

degeneration, it has been postulated that G6PD-deficient persons with red blood cell enzyme activities less than 15% of normal should avoid prolonged high-intensity physical exercise.<sup>4</sup> Two recent studies concluded that G6PD-deficient persons are able to perform mild to moderate exercise without higher oxidative stress than G6PD-nondeficients.<sup>5,6</sup> Of particular interest are 2 case reports describing adverse effects of exercise on G6PD-deficient persons. The first describes a 20-year-old G6PD-deficient man who was hospitalized for myalgia and myoglobinuria after intense exercise; he carried the same G6PD variant as our case.<sup>7</sup> The second is a 30-year-old pentathlon-trained athlete with G6PD deficiency who suffered from loss of consciousness and pigmenturia during the last meters of a 12-km competitive run.<sup>8</sup> Contrary to both these reports, our athlete showed no more severe signs of myalgia, myoglobinuria, or hemoglobinuria than other elite long-distance runners, although residual activity in erythrocytes was approximately 9%, and the calculated activity<sup>9</sup> in muscle cells was 13% of normal. We postulate that the athlete described here has developed a unique balance between his G6PD deficiency and exercise-induced disturbances of blood glutathione and lipid peroxidation that allows him to perform strenuous exercise.

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## To the editor:

### In vivo-activated CD103<sup>+</sup> Foxp3<sup>+</sup> Tregs: of men and mice

We read with interest the article by Zhao et al<sup>1</sup> on the specific abilities of murine CD103<sup>+</sup> T regulatory (Treg) cells to ameliorate ongoing chronic graft-versus-host disease (cGVHD). The authors report here that only in vivo-activated effector/memory-like Tregs that express high amounts of the integrin  $\alpha E$  (CD103) are able to efficiently control the disease in a murine model based on allogeneic transfer of DBA/2 donor cells into BALB/c hosts. In conclusion, the authors suggest that the infusion of human CD103<sup>+</sup> Tregs might also ameliorate cGVHD in patients, so that the reported findings may offer a new strategy for the treatment of cGVHD patients.

In principle, the use of effector/memory-like Treg cells may be indeed beneficial also in human therapy. We would like to point out, however, that the markers for this Treg subset are not compatible between humans and mice. CD103 is a well-established marker for murine effector/memory-like Treg cells, which have been activated in vivo by antigen in a specific context. In naive mice, up to 30% of CD25<sup>+</sup> FOXP3<sup>+</sup> lymphocytes in fact express CD103.<sup>2</sup> In contrast to this, cells with this marker are virtually absent among human CD25<sup>high</sup> Foxp3<sup>+</sup> Treg cells. It is expressed by less than 5% of human CD25<sup>high</sup> Foxp3<sup>+</sup> Treg cells in various tissues, including blood.<sup>3-6</sup> This applies also for  $\beta 7$ , the integrin  $\beta$ -chain binding to CD103, which is only poorly expressed by human Treg cells.<sup>3,7</sup> Although in CD4<sup>+</sup> T cells CD103 expression can be induced by different stimuli in vitro,<sup>5,8</sup> a significant expression of CD103 on human CD4<sup>+</sup> cells with regulatory potential ex vivo was reported only in tonsils, here, however, mostly on CD25<sup>-</sup> cells.<sup>5</sup>

Given the different expression of CD103 by Foxp3<sup>+</sup> Tregs in mice and humans, it is highly questionable whether the results obtained with murine CD103<sup>+</sup> Tregs can be translated 1:1 into a clinical setting. There is little doubt that equivalent cell subsets exist in humans and mice, and that effector/memory-like Treg cells fulfill analog functions in both species. Addressing these cells, however, requires considering species-specific differences. In humans, effector/memory-like Treg cells (T<sub>REM</sub>) are better defined by markers such as CCR6 or CD39.<sup>9,10</sup>

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## Response

### Is CD103 a good surface marker for in vivo-activated effector/memory Treg cells in patients with chronic inflammation? Unknown yet

We observed that the percentage of CD103<sup>+</sup> cells among CD4<sup>+</sup>FoxP3<sup>+</sup> regulatory T (Treg) cells of donor DBA/2 mice was less than 10% before transplantation, and the percentage of CD103<sup>+</sup> cells among donor-type CD4<sup>+</sup>Foxp3<sup>+</sup> Treg cells increased to more than 55% in BALB/c recipients with chronic graft-versus-host disease (GVHD) and 70% in recipients without chronic GVHD after hematopoietic cell transplantation (HCT).<sup>1</sup> We also observed that the percentage of CD103<sup>+</sup> cells among Foxp3<sup>+</sup>CD4<sup>+</sup> Treg cells in donor C57BL/6 mice was approximately 20% (mean ± SE: 20.7 ± 1.7%, N = 4) before HCT, and the percentage of CD103<sup>+</sup> Treg cells increased to around 40% (mean ± SE: 44.3 ± 2.0%, N = 4) in BALB/c recipients with acute GVHD. These results indicate that inflammation, especially chronic inflammation, can augment the expression of CD103 on mouse Foxp3<sup>+</sup> Treg cells, although the percentage of CD103<sup>+</sup> cells among Foxp3<sup>+</sup> Treg cells can differ up to certain levels in different mouse strains or different housing environments, because the percentage of CD103<sup>+</sup> Treg cells in our DBA/2 mice is around 10%, but the percentage of our C57BL/6 mice is around 20%; the percentage of CD103<sup>+</sup> Treg cells in our C57BL/6 mice is approximately 20%, but the percentage in C57BL/6 mice in Hühn's reports is approximately 30%.<sup>2</sup>

It has been well documented that the percentage of CD103<sup>+</sup> cells among Foxp3<sup>+</sup>CD4<sup>+</sup> Treg cells in healthy humans is low, around 5%.<sup>3-5</sup> However, it is still unknown whether Foxp3<sup>+</sup> Treg cells in human HCT recipients express CD103. It has been shown that the percentage of CD103<sup>+</sup> cells among CD25<sup>hi</sup>CD4<sup>+</sup> T cells was significantly higher than that among CD25<sup>-</sup>CD4<sup>+</sup> T cells in patients with multiple sclerosis.<sup>6</sup> In addition, in vitro TGF-β-induced Treg cells were all CD103<sup>+</sup>.<sup>7</sup> TGF-β secretion is usually up-regulated in patients with chronic inflammation.<sup>8</sup> Therefore, although most of the Foxp3<sup>+</sup> Treg cells in normal humans do not express CD103, we cannot exclude the possibility that Foxp3<sup>+</sup> Treg cells in patients with chronic inflammation (such as chronic GVHD) will up-regulate CD103 expression. Because it has been reported that effector/memory Foxp3<sup>+</sup> Treg cells express CCR6 in mice<sup>9</sup> and they express CD39 in multiple sclerosis patients,<sup>10</sup> comparison of expression of CD103, CCR6, and CD39 by Foxp3<sup>+</sup> Treg cells in patients with chronic inflammation is required to find out which one is best for identifying the Foxp3 Treg cells.

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