a long-term follow-up study designed to investigate the influence of serum triglyceride concentration upon outcome after stroke. An earlier study[138] had suggested an unexpected association between relatively high levels of cholesterol and better outcome following stroke. We sought to clarify the relationship between lipid concentration and stroke prognosis. Using the database maintained by the acute stroke unit of the Western Infirmary, 1312 non-diabetic patients presenting with computed tomography-confirmed acute stroke were identified. Data from fasting blood samples

drawn within 24 hours of admission for glucose, lipids and a standard battery of biochemical and hematological tests were analysed. Information on age, stroke type, admission blood pressure, smoking status, presence of atrial fibrillation, resolution time of symptoms and Oxfordshire Community Stroke Project[28] clinical classification was collated and incorporated into stepwise proportional hazards regression analysis to estimate the effect of these parameters on survival following stroke.

The results obtained suggest that triglyceride concentration is associated independently with survival of patients with acute stroke; those with relatively high triglyceride concentrations enjoyed a better prognosis than those with lower levels. Triglyceride concentration was found to be a better predictor of outcome than cholesterol.

The results presented raise a number of insights into the potential biological mechanisms underlying the relationship between lipids and stroke. The association of low triglyceride with relatively poor outcome following acute stroke suggests that there is no confounding effect of the inflammatory response as it is known that, in contrast with cholesterol concentration, triglyceride concentration does not fluctuate to a significant degree in the first week following ischaemic stroke.[141] The median fasting total serum triglyceride concentration in our cohort was 1.3 mmol/L. At this concentration, the majority of measured triglyceride circulates as very low density lipoprotein particles (VLDL). The results suggest that it is VLDL concentration which is predictive of stroke outcome, rather than concentration of other lipoprotein

particles. As VLDL particles also carry cholesterol, the previously-noted relationship between cholesterol concentration and stroke outcome is also consistent with this hypothesis. Further studies involving more detailed lipid analysis would be required for confirmation. All blood samples drawn were analysed in the fasting state within 24 hours of admission to the hospital, hence any confounding effect of stroke-induced dysphagia is unlikely. Pre-existing nutritional deficiency may account for the observed relationship between low triglyceride concentration and poor outcome; low serum triglyceride is a more sensitive indicator of poor nutritional status than low cholesterol concentration,[143] and patients with poor pre-stroke nutritional status may be less likely to enjoy a good outcome. A further prospective study using more detailed biochemical and anthropometric indices would be required to address this issue more fully.

8.05 Visible infarction after Lacunar Stroke

In chapter five the prognostic significance of appropriate visible cerebral infarction on x-ray computed tomography (CT) in patients with a clinical diagnosis of lacunar stroke is considered. Relatively few prognostic factors have been identified in this sub-group of stroke patients, and I sought to investigate whether patients with lacunar symptoms and visible infarction on CT had a poorer prognosis than those patients with similar symptoms and no appropriate CT lesion.

The computer database maintained by the Acute Stroke Unit of the Western Infirmary holds prospectively-acquired clinical, biochemical, radiological and

outcome data for all patients admitted to the unit. Using the database, we identified 633 patients admitted with lacunar stroke between 1990 and 1998. A total of 229 patients were excluded from the analysis due to incomplete follow-up, non ischaemic pathology or brain imaging other than CT. The remaining 404 patients were divided into two groups, depending on the appearance of the CT scan. Patients with an appropriately placed infarct on the CT scan classified as "CT positive". Those without such a lesion on the scan were classified as "CT negative".

A log-rank test was used to compare survival times in patients with and without visible infarction on CT; then effect of visible infarction on survival after correcting for other prognostic factors was examined using a proportional hazards model.

A comparison of six-month outcome with respect to presence or absence of infarction on CT was performed using a chi-squared test. Stepwise logistic regression was then used to adjust for significant prognostic factors before assessing the effect of CT findings on outcome at six months. Length of hospital stay in each group was compared using a Mann-Whitney test.

No significant difference in survival, outcome or length of hospital stay was identified between the two groups, indicating that the presence or absence of an appropriate ischaemic lesion on CT scan following ischaemic lacunar stroke does not confer prognostic information after correcting for other factors known to influence outcome.

Other studies have examined the prognostic significance of visible cerebral infarction on CT scan following ischaemic stroke,[158-160] however the issue remains clouded due to small sample sizes and methodological differences between studies. The specific question of whether visible infarction increases the risk of poor outcome following lacunar stroke after correction for confounding factors has not yet been fully addressed in the literature as previously published studies have either examined only relatively small groups of lacunar stroke patients,[158] have collected limited follow-up data[159] or have restricted the study to early imaging after stroke.[159,160] This study used a larger cohort of lacunar stroke patients than any yet published in the literature, and a single radiologist (unblinded to clinical information) interpreted the images. Although the cohort of patients studies was relatively large, one potentially-important confounding factor, the location of the lacunar lesion, was not examined. Lacunar syndromes may arise as a result of infarction in a variety of areas within both anterior (carotid) and posterior (vertebrobasilar) circulation territories. The study presented did not divide lacunar stroke patients according to the localisation of the lacunar lesion as the small numbers in each resultant sub-group would preclude any firm conclusions.

Reproducibility of radiological categorisation was not examined, and the absence of a measure of inter-observer variability could be criticised. Volumetric analysis of the radiological lesions was not performed as the small volumes involved would have resulted in the introduction of considerable error. Despite these weaknesses, the results obtained strengthen the conclusions of previous smaller studies which have reported similar findings

but which have been criticised on the basis of small sample size or failure to correct for known prognostic factors.

8.06 Diffusion MRI of Ischaemic Stroke at 1.0 Tesla

Chapter six describes the implementation of a novel diffusion-weighted magnetic resonance sequence on a standard 1.0 Tesla scanner. The potential advantages conferred by diffusion-weighted magnetic resonance techniques when applied to acute cerebral ischaemia have been recognised; however due to technical and financial constraints few centres in the United Kingdom use diffusion-weighted imaging to investigate stroke patients. We sought to further develop a pre-existing time-reversed steady-state (PSIF) diffusion-weighted sequence,[188] and investigate its clinical usefulness when implemented on a standard MRI scanner.

Having optimised the sequence, a series of patients with acute stroke were scanned with both conventional and diffusion-weighted techniques. It was found that abnormalities on diffusion-weighted images were seen more frequently than on conventional (T2-weighted) images, and that the combination of diffusion-weighted and conventional sequences yielded more diagnostic information than the conventional sequences alone. Addition of the diffusion sequence to the conventional sequences prolonged the duration of the scan by approximately ten minutes. Repeat brain imaging performed at an interval confirmed localisation of ischaemic damage by the early diffusion-weighted MRI scan.

With the advent of therapy for acute ischaemic stroke, the ability to depict ischaemic cerebral injury at an early stage has assumed increasing importance.[195] In the United Kingdom, CT scanning is currently the most widely-used brain imaging technique after stroke. Although CT accurately detects the presence of intracerebral haemorrhage, the changes seen on CT scan early after ischaemic stroke are subtle and may be absent for several hours. Early, accurate identification, localisation and quantification of brain injury with diffusion-weighted MRI will enable early diagnosis and may have implications both for clinical trial design and provision of future therapies.

Using diffusion-weighted imaging, the effect of intervention with novel neuroprotective agents upon infarct volume can be studied. This will allow the use of a more rational endpoint in efficacy studies of neuroprotective drugs, and the avoidance of the confounding factors which may influence measures of clinical outcome.

The ability to localise cerebral infarction in the acute phase may influence choice of neuroprotective therapy. The differing pathophysiological mechanisms which underlie white and grey matter ischaemia are likely to necessitate different pharmacological approaches to therapy; early diffusion-weighted imaging may enable selection of an appropriate agent.

A number of technical difficulties have been overcome to achieve the implementation of clinically-useful diffusion-weighted imaging on a standard 1.0 Tesla scanner; further development of the sequence will be required to enhance the research potential of the technique. Diffusion tensor imaging and generation of apparent diffusion coefficient (ADC) maps are both theoretically possible with PSIF-based sequences; further work in this area is planned.

8.07 Thrombolytic Therapy for Acute Ischaemic Stroke in the UK

Chapter seven reports the implementation of thrombolytic therapy, the only treatment shown to influence outcome after ischaemic stroke, in a United Kingdom stroke centre. The continuous, prospective audit of process and outcome undertaken over more than two years examined fifteen recipients of thrombolytic therapy, and although the number of patients studied was insufficient to permit conclusions on safety and efficacy relative to published studies, the demographics and clinical characteristics of the treated patients have been identified.

The small number of recipients of thrombolytic therapy over a two-year period reflects the low rate of early presentation to hospital with acute stroke. Very few patients currently present to hospital within three hours of onset of symptoms; the delay in presentation may arise as a result of failure to educate the public of the symptoms of stroke, failure of primary care physicians to promptly refer stroke patients to a specialist centre or reluctance of ambulance services to prioritise patients with stroke. Local variability in each

of these factors is likely, and the changes necessary to improve the numbers of patients presenting early after stroke will vary between regions.

The audit has identified the need for close liaison with radiologists to minimise the time spent waiting for CT brain examination. A locally-agreed protocol with the radiology department of the Western Infirmary allowed prioritisation of potential recipients of rt-PA over requests for "routine" CT examinations; this enables prompt acquisition of the necessary imaging prior to administration of thrombolytic therapy.

From the audit it is apparent that the patients treated in Glasgow were older and more severely affected by the stroke than those patients reported in the literature;[101-103] this difference again reflects patterns of presentation to hospital in that, at present, relatively few less severely affected patients reach hospital rapidly after onset of symptoms.

Although the use of thrombolytic therapy for acute ischaemic stroke is complicated by a number of logistic and clinical issues, we have found that patient outcomes and complication rates are similar to those reported by the NINDS investigators,[102] whose protocol was employed (with minor modifications) in our unit. The numbers treated to date are too small to justify formal statistical analysis. The implementation of a protocol for thrombolytic therapy with rt-PA in a specialist UK stroke centre appears safe, but a continuous prospective audit of outcome is required to ensure that patients

are not being exposed to unacceptable risk. Changes in the provision of acute care offered to stroke patients by relatives, primary care physicians, ambulance services and hospital staff will be required before thrombolytic therapy can be more widely used. These changes are likely to vary between regions and merit local investigation by all stroke centres intending to use thrombolytic therapy.

8.08 Conclusion

Acute treatment for ischaemic stroke is possible but at present its use is limited to a tiny minority of patients who present within three hours of onset of stroke to specialised stroke centres. More widespread implementation of diffusion-weighted magnetic resonance imaging may help in the identification of suitable recipients and allow safe use of thrombolytic therapy to be extended to other hospitals. This imaging modality may also facilitate the design and conduct of efficacy studies of other acute stroke therapies, for example intervention to control elevated blood glucose levels with sliding scale insulin. This technique has been shown to be feasible and safe; it could represent a further means of improving outcome after stroke and may be more widely applicable than other approaches due to the high proportion of stroke patients with hyperglycaemia early after symptom onset.

Secondary prevention of stroke should be considered in all stroke survivors. The importance of safe and effective blood pressure control is recognised however in patients with carotid disease blood pressure reduction carries the

risk of cerebral hypoperfusion. Angiotensin converting enzyme inhibitors have been shown to lower blood pressure without reducing cerebral blood flow in this group of patients; the generalised nature of atheromatous disease leads to the conclusion that screening for renal artery stenosis should be considered before they are used.

The role of lipid lowering therapy after stroke remains unclear. Data presented in this thesis suggest that the relationship between low triglyceride concentration and relatively poor outcome after stroke is stronger than the previously reported association between cholesterol concentration and outcome. This observation raises a number of questions concerning the precise effects of lipids and lipoprotein particles after stroke. Although the study presented can offer no immediate advice concerning treatment of lipids after stroke, it has accentuated the need for further mechanistic studies designed to clarify the observed relationships.

Appendix One

Magnetic Resonance Imaging Using PSIF

A1.1 Introduction

The technique used to provide the diffusion-weighted magnetic resonance images shown in chapter six is based upon a complex magnetic resonance sequence, termed a steady state free precession or reversed fast imaging with steady state precession ("PSIF") method. This appendix will provide a more detailed description of the basic mathematical and physical principles behind PSIF imaging.

The radiofrequency signal generated by tissue during a PSIF sequence contains two components: a free induction decay (FID) that arises as a result of the most recent radiofrequency pulse and a strongly T2 dependent spin-echo component that forms immediately prior to the upcoming pulse as a result of the cumulative effect of previous pulses. Because of the persistence of the second signal, it is possible to use additional strong gradients in the sequence to sensitise the image to the diffusion of water molecules. The spin-echo component is intrinsically weak, and image generation is therefore difficult using this component of the signal alone. Multiple acquisitions are therefore needed to generate an image with adequate signal-to noise ratio. The necessity for multiple acquisitions leads to a relatively long scan time in comparison with, for example, echo-planar techniques. The design of the sequence is such that the FID component of the signal is not used for imaging.

A1.2 Theory

A steady-state transverse magnetisation, M_{xy}, is generated when a succession of radiofrequency pulses, separated by a time interval TR, is applied to a tissue. The magnitude of the transverse magnetisation varies within each TR time interval as a function of tissue relaxation times and the flip angle of the radiofrequency pulses. From quantum theory any radiofrequency pulse flip angle can be described by the set of probabilities of any given proton in the tissue under study observing a 0°, 90° or 180° flip angle (Table a1.1). Therefore it is possible to construct combinations of these observed flip angles which we can visualise as components of the total signal. Typically it is appropriate to consider two components, since the others are of such low statistical probability. Let us consider M_{xy} which is comprised of two components. The first component represents the free induction decay generated when tipping the longitudinal magnetisation (M_z) by 90°. This signal decays toward the end of the time interval TR. The second component is termed the spin echo. In contrast to the FID signal, the spin echo arises as a result of the combined effect of two consecutive radiofrequency pulses. The 90° component of the first RF pulse tips M_z to become M_{xy}. This magnetisation dephases in the immediate TR interval until it is acted upon by the 180° component of the next RF pulse, which refocuses the dephased M_{xy} during the next TR cycle. The echo may therefore be thought to be M_{xy} undergoing a T2 decay of two TR cycles when it is collected immediately before the RF pulse of the next TR cycle.

A1.3 The PSIF Sequence

A labelled pulse sequence timing diagram of a non diffusion-weighted PSIF sequence is shown in figure a1.1. Each component of the sequence is described below.

A. A radiofrequency pulse is applied at a narrow frequency range to ensure that it is observed only by the desired tissue.

B. The slice select gradient selects a slice of tissue by sensitising it to the narrow range of frequencies within the radiofrequency pulse. The gradient resets immediately before application of the subsequent RF pulse.

C. To sample the data and produce an echo a read gradient is applied and reset after data acquisition.

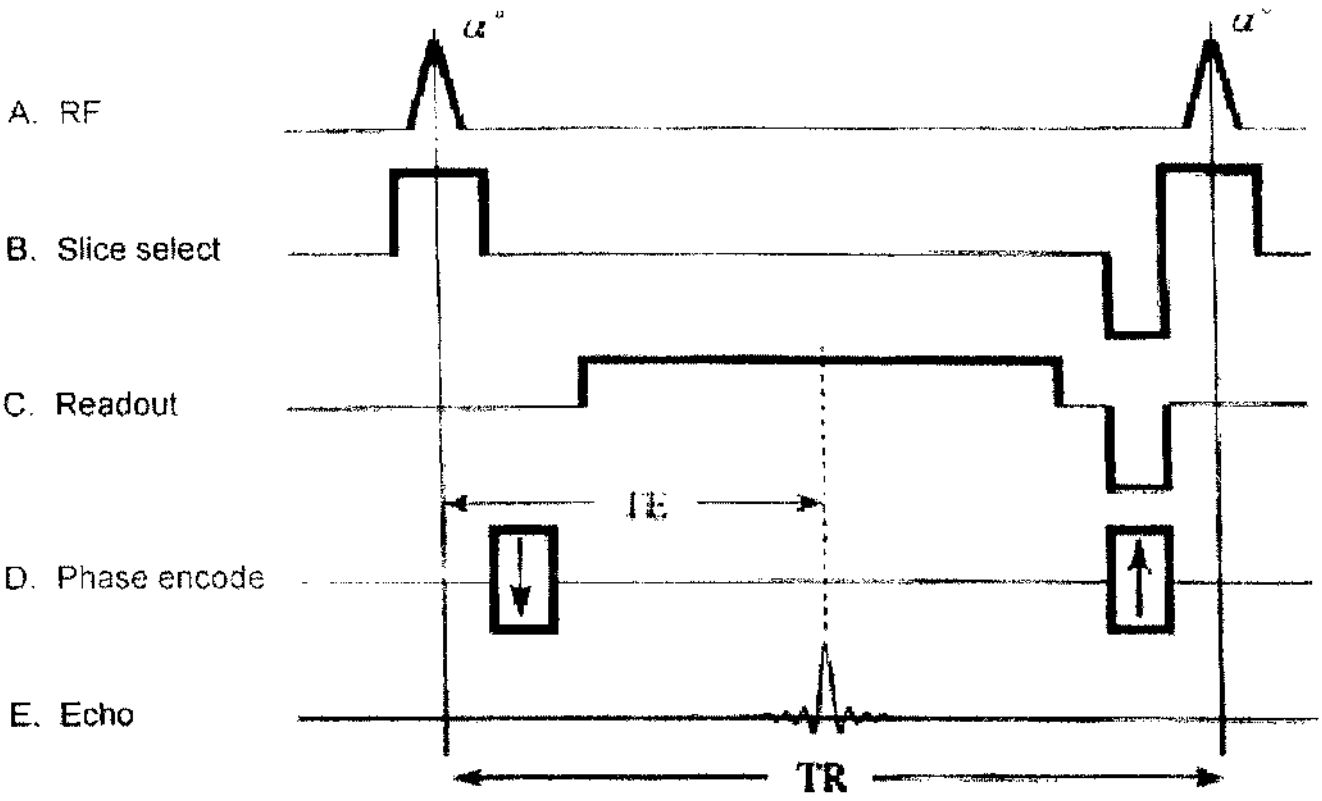
D. A range of phase encoding gradients in the phase direction is applied and then, after data collection, is reset by a gradient applied in the opposite direction.

E. The echo generated by the tissue under study is recorded. The echo time (TE) is defined as the time from the RF pulse to the centre of the echo. In the case of PSIF, the "effective TE" is much longer since several repeats of TR are required to produce an echo. TE is therefore much longer than TR, hence the image is T2 weighted.

Table A1.1 Quantum components of different flip angles

Flip angle	0°	90°	180°
0	1	0	0
180	0	0	1
90	0.5	1	0.5
45	0.854	0.707	0.146
135	0.146	0.707	0.854

Figure A1.1 Pulse sequence timing diagram of PSIF sequence



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