PRO: Regulatory T Cells Are Protective in Ischemic Stroke

Xiaoming Hu, MD, PhD; Peiying Li, MD, PhD; Jun Chen, MD

Immune disturbances play manifold roles in the pathogenesis of ischemic stroke. Regulatory mechanisms that continuously fine-tune the immune system are, therefore, essential for protection and recovery from stroke. Regulatory T cells (Tregs) is suggested to exert neuroprotection by Liesz et al on the basis of their findings that Tregs depletion with CD25 antibody exacerbates brain damage and worsens functional deficits 7 days after transient ischemic stroke. The endogenous Treg-afforded protection is late in onset, as Treg depletion failed to influence brain damage ≤4 days post ischemia. This late action is consistent with delayed Treg infiltration into the brain until 3 to 5 days after middle cerebral artery occlusion. Interestingly, anti-inflammatory effects of Tregs were observed before their appearance in the brain, suggesting early peripheral protective mechanisms of Tregs. Such anti-inflammatory effects, however, are not potent enough to reduce early brain damage, probably because of insufficient Treg numbers in the circulation. In support, clinical studies report that the number of circulating Tregs decreases transiently, but dramatically in the first 2 days after stroke. Therefore, adoptive transfer of Tregs soon after stroke may provide a novel immunotherapeutic strategy for stroke.

Surprisingly, Kleinschnitz et al recently demonstrated an opposite role for Tregs to exacerbate brain injury early after transient ischemia. The animal model they used was different from Liesz study in several aspects, including the duration of ischemia and methods for Treg depletion, which possibly contribute to the discrepancies between these 2 studies. For example, instead of using CD25 antibody for Treg depletion, Kleinschnitz used a DEREG transgenic mouse in which Foxp3+ cells are inducibly ablated. Neither CD25 nor Foxp3 is exclusively expressed on Tregs. Therefore, these 2 depletion protocols may differentially target a small portion of non-Tregs population and confound the data interpretation. Notably, another group using the DEREG mouse showed that Treg depletion has no effect on the ischemic brain 4 days after injury, which is against the Kleinschnitz study and tips the balance in favor of the Liesz study. Unfortunately, late stage effect of Tregs depletion was not addressed in this study. Apparently, more investigations are necessary to confirm the function of endogenous Tregs in ischemic stroke.

Given the potential protective effect of endogenous Tregs in the ischemic brain and the known therapeutic effects of Treg transplantation in ischemia/reperfusion models in many other organs, we explored the effect of Treg adoptive therapy in models of focal cerebral ischemia/reperfusion, in adherence with Stroke Therapy Academic Industry Roundtable (STAIR) recommendations. Studies conducted by 2 independent laboratories on 2 different species demonstrated that postischemic delivery of Tregs markedly reduced brain infarct size and improved neurological functions out to 4 weeks. Notably, the therapeutic time window of Tregs lasted 24 hours after the onset of ischemia, making it more applicable to humans who may not be treated in the clinic for many hours from symptom onset.

In contrast with the relatively late action of endogenous Tregs, transplanted Tregs led to an early attenuation of blood-brain barrier disruption and reduced infiltration of inflammatory cells into the lesioned brain. These effects result in an early reduction of infarct size at 3 days post ischemia. The distinct effects of transferred and endogenous Tregs on early stage ischemic brain injury may reflect that the therapeutic concentration of exogenous Tregs (2×10^6/mouse) greatly outnumbers endogenous circulating Tregs. In our hands, transplanted Tregs at a concentration <1×10^6 cells/mouse failed to offer any early protection. However, Kleinschnitz used even lower concentrations of Tregs (7.5×10^5/mouse) and observed exacerbation of ischemic damage. One possible interpretation of the discrepancies between our data and data of Kleinschnitz is that Tregs delivered before or after stroke exert opposing functions. Our protocol involved postischemic Treg delivery, whereas Kleinschnitz administered Tregs 24 hours before ischemic challenge. It may be counterproductive to increase Tregs in the immediate pre-ischemic period as the immune milieu before stroke may not be permissive for proper Treg action. However, a previous study contradicted this notion by priming Tregs in vivo by repetitive intranasal administration of recombinant E-selectin 10 days before ischemia reduced brain infarct volume and improved functional performance after permanent middle cerebral artery occlusion. The latter findings suggest that induction of Tregs long before stroke may also be protective. Therefore, the timing of Treg delivery seems to be critical for

Received April 2, 2013; accepted May 16, 2013.
The opinions expressed in this article are not necessarily those of the editors or of the American Heart Association. This article is Part 2 of a 3-part article. Parts 1 and 3 appear on pages e84 and e87, respectively.

From the Department of Neurology, Center of Cerebrovascular Disease Research, University of Pittsburgh School of Medicine, Pittsburgh, PA (X.H., P.L., J.C.); and Geriatric Research, Educational and Clinical Center, Veterans Affairs Pittsburgh Health Care System, Pittsburgh, PA (X.H., J.C.).

Correspondence to Jun Chen, MD, Department of Neurology, University of Pittsburgh Medical Center, Pittsburgh, PA. E-mail chenj2@upmc.edu

Stroke is available at http://stroke.ahajournals.org

DOI: 10.1161/STROKEAHA.113.001267
the direction of its impact on the ischemic brain, which warrants further investigation.

It is clear from the above-mentioned findings that research on the impact of Tregs in stroke still lies in its infancy. Although recent studies by us and Liesz et al strongly suggest that Tregs are protective in ischemic stroke, additional studies from independent laboratories are necessary to confirm the effectiveness and safety. The positive results in several phase I clinical trials about the effect of ex vivo–expanded Tregs in graft-versus-host disease and autoimmune diseases give further hope for this therapy to be advanced to the clinical practice.8

Disclosures
Dr Hu is supported by a Scientist Development Grant (13SDG14570025) from the American Heart Association. Dr Chen is supported by National Institutes of Health Grants NS36736, NS43802, and NS45048.

References


Key Words: regulatory T cells ■ stroke
PRO: Regulatory T Cells Are Protective in Ischemic Stroke
Xiaoming Hu, Peiying Li and Jun Chen

Stroke. 2013;44:e85-e86; originally published online July 2, 2013;
doi: 10.1161/STROKEAHA.113.001267

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://stroke.ahajournals.org/content/44/8/e85

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Stroke can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Stroke is online at:
http://stroke.ahajournals.org//subscriptions/