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I. Abstract: Recent success in achieving graft survival without chronic immunosuppression in patients receiving non-myeloablative conditioning followed by combined kidney and bone marrow transplantation (CKBMT) is very promising. However, only transient mixed chimerism was achieved in these patients and *in vitro* data suggest that the kidney itself contributed to tolerance induction. In cynomolgus monkeys, non-myeloablative recipient conditioning followed by CKBMT induces transient mixed hematopoietic chimerism and donor-specific renal allograft tolerance. However, transient chimerism has not been associated with tolerance to islet (without a donor kidney graft), heart or lung allografts in nonhuman primates. Rodent studies indicate that life-long mixed chimerism achieves robust, systemic tolerance permitting donor islet allograft acceptance. **We hypothesize that durable, rather than transient chimerism will be required to achieve robust, systemic tolerance that will allow donor islet allograft tolerance in monkeys and humans.** Moreover, studies in a Type 1 diabetes model (NOD mice) indicate that non-myeloablative establishment of durable mixed chimerism also tolerizes autoreactive T cells so that donor islets are not destroyed by recurrent autoimmunity. Thus, **durable mixed chimerism could cure Type 1 diabetes by allowing donor islet allograft tolerance and reversing autoimmunity**. However, a clinical protocol has not yet been developed that reliably achieves durable mixed chimerism across HLA barriers without graft-vs-host disease (GVHD). GVHD is especially severe following HLA-mismatched hematopoietic cell transplantation (HCT) and is unacceptable in applying HCT for islet allograft tolerance induction. In mice, infused recipient Tregs promote durable mixed chimerism and tolerance with minimal, non-toxic recipient conditioning, without GVHD. **We propose a study using this novel protocol to safely achieve durable mixed chimerism and robust allograft tolerance in cynomolgus monkeys**. Polyclonally expanded recipient Tregs can be cryopreserved in readiness for use whenever any deceased donor becomes available. Our preliminary studies show that expanded recipient natural Tregs sorted from cynomolgus monkey PBMCs can markedly augment the level and duration of chimerism in monkeys receiving an otherwise transient chimerism regimen that involves CyA for one month post-transplant. We now aim to replace CyA with a short course of rapamycin, which, in contrast to calcineurin inhibitors like CyA, favors the generation and expansion of Tregs over conventional T cells and is commonly used in islet transplantation protocols. In Type 1 diabetic humans, durable mixed chimerism will also prevent the autoimmune attack that currently impedes islet allograft survival. To begin to address our hypotheses, we propose studies in cynomolgus monkeys. **Success of this study will position us for non-human primate studies to achieve islet allograft tolerance, providing a direct conduit to clinical application in deceased donor islet transplantation**.