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# Introduction

## Background

Stroke, also known as cerebrovascular accident, is the third leading cause of death in Australia and is often referred to as the brain equivalent of the heart attack. Ischaemic stroke accounts for ~80% of all stroke cases and is the result of oxygen and nutrient deprivation to the brain caused by a narrowing or obstruction of the blood vessels. Conventional therapies for the management and prevention of stroke and its recurrence involve a combination of pharmacotherapy and rehabilitative therapies.

Traditional Chinese Medicine practitioners have been using plant and herbal medicine to treat a variety of illnesses for millennia. Recently, preliminary evidence has suggested that the use of herbal medicines can increase neurological recovery and enhance functional outcomes in stroke patient and thus may be beneficial in post-stroke rehabilitation. Nao Xin Qing (NXQ) is a standardised and patented herbal formula made from the extract of the *Diospyroskaki (D. Kaki L)* that has been used for numerous years in China for the treatment of cerebrovascular accidents and coronary artery disease. Preclinical studies have reported that the extract may be clinically useful for the treatment of cerebral atherosclerosis and transitory ischemia syndromes. A recent study from China showed that *D. Kaki* *L* extract effectively improved neurological outcomes and induce neuroprotective effects in individuals recovering from stroke. Although the preliminary evidence suggests that *D. Kaki L* may be clinically useful for a range of cerebrovascular conditions, there is a lack of evidence to support the efficacy of *D. Kaki L* extract products like NXQ in an Australian healthcare setting.

The researchers involved in this study propose to undertake a rigorous clinical trial to assess the clinical efficacy of NXQ treatment over 12-weeks in 88 adults who last suffered an ischaemic stroke up to 6-months but no less than 2-weeks ago. Global assessment of change, quality of life, cognitive and functional improvement, and safety will be assessed regularly over the period of the intervention (12 weeks) and follow-up (14 weeks).

This project is a collaborative effort involving researchers and clinicians across South Western Sydney Local Health District, Western Sydney University and the University of New South Wales. The results of this project will provide novel data regarding the efficacy and safety of NXQ for the treatment of Ischaemic stroke in an Australian healthcare setting. This project will also contribute to the growing body of evidence regarding the efficacy of herbal and complimentary medicines through national and international collaborations.

## Objectives

This multi-centre, double-blind, placebo controlled trial, will assess the effectiveness of NXQ for the symptomatic improvement of Ischaemic stroke in 88 participants who last suffered an ischaemic stroke up to 6-months but no less than 2-weeks. Specifically, the objectives of this study are to:

* determine the efficacy of NXQ on cognitive function, activities of daily living, and quality of life, and
* monitor the safety of NXQ as a treatment for Ischaemic stroke over 36 weeks

The investigators involved in this project hypothesise that a 12-week treatment of NXQ compared with placebo will be clinically effective and well-tolerated in participants with Ischaemic stroke.

## Efficacy Endpoints

Primary efficacy endpoints

The primary efficacy endpoint will be the difference between NXQ and the placebo groups for changes in National Institute of Health Stroke Scale (NIHSS) and Modified Rankin Scale (mRS) from baseline to 12- and 18-week follow up visits.

Secondary efficacy endpoints

The secondary efficacy endpoints of the study will be the differences between the NXQ and the placebo groups in the changes from baseline, 12- and 18-week follow up visits:

• Stroke-Specific Quality of Life (SSQOL) questionnaire

• Barthel Index (BI) for performance of daily living activities

• Lipid profile, hematology, coagulation index and inflammatory marker to assess the changes of vascular risk factors between baseline and the week-12 visit

• Montreal Cognitive Assessment (MoCA) to assess various aspects of cognitive and mental functions from baseline to 12-week follow up.

# Literature Review

Stroke, also known as cerebral vascular accident, is the brain equivalent to a heart attack. It is a disease with multiple overlapping risk factors, primarily of vascular origin, and is exacerbated by the ageing process and lifestyle factors such as dietary habits and smoking ([C. L. Allen & U. Bayraktutan, 2008](#_ENREF_2)). Stroke ranks as the third leading cause of death in Australia after coronary heart disease and many survivors are left with permanent disabilities ([Begg et al., 2007](#_ENREF_6); [Health & Welfare, 2019](#_ENREF_44)). It is estimated that over 375,000 Australians have suffered from stroke and one-third of these suffer from a disability which affects their daily life ([Health & Welfare, 2013](#_ENREF_43)).

Although the incidence of stroke has increased, increased awareness amongst the general public and health professionals regarding vascular risk factors and early symptom recognition has contributed to a 36.9% reduction in stroke mortality rates between 1999 and 2009 ([Go et al., 2013](#_ENREF_36)). As stroke survivorship is increasing, the cost of stroke treatment, including rehabilitation, is expected to increase proportionally ([Broderick, 2004](#_ENREF_13)). As of 2012, the stroke-related costs including prevention and rehabilitation were estimated to amount to $49.3b AUD ([Cadilhac, Carter, Thrift, & Dewey, 2009](#_ENREF_14); [NSF, 2013](#_ENREF_64)).

Stroke, by definition, is the rapidly developed clinical signs of focal (or global) disturbance of cerebral function, lasting more than 24 hours or leading to death, with no apparent cause other than of vascular origin (WHO, 1988). Stroke can be categorised into ischaemic and haemorrhagic sub-types where ischaemic stroke refers to a disturbance in blood supply to the brain and haemorrhagic stroke refers to a rupturing of blood vessels. Among the two, ischaemic stroke is the more common accounting for approximately 80% of all incidences ([Ekker et al., 2018](#_ENREF_31)), however the mortality rate is lower in comparison to haemorrhagic stroke ([Andersen, Olsen, Dehlendorff, & Kammersgaard, 2009](#_ENREF_4)). Although there are no defining features sufficient to clinically differentiate stroke type upon onset, resources are often directed to the treatment of ischaemic stroke due to its higher rate of incidences and potential therapeutic benefits whereas haemorrhagic stroke therapies are limited due to the rapid and extensive physiological damage, truncated therapeutic window and poorer short-term prognosis ([Musuka, Wilton, Traboulsi, & Hill, 2015](#_ENREF_60)).

## Ischaemic stroke

### Etiology and Risk Factors

Ischaemic stroke is caused by deprivation of oxygen and energy supply to the brain tissue and can be sub-categorised into thrombotic and embolic ischaemic stroke ([Lees, Bath, & Naylor, 2000](#_ENREF_53)). Thrombotic stroke occurs when a blood clot adhering to the vessel wall (mural occlusive thrombus) reaches a sufficient size to completely occlude cerebral arterial blood flow. The development of an occlusive thrombus due to atherosclerosis is a major risk factor of stroke ([Claire L Allen & Ulvi Bayraktutan, 2008](#_ENREF_3)).

Atherosclerosis is a condition where plaque builds up within an artery causing a narrowing of the blood vessel. The formation of this plaque has been shown to be associated with low-density lipoprotein cholesterol (LDL) and triglycerides levels however other factors such as hypertension, diabetes, smoking and aging have also been shown to contribute to the development of atherosclerosis ([Adams et al., 1993](#_ENREF_1); [Claire L Allen & Ulvi Bayraktutan, 2008](#_ENREF_3)). Atherosclerosis is a major risk factor for ischaemic stroke due to the development of atheroma which leads to increased plaque formation in the cerebral arteries. Atheroma can exacerbate the narrowing of the relatively small arteries in the brain resulting in increased blood pressure and reduced blood flow. Furthermore, should the fibrous cap of an atheroma ulcerate, ensuing thrombogenic material is released into the blood stream causing a thrombus to form ([Spagnoli, Bonanno, Sangiorgi, & Mauriello, 2007](#_ENREF_78)), thereby occluding the artery and causing ischaemic stroke. In addition to ischaemic stroke, atheroma produce enzymes that enlarge the arteries over time and this excessive widening may lead to the creation of an aneurysm with subsequent rupturing resulting in a haemorrhagic stroke ([Dzau, 1994](#_ENREF_30); [Kher & Marsh, 2004](#_ENREF_49)).

In contrast, embolic stroke, the most common form of ischaemic stroke, occurs when an embolus (e.g. a blood clot, atherosclerotic plaque, and rarely a fat globule or gas bubble) is carried by the bloodstream to the brain, where the larger arteries branch off into smaller vessels ([Ntaios & Hart, 2017](#_ENREF_65)). The embolus reaches a point in which it can travel no further thereby blocking a small cerebral artery opening and cutting off the blood supply to that area of the brain. Most blood clot emboli are caused by non-atherosclerosis factors such as atrial fibrillation (AF), where the two small upper chambers of the heart quiver causing the blood to pool and form clots ([Adams et al., 1993](#_ENREF_1); [Lees et al., 2000](#_ENREF_53)).

There exist multiple risk factors, both modifiable and non-modifiable which are associated with an increased risk of suffering ischaemic stroke. Modifiable risk factors include environmental and lifestyle factors such as high blood pressure, the consumption of calorie-dense diets high in fat and carbohydrates, insufficient physical activity and smoking ([C. L. Allen & U. Bayraktutan, 2008](#_ENREF_2)). These risks are typically managed with the help of healthcare professionals such as physicians, nurse practitioners, dietitians, and exercise therapists. Non-modifiable risk factors are related to hereditary or genetic processes such as ethnicity and ageing and cannot be altered ([Claire L Allen & Ulvi Bayraktutan, 2008](#_ENREF_3)).

### Pathophysiology

Ischaemic stroke often occurs as a result of a transient or permanent reduction in cerebral blood flow to a major arterial branch within the brain caused by the occlusion of arterial blood flow either by an embolus or local thrombosis ([Moustafa & Baron, 2008](#_ENREF_59)). The ensuing brain injury following the transient or permanent focal cerebral ischemia develops froming a complex series of pathophysiological events that evolve in time and space ([Dirnagl, Iadecola, & Moskowitz, 1999](#_ENREF_29)).

The human brain is the centre of the nervous system and exerts critical centralised control over most of the physiological and cognitive functions of the body. As a result, brain tissue requires relatively high consumption of oxygen and glucose, and depends almost exclusively on oxidative phosphorylation for energy production ([Dirnagl et al., 1999](#_ENREF_29)). Focal impairment of cerebral blood flow restricts the delivery of oxygen and glucose, leading to a functional failure of cellular membrane ion channels such as potassium, sodium and chloride pumps causing the plasma membrane of neurons to depolarise. The resultant cascade of pathophysiological processes such as excitotoxicity, peri-infarct depolarisation, inflammation and apoptosis either directly and/or indirectly lead to the death of neurons ([Dirnagl et al., 1999](#_ENREF_29); [Kehrer & Smith, 1994](#_ENREF_48)). The ensuing efflux of K+ and influx of Na+ and Cl- across the cell membrane following depolarisation causes increased water permeation into the neural cells via osmosis. The ensuing oedema can negatively affect the perfusion of blood to the regions surrounding the infract epicentre and can also have remote effects due to increased intracranial pressure, vascular compression and herniation ([Frank, 1995](#_ENREF_34)).

A further consequence of the membrane depolarisation is that the substantial influx of Ca2+ allows for excitatory amino acids to be released into the extracellular space ([Dirnagl et al., 1999](#_ENREF_29)). At the same time, the energy dependent processes such as presynaptic reuptake of excitatory amino acids are impeded, further increasing the accumulation of excitatory amino acids like glutamate in the extracellular space ([Dirnagl et al., 1999](#_ENREF_29)). The presence of these excitatory amino acids result in the activation of N-methyl-D-aspartate (NMDA) receptors and a specific glutamate receptor sub-type known as metabotropic receptors ([Dirnagl et al., 1999](#_ENREF_29)). The combined over-activation of these receptors from accumulated extracellular excitatory amino acids contribute to the cellular Ca2+ overload in the neural cell ([Dirnagl et al., 1999](#_ENREF_29); [Kehrer & Smith, 1994](#_ENREF_48)). This increase in Ca2+ is thought to initiate a cascade of cytoplasmatic and nuclear events that result in cellular destruction and activates inflammatory responses thereby generating free-radical species that overwhelm endogenous scavenging mechanisms ([Dirnagl et al., 1999](#_ENREF_29); [Ginsberg, 2008](#_ENREF_35)). These free-radical species damage the cellular membrane allowing further permeability of Ca2+ ions. Of these free-radical, reactive oxidation species (ROS) serve as important signalling molecules that trigger inflammation and apoptosis ([Coyle & Puttfarcken, 1993](#_ENREF_26)).

Studies have shown a strong correlation between neurodegenerative diseases including stroke and Alzheimer’s disease and ROS ([Coyle & Puttfarcken, 1993](#_ENREF_26)). It has been reported that there may be a surge in the generation of ROS during cerebral ischemia, particularly at the onset of reperfusion ([Kalogeris, Baines, Krenz, & Korthuis, 2012](#_ENREF_46)). ROS including superoxide, hydrogen peroxide and hydroxyl radicals are highly reactive and could break down cell membranes, damage DNA, create oxidation on proteins and amino acids and inactive specific anti-oxidant enzymes. Neurons in particular consist of abundant poly-unsaturated fatty acids and produce low amount of antioxidant enzymes and therefore are highly susceptible to ROS ([Kehrer & Smith, 1994](#_ENREF_48)). Moreover, cerebral ischemia could lead to increased level of excitatory amino acids such as glutamate, which generates more ROS and enhances cellular destruction ([van der Worp & van Gijn, 2007](#_ENREF_84)). As these effects are considerably more pronounced in tissue areas surrounding the focal point of the infarct, neuroprotection is therefore a critical yet relatively undeveloped therapeutic strategy for minimising neuron damage in ischaemic stroke patients.

Although recent research focuses on the survival of neurons via neuroprotection and anti-thrombotic therapies, little attention has been directed towards investigating the role of angiogenesis ([Krupinski, Kaluza, Kumar, Kumar, & Wang, 1994](#_ENREF_50)). During an ischaemic attack, a brain region with low perfusion where cells have lost their membrane potential is known as the “core” and is surrounded by an area called a penumbra where intermediate perfusion prevails and cell depolarise intermittently (peri-infarct depolarisation). By definition, a penumbra is an area where ischaemic tissue is potentially destined for infarction however is not yet irreversibly injured and is therefore an important target of acute therapy ([Dirnagl et al., 1999](#_ENREF_29)). Necrosis resulting from an ischaemic infarct is irreversible, however, studies have suggested that penumbra area may remain viable for many hours after an ischaemic event due to alternative blood supply from collateral arteries ([Dirnagl et al., 1999](#_ENREF_29)). A study by Krupinski et al. ([1994](#_ENREF_50)) investigating angiogenesis and ischaemic stroke reported that adequate perfusion through the penumbra can initiate angiogenesis. Evidence suggests that high blood vessel density is correlated with improved patient survival, independence and clinical outcome after stroke ([Choo, 1993](#_ENREF_20)). There is also a strong correlation between the extent of spontaneous neurological recovery and the volume of penumbra that escapes infarction ([Cheng, Al-Khoury, & Zivin, 2004](#_ENREF_19); [Dirnagl et al., 1999](#_ENREF_29)). Therefore, enhancing the angiogenic effect is another key aspect to improving clinical outcomes in stroke recovery.

## Conventional Management for Ischaemic Stroke

The overarching goal of stroke treatment is to increase the survival rate and reduce the dependency level of the patient ([Dirnagl et al., 1999](#_ENREF_29)). Conventional management for ischaemic stroke is comprised of three therapeutic strategies including prevention, general treatment and rehabilitation ([Ringleb et al., 2007](#_ENREF_71)). Current pharmacological therapies for prevention and treatment display considerable overlap. Anti-thrombotic therapy is a primary therapy employed as a treatment modality whereas neuroprotective strategies may be beneficial in both prevention and treatment ([Lansberg et al., 2012](#_ENREF_51)).

Pharmacological therapies for ischaemic stroke including anti-thrombotic medication are often time-sensitive (e.g. intravenous thrombolysis) and require rapid response to achieve the optimal clinical outcomes ([Ringleb et al., 2007](#_ENREF_71)). Anti-thrombotic therapies such as anti-platelet agents, anti-coagulants and thrombolytic drugs typically act on thrombosis or embolisms by thinning the blood, however they are associated with an increase in the incidence of haemorrhagic events ([Gubitz, Sandercock, & Counsell, 2008](#_ENREF_40); [Sandercock, Counsell, Gubitz, & Tseng, 2008](#_ENREF_72); [Wardlaw, Murray, Berge, & Del Zoppo, 2009](#_ENREF_86)).

 Neuroprotective therapies are currently considered a secondary modality due to the nature of ischaemic stroke ([Neuhaus, Couch, Hadley, & Buchan, 2017](#_ENREF_63)). The role of neuroprotection is considered an important therapeutic strategy in reducing neuron damage and dependency levels, and has been suggested to expand the treatment time window and reduce reperfusion injury following ischaemic stroke ([Cheng et al., 2004](#_ENREF_19); [Ginsberg, 2008](#_ENREF_35)). In addition to rehabilitation therapies, long-term pharmacotherapy is typically prescribed for the management of vascular and thrombotic risk factors with the goal of preventing the recurrence of stroke ([Ringleb et al., 2007](#_ENREF_71)).

Rehabilitation programs which include therapeutic therapies such as physiotherapy, occupational therapy, speech and language therapy are often implemented to assist stroke survivors in restoring functionality lost due to neurological damage ([Feigenson, 1979](#_ENREF_32)). The rehabilitative process is a long-term and continual process and depending on the severity of disability, many stroke survivors do not regain full functionality.

Stroke rehabilitation is centred around the recovery of motor and cognitive function, which lead to improved quality of life ([Hatem et al., 2016](#_ENREF_42)). Despite the variety of available pharmaceutical and adjunctive treatment options, there are shortfalls with each that have yet to be overcome ([Ringleb et al., 2007](#_ENREF_71); [van der Worp & van Gijn, 2007](#_ENREF_84)).

### Anti-thrombotic Therapy

General treatment usually occurs in a hospital setting after the onset of stroke. Treatment method varies depending on the severity and the time of treatment after onset ([Ringleb et al., 2007](#_ENREF_71)). The primary therapeutic aim is to minimise cerebral necrosis that is usually caused by arterial occlusion or thrombosis and prevent reoccurrence of ischaemic attack ([Ringleb et al., 2007](#_ENREF_71)).

Acute management such as intravenous thrombolysis with rtPA (Intravenous Tissue Plasminogen Activator) is currently given within first 4.5 hours after the onset of an ischaemic stroke to dissolve thrombotic embolisms ([Ringleb et al., 2007](#_ENREF_71)). This therapy has been found to decrease post-stroke dependency, despite an increased risk of cerebral haemorrhage (Wardlaw et al. ([2009](#_ENREF_86)). However, the majority of ischaemic stroke patients currently do not present within the narrow therapeutic time window for this type of therapy.

Anti-platelet treatment is considered as a valuable primary treatment and secondary preventative modality for ischaemic stroke ([Ringleb et al., 2007](#_ENREF_71)). In some patients, use of anti-platelet therapy may increase the risk of adverse effects including intracranial haemorrhage ([Sandercock et al., 2008](#_ENREF_72)). Whilst there are numerous anti-platelet pharmaceuticals available, aspirin is the most common anti-platelet agent administered for ischaemic stroke ([Mohr et al., 2001](#_ENREF_58); [NSF, 2013](#_ENREF_64); [Sandercock et al., 2008](#_ENREF_72)). Evidence based on the results of two large randomised, non-blinded, intervention studies indicate that aspirin is safe and effective when started within 48 hours post- stroke and is beneficial for acute ischaemic stroke ([Z. Chen et al., 2000](#_ENREF_18); [Sandercock et al., 2008](#_ENREF_72)).

However, long term use of aspirin may increases the risk of gastrointestinal bleeding and irritation and this adverse effect is likely to be more apparent in elderly individuals ([Guazzi, Brambilla, Reina, Tumminello, & Guazzi, 2003](#_ENREF_39); [Silagy et al., 1993](#_ENREF_77)).

Other pharmacotherapies for the treatment and prevention of ischaemic stroke include anti-coagulant agents such heparin and warfarin. This therapy is typically employed in patients with existing emboli such as deep vein thrombosis. However, there has been conflicting evidence regarding the efficacy of anti-coagulant medication for the treatment or prevention of non-cardioembolic ischaemic stroke. A review by Gubitz et al. ([2008](#_ENREF_40)), reported that anti-coagulants do not provide any significant overall short- or long-term reduction in death and disability. Conversely, older anti-coagulants such as warfarin appear to result in fewer recurrent ischaemic stroke incidents; however, this benefit is offset by a similarly-sized increased risk for intracranial haemorrhage ([Gubitz et al., 2008](#_ENREF_40); [Ringleb et al., 2007](#_ENREF_71)). Recently, non-vitamin K antagonist oral anticoagulants have begun to replace warfarin for the treatment and prevention of stroke due to their superior efficacy and safety ([Saraiva, 2018](#_ENREF_73)).

### Neuroprotection

The primary goal of neuroprotective therapy is to salvage the ischaemic penumbra ([Dirnagl et al., 1999](#_ENREF_29)). It has been well documented that abrupt deprivation of oxygen and glucose to neuronal tissues elicits a series of pathological cascades leading to the spread of neuronal death ([Ringleb et al., 2007](#_ENREF_71)). However, neuroprotective treatment has yet to be implemented in conventional treatment guidelines due to a lack of clinical evidence ([Patel & McMullen, 2017](#_ENREF_70); [Ringleb et al., 2007](#_ENREF_71)).

Neuroprotective agents, including glutamate receptor antagonist, calcium channel blockers, anti-inflammatory agents and free radical scavengers, designed to block these cascades, have been increasingly investigated in animal models of cerebral ischemia ([Cheng et al., 2004](#_ENREF_19)). Numerous agents have been found to reduce infarct size in rodents, rabbits, and primate stroke models ([Gorelick et al., 1999](#_ENREF_37); [Osuntokun, Bademosi, Akinkugbe, Oyediran, & Carlisle, 1979](#_ENREF_67)). However, the neuroprotective benefits observed in these laboratory experiments have not been repeated in subsequent clinical trials ([Cheng et al., 2004](#_ENREF_19)).

Whilst blocking glutamate receptors can protect against neurological excitotoxicity, these glutamate receptor antagonists exhibit serious adverse effects such as psychotomimesis, respiratory depression and cardiovascular dysregulation ([Dirnagl et al., 1999](#_ENREF_29)). Therefore the benefits of such a treatment found in clinical trials have been outweighed by these effects and long term preventative use is discouraged ([Dirnagl et al., 1999](#_ENREF_29)).

Clinical trials employing ion channel modulators including calcium channel antagonists, potassium channel activator and sodium channel blocker were all terminated prematurely due to lack of sufficient benefits ([Cheng et al., 2004](#_ENREF_19)).

The efficacy of free radical trapping agents such as Citicoline (cytidyldiphosphocholine) for improving stroke outcomes have been studied in preclinical and clinical trials. Citicoline is a phosphatidylcholine precursor that has membrane stabilization properties but might also have other neuroprotective attributes. Preclinical studies in animal stroke models showed that Citicoline improved neurological outcomes and reduced infarct size ([D'Orlando & Sandage Jr, 1995](#_ENREF_27); [Schäbitz et al., 1999](#_ENREF_74)) however, the clinical phase III trials could not replicate these findings despite employing a variety of dosing schedules ([W. Clark, Warach, Pettigrew, Gammans, & Sabounjian, 1997](#_ENREF_21); [W. M. Clark et al., 1999](#_ENREF_22)).

### Post-Stroke Rehabilitation

Even with optimal stroke unit care including thrombolysis, less than one third of patients recover fully from stroke ([Hacke et al., 2004](#_ENREF_41)). Rehabilitation aims to assist people with disabilities to reach and maintain optimal physical, intellectual, psychological and social function. It is a fundamental facet for post-stroke patient to increase independency and functional improvements ([Coleman et al., 2017](#_ENREF_23)). Current available rehabilitation modalities such as physiotherapy, occupational therapy, speech therapy and nursing could help improve functional outcomes for many stroke patients ([Feigenson, 1979](#_ENREF_32)). A meta-analysis concluded that continuous rehabilitation within the first year after being discharged from hospitalisation can reduce functional deterioration and improves activities of daily living ([Legg, 2004](#_ENREF_54)). However, this improvement is largely associated with the quality of the rehabilitation as opposed to the duration.

Despite these benefits, cognitive deficits are common following stroke and have a significant impact on quality of life of the stroke patients. There is limited evidence supporting the efficacy of specific memory and cognitive rehabilitation ([Das Nair & Lincoln, 2007](#_ENREF_28); [Merriman et al., 2019](#_ENREF_57)). This cognitive aspect of post- stroke rehabilitation is particularly important as cerebrovascular incidences have been shown to correlate with increased risks of severe cognitive deficits such as dementia ([Ivan et al., 2004](#_ENREF_45); [Tatemichi et al., 1994](#_ENREF_82)).

In summary, it is apparent that the conventional management of stroke remains insufficient as it narrowly focuses on dissolving/preventing emboli formation and a majority of the therapeutic benefits are time dependent and not applicable to the majority of acute stroke presentations. Consequently, there is an urgent need to develop new therapies, which are not only safer but also can treat the underlying pathological conditions associated with stroke.

## Traditional Chinese Herbal Medicine for Ischaemic Stroke

The cause of ischaemic stroke from a Traditional Chinese Medicine (TCM) perspective is referred to as an obstruction and/or stagnation. Preclinical studies have suggested that the use of herbal medicines can increase neurological recovery and enhance functional outcomes in stroke patient and thus may be beneficial in post-stroke rehabilitation ([Lee, Lee, Chang, Chien, & Lin, 2005](#_ENREF_52); [Liu et al., 2007](#_ENREF_56); [Shi, Hart, Sherman, & Tegeler, 1989](#_ENREF_76)).

Nao Xin Qing is a standardised and patented herbal extract from *D. Kaki L* that has been used for numerous years in China for the treatment of cerebrovascular accidents and coronary artery disease ([Bei, Peng, Ma, & Xu, 2004](#_ENREF_8); [Bei, Zang, et al., 2009](#_ENREF_11); [Cai & Yang, 2001](#_ENREF_15)). The key bioactive components of *D. Kaki* L. have been identified as flavonoids (e.g., quercetin and kaempferol) ([Bei, Peng, Ma, & Xu, 2005](#_ENREF_9)). Whilst this patented extract has been shown to induce anti-hypertensive and anti-oxidative effects ([Cai & Yang, 2001](#_ENREF_15); [Yu, Yu, & Guo, 1988](#_ENREF_95)), both clinical and preclinical studies have also suggested that the extract may therapeutically effective for the treatment of cerebral atherosclerosis, transitory ischemia, cerebral thrombogenesis, cerebral thrombosis sequel, cerebral embolism and with minimal adverse effects ([Bei et al., 2007](#_ENREF_10); [Cai & Yang, 2001](#_ENREF_15)).

### Chemical Definition and Preclinical Studies of NXQ

Laboratory studies have quantified more than 30 potentially bioactive compounds isolated from the leaves of *Diospyros* species. These can be divided into five main chemical groups including acids, biphenyls, flavonoids, polyphenols and triterpenoids ([Bei et al., 2004](#_ENREF_8), [2005](#_ENREF_9)). Amongst these chemical compounds, flavonoids are considered to be the key therapeutic components responsible for the observed pharmacological and clinical effects due to their biological effects including antioxidant, antiallergenic, anti-inflammatory, and vasodilatory actions ([Bei et al., 2004](#_ENREF_8); [Cao, Zhang, Bai, Wang, & Miao, 2012](#_ENREF_16)). The chemical composition of NXQ consists of more than 50% organic acids including protocatechuic acid, benzoic acid and scopoletin and over 25% flavonoids including quercetin and its glucosides; hyperin, isoquercitrin; kaempferol and astragalin ([Bei et al., 2004](#_ENREF_8)).

Previous research has demonstrated numerous pharmacological effects of *D. kaki L* extract such as improved cardiac and cerebral blood flow, anti-hypertensive effects, lipid lowering effects and radical salvaging capabilities ([Cao et al., 2012](#_ENREF_16)). A study conducted by Xin, Feng & Yao ([2007](#_ENREF_93)) investigated the effects of intravenous injections of *D. Kaki L* flavonoids and found a 30 to 40% improvement in aortic circulation in rabbits as well as significant dilation in the veins of the ears. These findings corroborate prior research by Y. Zhang, Wang & Xiao ([2004](#_ENREF_96)) who found that the leaves of *D. Kaki L* can improve overall circulation in anaesthetised dogs and can reduce oxygen consumption of cardiac muscles. Ou, Bei & Lai ([2003](#_ENREF_68)) found that the total flavonoid count can significantly inhibit reperfusion induced cell apoptosis. Furthermore, Ou, Liu & Bei ([2004](#_ENREF_69)) also suggested that *D. kaki L* can inhibit fibrin adhesion (scar tissues) on the adventitia stimulated by factors such as advanced glycation end-products (AGEs) and advanced oxidation protein products (AOPP).

 Cao et al. ([2012](#_ENREF_16)) studied the fibrinolytic function in an ischaemic brain injury model and suggested that *D. kaki L* flavonoids can increase tissue Plasminogen Activator (t-PA), which is involved in the breakdown of blood clots and reduction of Plasminogen activator inhibitor-1 (PAI-1) levels. Cao et al. ([2012](#_ENREF_16)) further demonstrated that the von Willebrand Factor (vWF), a blood glycoprotein involved in haemostasis, is reduced in ischaemic brain injury rats subjected to *D. kaki L* flavonoids. Reduction of vWF has been shown to be associated with the resolution of thrombosis during an ischaemic attack and restoration of damaged endothelial cells. Cao et al. ([2012](#_ENREF_16)) proposed that *D. Kaki L* flavonoids could reduce positive signalling of Intercellular Adhesion Molecule 1 (ICA-1), a immunoglobulin molecule associated with the release of inflammatory factors, and thereby act against atherosclerosis and reduce sustain haemostasis.

Bei, Li, et al. ([2009](#_ENREF_7)) proposed that the potential neuroprotective effects of NXQ and *D. Kaki L* flavonoids could activate L-type Ca2+ channels thus modulating intracellular Ca2+ level to protect neurons in the hippocampus. This action is complemented by the anti-oxidative ability of NXQ and *D. Kaki L* which positively inhibits ROS including hydrogen peroxide and may therefore reduce neuron damage.

Kameda et al. ([1987](#_ENREF_47)) first demonstrated the anti-hypertensive effects of *D. Kaki L* in hypertensive rats. The mechanism of action is thought be through inhibition of angiotensin –converting enzyme activity (Kameda et al. 1987). Tan, Lin & Zhang ([2009](#_ENREF_80)) have further investigated these antihypertensive effects and observed an increase in plasma nitric oxide (NO) and reduced over-production of platelets by essential thrombocytosis (ET) and reduced angiotensin II in rats. The results suggested that *D. Kaki L* flavonoids have potential to enhance and modulate endogenous vasodilators and vasoconstrictors, respectively.

### Clinical Studies of NXQ

Recently, several clinical trials were conducted in China assessing the clinical effects of NXQ. One study assessed the efficacy of 6 weeks of NXQ as an additional therapy to standard treatment for 35 adults with acute ischemic stroke and type 2 diabetes ([Tang et al., 2012](#_ENREF_81)). The findings of this study showed that NXQ positively improved C-reactive protein and blood lipids. The results suggested that the underlying mechanism of action may be linked to the active component flavonoids from *D. Kaki L* extract which elicit anti-ischaemic effects on cerebral and cardiac blood flow, and the neuroprotective effects were elicited through radical savaging and anti-atherosclerotic effects by lowering blood lipids.

Another study conducted by Wang et al. ([2012](#_ENREF_85)) compared NXQ in combination with standard therapy to standard therapy alone for improvements in disease-specific outcomes in patients with cerebro-cardiac syndrome cause by acute stroke. A total of 204 participants were included in the study, where 130 patients had suffered from ischaemic stroke, 61 participants had suffered from intracranial haemorrhages and 13 participants had suffered from subarachnoid haemorrhage. A standard dose of NXQ was given during the acute phase in addition to acute management therapy protocols for a 4-week period. The results suggested that the NXQ group had superior effects in improving cardiac function recovery including arrhythmia, changes in ST-T wave and abnormal cardiac enzyme production in comparison to control ([Wang et al., 2012](#_ENREF_85)).

Y. Chen, Shao, & Cheng ([2012](#_ENREF_17)) conducted a clinical trial where they compared NXQ as an adjunctive therapy with heparin versus heparin alone on 160 cerebral transient ischaemic attack patients for 3 to 7 days. The results indicated that both groups had prolonged partial thromboplastin (PT) time and active partial thromboplastin time (APTT) however NXQ showed significantly longer duration in both outcome measures when compared to controls. In addition, no significant adverse events were identified ([Y. Chen et al., 2012](#_ENREF_17)).

Although there are limited clinical studies assessing the effects of NXQ on cerebral ischaemic stroke, three further clinical trials were conducted assessing the effects of NXQ for ischaemic heart disease and angina which share a similar underlying pathological process with ischaemic stroke - atherosclerosis. A study conducted by Cai & Yang ([2001](#_ENREF_15)) in 48 atherosclerosis cases and 12 coronary artery disease induced angina cases and showed that NXQ can significantly lower blood lipids, and improve symptoms such as vertigo and headaches ([Cai & Yang, 2001](#_ENREF_15)). Furthermore, a recent study by D. Wu, Wu & Zheng ([2013](#_ENREF_90)) evaluated 90 patients with coronary artery disease and angina comparing NXQ against a standard vasodilator and revealed similar positive results. The authors reported significant results favouring the NXQ intervention for improving lipid levels and suggested that the significant reduction of cardiac stress, cardiac oxygen consumption, and improvements in oxygen supply to cardiac muscle was through aorta dilation ([D. Wu et al., 2013](#_ENREF_90)). This result was corroborated by Xiong, Tang & Zhou ([2013](#_ENREF_94)) who assessed the clinical effects of standard angina treatment including nitrates, aspirin, beta blockers, angiotensin-converting enzyme inhibitors and lipid lowering drugs compared to standard treatment combined with NXQ in 120 elderly patients with unstable angina. The results suggested that the addition of NXQ to standard treatment led to significant improvements in blood lipids levels and helped alleviate symptoms of angina ([Xiong et al., 2013](#_ENREF_94)).

Recently, an internal report from the manufacturer of NXQ reported the results of a randomised controlled trial assessing the effects of NXQ on ischaemic stroke for 12 weeks. The report showed that NXQ significantly improved NIHSS (Δ -6.79 ± 2.65; p < 0.0001). While placebo also improved NIHSS (Δ -3.18 ± 2.34; p <0.0001), there was a significant difference between NXQ and placebo groups favouring the NXQ group (p > 0.0001). Similar results were also observed for other outcome measures including SSQOL, mRS, and Barthel index in which NXQ performed significantly better than placebo (p < 0.0001 for all).

In summary, the preliminary clinical evidence suggests that NXQ may improve lipid levels and cardiac blood flow as well as relieve symptoms of angina. No severe adverse events were identified when NXQ was used in conjunction with anti-coagulant therapy. However, despite these positive results, the methodology of these trials did not appear to be rigorous and precise. At present there are no publicly available data reporting the efficacy of NXQ for ischaemic stroke outcomes, the considerable preclinical evidence provides a preliminary foundation and justification for further clinical investigations into the use of NXQ in ischaemic stroke.

## Rationale of the study

Ischaemic stroke accounts for more than 80% of all stroke incidences and have substantial disability rate ([Andersen et al., 2009](#_ENREF_4)). The current available pharmaceuticals administered after an acute ischaemic stroke such as anti-thrombotic agents are primarily aimed at secondary prevention. They are associated with increased risk of adverse events and have no neuroprotective benefits and no proven efficacy on rehabilitation. Preliminary studies have revealed the potential therapeutic effects of NXQ for ischaemic vascular diseases such as anti-lipidemic, neuroprotective, anti-inflammatory, anti-hypertensive and reduce haemorrhagic risks ([Bei et al., 2004](#_ENREF_8); [Cao et al., 2012](#_ENREF_16)).

This study will be conducted as a randomised, parallel, placebo control trial to determine the effectiveness and safety of NXQ in the cohort of ischaemic stroke patients during rehabilitation phase.

## Research Hypothesis

### Primary Hypothesis

A 12 week of NXQ treatment can improve neurological, motor and cognitive functional outcomes, and the quality of life of ischaemic stroke patients during the rehabilitation phase.

### Secondary Hypothesis

A 12 week of NXQ treatment can prevent the reoccurrence of ischaemic stroke and reduce vascular risk factors of stroke.

# Method

## Study Design

This study will be conducted as a two-arm randomized, double-blind, placebo controlled clinical trial of 36 weeks, including a 12-week intervention and 26-week follow-up.

For a two-arm trial, participants will be randomized after informed consent is obtained, into two parallel treatment groups:

a. Intervention group taking *NXQ* (Active)

b. Placebo control group (Placebo)

The trial will be double-blinded, such that the participants and investigators (including persons responsible for data collection, data management and data analysis) will not be aware of randomisation assignments. A run-in period is not required for this trial.

## Sample

**Sample Size and Randomisation**

This trial is a preliminary study to evaluate the effects of NXQ in comparison to placebo in patients with ischaemic stroke during rehabilitation phase

Thirty-seven participants per group provide 80% power to detect a detect a 2 unit difference in mean NIHSS score between treatment and control groups at follow-up, at the 0.05 significance level using a 2-sided independent samples test t-test and assuming a standard deviation of 3 units (G-Power software, University of Trier, Trier, Germany). The standard deviation of 3 units is consistent with a previous report involving a similar population and investigational product ([Wei et al., 2017](#_ENREF_87)). A 2-unit improvement in mean NIHSS score represents a non-controversial improvement, which has the potential to lead to changes in clinical care (despite any additional cost and/or pill load). The target sample size will therefore be 88 participants in total, 44 per group, which includes 20% dropout boundary.

Once the patients have been recruited, they will be randomly allocated to the NXQ intervention group or placebo control group by means of a computer randomisation package.

Randomisation will be conducted by a NICM Clinical Research Coordinator who is external to the research team. The research team will be concealed to the randomisation process. The NICM Clinical Research Coordinator will prepare individually assigned and sealed envelopes containing the participant's group allocation which will only be opened once the participant is confirmed to be eligible for the study. Participants will also be stratified based on disease type and treatment received.

*Inclusion Criteria*

To participate in this study, participants must:

* Be between the ages of 40-80 years;
* Be an outpatient diagnosed with atherosclerotic ischaemic stroke;
* Have suffered from a stroke no less than 2 weeks and no more than 3 months at screening;
* Have an NIHSS score equal or greater than 5 scores and equal or less than 25 scores;
* Have a premorbid Modified Rankin Scale (mRS) score between 0 and 2;
* Agree to take part in the study as evidenced by a personally signed and dated informed consent document indicating that the subject (or a legally acceptable representative if the subject is unable to provide consent), has been informed of all pertinent aspects of the study;
* Ability to read and communicate in the English language.

*Exclusion Criteria*

Participants will be excluded from this study if they have any of the following:

* Cerebral haemorrhagic stroke.
* Clinical assessment concluding the cause of ischaemic stroke from brain tumour, trauma, metabolic disorders, rheumatic valvular heart disease, and infectious cause of stroke.
* Participant has non-valvular atrial fibrillation.
* Concomitant clinical conditions affecting neural and motor function assessment including pre-existing dementia, inflammatory or non-inflammatory arthropathies or other medical disorders that result in a premorbid mRS of 3 or greater.
* Abnormal pathology test results: Cr > 1.5 times upper limit of normal (ULN); ALT, AST or ALP > 2 times ULN; PT > 3 second more than ULN; APTT > 10 seconds more than ULN; Plt < 100,000/mcL;
* Patients with severe depression or other psychiatric disorders that have not been stabilised for > 3 months prior to randomisation.
* Known allergy to the medication ingredients.
* Participant of another clinical trial within the past 3 months.
* Consuming *D. Kaki L* extract.
* Participant requires combination of dual antiplatelet therapy, for example, aspirin and clopidogrel with the exception of aspirin and dipyridample.

**Discontinuation of Participants from Treatment or Assessment**

Participants who enter the trial, finish their baseline examination, are allocated to randomised treatment and begin taking the blinded medication will be counted as withdrawal cases if, during the study if they:

* Become pregnant;
* Withdraw informed consent. Participants are free to discontinue their participation in the trial at any time, without prejudice to further treatment;
* Become lost to follow-up;
* Demonstrate significant protocol non-compliance as determined by the investigator; or
* The Investigator considers that it is not in their interest to continue the study.

The study may be discontinued at any time by the sponsor or the Coordinating Chief Investigator on the basis of new information regarding safety or efficacy. Additionally, the study may be terminated if progress is unsatisfactory. Withdrawn/discontinued participants will not be replaced.

**Procedures for discontinuation**

In case of premature termination or suspension of the trial, the coordinating chief investigator must inform the trial participants and ensure appropriate follow up and therapy. In addition, the regulatory authorities and ethics committee must be informed.

Participants may withdraw/discontinue from the study at will at any time without explanation. The participant may also be withdrawn at the discretion of the Investigator due to a safety concern or if judged non-compliant with trial procedures. Participants that withdraw/discontinue from the study will be asked to attend the end of treatment visit (Visit 8, Week 12) to monitor their safety, with a window period of up to 14 days of their withdrawal. The reason for withdrawal and date of withdrawal must be documented in the participant’s source data file and electronic Case Report Form (CRF). All AEs and SAEs must be followed until resolution or stabilisation unless, in the Investigator’s opinion, the condition is unlikely to resolve.

**Concomitant Treatment**

Participants may take routinely prescribed medications provided the relevant condition has been stable or controlled by optimal medication for more than 3 months except those drugs prohibited by the Exclusion Criteria. Participants are permitted to take aspirin, but cannot take aspirin in combination with clopidogrel. For other medications (including herbal medicine and nutritional supplements), which may have cognitive effects (e.g., cholinesterase inhibitors, beta blockers, angiotensin receptor blockers), the doses should be stable for at least 3 months for all medications prior to the commencement of the trial and remain stable for the duration of the study. The details of all medical treatments and health management plans will be recorded as part of the screening process and then, following enrolment, during every follow-up visit. Notification of a person’s enrolment in the trial and the implications for management will be communicated, with their consent, to their General Practitioner or treating Neurologist. It is anticipated that the presence or effect of any existing treatment confounders or effect modifiers will be equally distributed between groups due to the randomisation process.

To achieve the aim of this study, it is desirable that the participant group is relatively homogenous with regard to health profile and disease risk factor presentation to ensure internal validity of the results. However, a broad representation of Australian adults according to demographic and social factors will be sought to improve generalisability of the study findings for the community. Participants will not be excluded based on gender, ethnic, or social background.

## Study Protocol

*Figure 1:* Schematic trial protocol

Informed consent

Participant recruitment

Baseline/Randomisation (Week 0)

(n = 226)

Screening Health Measurements (Week - 2)

GROUP 1: *NXQ* (n=44)

GROUP 2: Placebo (n=44)

Follow up safety and outcome health measurements (Week 36)

Data Analysis

Safety and outcome health measurements (Weeks 2, 6, 12, 18)

Recruitment Protocol

A total of 88 Participants will be recruited through referrals by clinicians/practitioners involved in this trial and their associated hospitals, clinics and networks, advertising flyers at participating venues, contacting support groups, advertising via newsletters and websites of NICM Health Research Institute and Stroke Foundation Australia, and media advertising – Facebook, Twitter, Instagram, newspapers.

Initial contact will be made by phone or email to provide background information to potential participants and to identify if the candidate is potentially eligible. The referring clinicians will have knowledge of the inclusion / exclusion criteria to assist participants in deciding if they may be eligible to participate. They will also have copies of the participant information sheet, should potential participants require additional information before deciding to enrol for screening.

The CI based at Western Sydney University will have an overall responsibility for the recruitment efforts pertaining to this trial.Participants who self-refer through advertising, will be initially provided information over the phone or through email contact by a member of the research team based at Western Sydney University. The CI from Western Sydney University will be supported by the local research team of the PI at Liverpool Hospital who will have access to the consent form and participant information sheet which will be provided when participants express interest in the trial or subsequently when they are screened with the clinician.

Resources required for the recruitment phase of the trial include advertising materials, Health Practitioners Information Packages (information sheets for practitioners, information sheets for administration staff, information sheets and contact cards for potential participants). Participants may also be directly mailed information about the trial. Participants can take as much time as is required to consider their participation. Translator services may be engaged to facilitate communication for individuals from culturally and linguistically diverse backgrounds.

Screening Appointment Protocol

It is anticipated that 88 participants can be recruited over a 24-month period. The participants will be assessed and classified according to the NIHSS and mRS diagnostic criteria. Other assessments will include: history and examination, blood tests, and neuropsychological assessment if deemed necessary. Participants will be required to sign informed consent prior to the collection of any personal or health information at the screening visit.

Randomisation and Allocation Concealment Protocol

Randomisation will be conducted external to the primary research team (investigators listed at the start of this document) by a research officer of NICM Health Research Institute. This person will be responsible for producing computer generated randomisation treatment sequences, which randomly associate a randomisation number with either active or placebo treatment. NICM’s Clinical Trials Manager and Strategic Operations Manager are responsible for securely storing the randomisation allocation in a restricted access electronic file.

Randomisation numbers will be allocated in permuted blocks of 4 randomisation numbers starting from 01 to 88 with each block containing 2 active and 2 placebo assigned randomisation numbers. Sites will allocate the randomisation numbers in order of number sequence starting with the lowest number in each block and using all numbers in a block of 4 before starting with the next block of numbers. As soon as the randomisation number is assigned, it will be recorded on the screening log, in the participant source data file and electronic Case Report Form (eCRF). Details of any participants randomised out of sequence will be notified immediately to the Coordinating chief investigator.

Investigators and/or pharmacists may request to break the blind for a participant if there is a clinical need to know the treatment allocation for a subject e.g. to manage the treatment of a serious adverse event. The site’s Principal Investigator needs to discuss the reasons for breaking the blind with the Coordinating Investigator (or if unavailable, the Trial Coordinator). If this request is approved, it will be forwarded to the NICM Clinical Trials Manager. The NICM Senior Manager and Clinical Trials Manager will liaise with the Chief Investigator to process the code break request. When a participant allocation blinding is revealed, the participant ID, date, reason for opening, name of person and signature of who broke the seal, should be provided in writing. The Principal Investigator, Chief Investigator and Statistician will be notified of this information. The participants, principal investigators, research team members, trial medical practitioners, data collection nurses and other staff of the trial centres will have no knowledge of the randomisation assignments until follow-up outcome measures and data analysis are complete.

Treatment Description

Hutchison Whampoa Guangzhou Baiyunshan Chinese Medicine Co., Ltd has contracted Homart Pharmaceuticals Pty Ltd, an Australia-based pharmaceutical manufacturer to manufacture and package the actives and placebos for this trial. These will be manufactured in accordance with Australian Therapeutic Goods Administration (TGA) and Goods Manufacturing Practices (GMP) requirements and regulations.

GROUP 1

Active *NXQ* 0.41g tablet, composition:

* Diospyros Kaki leaf extract 50 mg (active component)
* Starch
* Sucrose Powder
* Magnesium stearate
* Microcrystalline cellulose

Participants in Group 1 will take 3 NXQ tablets, three times per day for 12 weeks.

GROUP 2

Placebos will be created by the same manufacturer to have similar colour, taste, texture and weight and will have no constituents which provide therapeutic effects. Participants in Group 2 will take 3 placebo tablets, three times per day for 12 weeks.

Medication Container Labelling Instructions

Each tablet container must state:

• Name of sponsor

• Pharmaceutical dosage form

• Batch/code number to identify contents and packaging operation (may be encoded for blinding purposes)

• Directions for use - take 3 tablets orally three times a day with meal.

• Statement “for clinical trial use only”

• A trial reference code

• Storage conditions

• Use-by or expiry date

• Statement “keep out of reach of children”

And any other items required to be consistent with TG048 (Therapeutic Goods Order TG048)

Clinical Laboratory Parameters and Abnormal Laboratory Test Results

 All participants will undergo screening and safety tests, which will be performed by a certified pathology laboratory. Hard copies of all results will be provided to the investigator and transferred electronically to the clinical database.

 The following laboratory tests will be performed at Screening, Baseline, Week 2, Week 6, and Week 12:

Haematology Haemoglobin (Hb):

* Haematocrit (Hct)
* Red Blood Cell Count (RBC)
* Platelets Count (Plt)
* White Blood Cell Count (including a five-part differential in absolute)

Coagulation testing:

* Prothrombin time (PT)
* Activated Partial Thromboplastin Time (APTT)
* Fibrinogen (FIB) Level

Liver Function:

* Total Bilirubin (TBil)
* SGOT (AST)
* SGPT (ALT)
* Alkaline Phosphatase (ALP)
* Albumin (ALB)
* Total Protein (TP)

Renal Function:

* Blood urea nitrogen (BUN)
* Creatinine (Cr)

Electrolytes:

* Sodium (Na)
* Potassium (K)
* Chloride (Cl)

 The results of all laboratory tests required by the protocol will be recorded in the participant’s source data file and in the subject’s CRF. All clinically important abnormal laboratory tests occurring during the study will be repeated at appropriate intervals until they return either to the subject’s baseline (i.e. the level recorded at Screening) or to a level deemed acceptable by the investigator and the clinical monitor (or his/her designated representative), or until a diagnosis that explains them is made.

Treatment Protocol

An investigator’s brochure will be provided to guide all clinical trial staff in protocol details.

Participants will take three tablets of active or placebo medications, orally, three times per day for 12 weeks. Medications will be dispensed at baseline and week 6 visits. Participants will attend clinic visits at screening, baseline (week 0), and at 2, 6, 12, and 18 weeks for progress and/or adverse event reporting with their site’s principal investigator. Participants and investigating staff will be blinded to treatment allocation until the end of the trial when the data analyses are completed, and the coding is unlocked. Assessments conducted between weeks -2 (screening) and 0 (baseline) will become the baseline assessments for the study. During the course of the study, assessments will be conducted at weeks 2, 6, 12, and 18 (follow-up) weeks after the commencement of treatment (See Table 1). Follow-up assessment questionnaires (36 weeks) will be completed after the treatment period and any adverse events will be closely monitored and reported. Participant’s general practitioner and usual specialist (if relevant) will be informed of the participant’s medication prescription and safety will be closely monitored during this period via telephone calls every three weeks, and participants will be requested to continue to keep their adverse event diary. Participants will also be asked to undergo identical pathology tests that have been conducted throughout the trial.

Medication containers will be labelled according to the randomisation schedule at an external location and transferred to all participating centres where they will be kept in a secure storage room and maintained below 30 degrees Celsius. At each appointment, an allocation of medication will be dispensed to participants by research personnel according to the pre-coded container labelling. Participants will be required to return the original containers and any unused medication for monitoring. Any unused medication will then be destroyed locally with approval of the coordinating chief investigator. All delivered and dispensed, unused and returned quantities will be recorded in a medication log.

**Study Procedures: Schedule of Assessments**

**Screening Visit (Week -2 to 0)**

Participants eligible for study recruitment, and their study partner/carer, will have the nature, purpose, and risks of the study explained to them by the investigator. Participants (or person responsible), and their study partner/carer, agreeing to participate in the study will sign the informed consent documents. A unique subject screening number will be issued at the time of consent. The screening visit should occur within 2 weeks prior to randomisation (week 0).

Procedures will be performed in the following order:

• Informed Consent

• Medical history, prior and concurrent medications, and demographics will be documented.

• Body weight and height will be measured.

• Physical examination.

• Blood pressure and pulse rate.

• NIHSS, and mRS examination.

• Blood samples taken and sent to central laboratory for liver function, renal function and electrolytes.

• Check concomitant medications and procedures, including any planned hospital admissions.

**Baseline visit (Week 0)**

Participants who meet all of the inclusion and none of the exclusion criteria will be scheduled to return to the site for their Baseline visit. At Baseline, the following will be performed:

• Check of inclusion and exclusion criteria.

• Check concomitant medications and procedures and note any changes in the CRF.

• Blood pressure and pulse rate.

• Blood samples taken and sent to central laboratory for haematology, coagulation, lipids profile and inflammatory markers.

• Complete assessment scales NIHSS, mRS, Barthel Index, and SSQOL questionnaire,

• Complete neurocognitive assessment MoCA

Week 2 Visit

• Record all AE/SAEs in the CRF

• Blood pressure and pulse rate.

• Blood samples taken and sent to central laboratory for haematology, coagulation, liver function, renal function and electrolytes.

Week 6 Visit

• Check concomitant medications and procedures and note any changes in the CRF.

• Record all AE/SAEs in the CRF

• Record compliance rates

• Body weight and height will be measured.

• Blood pressure and pulse rate.

• Haematology, coagulation, liver and renal function tests, and inflammatory markers (except TNF-a).

• Complete assessment scales NIHSS, mRS, Barthel Index, and SSQOL questionnaire.

• Complete neurocognitive assessment MoCA

Week 12 Visit

• Check concomitant medications and procedures and note any changes in the CRF.

• Record all AE/SAEs in the CRF

• Record compliance rates

• Body weight and height measurements.

• Blood pressure and pulse rate.

• Haematology, coagulation, liver and renal function tests, and inflammatory markers.

• Complete assessment scales NIHSS, mRS, Barthel Index, and SSQOL questionnaire.

• Complete neurocognitive assessment MoCA.

Week 18 Visit

• Complete assessment scales NIHSS, mRS, Barthel Index, and SSQOL questionnaire.

• Complete neurocognitive assessment MoCA.

• Blood pressure and pulse rate.

• Record all AE/SAEs in the CRF

• Record recurrence of ischaemic stroke incidence of cardiovascular disease or cerebrovascular disease.

Week 36 Visit

• Record recurrence of ischaemic stroke incidence of cardiovascular disease or cerebrovascular disease.

Drug Accountability

On an ongoing basis all study drugs will be reconciled against delivery, use and returned medication documents. Study drugs which are not used may be destroyed in a suitable facility locally after drug accountability has been completed and with confirmation from the coordinating chief investigator. To be compliant with the treatment protocol study, drug dosing must have a compliance rate of at least 70% over the 12-week study period. Treatment compliance will be calculated by dividing the actual number of tablets taken with expected number of tablets to be taken for the treatment period and multiplying by 100. Self-reported measures of compliance will involve asking participants and their study partner/carer to complete a diary between visits. The researchers will also ask about their adherence to the prescribed treatment regime at the week 2, 6 and 12 visits.

Data Management Protocol

All data records will be digitised and entered into REDCap, a secure electronic database. These files will be accessible to the CI, PI’s, and clinical trial assistants.

The following source data records will be utilised:

• Participant recruitment log

• Participant screening log

• Subject medication dispensing log

• Medication accountability log

• Informed consent documentation

• Clinic notes

• Expenditure records

• Participant case record forms for screening and data collection. These records will be dated and signed for correct coding, completeness, accuracy and legibility by the CI.

• Adverse event report form

# Ethical Considerations

## Ethical Guidelines

This research will be conducted in accordance with the Australian National Statement on Ethical Conduct in Research Involving Humans ([Council, 2007](#_ENREF_25)) and the ICH Guidelines for Good Clinical Practice (CPMP/ ICH/135/95).

Ethics approval will be sought from Human Research Ethics Committees (HREC) for all participating research institutions. The study and investigators will be accountable to these committees by way of ongoing update reports and by providing full access to source data documents.

The trial details will also be submitted to the TGA via CTN (Clinical Trial Notification) arrangements, which will include submission of notification fee and documents of ethical (HREC), sponsor, principal investigators and trial site approval.

## Safety and Tolerability of Interventions

Acute and long-term toxicity tests of NXQ Tablets have been previously undertaken on rats. The animal data described below were provided by the NXQ manufacturer and the results have not yet to be published.

In the long-term toxicity study, Sprague Dawley (SD) rats (50% male) were randomly allocated either a low-dose group (35g of crude drug/ kg), medium-dose group (70g crude drug/kg), or high-dose group (140g crude drug/kg). The drug was administered intra-gastrically continuously for 6 days per week over 26 weeks. The changes in appearance and general behaviour of the rats were observed daily throughout this period in addition to 4 -weeks drug withdrawal period. Haematological index and serum biochemical indices were measured on the weeks 12, 26, and end of the drug withdrawal period. The rats were then dissected and the histopathology changes were examined. The results from this study found no obvious change in the haematological index during the treatment period, except that total bilirubin was lowered in the medium-dose and high-dose groups. This change was found to recover after the drug withdrawal period and the author suggests that this change was related to NXQ but there is no significance impact on clinical toxicology. There were no significant changes on the haematological index during the treatment period in the low-dose group. There were no lesions and no delayed toxicity reactions in the organs could be identified in histo-pathologogical examinations. The researchers concluded that 35g/kg was the basic safe administration dose, which is equivalent to 16 times of the clinic dose for an adult.

In addition, a study explored the teratogenetic effects of *Diospyros kaki* extract in SD rats. Pregnant SD rats were randomly divided into five groups with 15-20 rats per group which included three experimental groups (low-dose 0.6g, medium-dose 1.8g and high-dose 3.84g/kg), one negative control group, and one positive control group. SD rats in this study were pregnant from days 7 to 16 and extracts of *Diospyros kaki* were given to the pregnant rats at a dose of 1.0 ml/100g by weight. Body weight and general condition of the rats were assessed on day 0, 7, 16, and 20. Potential poisonous embryotic effects were assessed on day 20, with foetal rats assessed by weight, body length, tail length as well as recording of any abnormalities in appearance. The results suggested that no significant differences were found across all experimental and control groups, concluding that there was no evidence of embryotic toxicity or teratogenicity was found in SD rats under these experimental conditions. In a chronic toxicity study of NXQ, no severe adverse reactions were identified in rats receiving 16 times of the clinical dose over 180 days. A slight decrease in total bilirubin level was found, which was alleviated within the 30-day post-treatment follow up period. The authors suggested that this phenomena may not be clinically relevant or threatening ([Tsai, 2012](#_ENREF_83)).

A clinical review conducted in China suggests that NXQ is highly regarded to be safe in clinical use with mild adverse reaction incidence of 0.5 to 2 percent. Most of the adverse reactions was allergic skin reaction which can be symptomatically treated by anti-histamine medication ([Z. Wu, Wu, P., Xuan, X., 2012](#_ENREF_92)).

No significant adverse reactions were reported in several trials of NXQ conducted in China for patients recovering from a cerebrovascular accident ([Y. Chen et al., 2012](#_ENREF_17); [Wang et al., 2012](#_ENREF_85); [D. Wu et al., 2013](#_ENREF_90); [L. Wu, Wang, M., Guang, W., Zhang, W., Ou, Q., 2008](#_ENREF_91)). Some mild adverse drug reactions (ADRs) such as palpitation, dizziness and tiredness were reported to be associated with NXQ treatment; these ADRs were generally alleviated after discontinuation of the medication ([Xiong et al., 2013](#_ENREF_94)).

A recent internal report from the manufacturers of NXQ reported that in a double-blind, placebo-controlled trial (217 participants in NXQ group; 218 participants in placebo group), 17 adverse events (n=9 in NXQ and n=8 in placebo group) were reported. These adverse events include dizziness, palpitation, sleep disturbance, common cold, abnormal pathology results (gamma-glutamyl transferase, routine urinalysis, and creatinine). There was no significant difference in prevalence of adverse events between NXQ and placebo groups (p > 0.05). No NXQ-related severe adverse events were reported in the study.

In conclusion, the preclinical and clinical studies of NXQ have not revealed any significant safety issues and the risk of harm to participants appears to be minimal. To minimise the risk, participants in this proposed clinical trial will be closely monitored. Safety pathology tests including full blood counts, liver function test, renal function test, hematology and coagulation index will be undertaken regularly throughout the trial (at baseline, 2 weeks, 6 weeks and end of treatment). NXQ has been listed and approved for human therapeutic consumption by the Therapeutic Goods Authority (TGA).

## Implications for Participants

Each participant will be required to contribute time to visit the allocated trial centre on six occasions over a 36-week period.

* + Screening consultation
	+ Baseline consultation
	+ 2-week safety review and adverse events recording
	+ 6-week review
	+ 12-week post-intervention
	+ 18-week follow-up.

The first occasion will involve a comprehensive consultation and health check-up with the principal investigator and a medical practitioner responsible for trial screening (including establishment of the diagnosis). Participants who do not fully meet the inclusion criteria will not be included in the study and will be notified of this outcome and any abnormal findings/test results from the screening. Their family doctors may also be contacted if the nature of these findings warrants this.

Subsequent visits for review and medication collection will be briefer. The participants will be required to have a blood sample taken on four occasions, which may involve brief discomfort but poses no significant health risks. Participants will need to refrain from caffeine and smoking on the morning before blood testing and avoid alcohol and exercise for 24 hours prior to testing. Participants will be reimbursed up to $30 for travel expenses and/or the time spent at each visit. In accordance with the above-mentioned ethical guidelines, a placebo will be utilised in this trial because there is no currently effective alternative medication for treatment of ischaemic stroke. In addition, participants will continue to take their current prescribed medication.

Participants will be insured for their involvement in clinical research through the University of Western Sydney.

## Informed Consent

The protocol of informed consent will be conducted according to guidelines and appropriate national regulations outlined in Section 4.1. Although colleagues may introduce the project to potential participants, the principal investigators must provide written information and verbally discuss the trial with each person.

Informed consent will involve detailed provision at the appropriate level of comprehension for each person, about the purpose, methods, demands, risks, inconveniences, discomforts, benefits and possible outcomes of the research. Each person must exercise voluntary choice to participate, without coercion or inducement and recognise their choice will not impact on their relationships with the research team or their ongoing health management, and be informed that they are free to withdraw from the trial at any time.

The principal investigator will determine the participant's capacity to consent at the screening visit via a discussion of the trial process, and their understanding of it. Both written and verbal explanations of the trial process will be given.

During the screening visit, the investigator must advise the participant and their carer of the possibility that the participant’s capacity to consent or participate in research may vary or be lost entirely. This matter will be discussed so the researchers can determine what course of action is to be taken at such times. It is important that the investigators agree to comply with the participant's wishes; unless these circumstances would prevent the investigator acting in the participant's best interest. The capacity to consent must also be considered at the end of the study, where the participant is due to complete the full 36-week study. The principal investigator will determine whether it is safe for the participant to be administered this medication, and whether they have the capacity to consent to the process. In cases where the participant cannot provide consent, the person responsible will be contacted to discuss the nature of this post-trial intervention and provided with the option to consent for this medication on the participant's behalf.

The participant, study partner/carer, and the principal investigator will sign two informed consent forms: one for the investigator and one for the participant to keep, in addition to their information sheet. The consent form will state the name and contact details for the principal investigator and study co-ordinators for each recruitment site, who are to be available 24 hours a day, if they have questions or concerns.

It is not envisaged that culturally sensitive issues other than those potentially encountered in the day-to-day running of a clinic would present. All personnel involved in the trial are experienced with conducting clinical practice and clinical research such that cultural needs and beliefs are recognised. If the consenting individual is unable to read the information provided, an impartial witness will be asked to observe the verbal discourse of the trial and witness the signing of the consent form.

## Anonymity/ Confidentiality/ Privacy

In accordance with ethical guidelines, the anonymity, confidentiality and privacy of participants will be protected.

The electronic clinical trial management data system and all electronic data will be password protected. Hardcopy source data records will be kept in a locked cabinet in the office of the principal investigator at each recruitment site during data collection and at completion of the study. The University of Western Sydney will keep source and electronic data for an indefinite period of time.

All publication material will refer to general trial results as aggregate data and no individual participant names or identifying information will be released.

Participants are welcome to view their personal data at any time during the trial.

# Analysis

## Outcome Measures

*The outcome measures to be acquired in this study are:*

*Primary*

NIHSS

The National Institutes of Health Stroke Scale (NIHSS) is a graded neurological examination rating speech and language, cognition, visual field deficits, motor and sensory impairments, and ataxia, which has become a standard part of the clinical assessments used in many recent interventional trials.

In a clinical setting, patients with a NIHSS score of ≤ 5 can be expected to return home and those with a NIHSS score ≥ 16 often go to a nursing facility whereas those with intermediate scores are often admitted to acute rehabilitation facilities ([Ortiz & L. Sacco, 2014](#_ENREF_66)).

This is to be assessed by neurologist or trained physician based on 11 sections scoring accordingly between 0 to 2, 0 to 3 and 0 to 4 of each of the sections ([Berger et al., 1999](#_ENREF_12); [Fischer et al., 2005](#_ENREF_33)).

Secondary objectives for this study will include the use of Modified Rankin Scale (mRS) will be used to assess the degree of global disability and the level of independence; Barthel Index (BI) to assess the performance of daily living activities.

mRS

The modified Rankin scale (mRS) commonly used scale, which measures the degree of disability or dependence in the daily activities of individuals who have suffered a stroke or other neurological disability. The scale ranges from 0-5 with 0 indicating no disability and 5 indicating severe disability ([Sulter, Steen, & De Keyser, 1999](#_ENREF_79)). While mRS is widely applied for evaluating stroke patient outcomes it is also widely used as valid end point in randomized clinical trials assessing therapeutic treatments ([Banks & Marotta, 2007](#_ENREF_5)).

mRS will be completed by a trained rater. Scores will be recorded according to patient’s degree of disability and dependence.

*Secondary*

SSQOL

The Stroke-Specific Quality of Life Scale (SSQOL) is a self-report questionnaire which consists of 49 items across the 12 domains of assessment including: energy, family roles, language, mobility, mood, personality, self-care, social roles, thinking, upper extremity function, vision, and work/productivity ([Lin, Fu, Wu, & Hsieh, 2011](#_ENREF_55)).

The SSQOL is a more comprehensive questionnaire compared to shorter variations of the tool such as the SF-36 and the Euro-QOL 5-D scales, which are commonly used in stroke trials but do not assess language, hand function, cognition, or vision ([Muus, Williams, & Ringsberg, 2007](#_ENREF_61); [Linda S Williams, Weinberger, Harris, & Biller, 1999](#_ENREF_88)).

SSQOL scales has shown to be a better valid and reliable to measure stroke-specific health related quality of life that is moderately responsive to changes in most domains during the first 3 months after stroke ([L. S. Williams, Weinberger, Harris, Clark, & Biller, 1999](#_ENREF_89)).

The SSQOL is a self-report questionnaire and will be completed by the participant with the aid of the investigator if necessary.

Barthel Scale

Barthel Scale, also known as the Barthel ADL Index, a widely used ordinal scale measuring performance in activities of daily living including feeding, bathing, grooming, dressing, toilet use, mobility and stairs ([Granger, Dewis, Peters, Sherwood, & Barrett, 1979](#_ENREF_38)).

The Barthel index has been used extensively to monitor functional changes in individuals receiving in-patient rehabilitation, mainly in predicting the functional outcomes related to stroke ([Collin, Wade, Davies, & Horne, 1988](#_ENREF_24)). The Barthel Scale produces a score from 0 to 100 with a score of 91 to 99 indicating ‘slight dependency’; 61-90 indicating ‘moderate dependency’; 21-60 indicating ‘severe dependency’; 0-20 indicating ‘total dependency’ ([Shah, Vanclay, & Cooper, 1989](#_ENREF_75)).

The Barthel Scale score will be assessed by a trained rater by asking the relevant questions and recording the scores from 0 to 100.

Hemorheology, Coagulation Index, Lipid Profile, Inflammatory Markers, Liver and Renal Function Test

Patients will be sent to a Laverty Pathology laboratory for blood sampling and pathology testing. The assessment of lipid profiles will include triglycerides, total serum cholesterol, high density lipoprotein cholesterol and low-density lipoprotein cholesterol. The assessment of inflammatory markers will include tumour necrosis factor alpha and c-reactive protein. Laverty Pathology is fully accredited under the National Association of Testing Authorities (NATA) and registered under the accreditation scheme of the Royal College of Pathologists of Australasia (RCPA).

A copy of the pathology results will be inserted into the participant file during the study which will be stored in a locked filing cabinet in a secure and lockable room, only accessed by the trial research staff. The email copy of results can only be accessed by a secure password only known to the Chief Investigator and Principle Researcher. Upon completion of the trial the pathology results will be securely and separately stored from the coded participant files, as the pathology results will be identifiable. These paper files will remain securely stored.

Montreal Cognitive Assessment (MoCA)

The MoCA is a brief cognitive screening tool with high sensitivity and specificity for detecting MCI as currently conceptualized in patients performing in the normal range on the Mini-Mental State Examination ([Nasreddine et al., 2005](#_ENREF_62)). The MoCA provides an overall score based on the following domains: short-term memory recall task, visuospatial abilities, executive function, attention, concentration, working memory, language, and orientation to time and place.

Safety Measures

General medical assessments including body temperature, heart rate, breathing, and blood pressure will be measured and recorded at each assessment visit.

The presence and frequency of adverse events will be recorded at each of assessment sessions on a paper-based form. Participants are encouraged to note down any adverse events during the trial period between the assessment time points and contact the research immediately via email or telephone. For classification of adverse events please refer to Appendix A.

Routine liver and renal function, and haematological and coagulation (PT, APPT and fibrinogen) testswill be assessed at screening, week 2, week 6 and week 12 of the trial period.

An independent Data Safety Monitoring Board (DSMB) will be established to periodically review and evaluate the accumulated study data for participants safety, study conduct and progress, and, when appropriate, efficacy. The DSMB will also make recommendations to the sponsor and/or Trial Steering Committee concerning the continuation, modification, or termination of the trial.

*Table 1: Data collection timeline*

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Exam** | **Screening** | **Baseline** | **Wk2** | **Wk 6** | **Wk 12** | **Wk 18** | **Wk 36** |
| **Screening** | Medical History | • |  |  |  |  |  |  |
| Physical Exam | • |  |  |  |  |  |  |
| **Primary** | NIHSS | • | • |  | • | • | • |  |
| mRS | • | • |  | • | • | • |  |
| **Secondary** | SSQOL |  | • |  | • | • | • |  |
| Barthel Index |  | • |  | • | • | • |  |
| Lipids Profile |  |  • |  |  | • |  |  |
| Haematology |  | • | • | • | • |  |  |
| Coagulation index |  | • | • | • | • |  |  |
| Blood Pressure |  | • |  | • | • |  |  |
| Inflammatory marker | C-RP |  | • |  | • | • |  |  |
| TNF-a |  | • |  |  | • |  |  |
| MoCA |  | • |  | • | • |  |  |
| **Safety** | Adverse event |  |  | • | • | • |  |  |
| Full blood count | • |  |  | • | • |  |  |
| LFT; RFT | • |  | • | • | • |  |  |
| **Long term efficacy** | Death; Reoccurrence of IS;  |  |  |  |  |  | • | • |

## Statistical Analysis

Safety Analyses

There will be no interim analyses with the exception of monitoring safety variables. The frequency, type and probable association of adverse events will be tallied by de-identified treatment group every 2-months for review by the study team. If there is any significant safety issue identified an additional DMC meeting will be called.

Preliminary Analyses and Data Cleaning

At completion of data collection, all variables and all logical pairs of variables will be subject to descriptive analyses using graphs, frequency counts and summary statistics. This will allow a) identification of unusual or unexpected results for data checking and b) familiarisation with the distributions and associations within the data set. Outcome variables which have severely non-symmetric distributions will be either transformed or categorised.

At the completion of data checking and correction, the data set will be locked for analysis.

Checking for homogeneity of study centres

As this is a multi-centre study, early analyses will address the question of whether or not there is heterogeneity between centres. Linear models will be fitted to each outcome measure in turn, with centre and centre by treatment added as fixed effects. Any statistically significant differences will be documented and explored further for potential confounding with demographic or medical history factors. If variation between centres cannot be explained (or is shown to be related to differences in study methods), the primary analysis will continue as planned, but will be followed by sensitivity analyses which will either stratify by or exclude the outlier site(s) (depending on the sample size of the site(s) involved).

Demographic and baseline characteristics

The demographic and medical characteristics of participants in each treatment group will be summarised using percentages or means and standard deviations. Pearson’s Chi-square and independent samples t-tests will be used to check for any statistically significant differences between groups. Results will be documented as p-values and, where necessary, addressed within the interpretation of study results.

Missing data and protocol violations

In the case of death, all measurements prior to death will be included in the analysis but all after death will be set to missing. To address withdrawals, loss to follow-up or non-compliance with the study analyses will be conducted on both an intention to treat (ITT) and per protocol (PP) basis. Withdrawing and non-compliant participants shall be encouraged to continue with data collection even if stopping treatment. Where data items are missing the last value carried forward method to replace missing data in the ITT analysis will be used. Participants who have significant deviations from the protocol will be removed from the PP analysis after the completion of the ITT analysis. Such significant deviations from the protocol will be determined and documented by the study clinician during the course of the study. Any deviation from randomisation, missing data and withdrawals will be fully reported for this purpose.

Primary efficacy analysis

The primary analysis will be linear mixed models through which we will test for differences between treatment groups on each outcome over time, with adjustment for random variation between treatment centres, with and without adjustment for other potentially important predicts (e.g. compliance, age, and gender). The random effects are at both the patient and site level. Non-linear changes over time will be tested by a) fitting time as categorical variable and b) testing for quadratic and cubic effects. Results will be reported as regression coefficients (or odds ratios if categorical) and associated 95% confidence intervals.

Subgroup analyses

Secondary analyses will be a repeat of the above stratified by disease type and, if necessary, with stratification by research centre. Data will be analysed using SAS and/or SPSS software.

# Timeline

*Table 1: Research Study Timeline*

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Month | 0-3 | 4-7 | 8-11 | 12-15 | 16-19 | 20-23 | 24-27 | 28-31 |
| Actives and placebo preparation |  |  |  |  |  |  |  |  |
| Engagement of staff, final preparation, ethics approval |  |  |  |  |  |  |  |  |
| Recruitment |  |  |  |  |  |  |  |  |
| Intervention and follow-up |  |  |  |  |  |  |  |  |
| Data entry and analysis |  |  |  |  |  |  |  |  |
| Publications, Final Report  |  |  |  |  |  |  |  |  |

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Appendix A

**Adverse Event Definitions:**

|  |  |
| --- | --- |
| **Adverse Event (AE) (Drug)**Any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment. An adverse event can therefore be any unfavourable and unintended sign, symptom, or disease temporally associated with the use of a medicinal (investigational/experimental) product, whether or not related to this product. | **Serious Adverse Event (SAE)**Any untoward medical occurrence that:* results in death;
* is life-threatening The term “life-threatening” refers to an event/reaction in which the patient was at risk of death at the time of the event/reaction; it does not refer to an event/reaction which hypothetically might have caused death if it were more severe;
* requires inpatient hospitalisation or prolongation of existing hospitalisation;
* results in persistent or significant disability/incapacity; is a congenital anomaly/birth defect;
* is a medically important event or reaction.

**Serious unexpected suspected adverse reaction**A serious adverse event (see definition above) for which there is some degree of probability that the event is an adverse reaction to the administered drug and the adverse reaction is unexpected.*NHMRC national statement on ethical conduct in human research 2007.**NHMRC Website:* [*http://www.nhmrc.gov.au/PUBLICATIONS/synopses/e72syn.htm*](http://www.nhmrc.gov.au/PUBLICATIONS/synopses/e72syn.htm)*Copy of statement:* [*http://www.nhmrc.gov.au/PUBLICATIONS/synopses/\_files/e72.pdf*](http://www.nhmrc.gov.au/PUBLICATIONS/synopses/_files/e72.pdf) |
| **Adverse Drug Reaction (ADR)**For unapproved medicines: all noxious and unintended responses to a medicinal product related to any dose should be considered ADVERSE DRUG REACTIONS. The phrase “responses to a medicinal product” means that a causal relationship between a medicinal product and an adverse event is at least a reasonable possibility, i.e. the relationship cannot be ruled out. |
| **An Unexpected Adverse Drug Reaction**is an adverse reaction, the nature or severity of which is not consistent with the applicable scientific information (e.g. Investigator’s Brochure for an unapproved investigational product or Product Information (PI) document or similar for an approved, marketed product).*The Australian Clinical Trials Handbook (March 2006),* [*http://tga.gov.au/ct/cthandbook.pdf*](http://tga.gov.au/ct/cthandbook.pdf) |

**.**

**The assignment of severity and causality for each AE should be made by the investigator responsible for the care of the participant based on the definitions in the tables below**

|  |
| --- |
| **Severity** |
| **Severity** | **Description** |
| **Mild** | An event that is easily tolerated by the participant, causing minimal discomfort and not interfering with every day activities. *Example Symptoms: diarrhoea or insomnia* |
| **Moderate** | An event that is sufficiently discomforting to interfere with normal everyday activities. |
| **Severe\*** | An event that prevents normal everyday activities.  |

|  |
| --- |
| **Relationship to Study Drug** |
| **Relationship** | **Description** |
| **Unrelated** | Where an event is not considered to be related to the study drug.. |
| **Possibly** | Although a relationship to the study drug cannot be completely ruled out, the nature of the event, the underlying disease, concomitant medication or temporal relationship make other explanations possible. |
| **Probably\*** | The temporal relationship and absence of a more likely explanation suggest the event could be related to the study drug. |
| **Definitely\*** | The known effects of the study drug or its therapeutic class, or based on challenge testing, suggest that study drug is the most likely cause. |
|
| **Not assessable\*** | When causality is, for one reason or another not accessible, e.g. because of insufficient evidence, conflicting data or poor documentation |
| **\*NOTE - Report all Possible, Definitely&Not assessable AE’s to PI’simmediately** |

**\* NOTE - Report ALL Severe AE’s to PI’s immediately**