Stroke is among the most frequent causes of permanent disability and death in adults, and its treatment is a clinical challenge because of the narrow window from the time when the symptoms appear until the initiation of medical care.\(^1\)\(^2\) Despite the advances in the past 20 years, the constricted time frame of 4.5 hours for thrombolysis and up to 6 hours for mechanical thrombectomy contributes to the ominous outcome of this condition.\(^1\) An estimate of 1.9 million neurons and 14 billion synapses is damaged for every minute a large vessel goes untreated in a patient with stroke.\(^1\) A recent study shows that treatment of ischemic stroke with early endovascular thrombectomy after intravenous administration of thrombolytic alteplase within 3 hours from the onset of symptoms resulted in faster and better reperfusion.\(^3\) However, only 15% to 60% of patients with stroke arrive at the hospital within 3 hours.\(^1\) Therefore, a better understanding of the mechanisms involved in brain ischemic injury will help to develop improved strategies for management and treatment.

The study presented by Gong et al\(^4\) in this issue of *Hypertension* proposes an interesting hypothesis to explain the activation pathways associated with ischemic injury. The authors used genetically manipulated mouse models, including adenoviruses encoding dominant-negative tumor growth factor-β–activated kinase 1 (TAK1) and 5Z-7-oxozeanol, a TAK1 inhibitor, to test their hypothesis.\(^4\) Gong et al proposes that neuronal tumor necrosis factor receptor–associated factor 3 (TRAF3) activates TAK1 during ischemic signaling cascades resulting in cell death, inflammation, and oxidative stress.\(^4\)

Treatments aiming to reduce brain ischemic injury should focus on improving brain tissue resistance to ischemic injury or diminishing the factors involved in tissue damage. Recently, was reported that activation of angiotensin-converting enzyme 2 provides neuroprotective effects in a rat model of ischemic stroke in a flow-independent manner.\(^5\) Angiotensin-converting enzyme 2 in conjunction with the downstream peptide angiotensin (1–7) and its Mas receptor may interact cooperatively to preserve brain cells and reduce ischemic injury.\(^5\) The cerebroprotection induced by angiotensin-converting enzyme 2/angiotensin (1–7)–Mas axis may include anti-inflammatory effects and reduction of oxidative stress.\(^4\) However, the exact mechanisms involved in angiotensin-converting enzyme 2 protective effects on brain ischemic injury are still under investigation. Other investigators reported that TAK1 is involved in several signaling pathways associated with ischemic injury, and its inhibition could be a potential candidate for neuroprotective interventions in ischemic stroke.\(^6\) Nevertheless, the pathways involved in activation of TAK1 during ischemic stroke are not fully understood.

The results presented by Gong et al\(^4\) identified TRAF3 as the central modulator involved in the generation of neuronal ischemic injury. Moreover, they provided new knowledge on the poorly understood pathways involved in TAK1 activation associated with neuronal ischemic injury. Previously, TRAF3 was reported as regulator of pathological cardiac hypertrophy and heart failure.\(^7\) TRAF3 is part of the TRAFs which represent a family of 7 members TRAF1–7.\(^7\) TRAFs are adaptor proteins mediating signal transduction involved in a wide range of biological functions, including inflammatory response, stress response, and cell survival, proliferation, differentiation, and death.\(^7\)

Gong et al\(^4\) observed that TRAF3 was significantly upregulated in neurons exposed to cerebral artery occlusion. Also, they found that transgenic mice overexpressing neuronal TRAF3 showed greater ischemic injury after artery occlusion in comparison with TRAF3-knockout mice.\(^4\) One of most notable finding of this study by Gong et al\(^4\) is the pathway involved in TAK1 activation via phosphorylation induced by TRAF3. Gong et al\(^4\) used an elegant experimental design, including cultured primary neurons and adenovirus harboring wild type of TAK1 (Adca-TAK1) or a dominant-negative TAK1 (Addn-TAK1), to investigate the link between TRAF3 and TAK1 activation. Gong et al\(^4\) demonstrated that neuronal ischemic injury was mediated by activation of TAK1, which sequentially activated Jun N-terminal kinase (JNK)/Jun proto oncogene (c-Jun), nuclear factor-kappa B (NF-κB), and Ras-related C3 botulinum toxin substrate 1 (Rac-1) resulting in neuronal death, inflammation, and oxidative stress, respectively.

Other investigators reported TRAF3 involvement in neuronal cell death not associated with ischemic injury.\(^8\) TRAF3 involvement was reported in the proapoptotic effect of high glucose on neuronal cells.\(^8\) The mechanisms proposed for this effect suggest that exposure to high glucose environment results in decreases expression of miR-322, which is a miRNA that represses TRAF3 translation.\(^8\) The decreased expression...
Barrier and neuronal cell apoptosis. These changes in brain barrier neuronal cells were induced by inflammatory cell recruitment, expression of proinflammatory mediators, and increased matrix metalloproteinase 2 and 9 activity. This study suggested that Akt/FoxO1 pathway could be involved in neuronal death, inflammation, and oxidative stress, respectively.

of miR-322 leads to overexpression of TRAF3 that results in neuronal death. The study suggests that changes in miR-322 are triggered by increased oxidative stress associated with high glucose levels.

TRAF5 and TRAF6 were also associated with brain ischemic injury. TRAF5 was reported as an essential mediator of ischemic brain infarction by disruption of brain blood barrier and neuronal cell apoptosis. These changes in brain blood barrier neuronal cells were induced by inflammatory cell recruitment, expression of proinflammatory mediators, and increased matrix metalloproteinase 2 and 9 activity. This study suggested that Akt/FoxO1 pathway could be involved in neuronal apoptosis observed in this model. The other member of TRAFs family associated with neuronal death was TRAF6, which was correlated with neuronal apoptosis in a rat model of cerebral ischemia reperfusion. TRAF6 participates in the regulation of apoptosis induced by inflammation via caspase-3 pathway. However, the exact mechanism(s) of caspase-3 activation is still under investigation. Several scientific reports are strongly supporting the association of TRAFs family members with neuronal apoptosis, and further investigation of the mechanisms involved is warranted.

The recovery of a patient after brain ischemic injury is always associated with the severity of neuronal apoptosis, inflammation, and oxidative stress observed after the ischemic event and during the reperfusion phase. Tackling these factors could be the key to improve recovery or even prevent injury after an ischemic event in stroke patients.

Gong et al proposed an interesting mechanistic pathway involved in brain ischemic injury, which includes neuronal apoptosis, inflammation, and oxidative stress. More importantly, this is the first report pointing to TRAF3 as the specific key initiator of this cascade in brain ischemic injury. Mechanistically, they reported that TRAF3 mediated activation of JNK as prodeath pathway, the NF-κB proinflammatory pathway, and the Rac-1 pro-oxidant pathway, via activation of TAK1 (Figure).

These findings added new information on the activation pathways involved in neuronal ischemic damage and provided potential new targets for treatments to reduce brain ischemic injury. Further studies should aim to translational approaches to explore proof of concept, causality effects, and validation of experimental finding to advance in this field.

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References
Neuron-Specific Tumor Necrosis Factor Receptor–Associated Factor 3 and Acute Ischemic Stroke
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