Regulatory T cells (Tregs) have been shown to play a critical role in immune homeostasis and in suppressing unwanted inflammatory responses toward self-antigens.\(^1,2\) Limited numbers of naturally occurring CD4\(^+\) CD25\(^+\) Tregs containing the Foxp3 transcription factor exist in peripheral blood, but represent only 5\%–10\% of the CD4\(^+\) T cell population. Tregs appear to localize to organs and tissues and play crucial roles at the site of inflammation and in the suppression of inflammation in local lymph nodes. In addition to natural Tregs that are selected in the thymus and appear selected for self-antigens expressed in the thymus, a second population of Tregs can be induced in peripheral Foxp3\(^-\) T cells by activation and appropriate cytokine conditions. The relative roles of these distinct Treg populations are still being resolved as are those of other less well defined regulatory populations. Evidence has initially accumulated regarding differing autoimmune diseases in which a reduction of Tregs or a loss of function plays a role, which has led to an interest in the potential therapeutic role of Tregs. Therefore, several approaches have been taken to expand the number of Tregs \textit{in vitro}, including various agonist strategies, including cytokines, and gene transfer approaches. However, because of the difficulties in Treg production and expansion \textit{in vivo}, expansion of Tregs for therapeutic use in humans is still an attractive option.

In this issue of \textit{JASN}, Kim et al.\(^3\) report that administering the IL-2/anti-IL-2 complex promotes the expansion of Tregs in renal ischemia-reperfusion injury (IRI), a mouse model of acute renal hypoxic injury leading to inflammation. The authors found that administration of the IL-2 complex induced a 3- to 5-fold expansion of Tregs in both spleen and kidney after IRI. Mice injected with the IL-2 complex, before or after IRI, exhibited improved renal function, less histologic renal injury, and attenuation of proinflammatory responses. Kim et al. further discovered that the expression of inflammatory molecules CCL2 and IL-6 was reduced by the IL-2 complex with decreased infiltration of neutrophils and macrophages. Tregs were crucial for this protection because depletion of Tregs with PC61, an anti-CD25 antibody, removed the protective effective of IL-2/anti-IL2. The IL-2 complex–mediated renal protection from IRI was not dependent on IL-10 and TGF-\(\beta\). This study provides evidence suggesting that \textit{in vivo} expansion of Tregs using IL-2/anti-IL-2 may be a promising approach for treating renal IRI and other autoimmune diseases. Kim et al. also suggest that Treg expansion can be highly effective after hypoxic injury, making the therapeutic use of Treg expansion much more efficacious than its limited prophylactic use. This study also suggests that Tregs may be acting through innate pathways, potentially using purine pathways to reduce inflammation through adenosine generation rather than just limiting effector T cells.\(^4\)

Treg treatment for autoimmune disease is currently entering clinical use. One of its major barriers is the difficulty of \textit{in vitro} selection and expansion of Tregs safely and in sufficient quantity for clinical use. Several cytokines are used to expand Tregs \textit{in vitro}. IL-2 and TGF-\(\beta\) have been shown to enhance growth, differentiation, and survival of Tregs and to protect cells from activation-induced cell death.\(^4,5\) The recent success of two clinical trials using low-dose IL-2 to expand Tregs \textit{in vivo} in trials of hepatitis C vasculitis and graft versus host disease is promising.\(^6,7\) In solid organ transplantation, the adoptive transfer of \textit{ex vivo}–generated Tregs was shown to be effective in a number of animal models.\(^8\) This led to the ONE Study, an ongoing series that incorporates a number of regulatory cell therapies, including Tregs for kidney transplantation (www.onestudy.org). Tregs have also shown effectiveness in animal models of graft versus host disease, leading to a number of proposed and ongoing human trials.\(^9\)

IL-2 plays a central role in activating CD4\(^+\) and CD8\(^+\) T cells and Tregs through the high-affinity IL-2 receptor composed of \(\alpha\beta\) and \(\gamma\) subunits.\(^10\) Tregs are also exquisitely dependent on IL-2 for survival. Studies have demonstrated that the potency of IL-2 can be enhanced with a specific IL-2 mAb clone (JES6-1), which leads to selective Treg expansion in mice and is used effectively in the study by Kim et al.\(^11,12\) This induces a temporary 3- to 4-fold increase in Treg numbers \textit{in vivo} and is protective in experimental models of allergy, experimental autoimmune encephalomyelitis, and islet transplantation.\(^11,13,14\) The utility of \textit{in vitro} Tregs created by gene transfer of Foxp3 in chronic proteinuria renal disease has also been demonstrated.\(^15\) Furthermore, the expansion of Tregs by the IL-2 complex as used in the study by Kim et al. and its potent clinical effect in limiting renal injury...
and proteinuria were shown in the well established model of Adriamycin nephropathy. The recent explanation of the mechanism of IL-2 superagonists suggests that compared with “superkines” such as mutant IL-2, which can activate T effectors better than Tregs without CD25 through the IL-2Rβ and IL2Rγ chains, the IL-2/IL-2 Ab complex functions primarily through CD25 and then the IL-2Rβ and IL2Rγ chains providing the selectivity for Tregs. These results suggest that in vivo expansion of Tregs using IL-2/anti-IL-2 is a possible therapeutic strategy for treating renal disease.

Additional studies must be performed to better understand the immunologic differences and risks between rodents and humans. Appropriate risk-minimization strategies are also crucial to make clinical application of Treg expansion by the IL-2 complex possible in the future. TGN1412, a superagonistic anti-human CD28 antibody (IgG4), showed efficacy in rodent models for treating autoimmune disease by Treg expansion. However, a clinical trial of this anti-CD28 antibody caused a massive cytokine storm and multiorgan failure through non-selective T cell activation in humans. This raises further questions about how best to design preclinical studies that can better predict the risks of novel immunotherapeutics in humans. Because the complex relies on CD25, effector cells expressing this receptor, including T cells and natural killer cells, are also capable of being expanded.

In summary, these studies demonstrate the important contribution of Treg therapy from IL-2 complex–induced Treg expansion in vivo. Despite the potential pitfalls of broad T cell activation shown previously with CD28 agonists, the IL-2/anti-IL-2 complex offers a promising pathway for Treg therapy in a number of conditions.

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DISCLOSURES

None.

REFERENCES
