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ORIGINAL ARTICLE

Immune tolerance via FCR001 cell therapy compared with maintenance immunosuppression for kidney transplantation: Real-world evidence analysis of safety and efficacy

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Abstract

While kidney transplantation (KTx) has traditionally required lifelong immunosuppression, an investigational stem cell therapy, FCR001, has been demonstrated to induce tolerance and eliminate the need for immunosuppression through the establishment of persistent mixed chimerism in a phase 2 clinical study. Real-world evidence (RWE) methods were employed to compare the safety and efficacy of non-myeloablative conditioning with FCR001 with standard of care [SOC] immunosuppression in a retrospective single-center analysis of outcomes among propensity score matched living-donor KTx receiving SOC (n = 144) or FCR001 (n = 36). Among the FCR001 recipients, 26 (72%) developed persistent chimerism allowing durable elimination of all immunosuppression. There was no significant difference in the composite primary endpoint (biopsy-proven acute rejection [BPAR], graft loss, or death) at 60 months (FCR001 27.8%, n = 10 and SOC 28.5%, n = 41; p = .9). FCR001 recipients demonstrated superior kidney function at 5 years (estimated glomerular filtration rate [eGFR] [mean ± standard deviation]: 64.1 ± 15.3) compared to SOC (51.7 ± 18.8; p = .02). At 5 years, FCR001 recipients experienced fewer complications including new-onset diabetes post-transplant, although two patients developed graft versus host disease. In conclusion, RWE demonstrated that KTx combined with non-myeloablative conditioning and FCR001 resulting in superior kidney function without increasing the risk of rejection, graft loss, or death among patients off immunosuppression.

KEYWORDS
immunosuppressive regimens, kidney transplantation: living donor, tolerance

1 | INTRODUCTION

Kidney transplantation is lifesaving; however, immunosuppression (IS) therapy has significant side effects and contributes to long-term nephrotoxicity and allograft loss.[1] Current immunosuppression regimens are associated with the development or exacerbation of significant comorbidities including hypertension, dyslipidemia, diabetes, malignancy, and life-threatening infection.[2–6] Furthermore, the requirement for daily immunosuppression treatment negatively impacts the patient's quality of life, is associated with substantial cost,

and contributes nonadherence which is a leading cause of premature allograft loss.[1,7] Consequently, despite marked improvements in early allograft survival and reduction in the rate of acute rejection, 10-year graft survival among living-donor kidney transplant (LDKT) recipients in the most recent annual report from the Scientific Registry of Transplant Recipients (SRTR) is only 65.5%, rendering the promise of "one kidney transplant for life" aspirational and out of reach for many patients.[8–10]

Real-world data, such as those originating from electronic health records and health care claims, are increasingly used to generate real-world evidence (RWE) that can contribute to treatment development programs investigating potential benefits and risks of medical products.[11,12] Through the use of modern statistical techniques, RWE can be used to create quasi-experimental analyses in which treated patients are matched with similar patients treated with existing clinical regimens to assess differences in patient outcome, resource utilization, and treatment-related complications. RWE can inform regulatory decision-making, including the approval of new treatments or expansion of indications for available treatments, as directed by recent guidance from the Food and Drug Administration.[11]

Combined hematopoietic stem cell transplant (HSCT) and kidney transplant from the same donor have been used to successfully establish donor-specific allograft tolerance in human leukocyte antigen (HLA)-matched or -mismatched LDKT recipients, allowing withdrawal of chronic IS without rejection of the kidney allograft.[13,14] Although HSCT was initially employed with ablative conditioning for patients with hematologic malignancies, novel approaches to tolerance induction in HSCT, including non-myeloablative conditioning regimens and innovative cell preparations, offer the promise of tolerance induction with less toxicity.[13,15,16] While early clinical trial data were promising, broader utilization of tolerance inducing strategies requires greater insight into the balance of the risks of the HCST therapy with the benefits of long term IS withdrawal and preservation of kidney allograft function.[17]

In single-arm clinical trials, facilitated allogeneic HSCT, which uses a defined combination of stem cells and immune cells, achieved persistent chimerism and donor-specific transplant tolerance, allowing for patients to be weaned off of IS *irrespective of the degree of HLA mismatch* in LDKT recipients.[13,18] FCR001 is an investigational allogeneic cell therapy derived from mobilized peripheral blood mononuclear cells from the same donor as the kidney allograft and contains hematopoietic stem cells, facilitating cells, and $\alpha\beta$ T cells. The facilitating cells promote survival, homing, and migration of stem cells to induce persistent mixed chimerism across HLA mismatch, including in unrelated recipients.[19] A two-center Phase 2 study (NCT00497926) demonstrated the ability of FCR001 to induce tolerance by establishing persistent chimerism. Patients were weaned from IS if their kidney function was stable, no donor-specific antibodies were detected, they had not experienced BPAR, kidney allograft biopsies were normal, and importantly, there was persistent mixed chimerism, defined as ≥50% T-cell chimerism for 6 months and beyond. In this trial, IS was completely and durably withdrawn in 26 of 37 patients across different levels of HLA mismatches in de novo LDKT recipients by 1-year posttransplant.[15,19]

To further inform clinicians and patients about the risks and benefits of this innovative treatment, a RWE investigation was developed to compare the outcome LDKT in recipients who received FCR001 (FCR-R) and with well-matched contemporaneously transplanted LDKT recipients receiving standard two- or three-drug IS treatment (standard of care [SOC]-R) at the same institution. A robust electronic data warehouse was queried to capture key clinical outcomes including death, graft loss, biopsy-proven acute rejection (BPAR), graft versus host disease, and allograft function.

2 | METHODS

2.1 | Research design and study population

This analysis utilized data extracted from the Northwestern Medicine Enterprise Data Warehouse (NMEDW), which includes Northwestern's electronic medical records, Northwestern's histocompatibility lab data, and the Organ Procurement and Transplantation Network (OPTN). The NMEDW was used to identify and characterize the outcomes among FCR-R and SOC-R treated patients using detailed clinical and laboratory data, administrative claims, and clinical record review/validation.

The FCR-R group included all 36 patients from the Phase 2 single-arm, uncontrolled trial of FCR001 at the Northwestern Memorial Hospital (NMH). The one FCR-R patient in the Phase 2 trial who was not treated at NMH was excluded. All FCR-R patients were ≥18 years of age and received LDKT between 2009 and 2016, the time frame of recruitment into the Phase 2 clinical trial.[16] At the time of this analysis (March 23, 2022), all surviving FCR-R patients from the Phase 2 study (33/37) remained in long-term follow-up and had >5 years of follow-up from the day the kidney transplant was received. The SOC-R cohort included patients ≥18 years old who received their transplant from a living donor aged 18 to 60 years old at NMH contemporaneously with LDKT recipients in Phase 2 FCR001 trial and met key inclusion/exclusion criteria of Phase 2 FCR001 trial. This study was approved by the ethics committee of Northwestern University (IRB ID STU00214597).

FCR-R patients received non-myeloablative conditioning (with fludarabine, cyclophosphamide, and low-dose total body irradiation [200 cGy]) and initiation of IS with tacrolimus and mycophenolate mofetil (as described previously[19]) over the 4 days prior to LDKT, followed by FCR001 infusion 24 h post-transplant. IS was tapered and withdrawn over 12 months according to the eligibility criteria for IS withdrawal.[19] No patient who received the conditioning regimen failed to progress to transplant. The FCR-R population was analyzed as an intent-to-treat arm and included the patients who failed to achieve persistent chimerism and tolerance despite receiving conditioning treatment and cell infusion and thus remained on IS. SOC-R patients were managed according to institutional post-transplant IS protocols

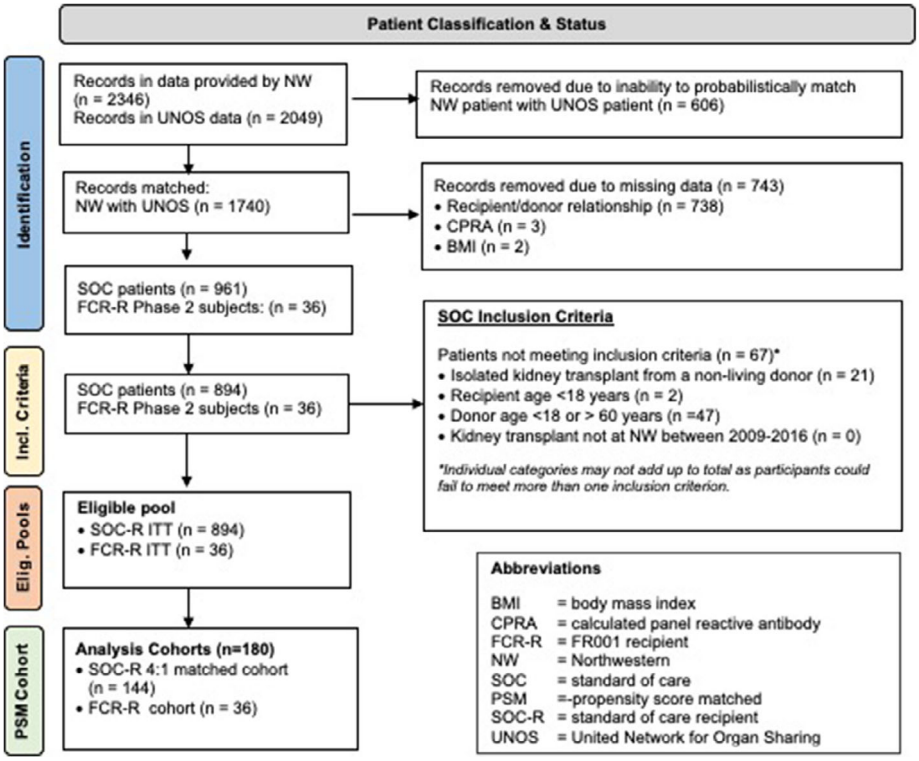


FIGURE 1 Patient eligibility and inclusion/exclusion criteria.

that included tacrolimus, mycophenolate, and antibody induction with or without corticosteroids.

2.2 | Matching

Propensity score matching (PSM) was used to identify a population of SOC-R patients that were comparable in baseline demographics and clinical characteristics to the FCR-R patients (Figure 1). The matching variables were selected with clinical input and a review of univariate analyses to identify characteristics potentially associated with the primary outcome (Table 1). The PSM scores were determined using a random forest model.[19] Propensity scores were used to create a 4:1 matched group ($n = 36$ FCR-R, $n = 144$ SOC-R) using a "greedy nearest neighbor" matching algorithm.[20] The balance of individual baseline characteristics between the FCR-R and SOC-R groups is reported using standardized mean differences (SMD), which was below .2 for the majority of characteristics (Table 2). Residual differences were adjusted for using a two-level nested mixed-effects regression model. The 4:1 PSM was chosen to maximize study power, while not compromising the balance in baseline characteristics between the matched SOC-R and FCR-R treatment groups.

2.3 | Endpoints

The primary endpoint was a composite of death, graft failure, or BPAR within 5 years of transplant. BPAR was defined as a biopsy result with either T-cell–mediated rejection of Banff Grade ≥1A or acute/active antibody-mediated rejection. Local pathology for all biopsies performed (both for cause and determined based on surveillance) were subjected to direct expert clinical review of the full text report. No re-interpretation of any allograft pathologic specimens was performed. Predefined secondary endpoints were derived from RWE including the individual components of the primary composite endpoint; kidney allograft function (defined as estimated glomerular filtration rate [eGFR] calculated with creatinine and albumin values, age, sex, and race based on the Modification of Diet in Renal Disease 4 [MDRD4] formula).[21] The mean eGFR was calculated in FCR-R and SOC-R patients post-transplant at months 1 (baseline), 6, 12, 24, 36, and 60. The incidence of adverse events (AEs) including infection, malignancy, hospitalization, hematologic abnormalities, thromboembolism, new-onset diabetes mellitus or dyslipidemia, and select cardiovascular events were determined using electronic medical records and international classification of diseases (ICD)−9/10, Healthcare Common Procedure Coding System (HCPCS), Diagnostic-Related Group (DRG), Systemized Nomenclature of Medicine (SNOMED), and Logical Observation Identifiers Names and Codes (LOINC) codes summarized in Supplemental Digital Content: Appendix. Direct chart review was not used consistent with RWE methodologies to define computable phenotypes.[12,22,23] Graft-vs-host disease was also evaluated in the FCR-R group using case review forms as there were no cases of graft-vs-host disease in SOC kidney transplant recipients. To increase the precision of these computable phenotypes, an adverse event was considered significant if the defined code associated with an inpatient hospitalization that lasted ≥2 days or was

TABLE 1 Propensity score matching classifications and definitions.

Covariates	Classification/definition
Age at kidney transplant	Years (within 5-year differences)
Sex (categorical)	Male or female
Race	White, non-White
Baseline BMI (categorical)	<25, ≥25 to < 30, ≥30 kg/m^2
Year of kidney transplant	Continuous (same year)
Primary cause of end-stage renal disease	Diabetes Hypertensive nephrosclerosis Other diagnoses (glomerular diseases; tubular and interstitial diseases; congenital, familial, metabolic; renovascular and vascular diseases; neoplasms) Other nonrelated diagnoses or missing diagnoses
Donor/recipient relationship	Related, unrelated
ABO compatibility (yes/no)	Donor and recipient combination
HLA mismatches	Number of HLA mismatches: 0–3, 4–6
Diabetes mellitus	Yes
Dialysis	Yes
Baseline CMV match	Donor vs. Recipient (D+/R−, D+/R+, D−/R−, D−/R+, missing)

Abbreviations: BMI, body mass index; CMV, cytomegalovirus; D+, donor positive; D−, donor negative; HLA, human leukocyte antigen; R+, recipient positive; R−, recipient negative.

listed on claims for ≥2 outpatient visits within the study period of interest.

2.4 | Statistical analysis

Time to the first event of the composite endpoint (time to death, graft loss (kidney), or BPAR) was assessed with Kaplan-Meier (KM) survival analysis. Subjects without an event were censored on their last day of clinical contact. Time to event by treatment group was reported with 95% confidence intervals. KM analyses were also performed on the individual endpoints. Log-rank tests were used to identify statistically significant differences. Welch's two-sample *t*-test was used to compare eGFRs between groups. Differences in mean change in eGFR from the end of month 1 to month 24 after LDKT between FCR-R and SOC-R cohorts were summarized by point estimates and 95% confidence intervals, and the effect was assessed using least-squares mean differences. Laboratory values and safety outcomes were descriptive and statistical analyses were not performed. Analyses were performed using R (version Rx64 4.1.1).

3 | RESULTS

3.1 | Patient disposition and demographics

Immunosuppression was successfully withdrawn from 26 of 36 FCR-R treated kidney transplant recipients, all of whom demonstrated persistent mixed chimerism. Outcomes for all 36 FCR-R patients were compared with 144 matched kidney recipients treated with SOC immunosuppression (SOC-R). The FCR-R and SOC-R cohorts were statistically well matched for race, primary cause of end-stage kidney disease, donor/recipient relationship, blood type (A, B, AB, and O) match, and HLA mismatches (standardized mean difference of −.1 to .1, Table 2). There were statistically significant differences in only in recipient age categorized in the proportion of recipients aged 41–45 (SMD = .3) and 56–60 (SMD = −.4).

3.2 | Death, graft loss, and BPAR

A similar proportion of FCR-R patients (27.8%, $n = 10$) and SOC-R patients (28.5%, $n = 41$) reached the composite endpoint of death, graft loss, or BPAR by 60 months post-transplant ($p = .9$) (Figure 2A). Within 5 years of transplant, there was no statistically significant increase in the risk of BPAR (FCR-R: 19.4% vs. SOC-R: 13.9%, $p = .34$) (Figure 2B). Banff classification for patients who experienced BPAR is shown in Table 3. None of the BPAR episodes occurred among FCR-R patients with stable chimerism and who had been weaned off IS. There were no significant differences in death or graft loss between FCR-R and SOC-R patients (Figure S1). The death occurred in <10% of patients in each group (8.3%, $n = 3$ FCR-R and 9.7%, $n = 14$ SOC-R; $p = .77$). Similarly, graft loss rates did not differ (FCR-R: 5.6% vs. SOC-R: 9%, $p = .52$).

3.3 | Kidney function

Mean eGFR values were similar in both groups from month 1 to month 24 ($p \geq .22$). Subsequently, renal function in the SOC-R population began to decline while FCR-R patients' renal function remained stable at 36 months (mean ± SD: FCR-R [$n = 19$]: 61.1 ± 17.6 mL/min/1.73m^2 vs. SOC-R [$n = 84$]: 57.4 ± 17.8 mL/min/1.73m2; $p = .41$). FCR-R patients had a statistically better graft function at 60 months (FCR-R: 64.1 ± 15.3 mL/min/1.73m2 vs. SOC-R: 51.7 ± 18.8 mL/min/1.73m2; $p = .02$) (Figure 3).

3.4 | Adverse events

FCR patients experienced fewer cardiometabolic complications associated with immunosuppression including new-onset type 2 diabetes mellitus (SOC-R: 20.8% compared to the FCR-R 7.7%) and new-onset dyslipidemia requiring medication treatment (SOC-R: 30.9% vs. FCR-R: 5.9%) (Figure 4A). Similarly, a higher proportion of patients

TABLE 2 Summary of post-propensity score matching baseline demographic and clinical characteristics.

	FCR-R ($n = 36$)[a] *n* (%)	SOC-R ($n = 144$) *n* (%)	Standardized mean difference
Age at kidney transplant (years)			
18–20	1 (2.8)	5 (3.5)	.0
21–25	3 (8.3)	9 (6.2)	.1
26–30	3 (8.3)	18 (12.5)	−.1
31–35	3 (8.3)	15 (10.4)	−.1
36–40	5 (13.9)	17 (11.8)	.1
41–45	7 (19.4)	12 (8.3)	.3
46–50	5 (13.9)	14 (9.7)	.1
51–55	5 (13.9)	19 (13.2)	.0
56–60	1 (2.8)	13 (9.0)	−.4
61–65	3 (8.3)	18 (12.5)	−.1
66–80	0	4 (2.8)	NA
Sex			
Male	30 (83.3)	108 (75.0)	.2
Race: White[b]	27 (75.0)	105 (72.9)	.0
BMI ≥30 kg/m^2 (obese) at baseline	7 (19.4)	33 (22.9)	−.1
Year of kidney transplant			
2009	4 (11.1)	24 (16.7)	−.2
2010	7 (19.4)	26 (18.1)	.0
2011	3 (8.3)	14 (9.7)	.0
2012	5 (13.9)	18 (12.5)	.0
2013	2 (5.6)	15 (10.4)	−.2
2014	6 (16.7)	13 (9.0)	.2
2015	4 (11.1)	14 (9.7)	.0
2016	5 (13.9)	20 (13.9)	.0
Primary cause of ESRD[c]			
Diabetes	3 (8.3)	12 (8.3)	.0
Hypertensive nephrosclerosis	3 (8.3)	13 (9.0)	.0
Other diagnoses[d]	28 (77.8)	113 (78.5)	.0
Other unrelated diagnosis group or missing	2 (5.6)	6 (4.2)	.1
Related donor/recipient: yes	20 (55.6)	81 (56.2)	.0
ABO compatible: yes	35 (97.2)	143 (99.3)	−.1
HLA mismatches			
0–3	14 (38.9)	60 (41.7)	−.1
4–6	19 (52.8)	75 (52.1)	.0
Missing	3 (8.3)	9 (6.2)	.1
CMV serostatus			
D+/R−	6 (16.7)	28 (19.4)	−.1
D+/R+	8 (22.2)	43 (29.9)	−.2
D−/R−	16 (44.4)	45 (31.2)	.3
D−/R+	5 (13.9)	26 (18.1)	−.1
Missing	1 (2.8)	2 (1.4)	.1

(Continues)

TABLE 2 (Continued)

	FCR-R ($n = 36$)[a] n (%)	SOC-R ($n = 144$) n (%)	Standardized mean difference
Comorbidities[c]			
Diabetes mellitus	10 (27.8)	45 (31.2)	−.1
Dialysis	25 (69.4)	87 (60.4)	.2

Note: NA is presented for the standardized difference for categories where cell counts were 0 and therefore not calculable.
Abbreviations: ABO, A, B, AB, and O blood types; BMI, body mass index; CMV, cytomegalovirus; D−, donor negative; D+, donor positive; ESRD, end-stage renal disease; FCR-R, FCR001 recipient; HLA, human leukocyte antigen; NA, not applicable; R−, recipient negative; R+, recipient positive; SOC-R, standard of care recipient.
[a]Denominator for percentages is the column total unless otherwise specified.
[b]Propensity score matching used two categories for race (White vs. non-White). Percentages will not add up to 100%.
[c]Percentages may not add to 100%, because patients can have multiple causes of ESRD/comorbidities.
[d]Glomerular diseases; tubular and interstitial diseases; congenital, familial, and metabolic diseases; renovascular and vascular diseases; neoplasms.

TABLE 3 Banff grades among those who experienced rejection (BPAR).

	FCR-R ($n = 7$)[a]	Matched SOC-R ($n = 20$)[a]
Banff grade (categories), *n* (%)		
1A/1B	4 (57.1)	13 (65.0)
2A/2B	2 (28.6)	3 (15.0)
3	1 (14.3)	1 (5.0)
AMR/ABMR	0	2 (10.0)
Missing	0	1 (5.0)

Abbreviations: AMR/ABMR, acute/active antibody-mediated rejection; BPAR, biopsy-proven acute rejection; FCR-R, FCR001 recipient; SOC-R, standard of care recipient.
[a]Denominator for percentages is the number who experienced BPAR.

in the SOC-R cohort had cholesterol, low-density lipoprotein, and white blood cell values falling outside of the normal range at 24 months post-transplant (14.6%, 68.8%, and 44.8%, respectively) compared with those in the FCR-R cohort (13.6%, 50.0%, and 9.1%). Deep vein thrombosis AEs were more frequent in FCR-R patients relative to SOC-R patients (25.0% vs. 6.9%, respectively). There were no differences observed in the incidence of stroke or intracerebral hemorrhage (2.8% vs. 2.8%) or pulmonary embolism (2.1% vs. 2.8%) (Figure 4B). Finally, while there were higher rates of acute myocardial infarction in the FCR-R group (FCR-R: 2.8% vs. SOC-R: 1.4%), heart failure was more common in the SOC-R (FCR-R: 5.6% vs. SOC-R: 10.4%) Two FCR-R experienced graft-versus-host disease (GVHD), neither of which was fatal. No patients in the SOC group experienced GVHD.

Hematologic AEs that required intervention (anemia, neutropenia, thrombocytopenia, pancytopenia, and leucopenia) were more frequent in the FCR-R cohort than in the SOC-R cohort across the entire follow-up period. As expected, given the conditioning regimen in FCR-R patients, neutropenia was more common in the FCR-R cohort (61.1%), than in the SOC-R cohort (10.4%). FCR-R patients also experienced increased rates of anemia (36.1% vs. 5.6%) and thrombocytopenia (38.9% vs. .7%); however, most of these events occurred within 6 months of the transplant. No FCR-R patients experienced a hematologic AE at 13−24 months post-transplant, whereas SOC-R patients experienced low levels of hematologic AEs throughout the follow-up period (Figure 4B).

The incidence of any infection was initially greater in the FCR-R group; however, the frequency of infections in the FCR-R group continued to decline throughout the follow-up period. The incidence of infections in the SOC-R patients started at a lower level than for FCR-R (0−6 months), but was higher by 13−24 months, and was notably more common than in the FCR-R group at the end of the follow-up period (24+ months). The infection rate was therefore greater in the FCR-R group 0−24 months and was higher in the SOC-R group beyond 24 months (Figure 4C).

Skin cancer was reported in 2 FCR-R patients (5.6%) and 4 SOC-R patients (2.8%). Hematologic cancer occurred in 1 FCR-R patient (2.8%) and 2 SOC-R patients (1.4%). Other solid organ tumors developed in 2 FCR-R patients (5.6%) and 6 SOC-R patients (4.2%). Post-transplant lymphoproliferative disease occurred in 2 SOC-R patients (1.4%) but was not reported in the FCR-R cohort (Figure 4D).

4 | DISCUSSION

A RWE evaluation comparing the outcomes of HLA mismatched patients treated with FCR001, 70% of whom were weaned off immunosuppression, demonstrated equivalent rates of death, graft failure, and BPAR when compared with to patients who remained on chronic immunsouppresion.[15,19] FCR001 treatment was associated with preserved kidney allograft function, whereas SOC patients experienced to a progressive decline in mean eGFR ($p < .02$). Two of the FCR patients experienced non-fatal GVHD. This RWE evaluation suggests that non-myeloablative induction and donor-derived FCR001 infusion was not associated with an increased risk of early graft loss or rejection. Furthermore, FCR001 with selected immunosuppression withdrawal in chimeric resulted in superior allograft function in an intent-to-treat

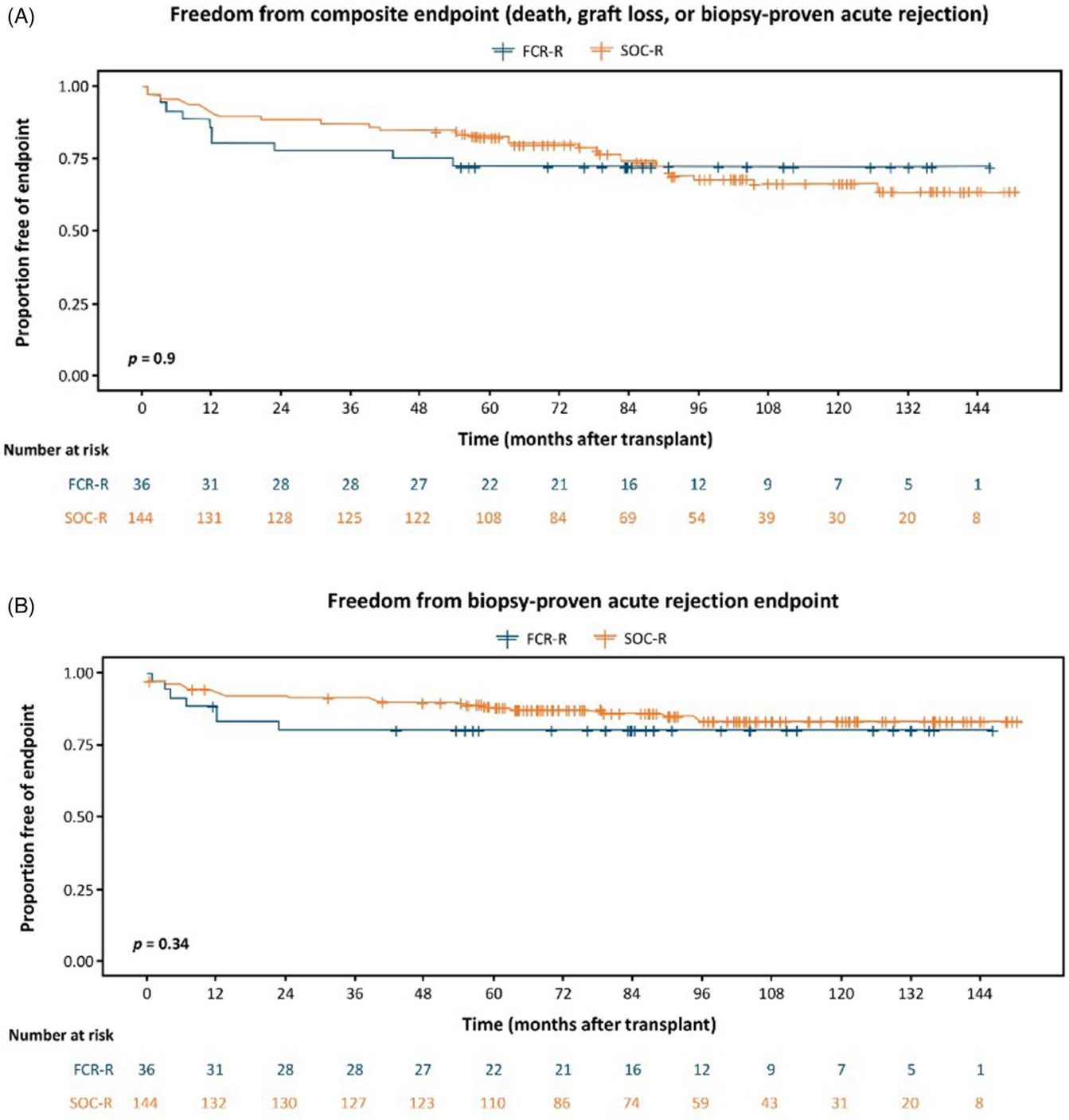
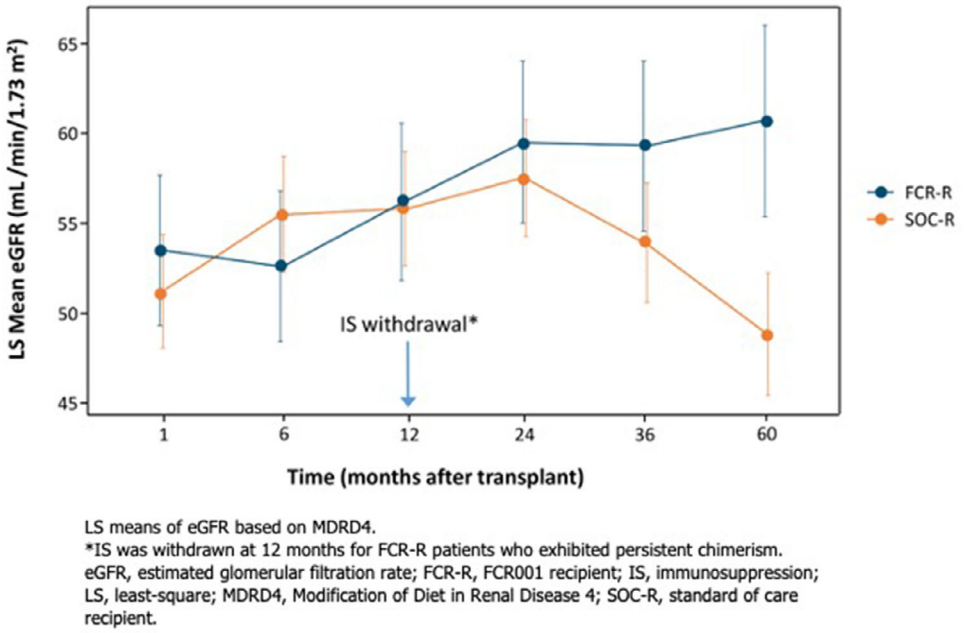


FIGURE 2 (A) Composite death, graft loss, or biopsy-proven acute rejection. BPAR, biopsy-proven acute rejection; FCR-R, FCR001 recipient; SOC-R, standard of care recipient. (B) Incidence of biopsy proven rejection. BPAR, biopsy-proven acute rejection; FCR-R, FCR001 recipient; SOC-R, standard of care recipient.

analysis, which may reduce the long-term graft loss resulting from chronic exposure to calcineurin inhibitors.

Immunosuppression therapy increases the risk of type 2 diabetes and dyslipidemia, contributing to cardiovascular disease which is the leading cause of post-transplant mortality.[5,6,24–27] FCR001 treatment was associated with a lower incidence of new-onset type 2 diabetes and dyslipidemia than SOC-R maintained on chronic immunosuppression. In addition, FCR001 treatment was associated with a lower risk of de novo cardiometabolic complications compared with SOC treatment. As expected, following the non-myeloablative conditioning regimen, the incidence of infections was initially higher in FCR-R patients. However, as immunosuppression was successfully weaned, the rate of infection decreased. By 24 months, there were fewer serious infections requiring hospitalization or multiple outpatient treatments among



LS means of eGFR based on MDRD4.
*IS was withdrawn at 12 months for FCR-R patients who exhibited persistent chimerism.
eGFR, estimated glomerular filtration rate; FCR-R, FCR001 recipient; IS, immunosuppression; LS, least-square; MDRD4, Modification of Diet in Renal Disease 4; SOC-R, standard of care recipient.

FIGURE 3 Mean renal function measurement after transplant. LS means of eGFR based on MDRD4. *IS was withdrawn at 12 months for FCR-R patients who exhibited persistent chimerism. eGFR, estimated glomerular filtration rate; FCR-R, FCR001 recipient; IS, immunosuppression; LS, least-square; MDRD4, modification of diet in renal disease 4; SOC-R, standard of care recipient.

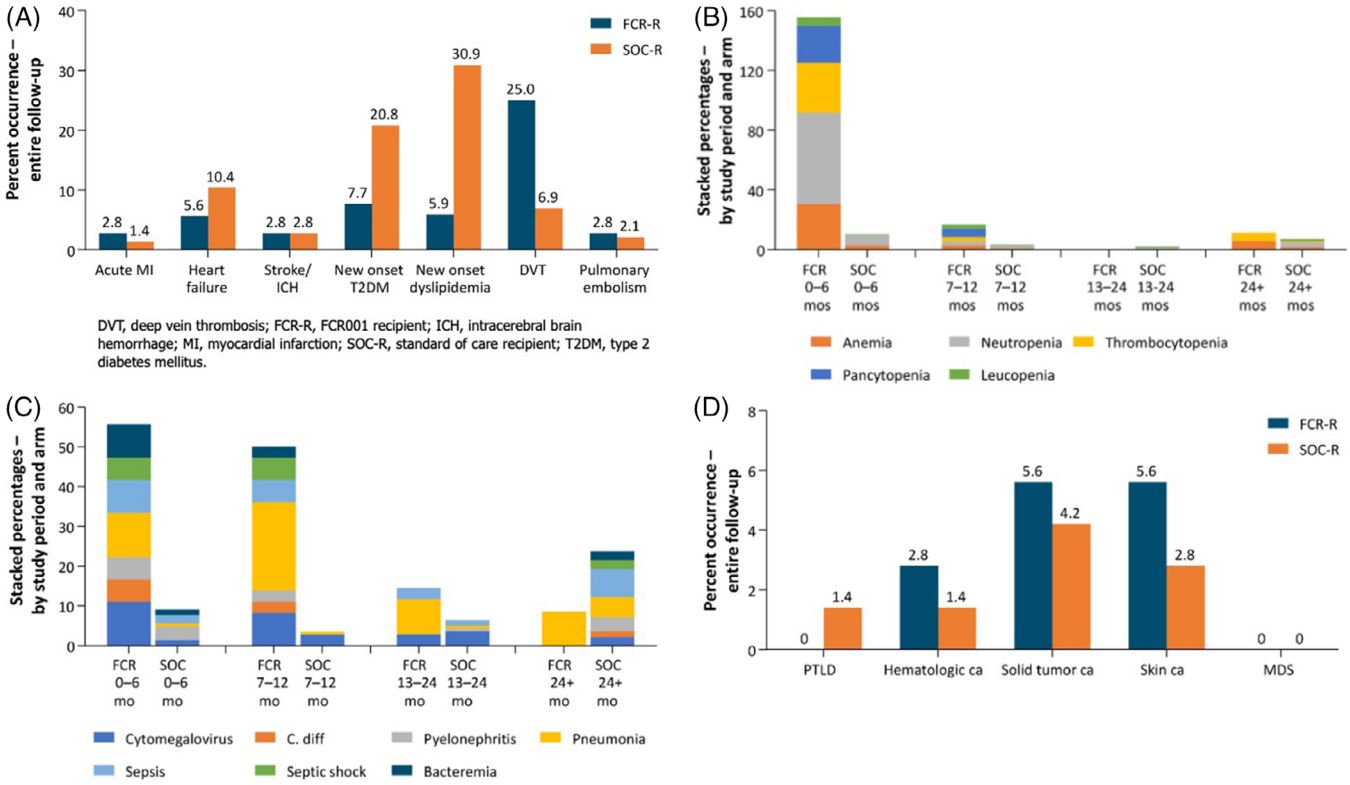


FIGURE 4 (A) Cardiometabolic adverse events after kidney transplant. DVT, deep vein thrombosis; FCR-R, FCR001 recipient; ICH, intracerebral brain hemorrhage; MI, myocardial infarction; SOC-R, standard of care recipient; T2DM, type 2 diabetes mellitus. (B) Hematologic disorders after transplant. (C) Infection occurrence post-transplant. C. diff, Clostridioides difficile; FCR, FCR001; mos., months; SOC, standard of care recipient. (D) Malignancies over the entire follow-up period. ca, cancer; FCR-R, FCR001 recipient; MDS, myelodysplastic syndrome; PTLD, post-transplant lymphoproliferative disease; SOC-R, standard of care recipient.

FCR-R than in SOC-R patients. FCR-R patients experienced numerically higher rates of solid-tumor, hematologic, and skin cancers over the entire follow-up period compared to the SOC-R group; however, there were no cases of PTLD or MDS in the FCR-R group. As no statistical analysis of these outcomes was performed, these findings merit further research to assess the benefits and risks of FCR001.

There is a myriad of potential benefits of the elimination of IS including improvements in quality of life through a reduction in pill burden and treatment complexity, avoidance of cardiometabolic disease, decreased cost, and reduced risk of infection. Furthermore, even with state-of-the-art immunosuppression, 30% of surviving LDKT patients will lose their allografts by 10 years, resulting in the need for resumption of renal replacement therapy or retransplantation, further increasing the already excessive waiting list for a kidney transplant and increasing the long term cost of care.[9,28,29] Successful tolerance induction in HLA-mismatched patients would expand transplant access in patients without a closely matched donor and attenuate the poorer outcomes associated with greater HLA mismatch seen in standard LDKT.[30,31]

RWE is an important complement to evidence derived from clinical trials. Benefits of RWE include longer follow-up, a diverse population, and reduced cost.[12,23] The NMEDW is particularly valuable for these studies, as it includes data from diverse sources including clinical records, laboratory data, biopsy reports, and linked transplant registry data. These detailed data provide insight into the functional outcome of SOC-R patients not available in studies based exclusively on administrative claims.[22] Among the threats to the validity of RWE data is the risk that clinical trial patients are significantly distinct from non-trial participants. This study utilizes robust PSM algorithms to ensure that patients selected for the SOC study are as comparable to the trial population as possible. The groups had nearly identical donor and recipient characteristics, degree of HLA mismatches, and serologic matching. Furthermore, all patients received care from the same surgical and medical teams.

Several limitations are important to consider within this RWE examination. First, we utilized ICD-9/ICD-10 diagnosis codes to identify clinical complications. To reduce the risk of misclassification, we required two outpatient claims or one inpatient claim prior to inclusion. In addition, we validated this methodology to determine a computed phenotype using adverse events reported in the clinical CRM for the FCR-001 patients. Thus, we were able to confirm that the RWE methodology accurately captured the know AEs in the FCR-R trial patients and could be then applied to the larger population of SOC-R patients. Second, all patients were drawn from only one treatment site, which may limit generalizability. While it is possible that SOC-R outcomes may be different at other transplant centers, a comparison of Northwestern Medicine post-transplant outcomes with national average outcomes suggests no systematic differences.

Third, post-transplant complications beyond death or graft failure, which are captured in the national transplant registry, may be missed for patients treated outside of Northwestern Medicine or for complications that are inaccurately captured using ICD 9/10 codes. This includes viral infections that do not result in hospitalizations such as BK polyomavirus and may be noted only in laboratory results that were not included in the data warehouse. This limitation in RWE identification of complications is more likely to impact SOC patients, who may have received routine follow-up post-transplant care, including laboratory draws, outside of Northwestern Medicine, while most FCR-R care was provided internally as this was a clinical trial. In general, poor data capture of complications would bias the study by suggesting an increased incidence of complications in FCR-R patients and the lack of significant differences suggests that the findings are likely robust. Fifth, statistical tests for differences in rates for selected post-transplant complications were not performed, given concern about multiple comparisons for the same data and the potential for a type 2 error. Finally, kidney function measurements were made using the MDRD4 equation from serum creatinine using the existing equations which incorporated the patient race. This was applied equally in both groups and is unlikely to bias the comparisons as patients' race and ethnicity were well matched.

5 | CONCLUSION

FCR001 therapy allowed more than 70% of LDKT treated in a Phase 2 clinical trial to be safely weaned and withdrawn from chronic IS using a robust RWE approach to ascertain long term follow-up. Comparison of well-matched patients with all FCR001 treated patients (regardless of chimerism status) demonstrated resulted in preserved kidney graft function without an increase in the risk of acute rejection or early graft loss. Despite the need for non-myeloablative conditioning, the overall rate of infectious and malignant complications was similar in both groups, although there were two cases of non-fatal GVHD in the Phase 2 trial. The potential long-term benefits to patients and the healthcare system could be dramatic, with fewer IS-associated adverse events, a lower risk of graft loss resulting in return to dialysis or retransplantation, improvements in patients' quality of life, and lower cost of care. The significant clinical benefits demonstrated in this analysis support the merit of further clinical investigation of FCR001 to ensure that potential risks of combined stem cell and organ transplant are justified by the opportunity for lifelong allograft function without the burden of chronic immunosuppression.

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CONFLICT OF INTEREST STATEMENT

The authors of this manuscript have conflicts of interest to disclose as described by the *American Journal of Transplantation*. N.K. is the Chief Medical Officer of Talaris Therapeutics. D.L. is an employee of Talaris Therapeutics. J.R.L. has received research support from Talaris Therapeutics. M.R. was an employee of Evidera, PPD at the time this research was conducted. D.S. is an employee of Evidera, PPD. S.T.I. is the founder, a Senior Scientific Advisor, a member of the Scientific Advisory Board, and an independent member of the Board of Directors at

Talaris Therapeutics. L.C. reports a consulting relationship with Talaris Therapeutics. D.A.A. reports consulting relationships with Talaris Therapeutics, CareDX, and SpecialistDirect. B.H. has no conflicts of interest to disclose.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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