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Phase 1–2 Trial of AAVS3 Gene Therapy in Patients with Hemophilia B

TO THE EDITOR: Chowdary and colleagues (July 21 issue)[1] present data on an adeno-associated virus (AAV)–based gene therapy with FLT180a (verbrinacogene setparvovec) in 10 patients with hemophilia B. The patients received one of four doses of FLT180a: 3.84×10^{11} vector genomes (vg) per kilogram of body weight, 6.40×10^{11} vg per kilogram, 8.32×10^{11} vg per kilogram, or 1.28×10^{12} vg per kilogram. Most interestingly, the level of factor IX coagulant activity in the 4 patients receiving the highest dose reached an initial peak of approximately 200 IU per deciliter and later showed a notable decline up to 9 to 12 months after vector genome infusion. The decline in factor IX activity to approximately 30 to 55 IU per deciliter coincides with the stoppage of or reduction in immune suppression.

The immune response against AAV gene therapy is complex and can be triggered by vector capsid, genomes (number of CpG dinucleotides), and protein products.[2,3] FLT180a contains only five CpG dinucleotides, and the vector capsid should have been metabolized by the time of decline in factor IX activity; therefore, these factors are unlikely candidates to account for this late-onset decline. The protein product has been discussed in the context of the nonphysiologic expression of factor VIII protein in hepatocytes.[4] However, factor IX is made endogenously by the hepatocytes. Thus, the late onset of a decline in factor IX activity in the FLT180a study remains mysterious but has a clear correlation with the number of infused vector genomes.

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THE AUTHORS REPLY: We agree with Oldenburg that the pattern of late-onset declines in factor IX levels in the final four patients is notable. We also agree that the immune response to AAV gene therapy is complex and that the causes of these declines are unknown. However, the statement that the final four patients (Patients 7 to 10) received the highest dose is inaccurate. As Figure 1 in our article shows, the final four patients received a dose of 8.32×10^{11} vg per kilogram, which was less than the dose of 1.28×10^{12} vg per kilogram received by both Patients 3 and 6. The two patients who did receive the highest dose did not have the late declines in factor IX activity that was observed in the final four patients (Fig. 2 in our article). Therefore, it is incor-

rect to say that the late-onset decline in factor IX activity "has a clear correlation with the number of infused vector genomes."

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Genetically Modified Porcine-to-Human Cardiac Xenotransplantation

TO THE EDITOR: In their pioneering work, Griffith et al. (July 7 issue)[1] denuded the porcine xenograft of the most immunogenic epitopes before transplantation. However, overzealous attempts at achieving immunologically inert nonhuman organs for transplantation may paradoxically promote rejection. There has been a recent realization that transplant rejection is mediated not only by host recognition of foreign antigens but also by failure of host immunocytes to recognize self-antigens on the xenograft.[2,3] This is the aptly named "missing-self" phenomenon.[2,3]

Tumors attempt to evade immunosurveillance by adopting a hypoimmunogenic state; they are therefore characterized by a paucity of antigen expression. However, the host overcomes this characteristic of tumors by means of natural killer (NK) cell–mediated destruction of cells that do not express self-antigens.[4] Recent studies have shown the primacy of the missing-self phenomenon in organ rejection.[2,3] Some of the histologic features described by Griffith et al. reflect those observed in this mode of graft rejection. Mammalian target of rapamycin (mTOR) inhibitors such as sirolimus (also called rapamycin) have been shown to abrogate NK cell function and to impair missing-self rejection[2]; through a linked or separate mechanism, they also prevent cardiac hypertrophy in the context of xenotransplantation.[5,6] Substantial hypertrophy was observed here. It is possible that mTOR inhibition is required for successful xenotransplantation.

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No potential conflict of interest relevant to this letter was reported.

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TO THE EDITOR: The report by the team that conducted porcine-to-human transplantation suggests that they rightly focused on preventing rejection, but one could argue that the heart they used was not able to sustain itself for a more extended period because the porcine heart is probably not a good fit for the human cardiac vasculature. The postmortem findings in the heart could be a reflection of decompensated ventricles, as reflected by the loss of myocardial integrity. The organization of the cardiac musculature in a helical fashion in the ventricles along with other spatial features are essential for sustained long-term cardiac function.[1-3] Future attempts at xenotransplantation may require consideration of cardiac models[4] that replicate the ventricular