1. Introduction

Ischemic brain stroke causes interruption of blood supply to brain through carotid circulation. Around 83% of stroke patients have ischemic type of brain stroke and remaining have haemorrhagic stroke due to rupture of blood vessel. Numbers of experimental studies have revealed that diabetes can worsen cerebral ischemia and therefore it is considered as one of the modifiable risk factor for ischemic brain stroke. It is well reported that DM raises the susceptibility of an individual’s to hypertension, atherosclerosis, dyslipidaemia and therefore it increases risk for brain stroke 2-3 times more [Goldstein LB et al., 2006]. Blood sugar control has been shown to reduce diabetic macrovascular complication like brain stroke [Boden-Albala B et al., 2008].

People who have diabetes are two to four times more likely to have a stroke than people who do not have diabetes. They also tend to develop heart disease or have strokes at an earlier age than people without diabetes. Various epidemiological studies showed that long-term diabetes increase the risk of cerebral ischemia by 2-4 times as compared with non-diabetic population. Animal models correlated these clinical findings and provided base for the pathophysiological effect of diabetes on cerebral ischemia. High plasma glucose level is a key factor for the poor outcome observed after cerebral ischemia in diabetic patients [Dave KR et al., 2011].

It is also reported that ischemic stroke increases the risk for developing Alzheimer’s disease (AD) and dementia. Patients with AD have high risk for ischemic as well as haemorrhagic stroke [De la Torre JC, 2002; Chi NF et al., 2013]. Various epidemiological studies have shown that AD risk factors involve vascular basis. Practically all the agents that have been reported to slow down development of AD, increases or improve cerebral blood flow. Cerebral hypo-perfusion leads to cognitive and neurodegenerative pathogenesis in AD. The AD associated dementia and ischemic stroke are known to increase at comparable rate with age. It is also suggested that vascular risk factors linked to stroke in elders significantly increase the risk for developing AD. These include coronary artery disease, atherosclerosis, atrial fibrillation, diabetes mellitus and hypertension. Around 60-90% of AD patients have cerebrovascular pathology including amyloid induced angiopathy, endothelial degeneration and infarctions.

Nature of a true relationship between stroke and AD has been little explored and higher incidence of stroke in DM and AD person is still under investigation. Probable detrimental reasons are higher ROS formation and change in blood vessel vasculature [Martini SR &
Prolonged or acute hyperglycaemia and neuroinflammatory causes associated with β-amyloid plaques are responsible for endothelial dysfunction which leads to reduction in cerebral blood flow. It also involves apoptosis, necrosis and excitotoxicity (excessive activation of glutamate receptors) [Mehta SL, Manhas N & Raghubir R, 2007]. Present treatment approach for ischemic brain stroke uses tissue plasminogen activators (TPAs), anti-coagulants and anti-platelets for their thrombolytic effects. Minocycline, growth factors and other free radical scavengers have been reported for their neuroprotective effects in stroke while anti-hypertensive, anti-diabetics and anti-dyslipidemic drugs have shown their beneficial effect in the prevention of stroke [Kikuchi K et al., 2012]. For hypertensive patients with ischemic stroke, choice of agents are angiotensin converting enzyme (ACE) inhibitors [Hack W, Kaste M & Bogousslavsky J, 2003]. Statins and anti-platelet agents are recommended for ischemic patients with hypercholesterolemia and coronary artery disease. Some of the training therapies such as cognitive functions, training of skills and of sensory motor functions are recommended to improve quality of life of ischemic patients [Steultjens E & Dekker J, 2003]. However these agents have number of side effects and therefore there is need to find newer cellular targets and molecules which provides neuroprotective effects in cerebrovascular stroke. Present treatment approach is not capable to provide neuroprotection in ischemic patients with DM and AD. Because in both of these associated diseases, ROS turnover is high through neurovascular pathogenesis, nuclear factor κB (NF-κB) activation and caspase-3 mediated apoptosis of neurons [Tsuruta R et al., 2010; Dludla PV et al., 2017]. Various types of ion channels for Na⁺, K⁺, Ca²⁺ and Cl⁻ exist in CNS. These channels are involved in regulating various physiological actions such as action potential generation, release of neurotransmitters, regulation of synaptic plasticity and excitotoxicity. Therefore their dysfunction could results into various neurological disorders such as Parkinsonism, AD, brain stroke and Huntington’s disease. Targeting these ion channels could provide neuroprotective actions in mentioned diseases. Potassium ion channels are one of them in CNS. Neuroprotective role of K⁺ channels have been already established through pre-clinical in-vitro and in-vivo experiments [Clark AG, Booth SE & Morrow JA, 2003]. Various in-vitro and in-vivo experimental studies have reported that ion channel modulators particularly potassium channel openers (KCOs) provides neuroprotection in ischemic injury to brain. Adenosine triphosphate sensitive potassium channel (KATP) openers such as cromakalim [Wang S et al., 2010], nicorandil [Akao M et al., 2002] and cinnarizine [Abdel-
Salam OME, 2007] showed neuroprotective actions through either antioxidant or anti-apoptotic or antiinflammatory actions in rats.

As none of the KCOs reported their neuroprotective role in stroke related conditions such as DM and AD, objectives of our study were to evaluate in-vitro antioxidant activity and in-vitro neuroprotective activity in glioblastoma cell line as well as in-vivo neuroprotective activity of potassium channel openers in ischemic brain stroke related disorders DM and AD. To find out mechanism of neuroprotection by KCOs, we induced ischemic type of brain stroke using transient middle cerebral artery occlusion (tMCAO) method in diabetic animals and animals treated with AlCl$_3$ to induce dementia like AD.

DM was induced by administration of streptozotocin (STZ) and AD type dementia was induced with aluminium chloride (AlCl$_3$). Various parameters measured in STZ induced DM ischemic models are serum glucose level, neurological score, infarct volume, physical parameters, serum lipid profile, level of ketone bodies, oxidative stress parameters, caspase-3 activity and VEGF (vascular endothelial growth factor) level. Three potassium channel openers viz- cromakalim, nicorandil, cinnarizine, potassium channel blocker glibenclamide, standard antihypertensive agent telmisartan and antioxidant vitamin E were given as treatment in diabetic ischemic animals. Similarly in second model AD like symptoms were induced by AlCl$_3$ along with ischemic stroke. The animals were treated with three potassium channel openers cromakalim, nicorandil, cinnarizine and standard vitamin E. In this model neurological score, infarct volume, physical parameters such as body weight changes and brain oxidative stress parameters, caspase-3 activity and acetylcholine esterase activity (AchE) were measured.

Results of our study showed that all three KCOs showed dose dependant in-vitro antioxidant activity and in-vitro neuroprotective activity in neuronal cell line. In in-vivo study, more consistent antioxidant, anti-apoptotic and neuroprotective activity was obtained with two potassium channel openers cromakalim and nicorandil as compare to cinnarizine. These effects were parallel to the standard drug vitamin E. Therefore it may be suggested that KCOs produced neuroprotective action through antioxidant and anti-apoptotic action in ischemic stroke related condition DM and AD.