A PHASE II, OPEN-LABEL SAFETY AND TOLERABILITY STUDY OF A RENAL AUTOLOGOUS CELL THERAPY (REACT) IN PATIENTS WITH TYPE 2 DIABETES AND CHRONIC KIDNEY DISEASE (REGEN-003)

Protocol Number:	REGEN-003
IND Number:	16482
Development Phase:	Phase II
Investigational Therapy Name:	REACT (Renal Autologous Cell Therapy)
Brief Description:	Multi-center, prospective, open-label, single arm study. After kidney biopsy all subjects will receive up to 2 injections of REACT (made from expanded autologous selected renal cells) into the biopsied kidney beginning as soon as REACT can be prepared, given 6 months (+4 weeks) apart.
Version Number:	1.7
Date of Issue:	January 12, 2021
Sponsor:	ProKidney 8020 Arco Corporate Drive, Suite 118 Raleigh, NC 27617 Phone: +1 (919) 294-4521 Email: info@ProKidney.com
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PROTOCOL SIGNATURE PAGE

REGEN-003

Protocol Version: Version 1.7; January 12, 2021

By signing below, I agree to the following:

- ✓ I have received and read Protocol REGEN-003: *"A PHASE II, OPEN-LABEL SAFETY AND TOLERABILITY STUDY OF A RENAL AUTOLOGOUS CELL THERAPY (REACT) IN PATIENTS WITH TYPE 2 DIABETES AND CHRONIC KIDNEY DISEASE (REGEN-003)"*
- ✓ In my formal capacity as Investigator, I understand that my duties include ensuring the safety of all study subjects as well as conducting the study in accordance with all stipulations of the protocol as specified in both the clinical and administrative sections, including all statements regarding confidentiality.
- ✓ I understand that no deviation from, or changes to the Protocol will take place without prior agreement from the Sponsor and documented approval from the Institutional Review Board or Ethics Committee, except where necessary to eliminate an immediate hazard(s) to the study participants.
- ✓ This study will be conducted in compliance with the protocol, in accordance with ICH E6 Harmonized Tripartite Guideline (ICH-GCP), in general agreement with the most recent version of the Declaration of Helsinki, and in accordance with all applicable United States and European regulations.
- ✓ I agree to ensure that all staff members at this site who are involved in the conduct of this study understand their obligations in meeting the above commitments.

_____

Printed Name of Investigator

_____ _____

Signature of Investigator Date

Ashley H. Johns

Printed Name of Sponsor's Representative

Ashley H. Johns 12 January 2021

Signature of Sponsor's Representative Date

PROTOCOL SYNOPSIS

Title: A PHASE II, OPEN-LABEL SAFETY AND TOLERABILITY STUDY OF A RENAL AUTOLOGOUS CELL THERAPY (REACT) IN PATIENTS WITH TYPE 2 DIABETES AND CHRONIC KIDNEY DISEASE (REGEN-003)
Therapeutic Product: REACT is made from expanded autologous selected renal cells (SRC) obtained from each individual subject's kidney biopsy. To manufacture REACT, kidney biopsy tissue from each enrolled subject will be sent to ProKidney, where renal cells will be expanded and SRC selected. SRC will be formulated in a gelatin-based hydrogel at a concentration of 100 x 10^6 cells/mL, packaged in a 10 mL syringe, and shipped to the clinical site for use.
Study Objectives: **Primary Objective:** The primary objective of the study is to assess the safety of REACT injected in one recipient kidney. • Primary Outcome Measures: procedure and/or product related adverse events (AEs) through 24 months post-injection. **Secondary Objective:** The secondary objective of the study is to assess the safety and tolerability of REACT administration by assessing renal-specific AEs over a 24-month period following injection. • Secondary Outcome Measures: renal-specific laboratory assessments through 24 months post-injection. **Exploratory Objective:** Exploratory objectives of the study are designed to assess the impact of REACT on renal function over a 24-month period following injection and on the Quality of Life. • Exploratory Outcome Measures: 1) clinical diagnostic and laboratory assessments of renal structure and function (including eGFR, serum creatinine, and proteinuria) to assess changes in the rate of progression of renal disease, 2) patient-reported outcomes from the Kidney Disease Quality of Life (KDQOL) and EQ-5D-5L surveys obtained at baseline (i.e., after randomization, but before REACT injection) through 24 months after the last REACT injection.
Study Design: Multi-center, prospective, open-label, single-group study. All subjects will be treated with two REACT injections 6 months (+4 weeks) apart after biopsy.
Randomization: Open-label, non-randomized.
Control Group: Each subject will serve as his or her own control; the patient's previous medical history, which must include a minimum 6-month period of observation of renal function, will serve as the control for rate of progression of renal insufficiency.
Sample Size: Up to 10 patients will be treated with REACT. As this is a Phase II safety study, robust statistical analysis will not be performed. Therefore, the sample size proposed for this study is a size typical for in Phase 1 studies, allowing for identification of safety outcomes in a limited population.
Study Population: Male or female patients 30 to 65 years of age with CKD and eGFR between 14 and 20 mL/min/1.73m^2. Patients should have sufficient historical clinical data to determine their individual rate of CKD disease progression.

Inclusion Criteria: Unless otherwise noted, subjects must satisfy each inclusion criterion to participate in the study. Inclusion criteria will be assessed at the Screening Visit, prior to renal biopsy, and before each REACT injection unless otherwise specified.

1. The subject is male or female, 30 to 65 years of age on the date of informed consent.
2. The subject has an established diagnosis of T2DM.
3. The subject has an established diagnosis of diabetic nephropathy as the underlying cause of renal disease.
4. The subject has an established diagnosis of CKD not requiring renal dialysis, defined as having an eGFR between 14 and 20 mL/min/1.73m^2 inclusive at the Screening Visit and prior to REACT injection.
5. The subject has blood pressure less than 150/90 at the Screening Visit, prior to renal biopsy, and prior to REACT injection(s). At the time of the biopsy and injections, the subject's BP should not be significantly below the previously recorded stable pressure.
6. The subject has stable blood pressure and is maintained on a stable anti-hypertensive medication regimen, if treatment for hypertension is necessary. If treatment includes an angiotensin-converting-enzyme inhibitor (ACEI) or an angiotensin receptor blocker (ARB), that treatment must have been initiated at least 8 weeks prior to renal biopsy. Treatment must be stable during the 6-week period immediately prior to REACT injection. Stable treatment is defined as dose adjustment to no less than one half of the current dosage or to no more than 2 times the current dosage. Dose interruptions up to 7 days due to medical necessity are allowed.
7. A minimum of 3 measurements of eGFR or sCr should be obtained at least 3 months apart prior to the Screening Visit or within the previous 24 months to define the rate of progression of CKD. The subject should have adequate, historical clinical data to provide a reasonable estimate of the rate of progression of CKD. The Medical Monitor may be consulted to ensure there is sufficient data.
8. The subject is willing and able to refrain from NSAID consumption (including aspirin) as well as clopidogrel, prasugrel, or other platelet inhibitors during the period beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).
9. The subject is willing and able to refrain from consumption of fish oil and platelet aggregation inhibitors, such as dipryridamole (i.e., Persantine®), during the period beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).
10. The subject is willing and able to cooperate with all aspects of the protocol.
11. The subject is willing and able to provide signed informed consent.

Exclusion Criteria: Subjects who satisfy any exclusion criterion listed below are not eligible to participate in the study. Exclusion criteria will be assessed at the Screening Visit, before renal biopsy, and before each REACT injection unless otherwise noted.

1. The subject has a history of type 1 diabetes mellitus.
2. The subject has a history of renal transplantation.
3. The subject has a serum HbA_{1c} level greater than 10% at the Screening Visit.

4. The subject has uncontrolled diabetes (defined as metabolically unstable by the Investigator).
5. The subject has hemoglobin levels less than 9 g/dL prior to each REACT injection.
6. The subject has abnormal coagulation status as measured by activated partial thromboplastin time (APTT), prothrombin time international normalized ratio (PT INR), and/or platelet count at the Screening Visit.
7. The subject has a bleeding disorder(s) or is taking anticoagulants, such as Coumadin® (warfarin) or direct thrombin inhibitors that, in the judgment of the Investigator, would interfere with the performance of study procedures.
8. The subject has small kidneys (average size less than 9 cm) or has only one kidney, as assessed by ultrasound and/or MRI prior to renal biopsy, unless earlier radiology reports (generated within 1 year of the Screening Visit) are made available to confirm kidney size and number.
9. The subject has a known allergy or contraindication(s), or has experienced severe systemic reaction(s) to kanamycin or structurally similar aminoglycoside antibiotic(s)
10. The subject has a history of anaphylactic or severe systemic reaction(s) or contraindication(s) to human blood products or materials of animal origin (e.g., bovine, porcine).
11. The subject is not a good candidate to undergo percutaneous REACT injection, in the judgment of the surgeon or physician who will perform the procedure. This includes individuals who are morbidly obese (defined as BMI greater than 45 kg/m^2), have excessive fat surrounding the kidney, or who are otherwise at excessive risk for serious complications.
12. The subject has a history of severe systemic reaction(s) or any contraindication to local anesthetics or sedatives.
13. The subject has a clinically significant infection requiring parenteral antibiotics within 6 weeks of REACT injection.
14. The subject has acute kidney injury or has experienced a rapid decline in renal function during the last 3 months prior to REACT injection.
15. The subject has any of the following conditions prior to REACT injection: renal tumors, polycystic kidney disease, anatomic abnormalities that would interfere with the REACT injection procedure or evidence of a urinary tract infection. *Note*: anatomic abnormalities are not exclusionary if the kidney remains accessible and meets the criteria to receive the REACT injection.
16. The subject has incapacitating cardiac and/or pulmonary disorders.
17. The subject has a history of cancer within the past 3 years (excluding non-melanoma skin cancer and carcinoma in situ of the cervix).
18. The subject has clinically significant hepatic disease (ALT or AST greater than 3 times the upper limit of normal) as assessed at the Screening Visit.
19. The subject is positive for active infection with Hepatitis B Virus (HBV), or Hepatitis C Virus (HCV), and/or Human Immunodeficiency Virus (HIV) as assessed at the Screening Visit.
20. The subject has a history of active tuberculosis (TB) requiring treatment within the past 3 years.

21. The subject is immunocompromised or is receiving immunosuppressive agents, including individuals treated for chronic glomerulonephritis within 3 months of REACT injection.
    Note: inhaled corticosteroids and chronic low-dose corticosteroids (less than or equal to 7.5 mg per day) are permitted as are brief pulsed corticosteroids for intermittent symptoms (e.g., asthma).
22. The subject has a life expectancy less than 2 years.
23. The female subject is pregnant, lactating (breast feeding), or planning a pregnancy during the course of the study. Or, the female subject is of child-bearing potential and is not using a highly effective method(s) of birth control, including sexual abstinence. Or, the female subject is unwilling to continue using a highly effective method of birth control throughout the duration of the study.
    Note: A highly effective method of birth control is defined as one that results in a low failure rate (i.e., less than one percent per year) when used consistently and correctly, such as implants, injectables, combined oral contraceptives, some intrauterine devices (IUDs), sexual abstinence, or a vasectomized partner.
24. The subject has a history of active alcohol and/or drug abuse that, in the judgment of the Investigator, would impair the subject's ability to comply with the protocol.
25. The subject's health status would, in the judgment of the Investigator, be jeopardized by participating in the study.
26. The subject has used an investigational product within 3 months prior to REACT injection without receiving written consent from the Medical Monitor.

Number of Sites: Up to 10 clinical centers will be included in the study.

Study Duration: treatment to begin as soon as the REACT product is made available and assuming a one-month interval prior to receiving the first REACT injection, and assuming a 6-month interval before receiving the second injection, plus a 24 month follow up period after the final injection, the duration of treatment would be:

- 31 months for a series of 2 REACT injections

Study Enrollment: up to 10 subjects will be enrolled into the study. Patients who complete screening procedures satisfying all I/E criteria will be enrolled into the study immediately prior to the biopsy. Patients who do not meet all criteria before the biopsy is taken will be considered screen failures. Patients who have a biopsy but are not injected for whatever reason will be discontinued from the study and may be replaced. Once a patient has been injected, the patient will have completed treatment and every effort should be made to ensure the patient completes all follow-up visits.

Investigational Plan:

Screening: Subjects who satisfy eligibility criteria and provide written informed consent may be entered into the study. The subject should have adequate, historical clinical data to provide a reasonable estimate of the rate of progression of CKD (re: Inclusion 7). Screening procedures include a full physical exam, ECG, and laboratory assessments (hematology, clinical chemistry, and urinalysis). An ultrasound will be performed to confirm the presence of two kidneys with normal anatomic features. A MRI study will determine kidney size and volume.

Renal Biopsy: Three days or less before undergoing renal biopsy, enrolled subjects will report to the clinic and undergo an interim physical exam along with an ECG and renal MRI (if not completed during or after the Screening Visit). Laboratory tests, including renal

function, hemoglobin, and a pregnancy test for females will also be performed. Eligible subjects satisfying all inclusions and exclusion criteria will be admitted to the hospital / clinical research center to undergo a kidney biopsy. A minimum of 2 tissue cores measuring at 1.5cm a piece must be collected using a 16-gauge biopsy needle to provide sufficient material for the manufacture of REACT. Subjects who do not experience complications from the biopsy may be discharged the same day consistent with site standard practice. Each individual subject's kidney biopsy tissue will be sent to ProKidney.

REACT Injection: Ten to 14 days before the scheduled injection date, subjects will undergo an interim physical exam for ongoing verification of inclusion and exclusion criteria. Subjects also will undergo renal scintigraphy (i.e., split kidney function scan) to find out what percentage each kidney contributes to total baseline kidney function. On the day of the scheduled REACT injection, eligible subjects will be admitted into the hospital / clinical research unit. After warming to liquefy the hydrogel, REACT will be injected into the same kidney that was previously biopsied using a percutaneous approach. This procedure will follow a standardized technique, such as that used in the ablation of renal masses by radiofrequency or cryogenic methods. Subjects without complications may be discharged the same day consistent with site standard practice. An ultrasound will be performed the day after injection to detect possible, subclinical AEs. It is anticipated that all subjects will receive 2 REACT injections given 6 months (+4 weeks) apart. The first and second injections will occur in the same kidney in which the biopsy was taken. Therefore, only one kidney will be used for the duration of this study.

Follow-up: Subjects will complete follow-up evaluations on Days 1, 7, 14, 28 (± 3 days) and Months 2, 3, 4 and 5 (± 7 days) after the first REACT injection, and on Days 1, 7, 14, 28 (± 3 days) and Months 2 and 3 (± 7 days) after the second REACT injection. Depending on when the second injection is administered (i.e., at 6 months [+4 weeks]), subjects may undergo evaluations at 6 months after the first REACT injection. Following the final REACT injection, subjects will complete long-term, follow-up assessments of safety and efficacy through 6, 9, 12, 15-, 18-, 21-, and 24-months post-treatment.

Safety Monitoring: Hemorrhage following REACT injection is a known and foreseeable risk to subjects participating in this study. Therefore, hemoglobin will be measured by the site's local laboratory at the following times: a) before, b) after procedure per site standard practice.

Investigational Product, Dosage and Mode of Administration:

Investigational Product: REACT is made from expanded autologous selected renal cells obtained from each individual subject's kidney biopsy. To manufacture REACT, biopsy tissue from each enrolled subject will be sent to ProKidney, in whose facilities renal cells will be expanded and SRC selected. SRC will be formulated in a gelatin-based hydrogel at a concentration of 100 x 10^6 cells/mL, packaged in a 10 mL syringe, and shipped to the clinical site.

Dosage: The volume of REACT to be administered will be determined by pre-procedure MRI volumetric 3D evaluation or ellipsoid formula (Length x width AP plane x width Transverse plan x .62). Based on pre-clinical data, the dose of REACT will be 3 x 10^6 cells/g estimated kidney weight (g KW^{est}). Since the concentration of SRC per mL of REACT is 100 x 10^6 cells/mL, the dosing volume will be 3.0 mL for each 100 g of kidney weight. Using this dosing paradigm, the following table shows the dosing volume and number of SRC to be delivered relative to estimated kidney weight. The maximum volume of REACT injected into the biopsied kidney will be 8.0 mL.

Estimated Kidney Weight (g KW^{est})*		Dosing Volume (mL)	SRC Delivered (Number of Cells x 10^6)
Median Weight (g)	Weight Range (g)		
100	95 – 108	3.0	300
117	109 – 125	3.5	350
133	126 – 141	4.0	400
150	142 – 158	4.5	450
167	159 – 175	5.0	500
183	176 – 191	5.5	550
200	192 – 208	6.0	600
217	209 – 225	6.5	650
233	226 – 241	7.0	700
250	242 – 258	7.5	750
— — —	>259	8.0	800

*Kidney weight will be estimated from the results of an MRI study performed on or after the Screening Visit until Day 0 (renal biopsy).

Investigational Product, Dosage and Mode of Administration: Dosage *(continued)*

It is anticipated that all subjects will receive two planned REACT injections to allow dose-finding and evaluate the duration of effects. The first and second injections will occur in the same kidney in which the biopsy was taken. In some cases, a subject or the Investigator may decide to postpone or withhold the second REACT injection. For example, if there appears to be any untoward safety risk, or rapid deterioration of renal function, or the development of uncontrolled diabetes or uncontrolled hypertension, or the development of a malignancy or an intercurrent infection, then the second REACT injection should not be administered.

Mode of Administration: REACT will be injected into the biopsied kidney using a percutaneous approach. The percutaneous method will employ a standardized technique (such as that utilized in the ablation of renal masses by radiofrequency or cryogenic methods).

Data Safety Monitoring Board: A Data Safety Monitoring Board (DSMB) will be chartered to oversee subject safety, especially as it relates to unexpected investigational product-related events. The DSMB will minimally consist of 3 members who have expertise directly related to protocol-specified activities. It will function independently, and its members will have no other engagement with ProKidney. The DSMB will meet by teleconference at regular intervals, depending on the speed of subject enrollment and the amount of new data generated. The DSMB will advise ProKidney on aspects concerning the safety of subjects participating in the clinical trial. Apart from reviewing study data, the DSMB will consider feedback from the Sponsor and Investigators. The DSMB will share its recommendations with the study centers, Institutional Review Boards/Ethics Committees, and regulatory authorities, as appropriate. Other specific activities and responsibilities of the DSMB will be detailed in the DSMB charter.

Statistical Analysis Methods: Statistical analyses will be primarily descriptive in nature and no statistical hypothesis testing is planned for the study. Unless otherwise specified, continuous variables will be summarized by presenting the number of non-missing observations (n), mean, standard deviation, median, minimum, and maximum. Categorical variables will be summarized by presenting frequency count and percentage for each category.

Table 1: Time and Events Table

Clinical Assessment:	Screening Visit	Renal Biopsy		Optional*	Preparation and Shipment of REACT Product	First REACT Injection			Follow-up First REACT Injection				Optional[b]	Interval Between REACT Injections = 6 Months (+) 4 Weeks**	Last REACT[c] Injection			Follow-up Last REACT Injection				Follow-Up Long-Term	
	Day – 60 to -3[a]	Day -3 to -1	Day 0 Biopsy	Day 1 Follow-up		Day –14 to – 10	Day 0 REACT	Day 1 Follow-up	Day 7 (±) 3 days	Day 14 (±) 3 days	Day 28 (±) 3 day	Months 2, 3, 4, and 5 (±) 7 days	Month 6 (±) 7 days		Day** –14 to – 10	Day 0 REACT	Day 1 Follow-up	Day 7 (±) 3 days	Day 14 (±) 3 days	Day 28 (±) 3 days	Month 2 Month 3 (±) 7 days	Months 6, 9, 12,15,18, 21 (±) 7 days	EOS[d] Month 24 (±) 7 days
Obtain Informed Consent[e]	X																						
Verify I/E Criteria	X	X	X			X	X								X	X							
Obtain Demographic Data	X																						
Obtain Medical History	X	X																					
Record ConMeds	X	X	X	X		X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X
Perform Comprehensive PE[f]	X																						
Perform Interim PE[f]		X				X					X				X					X		X	X
Measure Vital Signs[g]	X	X	X	X		X	X[h]	X	X	X	X	X	X		X	X[h]	X	X	X	X	X	X	X
Conduct Laboratory Tests[i]	X	X	X	X		X	X	X		X	X	X	X		X	X	X		X	X	X	X	X
Perform 12-lead ECG	X	X				X					X				X					X		X	X
Perform Ultrasound	X[j]		X[k]	X[k]		X[j]	X[k]	X[k]							X[j]	X[k]	X[k]						X[j]
Perform MRI Study	→→→	X[l]																					X
Perform Renal Scintigraphy						X[m]									X[m]							X[m]	X[m]
Admit to Hospital			X[p]				X[p]									X[p]							
Perform Kidney Biopsy			X																				
Monitor /Record AEs			X	X		X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X
Inject Autologous REACT							X[n]									X[n]							
CT Scan[o]							X									X							
Discharge			→→→X[p]				→→→X[p]									→→→X[p]							
Funduscopic Exam of Retinae	X																					X[q]	X
Administer KDQOL Survey	X						X				X	X	X			X				X	X	X	X
Administer EQ-5D-5L Survey	X						X				X	X	X			X				X	X	X	X

Abbreviations: AE (adverse event); ConMeds (concomitant medications); DSMB (Data Safety and Monitoring Board); ECG (electrocardiogram); EOS (End-of-Study Visit); I /E (inclusion /exclusion); KDQOL (Kidney Disease Quality of Life Survey); MRI (magnetic resonance imaging); REACT (Renal Autologous Cell Therapy); PE (physical examination).

Notes:

* Day 1follow-up visit should only be conducted if there is a safety event identified during or after the kidney biopsy.

** Every attempt should be made to ensure the second REACT injection is administered 6 months (+4 weeks) after the first injection. In the event that the subject cannot schedule his/her second REACT injection 6 months after receiving the first injection or cannot keep his/her scheduled 6-month visit, ProKidney and the Medical Monitor must be notified immediately. The expectation is that the subject and the clinical center will accommodate the Sponsor's preference that the series of 2 REACT injections should be administered no more than 6 months apart.

a. If the screening assessment falls outside of the 60-day window before renal biopsy, re-screening will be performed as described in Screening (Section 6.1).
b. Because the second REACT injection will occur 6 months (+4 weeks) after the first injection, the 6-month visit may not be scheduled.
c. In the event that a second REACT injection will not be administered, the subject will undergo all follow-up assessments after the last REACT injection at the 24-month EOS Visit.
d. The EOS Visit will take place 24 months after the last REACT injection, or when the subject is terminated from the study by the Investigator (Section 8.6) or when the subject voluntarily discontinues from the study (Section 5.4).
e. The Informed Consent Form must be signed and dated prior to conducting any study-specific procedures, including those at the Screening Visit. Signing the Informed Consent Form starts the 60-day timeline for screening.
f. The PE and interim PE are described in Section 7.2.2.
g. Vital signs include heart rate, resting blood pressure, respiration rate, and body temperature. (Section 7.2.1). At the Pre-Biopsy Visit (Day -3 to Day -1) and the Pre-Injection Visit (Day -14 to Day -10) three BP measurements will be taken and the average will be used to satisfy entry criteria.
h. Vital signs (included are heart rate, blood pressure, and respiration rate) will be measured throughout the procedure. Temperature is not required to be recorded throughout the procedure.
i. Refer to Table 2 for a schedule of laboratory assessments.
j. Ultrasound will be performed at the Screening Visit to verify subject eligibility and to obtain baseline echogenicity reading. Subsequent Ultrasounds will monitor echogenicity.
k. Ultrasound will be performed following the in-patient renal biopsy on Day 0 and Day 1, and following the in-patient REACT injection(s) on Day 0 and Day 1 with the aim of monitoring possible, subclinical AEs.
l. A MRI study without contrast will be performed at the Screening Visit through Day -1 before renal biopsy to determine kidney size and volume.
m. Renal scintigraphy will be performed before the first REACT injection, before the last REACT injection, at the 6-Month Visit after the last REACT injection, at the 12-Month Visit after the last REACT injection, and at the month 24/EOS Visit.
n. The REACT preparation will be handled and injected according to procedures described in the Study Reference Manual.
o. CT Scan must be used in conjunction with ultrasound for the REACT injection procedure.
p. Subjects may be admitted to hospital per site standard practice. Subjects who do not experience complications may be discharged the same day consistent with site standard practice.
q. Subjects will undergo a funduscopic exam with retinal photography of both retinae to monitor diabetic retinopathy at screening, month 12, and month 24/EOS visit.

Table 2: Laboratory Time and Events Table

Laboratory Evaluation:	Screening Visit	Renal Biopsy		Optional*	Preparation and Shipment of REACT Product	First REACT Injection			Follow-up First REACT Injection			Optional[b]	Interval Between REACT Injections = 6 Months (+) 4 Weeks**	Last REAC T[c] Injection			Follow-up Last REACT Injection			Follow-up Long-Term	
	Day -60 to -3[a]	Day -3 to -1	Day 0 Biopsy	Day 1 Follow-up		Day -14 to -10	Day 0 REACT Injection	Day 1 Follow-up	Day 14 (±) 3 days	Day 28 (±) 3 days	Months 2, 3, 4, and 5 (±) 7 days	Month 6 (±) 7 days		Day** -14 to -10	Day 0 REACT Injection	Day 1 Follow-up	Day 14 (±) 3 days	Day 28 (±) 3 days	Month 2 Month 3 (±) 7 days	Months 6, 9, 12, 15, 18, 21 (±) 7 days	EOS[d] Month 24 (±) 7 days
Clinical Chemistry																					
Standard panel	X	X		X		X	X	X	X	X	X	X		X	X	X	X	X	X	X	X
Renal analytes	X	X		X		X	X	X	X	X	X	X		X	X	X	X	X	X	X	X
Lipid panel	X					X								X							X
Pregnancy test[e]	X	X				X								X							X
FSH test[f]	X																				
Serology																					
HIV, HBV, HCV	X																				
Hematology																					
Standard cell counts/indices	X	X		X		X		X	X	X	X	X		X		X	X	X	X	X	X
Hemoglobin, Hematocrit[g]			X[h]				X[h]								X[h]						
Coagulation Status																					
Platelet count	X	X				X								X							
APTT	X	X				X								X							
PT-INR	X	X				X								X							
Urine Collection																					
Standard panel (Micro and Macro)	X	X		X		X	X	X	X	X	X	X		X	X	X	X	X	X	X	X
Urine Chemistry		X		X		X	X	X		X				X	X	X	X	X			
24-hour	X										X	X							X	X	X
Test stick (Spot Urine)		X[i]	X[i]	X[i]		X[i]	X[i]	X[i]				X		X[i]	X[i]	X[i]					
Additional Tests																					
HbA_{1c}	X																			X	X
Drugs of abuse	X					X								X							
iPTH	X	X				X				X	X	X		X				X	X	X	X
ß2-microglobulin[j]	X	X				X				X		X		X				X		X	X
NGAL	X	X				X				X	X	X		X				X		X	X
Research (reserve) samples[k]	X	X				X				X	X	X		X				X	X	X	X

Abbreviations: APTT (Activated Partial Thromboplastin Time); FSH (Follicle Stimulating Hormone; HbA_{1c} (glycosylated hemoglobin); HIV (Human Immunodeficiency Virus); HBV (Hepatitis B Virus); HCV (Hepatitis C Virus); NGAL (Neutrophil Gelatinase-Associated Lipocalin); REACT (Renal Autologous Cell Therapy); i PTH (Parathyroid Hormone, intact); PT-INR (Prothrombin Time-International Normalized Ratio)

Notes:

* Day 1 follow-up visit should only be conducted if there is a safety event identified during or after the kidney biopsy.

** Every attempt should be made to ensure the second REACT injection is administered 6 months (+ 4 weeks) after the first injection. In the event that the subject cannot schedule his/her second REACT injection 6 months after receiving the first injection, or cannot keep his/her scheduled 6-month visit, ProKidney and the Medical Monitor must be notified immediately. The expectation is that the subject and the clinical center will accommodate the Sponsor's preference that the series of 2 REACT injections should be administered no more than 6 months apart.

a. If the screening assessment falls outside of the 60-day window before renal biopsy, re-screening will be performed as described in Screening (Section 6.1).
b. Because the second REACT injection will occur 6 months (+4 weeks) after the first injection, the 6-month visit may not be scheduled.
c. In the event that a second REACT injection will not be administered, the subject will undergo all follow-up assessments after the last REACT injection at the 24-month EOS Visit.
d. The EOS Visit will take place 24 months after the last REACT injection, or when the subject is terminated from the study by the Investigator (Section 8.6), or when the subject voluntarily discontinues from the study (Section 5.4).
e. The clinic will perform a urine dip-strip pregnancy test. If positive, then a confirmatory test will be performed by the central laboratory.
f. Post-menopausal women with a confirmatory FSH test do not have to undergo pregnancy testing throughout the study.
g. Within 48 hours before Days 0 for renal biopsy and REACT injection(s), hemoglobin levels will be verified as > 9 g/dL per site standard practices.
h. On Days 0 for renal biopsy and REACT treatment (s), hemoglobin and hematocrit will be measured before and after procedure per site standard practice at the local lab. These samples will be processed by the site's local laboratory to accelerate notification of results and subsequent decisions affecting clinical care. Additionally, blood samples for hemoglobin and hematocrit after procedure will be sent to the central laboratory where results can be entered into the study database.
i. Prior to, day of, and day after biopsy and REACT injection(s), microscopic urinalysis will be performed using a dip (test) stick
j. β2-microglobulin will be assessed in both serum and urine samples.
k. Research samples (serum/plasma and urine) will be collected, frozen, and stored for the evaluation of novel biomarkers.

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Table 3: Abbreviations and Specialist Terms

Abbreviation or Specialist Term	Explanation
ACEI	Angiotensin-Converting-Enzyme Inhibitor
AE	Adverse Event
ALT	Alanine Transaminase
APTT	Activated Partial Thromboplastin Time
ARB	Angiotensin Receptor Blocker
AST	Aspartate Transaminase
BMI	Body Mass Index
BP	Blood Pressure
BUN	Blood Urea Nitrogen
CKD	Chronic Kidney Disease
ConMed(s)	Concomitant Medication(s)
CRF	Case Report Form
CRP	C-Reactive Protein
CTCAE	Common Terminology Criteria for Adverse Events
DMSA	Dimercaptosuccinic acid
DSMB	Data Safety Monitoring Board
ECG	Electrocardiogram
EC	Ethics Committee
eGFR	Estimated Glomerular Filtration Rate
EOS	End-of-Study
ESRD	End Stage Renal Disease
FDA	Food And Drug Administration
g	Gram(s)
GCP	Good Clinical Practice
GFR	Glomerular Filtration Rate
$g\ KW^{est}$	Gram(s) of Estimated Kidney Weight
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
Hb	Hemoglobin
HbA_{1c}	Glycosylated Hemoglobin
HBV	Hepatitis B Virus
HCV	Hepatitis C Virus
HIV	Human Immunodeficiency Virus
HR-QoL	Health-Related Quality of Life
IB	Investigator's Brochure
ICH	International Conference On Harmonization

Abbreviation or Specialist Term	Explanation
I /E	Inclusion /Exclusion
IND	Investigational New Drug
INR	International Normalization Ratio
iPTH	Intact Parathyroid Hormone
KDQOL	Kidney Disease Quality-of-Life Survey
MedDRA	Medical Dictionary for Regulatory Activities
mo	Month
MRI	Magnetic Resonance Imaging
NCI	National Cancer Institute
NOAEL	No-Observed-Adverse-Effect-Level
NSAIDs	Non-Steroidal Anti-Inflammatory Drugs
Nx	Nephrectomy
PE	Physical Examination
PI	Principal Investigator
PT	Prothrombin Time
PT-INR	Prothrombin Time-International Normalization Ratio
QA	Quality Assurance
RBC	Red Blood Cell
REACT	Renal Autologous Cell Therapy
SAE /SAR	Serious Adverse Event /Serious Adverse Reaction
sCr	Serum Creatinine
SPIO	Superparamagnetic Iron Oxide
SRC	Selected Renal Cells
SUSAR	Suspect, Unexpected Serious Adverse Reaction
T2DM	Type 2 Diabetes Mellitus
TB	Tuberculosis
UACR	Urinary Albumin /Creatinine Ratio
WBC	White Blood Cell

1. INTRODUCTION AND BACKGROUND

1.1. Chronic Kidney Disease

1.1.1. Background

Chronic Kidney Disease (CKD) is characterized by progressive nephropathy that without therapeutic intervention will worsen until the patient reaches end stage renal disease (ESRD). It is defined as reduced kidney function, demonstrated by decreased estimated glomerular filtration (eGFR) or evidence of kidney damage, such as increased excretion of urinary albumin. Global prevalence of CKD is estimated at 8-16%.[1] CKD is associated with considerable morbidity, such as diabetes mellitus,[2] and is often accompanied by adverse outcomes owing to underlying disease states or and/or risk factors such as hypertension and renovascular disease.[3] Ninety-seven percent of patients with moderate to severe CKD have mostly asymptomatic Stage 3 disease, but even this stage of CKD bears a two- to four-fold rise in cardiovascular disease risk along with a significant increase in all-cause mortality.[4, 5] Only a small proportion of patients progress to ESRD (i.e., Stage 5 disease); even with costly treatments, however, patients with ESRD experience substantial morbidity and mortality.[4] To survive, ESRD patients require renal replacement therapy (dialysis or kidney transplantation). Preventing or delaying adverse outcomes of CKD via early intervention is the primary strategy in CKD management.[6] Nevertheless, early treatments have been less than optimal, resulting in a significant unmet medical need for improved interventional strategies to manage CKD and delay progression to ESRD.

1.1.2. Progressive Staging of CKD

The major causes of CKD are diabetes and hypertension. Nearly half of all CKD cases arise from diabetes with or without hypertension.[2,5] The incidence of CKD continues to increase, primarily due to the increased incidence of Type 2 Diabetes Mellitus (T2DM).[2]

Staging and grading of kidney function is most often quantified by the estimated glomerular filtration rate (GFR), which is defined as "the volume of plasma from which a given substance is completely cleared by glomerular filtration per unit time".[7] Creatinine clearance is the principal endogenous marker that is used to measure GFR. In February 2002, with the aim of providing a uniform definition of CKD, the Kidney Disease Outcomes Quality Initiative of the US Kidney Foundation defined CKD and the various stages of CKD[8] (Table 4). For example, the threshold for CKD is 60 mL/min/1.73m^2, which is less than one-half of the GFR for a normal, young adult male (120-130 mL/min/1.73m^2).[9] Additionally, there is a heightened risk for CKD complications when the GFR is 60 mL/min/1.73m^2 and lower.

Table 4 defines CKD stages according to GFR measurements, and the relative prevalence and clinical action taken at each stage.[13] The initial stage of diabetic nephropathy (Stage 1) occurs over a period of several years and is characterized by microalbuminuria (30-300 mg/24 hr.) followed by macroalbuminuria (> 300 mg/24 hr.). As the ability of the kidney to filter blood waste products declines, serum creatinine rises. With increasing kidney damage (Stages 2-4), rising blood pressure further exacerbates kidney disease. When the kidneys cease to function entirely (Stage 5 [ESRD]), renal replacement therapy (dialysis or transplantation) is required.

Table 4: Summary of Classification and Prevalence Estimates for CKD

Stage*	Description	GFR (mL/min/1.73m^2)
1	Kidney damage with normal or increased GFR	≥90
2	Kidney damage with mild decrease in GFR	60-89
3	Moderate decrease in GFR	30-59
4	Severe decrease in GFR	15-29
5	Kidney failure	<15 (or dialysis)

* Source: National Kidney Foundation, 2002. National Kidney Foundation. 2002. KDOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. *Am J Kidney Dis.* 39:S1-266.

All-cause mortality rates were shown to increase as GFR declined; mortality rates were highest at Stages 4-5 of CKD.[10,11,12] Populations defined as having an eGFR <60 mL/min/1.73m^2 consistently exhibited a higher mortality rate than comparator groups where there was no evidence of CKD.

1.1.3. Standard-of-Care in CKD

Treatment of patients with CKD is focused on slowing progression and preparing for kidney failure /replacement. For many patients, CKD occurs as part of a complex comorbidity cluster, especially with cardiovascular disease and T2DM.

Increased risk of cardiovascular disease can be a complication of CKD or an independent comorbidity associated with T2DM.[14] Collectively, the aim is to lower cardiovascular risk and prevent or slow the progression of kidney failure via administration of: 1) angiotensin converting enzyme inhibitors (ACEI) and/or angiotensin receptor blockers (ARB) to decrease proteinuria and control hypertension; 2) insulin and anti-diabetic agents for glycemic control (e.g., reduced serum hemoglobin A1c); and, 3) statin therapy to counter dyslipidemia.

When a patient reaches ESRD, renal replacement therapy (i.e., dialysis or transplantation) is indicated. The vast majority of Stage 5 patients receive hemodialysis.[15] Dialysis replaces about 5-15% of kidney function, depending on the intensity and frequency of use; dialysis also helps to restore fluid and electrolyte balance when kidneys fail. However, the life-expectancy of an ESRD patient initiating hemodialysis is only 4-5 years.[16] Additionally, hemodialysis has been associated with multiple, serious complications as well as interference with quality of life, such as the need to undergo dialysis up to three times per week. Although kidney transplantation remains the most effective form of therapy at this time; there is a chronic shortage of organs. If a patient is able to secure a kidney for transplantation, long-term immunosuppressive therapy is required to prevent rejection. Use of these regimens results in a higher incidence of infection and, over the long term, some types of cancer.[17] Taken together, there is a critical medical need for improved therapies for CKD which could dramatically slow the progression of disease and significantly delay the need for renal transplantation.

1.2. Scientific Rationale

ProKidney is developing Renal Autologous Cell Therapy (REACT) based on extensive scientific evaluation of its biologically active component known as SRC (i.e., homologous, autologous selected renal cells). This cell population is naturally involved in renal repair and regeneration.[18,19,20,21,22,23,24,25,26,27,28,29,30,31,32] Therapeutic intervention with REACT is intended to

improve renal function in subjects with CKD and T2DM and delay the need for renal dialysis or transplantation which, based on the current standard-of-care, is inevitable for ESRD.

1.3. Non-clinical Pharmacology Studies

In a series of pre-clinical studies, Tengion (a former regenerative medicine company) defined the pharmacological characteristics of SRC and delayed the progression of experimental models of CKD by augmenting renal structure and function.[18,19,33,34,35,36] Tengion subsequently conducted safety pharmacology and GLP toxicology studies. An overview of these non-clinical studies is presented in Table 5 (Refer to the Investigator's Brochure for more detailed information).

Table 5: Summary of Non-clinical Pharmacology and Toxicology Studies

Model	Number Animals	Study Length	Dose per Kidney Weight[a] 10^6Cell/g	Number Kidneys Injected	Total Kidney Weight (g) Implanted[a]	Total[b] Dose (mL)	Total SRC x 10^6	SRC Conc. 10^6/mL	Study
Pharmacology (efficacy, kinetics, migration, and persistence)									
5/6 Nephrectomy Lewis Rat[d]	*3*	*6 mo*	*5 - 10*	*1 (1)*	*1*	*0.1*	*5-10*	*50-100*	***1***
70% Nephrectomy Canine[d]	*4*	*10 mo*	*6*	*1 (2)*	*57.7*	*5*	*334*	*66.8*	***2***
ZSF-1[e]	*7*	*12 mo*	*3*	*2 (2)*	*3.2*	*0.4*	*10*	*25*	***3***
5/6 Nephrectomy Lewis Rat[e]	*77*	*4 days*	*5-15*	*1 (1)*	*1*	*0.1*	*5-15*	*50-150*	***4***
Canine[e]	*1*	*30 min*	*12.5*	*2 (2)*	*120*	*10*	*1500*	*150*	***5***
Canine[e]	*4*	*30 min*	*1.5-9.2*	*2 (1-2)*	*120*	*2.5-10*	*92.7-553.5*	*37-55*	***6***
GLP Toxicology									
ZSF-1 rat[d]	*5M/5F* *5M/5F* *5M/5F* *5M/5F*	*3 mo* *6 mo* *3 mo* *6 mo*	*3.13* *3.13* *6.25* *6.25*	*2 (2)* *2 (2)* *2 (2)* *2 (2)*	*2* *2* *2* *2*	*.25* *.25* *.25* *.25*	*6.25* *6.25* *12.5* *12.5*	*25* *25* *50* *50*	***7***
Canine[e]	*2M/2F* *2M/2F* *2M/2F* *2M/2F*	*1 mo* *3 mo* *1 mo* *3 mo*	*2.75* *2.75* *11.0* *11.0*	*1 (1)* *1 (1)* *2 (1)* *2 (1)*	*60* *60* *120* *120*	*3* *3* *12* *12*	*330* *330* *1320* *1320*	*110* *110* *110* *110*	***8***
Canine[c,e] *0 Months*	*2M/2F*	*n/a*	*5.5*	*2 (2)*	*120*	*6*	*660*	*110*	***9***
3 Months		*6 mo*	*5.5*	*2 (2)*	*120*	*6*	*660*	*110*	***10***

Notes:
a. Estimated kidney weight based on animal model. Actual weights are listed in the study reports where applicable.
b. Dose refers to SRC or REACT.
c. Two doses of REACT were administered, one at 0 months and the second at 3 months.
d. Delivered REACT to rodents using syringe affixed to sharp needle that pierced the capsule and deposited REACT.
e. Delivered REACT to dogs using cannula to pierce the capsule plus blunt-end delivery cannula to deposit REACT.

1.3.1. Pharmacodynamics

Proof of principle for SRC as the biologically active component of REACT was established in multiple animal models of CKD. For example, the 5/6th nephrectomy (Nx) rodent mass reduction model of CKD allowed for an optimized selection of a therapeutically relevant SRC cell population. A 70% Nx canine model of CKD confirmed SRC activity in a large mammal, while the ZSF-1 rat

served as proof-of-principle for demonstrating the effects of SRC in a model relevant to T2DM.[37] SRC delivered directly into the kidney cortex in multiple experimental models of CKD induced a regenerative response through direct engraftment or tissue replacement, and also induced secretory factors via a putative paracrine mechanism.[18,21,22,24, 26]

This intervention strategy significantly improved survival, stabilized disease progression, and extended the longevity in both the 5/6th Nx model and the ZSF-1 rodent models of CKD. Morphological normalization of multiple nephron structures was accompanied by functional improvements, including glomerular filtration, tubular protein handling, electrolyte balance, and the ability to concentrate urine. Lowered blood pressure and reduced levels of circulating renin were also observed in the ZSF-1 rat model. The observed functional improvements following SRC treatment were accompanied by significant reductions in glomerular sclerosis, tubular degeneration and interstitial inflammation and fibrosis. No toxicologically significant in-life, clinical pathology, or histological changes were noted in the target organ or other tissues. Based on results from multiple pre-clinical studies conducted in different CKD animal models, SRC (i.e., active component of REACT) were effective in significantly delaying progression of CKD when implanted in the diseased organ prior to irreversible nephropathy. These results provide a rationale for investigating the effects of this cell-based intervention in patients prior to ESRD.

1.3.2. Safety Pharmacology

1.3.2.1. Extra-Renal Activity

REACT (i.e., SRC formulated in a gelatin-based hydrogel) was administered in various rat and canine models to assess immediate cardiovascular and respiratory pharmacologic effects. The acute effects of lower and higher SRC concentrations formulated in varying percentages of gelatin (0.75-1.0%) were evaluated in the rodent 5/6th Nx model. Potential changes in blood pressure were assessed immediately before, during, and shortly after REACT delivery in the normal canine model. No studies on the effects of REACT on the central nervous system were performed since: 1) animals exhibited normal behavior before, during, and after REACT injection; 2) no effects on the central nervous system were expected from an investigational product containing intact renal cells; and, 3) REACT was delivered into the kidney.

1.3.2.2. Hemodynamic Effects

Rats in the 5/6th Nx study (Study #4) received REACT or vehicle control, and potential hemodynamic effects were monitored over 4 days. Among the 77 animals treated in this REACT formulation study, 16 animals experienced apnea during or immediately after REACT delivery. A total of 9 animals died; the causes of death were classified as apnea (n=3), renal hemorrhage (n=2), and deaths associated with CKD (n=4). Six of the 16 animals that experienced apnea were not pre-treated with atropine; of these animals that experienced apnea, two died under the influence of anesthesia prior to the use of atropine. Ten of the 16 animals that experienced apnea were treated with atropine, and all recovered from the surgical procedure and REACT injection.

On the other hand, apnea, renal hemorrhage, and deaths that occurred in the 5/6th Nx rat study were not observed in the ZSF-1 rat study, or in the canine pharmacology study, or in two (intact) canine pilot studies that assessed the short-term effects of volume administration on blood pressure. Taken together, this model-specific hemodynamic response can be potentially attributed to: 1) altered hemodynamics of the severely mass-reduced rodent remnant kidney [38] 2) transient changes in kidney interstitial pressure administration triggering a central autonomic response;[39 40] and, 3) under perfusion of tissue or acute hypoxia from bleeding following delivery into the kidney. Pre-treatment

with atropine, a competitive antagonist of the parasympathetic nervous system, helped mitigate the model-specific hemodynamic changes. The effect of atropine suggests a possible autonomic response to REACT delivery that was specific to the severely mass-reduced, 5/6th Nx rodent model of CKD.

1.3.2.3. Dose Volume

Using a range of REACT doses, volumes, and concentrations, (Study #5), the normal canine was selected to evaluate blood pressure immediately before, during, and after REACT delivery into the kidney. In this study, each pole of each kidney received 2.5 mL of REACT; therefore, a total of 10 mL/120 g, or 0.083 mL/g, was delivered at a dosage of 12.5 x 10^6 cells /gram of kidney mass. REACT treatment was well tolerated; there were no adverse systemic effects (physical or serological), nor were there any significant toxicological or histomorphological changes indicative of kidney injury or other tissue injury as a result of REACT delivery. In contrast to the severe mass reduction rodent model of CKD, no apnea, renal hemorrhage, or deaths were noted.

1.3.3. Kinetics, Migration, and Persistence

As with other cell-based therapies targeting soft organs, data on biodistribution of the investigational product has been limited. To that end, three additional studies provide evidence concerning the potential migration and persistence of REACT within the kidney at selected sampling times post-delivery.[24,25,26] The results of these studies are summarized in this section; detailed information is provided in the Investigator's Brochure.

1.3.3.1. ZSF-1 Rat

SRCs were labeled with the Rhodamine-B superparamagnetic iron oxide (SPIO) particle. This contrast agent is specifically formulated for cell labeling and is readily internalized by non-phagocytic cells. SPIO-labeled cells were administered to the ZSF-1 rat kidney. Twenty-four hours after delivery, SPIO-labeled cells were detected by MRI and whole organ optical imaging. In addition, ZSF-1 rats received SRCs labeled with CelSense-19F, which were quantified by Nuclear Magnetic Resonance at 3 hr., 24 hr., and 7 days after implantation.[24,25]

Both acute ZSF-1 detection and long-term donor cell detection using the 5/6th Nx model of CKD showed significant retention of SPIO-labeled cells. Clinically relevant MRI detection at 24 hr. following cell delivery revealed a region at the anterior pole of the kidney where SPIO-labeled cells had been injected. Sectioning of the whole kidney and staining with Prussian blue demonstrated a bolus of iron labeled SPIO cells migrating and distributing from the cortical injection site, which confirmed their presence in tubular and peritubular spaces of the renal cortex and medulla.

Likewise, whole organ fluorescent imaging highlighted cell detection at and around the site of injection located at the upper cortex of the anterior pole of the ZSF-1 rat kidney. Detection of 19F-labeled SRC at 3 and 24 hr. after delivery confirmed nearly 100% retention in the kidney. After 7 days, detection of 19F-labeled SRC was diminished by an order of magnitude, consistent with continuous urinary excretion.

1.3.3.2. Porcine Non-GLP Analysis of Non-Renal Tissue

In a pre-clinical, non-GLP study, SPIO-labeled SRC were delivered to the kidneys of living swine (n=11). Cellular distribution was monitored over the course of the 30-day study period using MRI. Labelled SRC were distributed in two major compartments: urinary bladder and renal parenchyma.

The major route of excretion was urine. Notably, there was no evidence of ectopic SRC migration or site-specific engraftment at non-target organ sites.[41]

1.3.3.3. Canine Non-GLP Analysis of Non-Renal Tissue

In a pre-clinical, non-GLP study, SPIO-labeled SRC were delivered to the kidneys of living canine hosts. Cellular distribution was monitored at 30 minutes post-injection via MRI. Consistent with observations from the living porcine model demonstrating that injected SPIO-labeled SRC was retained in renal parenchyma or excreted in urine, SPIO-labeled SRC likewise were retained within the renal parenchyma at the injection site after 30 minutes.

1.3.3.4. Conclusions

- SRC are distributed at the site of injection (renal parenchyma) and excreted via the urine, based on SRC labeling studies with SPIO and CelSense-19F.
- SRC delivered into rat, swine, and canine kidneys were not detected in non-target organs (other than urinary tract during excretion), based on extensive histological evaluation.
- Based on reports concerning allogenic mesenchymal stem cells as well as published data concerning the safety of autologous mesenchymal stem cells in clinical trials, autologous REACT-related materials should also not give rise to ectopic tissue growth, organ dysfunction, or tumor development.[42,43,44,45,46]

1.4. Toxicology Studies

To assess the safety of REACT, three GLP safety studies were conducted, i.e., one study was conducted in the rat ZSF-1 disease model of CKD and the other two studies were conducted in normal canines. A brief summary is presented below; detailed information is provided in the Investigator's Brochure.

1.4.1. ZSF-1 Rat Single Dose Study

The purpose of this study was to assess the safety of a single administration of REACT in ZSF-1 rats, a model of uncontrolled metabolic syndrome including T2DM, hypertension, and severe obesity. The rats received: 1) high dose REACT; 2) low dose REACT; 3) sham; or, 4) biomaterial only. Each animal received 4 injections of test article, one into each pole of each kidney. The results were assessed at 3- and 6-months post-treatment.

1.4.1.1. Renal-Related Findings

No treatment-related kidney findings were noted following evaluation of 8 areas of each kidney (3 stains per area), including assessment and scoring of 150 glomeruli per kidney. Apart from changes related to implantation and/or injection site linear scars, all kidneys were considered normal within the context of the disease model. No test article-related kidney finding was observed at 3- or 6-month post-treatment. All macroscopic and microscopic kidney changes were considered related to the natural progression of renal disease in the ZSF-1 obese rat, or to the injection procedure.

Kidney changes in all groups were more severe in males, and consistent with differences in the disease stage between genders. Overall, there was an apparent trend of lower renal histological severity scores (i.e., lower glomerular injury score, tubule-interstitial injury score, and global nephron score) that was consistently noted in the low-concentration REACT treatment group when compared to the Sham control group 6 months post-procedure.

Based on the absence of differences across study groups, the No-Observed-Adverse-Effect-Level (NOAEL) was the high dose, 6.25 x 10^6 cells /g KW^{est}.

1.4.1.2. Non-Renal Findings

No REACT safety-related findings were observed in non-target tissues. No ureteral or bladder (primary routes of REACT excretion) REACT-related changes were observed. There were no REACT- related effects, and no observable REACT cellular materials in any of the draining (lymph nodes) or filtering (liver, lung, spleen) tissues examined.

1.4.1.3. Clinical Pathology

The results of clinical laboratory tests (including hematology, clinical chemistry, and special urinalysis panels) were evaluated for differences between baseline and end of study (3- or 6-months post-treatment), and between treated and control groups. No REACT-related clinically significant laboratory abnormalities were identified.

1.4.1.4. Conclusions:

- All animals survived to the end of study (3- or 6-months post-treatment).
- There were no significant safety-related clinical pathology findings or safety-related findings of toxicological significance attributable to treatment.
- No REACT-related clinically significant laboratory abnormalities were identified.
- The observed NOAEL was 6.25 x 10^6 cells /g KW^{est}.

1.4.2. Initial Single Dose Canine Study

The initial canine toxicology study assessed the safety of a single administration of two different doses of REACT compared to sham treatment or treatment with the biomaterial. The test article was delivered into one pole of each kidney. A total of 32 mongrel dogs were entered into the study; 16 were assessed at one month and 16 were assessed at 3 months.

1.4.2.1. General Results

All 32 animals survived to their designated termination time point at one or 3 months after treatment. The animals appeared to be in good health throughout the study. There were no significant clinical pathology findings. There were no signs of renal insufficiency (azotemia), and there were no indications of decreased GFR.

1.4.2.2. Renal-Related Results

No REACT delivery-related macroscopic or microscopic findings were observed at the one- or 3-month endpoint. No treatment-related kidney findings were noted following enhanced evaluation of 8 areas of each kidney (3 stains per area), including evaluation and scoring of 150 glomeruli per kidney. Apart from changes related to injection site scars all kidneys were normal. All macroscopic and microscopic kidney changes were considered background findings or related to the injection procedures.

1.4.2.3. Non-Renal Findings

No test article-related findings were identified in other (non-kidney) tissues. All macroscopic and microscopic changes were considered background changes and within normal limits.

1.4.2.4. Procedure-Related Findings

The most common abnormalities included swelling at the incision sites (seroma formation) and weight loss at study termination. With regard to incision site swelling, ten of sixteen (10/16) animals had sterile seroma formation post-implantation, and nine of sixteen (9/16) post treatment. The animals had varying degrees of swelling at their retroperitoneal incisions and were treated as deemed necessary by a veterinarian.

Numerous animals had mild inappetence following REACT administration (29 of 32) and treatment procedures (18 of 32). Most animals (28 of 32) experienced weight loss from baseline (prior to implantation) to termination. Of note, a greater amount of weight loss occurred between baseline (2 weeks prior to treatment) and treatment (Day 0; renal injections) than between treatment and termination. Weight loss also occurred across all treatment groups and was judged to be related to the stressful nature of the study.

1.4.2.5. Conclusions

- All animals survived to their designated termination time point and were in good general health throughout the study based on clinical pathology, urinalysis, and veterinary assessment.
- Neither the low or high dose of REACT produced macroscopic or microscopic adverse effects at one- or 3-months post-treatment, similar to observations following sham treatment or treatment with the biomaterial.
- Pathological evaluation showed no REACT safety-related (macroscopic or microscopic) findings in the target (kidney) or non-target organs examined.
- Based on anatomic pathology, the observed NOAEL was the higher concentration tested, i.e., 11.7 x 10^6 cells/g KW^{est}.

1.4.3. Repeat Dose Canine Study

The second canine toxicology study assessed the safety of administering two repeat doses of REACT. Each dose was delivered into both kidneys at baseline (time zero) and 3 months. All animals were subjected to two renal biopsies per kidney 4 to 6 weeks prior to the baseline injection procedure. Control animals were injected with PBS. Animals were monitored for 6 months following the baseline injection.

1.4.3.1. Study Results

All 8 animals were in good clinical health throughout the study and survived to their designated termination time point at 6 months. There was mild or insignificant weight loss in 5 of 8 animals, with 2 animals losing >3% body weight for the duration of the study. Greater weight loss occurred between the renal implantation and initial treatment than between the initial treatment and termination. The clinical pathology and urinalysis data revealed no abnormal trends. There were no signs of renal insufficiency, and no indications of decreased GFR.

1.4.3.2. Kidney-Related Findings

No REACT implantation safety-related macroscopic or microscopic findings were observed at the 6-month time point. No treatment-related kidney findings were noted following enhanced evaluation of 8 areas of each kidney (3 stains per area), including evaluation and scoring of 150 glomeruli per kidney. All kidneys appeared normal, apart from changes related to injection site

scars (fibrosis/chronic inflammation in the capsule; linear fibrosis/chronic inflammation and inflammatory cells in the cortex/medulla).

1.4.3.3. Non-Kidney Related Findings

No test article safety-related findings were identified in non-target tissues. All macroscopic and microscopic changes were considered background changes and thus, considered within normal limits.

1.4.3.4. Conclusions

- All animals survived to their designated termination time point, and appeared in good health based on clinical pathology, urinalysis, and veterinary assessment data.
- Pathological assessment showed no REACT safety-related (macroscopic or microscopic) findings in either the target organ (kidney) or non-target organs examined.
- At the 6-month time point, no detrimental effects of two repeat doses of REACT were observed in comparison to the control animals injected with PBS.

1.5. Non-clinical Conclusions

Evidence from multiple animal studies over a wide range of doses (3 to 15 million SRC/g of kidney tissue implanted), and extended periods of time post-REACT treatment (up to one year), including three GLP studies, indicates that the potential risk of complications from REACT delivery into the kidney is similar to the potential risk of complications associated with standard renal biopsy practice.[47,48,49]

Apart from changes related to injection procedures and cardiovascular findings specific to 5/6th nephrectomy rodent mass reduction model of CKD, no unanticipated in-life, hematological, urological, serological, or histological changes were found in the target organ or non-target tissues following delivery of REACT.

1.6. Phase 1 Clinical Trial: Interim Results

1.6.1. Background

In April 2013, a first-in-human clinical trial was initiated at the Karolinska University Hospital Huddinge in Stockholm, Sweden: A Phase 1, Open-Label Safety and Delivery Optimization Study of Renal Autologous Cell Therapy (REACT) in Patients with Chronic Kidney Disease (RMTX-CL001). This is a Phase 1, open-label, safety and delivery optimization study of REACT implanted into subjects with CKD. REACT will be manufactured from SRC obtained from a subject's renal biopsy, formulated with gelatin biomaterial, and injected back into the subject's left kidney. The primary objective is to assess the safety and optimal delivery of REACT implanted at one site in a recipient kidney as measured by procedure- and/or product-related adverse events (AEs) through 12 months post-treatment. The secondary objective is to assess renal function by comparing the results of laboratory tests from baseline through 12 months following REACT injection, followed by an additional observational period of 18 months. Each subject's baseline rate of CKD disease progression serves as his/her own "control" to monitor for changes in renal insufficiency over time. Six subjects, recruited from the Karolinska University Hospital, were enrolled into the study. In addition, one subject was enrolled in the study at the University of North Carolina.

1.6.2. Adverse Events

Among a cohort of 7 male subjects, 53 to 70 years of age, with pre-dialysis diabetic nephropathy (Stage 3b/4 of CKD), all subjects recovered from the laparoscopic REACT delivery procedure without immediate perioperative complications. Notably, no subject experienced hematuria, which was prospectively considered to be the most likely untoward event. One subject developed an intestinal volvulus on Day 2 after REACT injection, and required a partial colonic resection that was complicated by anastomotic hemorrhage. In the judgment of the Investigator, this event was not related to the investigational product or the procedure. One subject experienced a skin infection that was associated with the laparoscopic implantation procedure. Another subject recovered from the surgical procedure with inflammation of the respiratory tract. All serious adverse events (SAEs) associated with the clinical trial are presented in Table 6.

Eight of the nine SAEs were considered possibly related to the implant surgery. There were no AEs or SAEs considered related to the biopsy procedure. To date, no delayed or late-onset adverse reactions related to REACT or other study procedures have been identified (e.g., negative immune mediated reactions). Based on the current data, the highest risk associated with REACT treatment appears to be from the implant surgery. Consequently, steps are being taken to decrease the duration of the surgical procedure and improve surgical outcomes.

Table 6: Serious Adverse Events Reported in Study RMTX-CL001

Subject Number	MedDRA Preferred Term	Intensity	Time to	Outcome of SAE	Relationship to REACT
001-002	Fatigue	Mild	4 days	Recovered /Resolved	Not Related
001-001	Fatigue	Mild	5 days	Recovered /Resolved	Not Related
001-001	Postoperative wound infection	Mild	0 days	Recovered /Resolved	Possibly Related
001-002	Pneumonia	Moderate	1 days	Recovered /Resolved	Possibly Related
001-003	Urinary tract infection	Mild	7 days	Recovered /Resolved	Not Related
001-004	Fatigue	Mild	4 days	Recovered /Resolved	Not Related
001-005	Volvulus	Moderate	2 days	Recovered /Resolved	Not Related
001-003	Fluid retention	Moderate	1 days	Recovered /Resolved	Not Related
001-005	Anastomotic hemorrhage	Moderate	0 days	Recovered /Resolved	Not Related

Abbreviations: MedDRA (Medical Dictionary for Regulatory Activities); Renal Autologous Cell Therapy (REACT); SAE (Serious Adverse Event)

1.6.3. Estimated Glomerular Filtration Rate (eGFR)

Seven male patients with T2DM and Stage 3b/4 of CKD were implanted with REACT in the left kidney. Pre-implantation information from these subjects indicated that their average decline in eGFR was 6.1 ml/min/year. Following REACT treatment, eGFR decline for the combined group (all subjects) was -3.1 ml/min/year (green line in Figure 1).

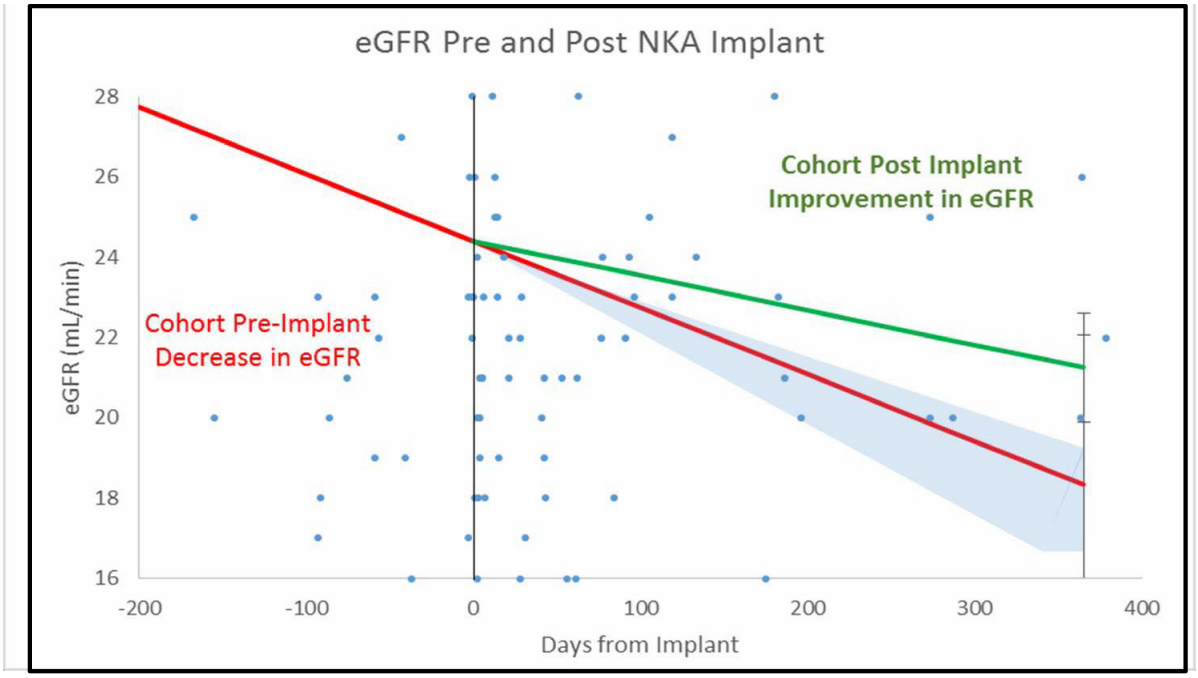

Figure 1: Estimated Glomerular Filtration Rate Pre- and Post-REACT Treatment (RMTX-CL001)

After monitoring the potential impact of REACT treatment on CKD progression in this cohort for approximately one year, the expected decline in renal function appears to have been modified by a single injection of REACT into a single kidney. In Figure 1, a comparison of eGFR following REACT treatment (green line) versus eGFR before REACT treatment (red-line) showed that 6 of 7 subjects had a reduction in the rate of eGFR decline post- treatment. The annual rate of change for eGFR, before and after REACT treatment, is presented for each subject in Table 7.

Table 7: Estimated Glomerular Filtration Rate by Subject (RMTX-CL001)

Subject Number	Change in eGFR (mL/min/year)	
	Pre-REACT Treatment	Post-REACT Treatment
001-001	-14.8	1.5
001-002	-0.2	-1.3
001-003	-6.7	-5.9
001-004	-16.3	-7.5
001-005	-3.9	-2.6
001-006	-11.4	-5.9
001-007	-7.7	1.4

1.6.4. Serum Creatinine

Pre-treatment levels of serum creatinine (sCr) were generally elevated in this cohort, which would be expected from subjects with T2DM and moderate to severe renal insufficiency (Stage 3b/4 of

CKD). The annual rate of change for sCr, before and after REACT treatment, is presented for each subject in Table 8. All subjects demonstrated a reduction in their individual rate of increase for sCr following REACT treatment compared to the rate of sCr increase that had been observed before REACT treatment.

Table 8: Serum Creatinine by Subject (RMTX-CL001)

Subject Number	Change in Serum Creatinine (µmole/L/year)	
	Pre-REACT Treatment	Post-REACT Treatment
001-001	153	-41
001-002	17	-21
001-003	214	200
001-004	16	-39
001-005	69	23
001-006	216	95
001-007	48	-40

The collective pre-treatment level of sCr for this cohort was >100 µmole/L/yr. Following REACT treatment, sCr decreased to <50 µmole/L/yr. As shown in Figure 2, a comparison of sCr after REACT treatment (green line) *versus* sCr before REACT treatment (red-line) showed that the cohort experienced a reduction in the rate of increase for sCr post-REACT treatment. This change was consistent for each subject.

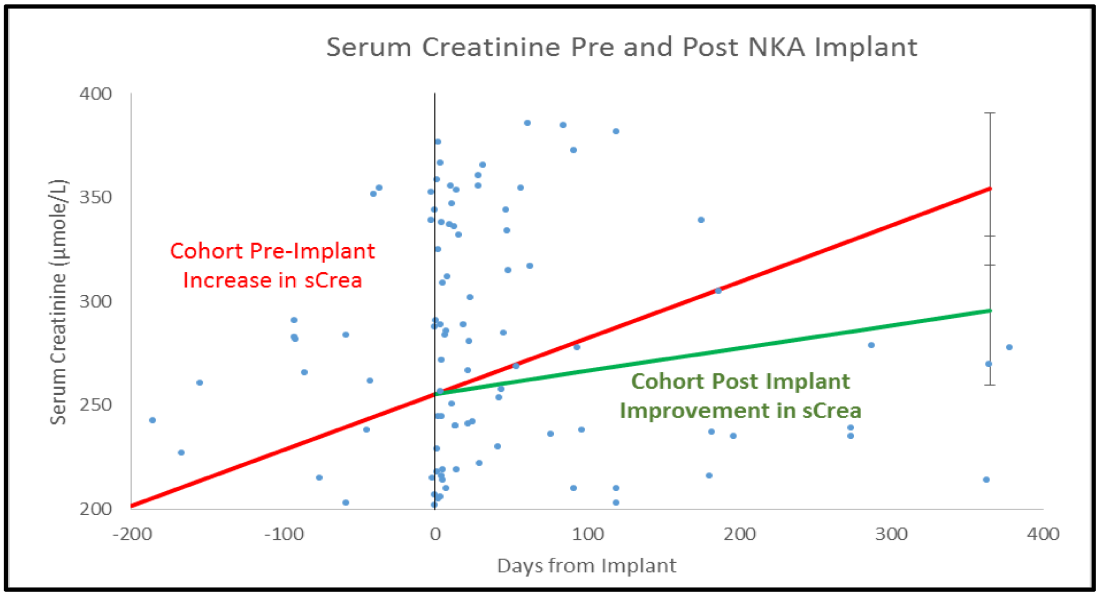

Figure 2: Serum Creatinine Pre- and Post-REACT Treatment (RMTX-CL001)

1.5.1. Kidney Cortical Thickness

Patients suffering from chronic kidney disease undergo a thinning of the functional portion of the kidney, i.e., the cortex. Renal cortical thickness is reduced in CKD as a result of fibrosis and

scarring as the disease progresses. An increase in cortical thickness was associated with kidney regeneration in pre-clinical studies of REACT and was confirmed histologically in all 4 animal species studied (refer to the Investigator's Brochure). In the clinical trials TNG-CL010 and TNG-CL011, cortical thickness was evaluated using imaging technologies; no biopsies were taken to confirm the basis for the increased thickness. Cortical thickness was measured in both the right and left kidney to determine if the implanted left kidney exhibited any change in cortical thickness that could be attributed to REACT implantation. The right kidney served as a non-implanted control.

On average, cortical thickness increased in the left kidney from 14 mm at baseline to approximately 16 mm after one year of REACT treatment. This change in cortical thickness was not sufficient to cause an increase in the total volume of the left kidney (data not presented). No change in cortical thickness was observed in the right kidney cortex.

1.6.5. Hemoglobin

CKD can be associated with anemia due to an alteration in renal erythropoietin production as well as metabolic abnormalities resulting from chronic uremia.[50] In the clinical trial RMTX- CL001, 3 of 7 subjects exhibited improvement in hemoglobin levels after REACT treatment, while the remaining 4 subjects maintained normal levels during the study.

1.6.6. Blood Pressure

Blood pressure was monitored during the course of clinical trials TNG-CL010 and TNG-CL011. Subjects received medication to control their blood pressure. Notably, intake of antihypertensive medication was reduced in 3 of 6 subjects during the first six months following REACT treatment.[51]

1.7. Potential Risks

In general, potential risks associated with the clinical use of REACT can be broadly divided into 3 categories: kidney biopsy, REACT product, and delivery into the recipient kidney. An assessment of potential risks associated with each of these steps is presented in this Section.

At this time, there are no specific warnings or precautions associated with the use of REACT. However, warnings and precautions for the renal biopsy and the percutaneous injection procedure must be considered with use of this product. The risks of renal biopsy have been well characterized in the 100 years that this procedure has been used and developed. Percutaneous needle instrumentation of the kidney has a shorter history.

The risks of renal biopsy and percutaneous needle kidney injection include:

1. Pain in flank / injection/biopsy site
2. Bleeding at injection/biopsy site which may occur around the kidney or anywhere along the needle track and which may be sufficient to entail clinically significant anemia, acute kidney injury (AKI), hematoma, and in the case of subcapsular bleeding, a "Page kidney" and acute hypertension
3. Surgical damage to the kidney from needle injury, as well as injuries of other structures that include connective tissues, bone, and intra-abdominal viscera

1.7.1. Potential Risks Associated with Renal Biopsy

Autologous kidney cells will be obtained from individual subjects via a kidney biopsy performed according to standard medical practice [47,48,49] and consistent with standard operating procedures at participating hospitals /medical institutions. A minimum of 2 tissue cores from a single kidney

biopsy is needed to obtain sufficient renal cortical tissue for the production of REACT. A 16-gauge biopsy needle measuring approximately 10 mm in length will remove 0.01-0.02% of the average total volume of the diseased kidney. Since approximately 0.001% of the total number of renal glomeruli will be harvested,[49] the biopsy is not expected to adversely affect kidney function.

Kidney biopsies for diagnostic procedures are of low risk and often conducted under sedation on an outpatient basis in the US.[52,53] When performed by qualified interventional physicians, a renal biopsy properly targeted towards the cortex produces limited renal damage.[45] On the other hand, reports of kidney damage at the biopsy site describe vascular injury and varying degrees of ischemia and infarction. The severity of damage depends on the size and number of vessels injured during the biopsy procedure.[54]

Hemorrhage is the most common AE associated with a routine kidney biopsy. Nearly all patients experience microscopic hematuria as a result of the biopsy, but this is not clinically significant.[48,53] On the other hand, gross hematuria occurs in 3-9% of patients,[52,53] and generally resolves by 24 hr. post-biopsy. The most serious complication is severe bleeding that requires transfusion and/or results in patient death. Transfusions are needed in less than 1% of renal biopsies, and death occurs in less than 0.01% of cases.[55,56,57]

1.7.2. Potential Risks Associated with REACT Product

The investigational product, REACT, is composed of autologous renal cells obtained from the same subject via kidney biopsy. Based on experience with autologous stem cell transplantation, the risk of an immune response (e.g., graft rejection) caused by REACT injection into the kidney seems unlikely.

Since the kidney is a highly perfused organ, it is doubtful that the implanted SRC will remain localized at the injection site. The three locations considered to be the most likely destinations of migrated SRC are: 1) the sub-capsular space; 2) the systemic circulation; and, 3) the urinary tract. Leakage of SRC into the sub-capsular space is not expected to pose a risk to the subject.

For example, the sub-capsular space is commonly used to implant endocrine tissue, such as islet cells.[58] The renal capsule also serves as a niche for native stem cells capable of migrating into the renal parenchyma.[59] Additionally, direct injection into the kidney mitigates the possible entry of SRC into the systemic circulation by providing a natural route of elimination via the urinary tract. Furthermore, intravenous administration of heterologous, allogeneic stem cells (i.e., mesenchymal stem cells) has been evaluated in clinical trials and yielded no significant risk to subjects.[42,43,44,45,46]

Porcine Skin Type B gelatin used in the formulation of REACT meets Pharmaceutical and Edible Gelatin Monograph (European Pharmacopeia 7.0, US Pharmacopeia-National Formulary USP35 NF30) requirements. Gelatin is widely used in pharmaceutical and medical applications, including cellular transplantation for regenerative products. Gelatin would not be expected to cause adverse effects in study subjects based on its biocompatible nature, widespread use, and results of GLP toxicology studies with REACT-containing porcine gelatin. Additional information on the porcine gelatin used to manufacture REACT is provided in the Investigator's Brochure.

1.7.3. Potential Risks Associated with REACT Treatment

A percutaneous technique will be used to access the kidney for REACT delivery. The percutaneous approach has been used for over a decade in ablation of renal masses. A concise review of this method can be found in Salagierski and Salagierski (2010).[60] Safety measures will be executed

during REACT treatment and post-surgical follow-up to reduce the potential for excessive bleeding and other AEs. Patients will be closely monitored as discussed in Section 6.

Cain and coworkers (1976)[61] reported that renal cell homogenates injected into rodent kidneys produced no significant AEs. Similarly, the observed morphological effects following REACT delivery into the kidney were consistent with those reported for repeated kidney biopsies taken from canines, i.e., the presence of a mature connective tissue track with no functional deficits linked to minimal structural changes.[54] Increasing intracapsular kidney water volume in canines can elevate intra-kidney pressure as well as transient increases in kidney weight and systemic blood pressure.[39,40] However, in the pilot canine studies that assessed the short-term effects of volume administration on blood pressure, there were no adverse effects on blood pressure following volume escalation up to 6 mL per kidney of REACT.

1.8. Potential Benefits

The potential to achieve clinically significant improvement in CKD is supported by studies that tested REACT in pre-clinical animal models of kidney insufficiency, i.e., surgical models for decreased kidney function in otherwise healthy rats and dogs plus the ZSF-1 rat model of T2DM. The main finding was that REACT significantly decreased the rate of structural and functional deterioration in already compromised kidneys to an extent that was clinically relevant in the animal model. Therefore, the potential exists for subjects participating in this clinical trial to realize therapeutic benefit from REACT treatment, such as a possible reduction in the rate of progression of CKD.

2. PHASE II TRIAL OBJECTIVES AND PURPOSE

ProKidney is currently developing a regenerative cell-based product, Renal Autologous Cell Therapy (REACT), with the aim of improving renal function in subjects who have CKD and T2DM. Therapeutic intervention with REACT is intended to delay the need for renal replacement therapy (dialysis or transplant) which, based on the current standard-of-care, is inevitable for patients with end-stage CKD. The purpose of the present study is to compare the safety and efficacy of up to 2 injections of REACT given 6 months (+4 weeks) apart (maximum). Each subject's annual rate of renal decline, based on adequate historical clinical data from 24 months prior to the Screening Visit, will serve as a comparator to monitor the rate of progression of renal insufficiency.

We propose that REACT treatment will reduce the rate (slope) of eGFR decline and improve renal function over the 24-month period following the last REACT injection.

2.1. Primary Objective

To assess the safety of REACT injected in one recipient kidney.

- ***Primary Endpoint:*** Procedure and/or product related AEs through 24 months post-injection.

2.2. Secondary Objective

To assess the safety and tolerability of REACT administration by assessing renal-specific AEs over a 24-month period following injection.

- ***Secondary Endpoints:*** Renal-specific laboratory assessments through 24 months post injection.

2.3. Exploratory Objective

To assess the impact of REACT on renal function over a 24-month period following injection.

- ***Exploratory Endpoint:*** Clinical diagnostic and laboratory assessments of renal structure and function (including eGFR, serum creatinine, and proteinuria) to assess changes in the rate of progression of renal disease.

3. INVESTIGATIONAL PRODUCT

3.1. Description of Study Drug

REACT is an injectable product composed of SRC formulated in a biomaterial (gelatin-based hydrogel). Table 9 presents an overview of the investigational product. Refer to the Investigator's Brochure for a detailed description of SRC and REACT as well as the manufacturing process.

Table 9: Investigational Product

	Investigational Product
Product Name:	Renal Autologous Cell Therapy (REACT)
Dosage Form:	Renal cells obtained from autologous kidney biopsy tissue will be expanded and SRC selected. SRC will be formulated in a gelatin-based hydrogel at a concentration of 100 x 10^6 cells/mL. This sterile cell preparation (REACT) will be contained in a sterile 10 mL syringe and shipped to the clinical site for use.
Unit Dose	The dose of REACT will be adjusted to 3 x 10^6 cells/g estimated kidney weight determined from MRI study.
Route of Administration	Percutaneous injection into the cortex of the biopsied kidney
Physical Description	Sterile, labelled, 10 mL syringe containing up to 8 mL of REACT
Manufacturer	ProKidney, Winston-Salem, North Carolina, USA

3.2. Procurement and Manufacture of REACT

REACT will be manufactured in a GMP facility at ProKidney located in Winston-Salem, North Carolina, USA.

3.2.1. Biopsy

The biopsy material will be collected using standard surgical techniques to assess the left or right kidney. A minimum of 2 tissue cores each measuring in 1.5cm must be collected using a 16-gauge biopsy needle to provide sufficient material for the manufacture of autologous REACT. When the biopsy material is received at ProKidney, the samples will be labeled and strict documentation measures followed to insure that product traceability is maintained. ProKidney will notify the site concerning the adequacy and quality of the biopsy sample for the manufacture of REACT and confirm the scheduled date for REACT injection. If the biopsy material cannot be used, the subject should be discontinued from the study.

3.2.2. Selection of SRC

Approximately 4 weeks prior to the subject's planned REACT treatment, autologous renal cells will be removed from the vapor phase of a liquid nitrogen freezer, thawed, and isolated from kidney tissue by enzymatic digestion. Cells will be cultured and expanded using standard techniques.

The cell culture medium is designed to expand primary renal cells and does not contain any differentiation factors. Harvested renal cells will be subjected to density gradient separation to obtain SRC, which are composed primarily of renal epithelial cells known for their regenerative

potential.[62] Other parenchymal (vascular) and stromal (collecting duct) cells may be sparsely present in the autologous SRC population.

If sufficient cells are available, the same biopsy material will be used to make additional REACT preparations for research studies and stored, under GMP conditions, in the vapor phase of a liquid nitrogen freezer.

It is anticipated that all subjects will receive a series of 2 REACT injections. The time and events table shows that the series of 2 REACT injections will be administered 6 months apart with a study visit window of 4 weeks. Regardless, **every attempt should be made to ensure that the second REACT injection is administered 6 months after the first injection**. ProKidney will notify the site to obtain information about the scheduled date for the second injection.

3.2.3. Formulation

SRC will be formulated in a gelatin-based hydrogel to improve stability during transport and delivery upon injection into the renal cortex. Porcine gelatin will be dissolved in buffer to form the thermally responsive hydrogel. Although fluid at room temperature, this biomaterial gels when cooled to refrigerated temperature (2 to 8ºC). Prior to injection, the REACT investigational product must be warmed to ≥20C up to 26°C to liquefy the hydrogel.

3.2.4. REACT Product for Injection

Ten to 14 days prior to the scheduled date for REACT injection, the subject will report to the clinic and undergo assessments to verify continued eligibility. If the subject does not qualify for REACT injection, the Investigator and the Sponsor will discuss possible options, for example, if there is sufficient stability to attempt an REACT injection at a future date. If the subject still qualifies, the REACT product will be manufactured and shipped to the clinical center. It is the responsibility of the site to ensure REACT shipments can be delivered directly to site personnel. REACT will be injected into the biopsied kidney of eligible subjects using a percutaneous approach. The percutaneous method will employ a standardized technique (such as that utilized in the ablation of renal masses by radiofrequency or cryogenic methods).[60]

It is anticipated that all subjects will receive two planned REACT injections. However, if there appears to be any untoward safety risk, or rapid deterioration of renal function, or development of uncontrolled diabetes or uncontrolled hypertension, or development of a malignancy or an intercurrent infection, then the second REACT injection should not be administered.

Renal cells that may have been frozen but not used to manufacture REACT will be remain in the vapor phase of a liquid nitrogen freezer at ProKidney until the EOS Visit. At that time, if these renal cells are no longer needed, they will be de-identified of all personal information and stored in the vapor phase of a liquid nitrogen freezer for a maximum of 5 years. The aim is to test these renal cells in laboratory research studies. During the informed consent process, each subject must provide written consent for the storage and future use of autologous cells not used for REACT injection. Subjects must have the option of having these cells destroyed upon study completion.

3.2.5. REACT Dose

The dose of REACT for subjects in the Phase 1 clinical trials (TNG-CL010 and TNG-CL011) was 3 x 10^6 SRC /g estimated kidney weight (g KW^{est}). Similarly, in the present study, each REACT injection will contain 3 x 10^6 cells/g KW^{est}. Since the concentration of SRC is 100 x 10^6 cells/mL of REACT, the dosing volume will be 3.0 mL for each 100 g of kidney weight. The volume of REACT to be administered will be determined by pre-procedure MRI volumetric 3D evaluation or

ellipsoid formula (Length x width AP plane x width Transverse plan x .62). Examples of dosing volumes based on estimated kidney weight are shown in Table 10.

Table 10: REACT Dosing Relative to Estimated Kidney Weight

Estimated Kidney Weight (g KW^{est})[a,b]		REACT Dosing Volume (mL)	SRC Delivered (Number of Cells x
Median Weight (g)	Weight Range (g)		
100	95 – 108	3.0	300
117	109 – 125	3.5	350
133	126 – 141	4.0	400
150	142 – 158	4.5	450
167	159 – 175	5.0	500
183	176 – 191	5.5	550
200	192 – 208	6.0	600
217	209 – 225	6.5	650
233	226 – 241	7.0	700
250	242 – 258	7.5	750
— — —	>259	8.0[c]	800

Abbreviations: Estimated Kidney Weight (g KW^{est}); SRC (Selected Renal Cells).
Notes:
a. The dose of REACT will be 3 x 10^6 cells/g estimated kidney weight.
b. Kidney weight will be estimated from an MRI study performed before renal biopsy.
c. 8 mL will be the maximum dosing volume (mL).

The dose of REACT will be based on kidney volume calculated via MRI. In contrast to other methods, measurements of renal volume using MRI are more accurate, and acquire true tomographic data along any orientation without the risk of ionizing radiation or nephrotoxic contrast agents. Renal volume measurements (mL) estimated from MRI are approximately 92 to 97% of dry weight measurements in grams for isolated organs trimmed of perirenal fat. As a conservative approach, the REACT dose will be calculated using a conversion of one g equals one mL. The volume of REACT to be administered will be determined by pre-procedure MRI volumetric 3D evaluation or ellipsoid formula (Length x width AP plane x width Transverse plan x .62). This ensures that subjects will not receive REACT doses higher than those previously tested in animal studies.

3.2.5.1. Rationale for Two REACT Injections

It is anticipated that all subjects will receive two planned REACT injections to allow dose-finding and evaluate the duration of effects. The scientific rationale, based on non-clinical studies, is that the biologically active component of REACT (homologous, autologous, SRC) delays progression of experimental models of CKD by augmenting renal structure and function.[18,19,33,34,35,36] As a result, the more cells that can be infused, the greater the potential improvement in renal function.

The total number of cells that can be delivered into a kidney at one time is limited by the size of the kidney, however, as well as the inelasticity of the renal capsule. Consequently, it may be possible to improve therapeutic benefit by administering greater numbers of SRC via a second injection, given after cells from the first injection have become incorporated into the kidney.

Apart from increasing SRC numbers by administering 2 REACT injections into the same kidney, the duration of effects can be evaluated. The processes by which functional nephrons become disabled in kidneys with CKD may, over time, adversely affect “new” cells delivered via REACT

injection. Consequently, REACT might not result in long-term, therapeutic benefit. Exploring the effects from a second REACT injection, given at an appropriate interval after the first injection, would address this question.

In the present study, subjects will be administered a second REACT injection 6 months after the first injection, with a study visit window of 4 weeks. Regardless, **every attempt should be made to ensure that the second REACT injection is administered 6 months after the first injection**. In the event that the subject cannot schedule his/her second REACT injection 6 months after the first injection, ProKidney and the Medical Monitor must be notified immediately.

However, if there appears to be any untoward safety risk, or rapid deterioration of renal function, or development of uncontrolled diabetes or uncontrolled hypertension, or development of a malignancy or an intercurrent infection, then the second REACT injection should not be administered.

3.2.5.2. Safety of Two REACT Injections

To assess the safety of administering two doses of REACT into the biopsied kidney, a canine GLP toxicology study was conducted (Refer to Section 1.4.2). Similar to the clinical study design, study animals (n=8) underwent renal biopsies at 4 to 6 weeks prior to baseline. Each dose was delivered into both kidneys at baseline and 3 months; animals were observed for 6 months following the baseline injection. While control animals received PBS, REACT-treated animals received a two-fold greater dose than that used in the present clinical study.

Briefly, no detrimental effects of two doses of REACT into the biopsied kidney were observed in comparison to control animals 6 months after baseline treatment. Pathological assessment showed no REACT safety-related (macroscopic or microscopic) findings in either the target organ (kidney) or non-target organs examined. No treatment-related kidney findings were noted following enhanced evaluation of 8 areas of each kidney (3 stains per area), including assessment and scoring of 150 glomeruli per kidney. All kidneys appeared normal, apart from changes related to injection site scars. There were no signs of renal insufficiency, and no indications of decreased GFR. Detailed information is provided in the Investigator's Brochure.

3.3. Study Drug Packaging

The product delivery system consists of 3 components:

1. 10 mL standard, Luer-Lok® syringe
2. Package for containment of the syringe
3. REACT shipping container for transportation of the package to the clinical site

The syringe containing REACT will be shipped to the clinical site encased in a package designed to maintain integrity of the product as well as sterility of the product and syringe. A representative image of the product delivery system is shown in Figure 3.

Figure 3: Product Delivery System

The product delivery system is made from components listed in Table 11. Materials that come into contact with the REACT product are USP class VI or equivalent. The syringe, tubing and ancillary parts will be obtained from vendors listed in Table 11 or other vendors that satisfy the biocompatibility classification and product compatibility testing requirements. The syringe is pre-sterilized in the package by gamma sterilization. After filling, the tubing is sealed and cut.

Table 11: Product Delivery System Components

Components	Vendor	Production Material		REACT Contact	Biocompatibility Test Reference*
Syringe	Merit Medical or Becton-Dickinson	Polycarbonate, Silicone or Polypropylene, Silicone		Direct	ISO 10993 USP Class VI
Tubing	Saint-Gobain Performance Plastics	Polyvinyl Chloride		Direct	Ph Eur 3.1.1.2 ISO 10993 USP Class VI
Luer-Lok® Fittings	PAW BioScience	Polyethylene		Direct	USP Class VI
	Value Plastics	Polypropylene MABS		Direct	USP Class VI

* Additional testing has been performed (Cytotoxicity, MEM Elution, in vitro: USP <87>; Rabbit Blood Cell Hemolysis: ASTM F756-00; Physicochemical Test for Plastics: USP <661>).

3.4. Study Drug Label

The REACT product is made from expanded autologous SRC obtained from each individual subject's kidney biopsy and is, therefore, subject specific. Each package containing the syringe will have affixed to it a label containing the following: "FOR AUTOLOGOUS USE ONLY". In addition, the label will indicate that this drug (REACT) is for "Investigational Use ONLY".

3.5. Study Drug Transportation

All biopsy specimens will be transported to ProKidney using packaging mandated in the Code of Federal Regulations (42 CFR Part 72) and according to individual carrier guidelines.

Because the REACT hydrogel formulation must maintain a temperature from 2 to 8ºC during shipping, REACT product will be transported from ProKidney to the clinical site in a shipping container validated to maintain temperature at 2 to 8oC. The REACT package will be placed in a plastic outer containment bag and then in a refrigerated shipping container from Minnesota Thermal Sciences. A temperature recorder is also included in the shipping container. A representative image of the shipping container is shown in Figure 4.

Figure 4: REACT Shipping Container

When the shipping container arrives at the clinic in time for a scheduled injection, please open the package to confirm the patient ID and that no leaks have occurred. The REACT is to be left in the shipping container use for injection. Please follow the checklist included with the shipment for further details. The day of injection the inner REACT package will be removed from the shipping container and equilibrated to controlled room temperature (≥ 20ºC up to 26ºC) per instructions provided in the Study Reference Manual. Two individuals will independently verify identifying information in the presence of the subject, thereby confirming that the information is correctly matched to the specific study participant.

Once the hydrogel becomes liquid, the surgical assistant will open the container in a sterile field and transfer the syringe to the physician who will perform the percutaneous injection of REACT into the renal cortex of the biopsied kidney.

3.5.1. Study Drug Accountability

Investigational drug product accountability and traceability is ultimately the responsibility of the Investigator once it is released by the Sponsor. However, this responsibility may be delegated to a suitably qualified individual who has had appropriate study-specific training and whose name is listed on the Delegation of Responsibility Log for this task.

Detailed records will be maintained to allow for accurate accountability of the Investigational drug product in accordance with applicable Sponsor and clinical site procedures. These records will include details about the transfer of renal biopsy specimens from the clinical site to ProKidney, transfer of REACT drug product from ProKidney to the clinical site, injection of study drug to

autologous subjects, the number of stored specimens remaining at ProKidney LLC, and disposal of unused materials.

3.5.2. Study Drug Handling and Disposal

Since transmission of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood, and blood products; appropriate blood and secretion precautions will be employed by all personnel in the shipping and handling of all specimens for this study, as currently recommended by the Centers for Disease Control and Prevention and the National Institutes of Health.

All material containing investigational drug product will be treated and disposed of as hazardous waste in accordance with governing regulations and clinical site procedures.

3.5.2.1. Disposition of Stored Specimens

Specimens will be stored in the vapor phase of a liquid nitrogen freezer at ProKidney. Only Investigators or suitably qualified individuals who have received appropriate study-specific training and whose names are listed on the Delegation of Responsibility Log will have access to these specimens.

3.5.2.2. Unused Specimens

Renal cells that may have been frozen but not used to manufacture REACT will be remain in the vapor phase of a liquid nitrogen freezer at ProKidney until the EOS Visit. At that time, if these renal cells are no longer needed, they will be de-identified of all personal information and stored in the vapor phase of a liquid nitrogen freezer for a maximum of 5 years. The aim is to test these renal cells in laboratory research studies.

During the informed consent process, each subject must be provided the option *not* to store or use their unused renal cells and research samples (as described in Table 2), and must provide written consent for the storage and future use of autologous cells not used for REACT injection if they consent to the long-term storage of unused specimens. Additionally, subjects may decide at any point during the study *not* to have their unused renal cells stored. In this case, the Investigator will destroy all known remaining samples attributed to that subject and report what was done to both the subject and to the IRB. Likewise, subjects must have the option of having their cells destroyed upon completion of the study.

4. INVESTIGATIONAL PLAN

4.1. Study Administrative Structure

Figure 5 depicts the administrative structure for the study.

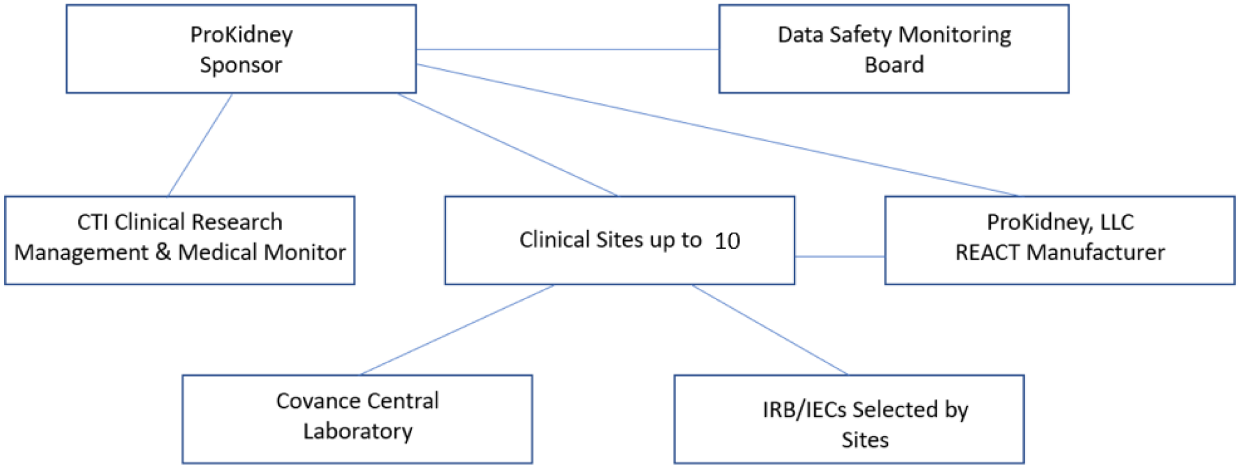

Figure 5: Study Administrative Structure

ProKidney is the Sponsor of this study. The following organizations will provide services for this study under contracts with the Sponsor:

- ProKidney LLC, Winston Salem, North Carolina, USA, is the contract manufacturer of the investigational drug product, REACT.
- Covance (www.covance.com/central_lab/info.php) will provide central laboratory services for the study.
- CTI Clinical Research Consulting Services, Covington, Kentucky, USA will provide regulatory, medical writing, statistical, clinical research management and medical monitoring for the study.

4.2. Overall Study Design

An overview of the study flow is shown in the diagram below.

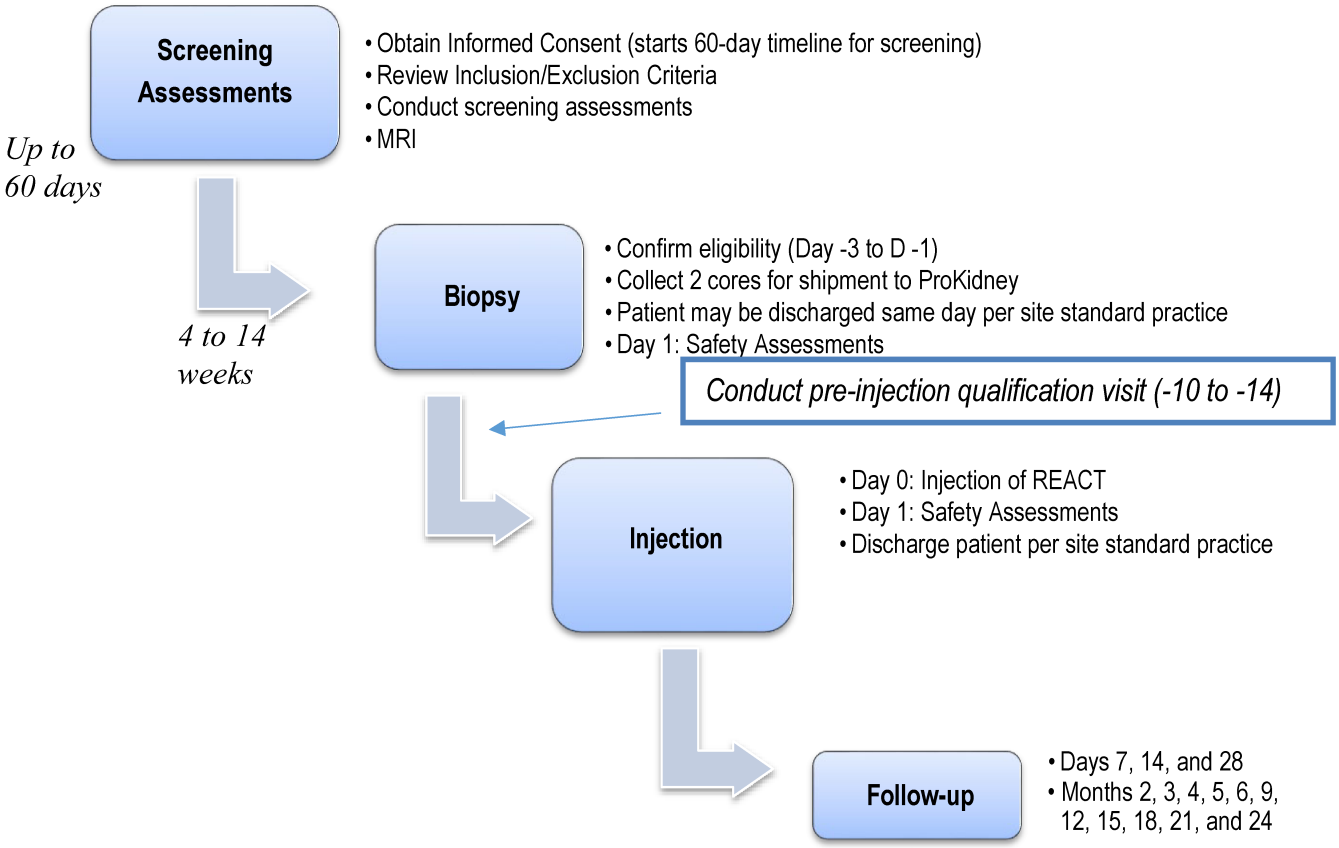

Figure 6: Study Design

After patients have signed the ICF, they should be screened for entry into the study. Screening assessments include laboratory assessments, physical examination, and an ECG and MRI study, all of which must be performed before the biopsy is taken. If the patient meets all I/E criteria, a biopsy of the left or right kidney should be taken within 60 days of the first screening assessment.

During the biopsy procedure, two tissue cores should be collected and sent to ProKidney for manufacture of REACT. If the patient experiences significant AEs/SAEs following biopsy (e.g., excessive bleeding, development of AV fistula) that, in the opinion of the PI would preclude safe injection, then the patient should be discontinued from the study.

One to two weeks after receipt at ProKidney's GMP facility in North Carolina, USA, ProKidney will notify the site if the tissue received was of sufficient size and quality for manufacture of REACT. If results are positive, the site should confirm the scheduled date of injection. If the biopsy was not able to be used for manufacture of REACT (for whatever reason), the patient should be discontinued from the study.

Ten to 14 days before the scheduled injection, the patient should report to the clinic for a pre-injection qualification visit including final review of I/E criteria and a renal scintigraphy study. If the patient is still eligible for injection, the site will notify ProKidney to manufacture REACT product from frozen renal cells. On the day of injection (Day 0), the patient should arrive at the hospital and receive an REACT injection into the kidney that was biopsied.

4.3. Number of Subjects

Up to 10 subjects who complete screening procedures and satisfy all inclusion and exclusion criteria will be enrolled.

4.4. Treatment Compliance

Eligible subjects will receive their autologous REACT preparations via a series of up to two injections. The investigational product will be administered into the biopsied kidney using a percutaneous approach. REACT product preparation and dosing procedures are specified in this protocol as well as the Study Reference Manual.

It is anticipated that all subjects will receive two REACT injections. In some cases, a subject or the Investigator may elect to delay or withhold the second REACT injection. For example, if there appears to be any untoward safety risk, or if the subject's health status would, in the judgment of the Investigator, be jeopardized, then the second REACT injection should not be administered.

4.5. Study Duration

Subjects will begin their series of REACT injection(s) as soon as the autologous REACT preparation is made available. Assuming a one-month interval prior to the first REACT injection, and assuming a 6-month interval before the second injection, plus a 24-month follow-up period after the final injection, the study duration would be:

- 31 months for a series of 2 REACT injections

5. STUDY POPULATION

5.1. Subject Inclusion Criteria

Unless otherwise noted, subjects must satisfy each inclusion criterion to participate in the study. Inclusion criteria will be assessed at the Screening Visit, prior to renal biopsy, and before each REACT injection unless otherwise specified.

1. The subject is male or female, 30 to 65 years of age on the date of informed consent.
2. The subject has an established diagnosis of T2DM.
3. The subject has an established diagnosis of diabetic nephropathy as the underlying cause of renal disease.
4. The subject has an established diagnosis of CKD not requiring renal dialysis, defined as having an eGFR between 14 and 20 mL/min/1.73m^2 inclusive at the Screening Visit and prior to REACT injection.
5. The subject has blood pressure less than 150/90 at the Screening Visit, prior to renal biopsy, and prior to REACT injection(s). At the time of the biopsy and injections, the subject's BP should not be significantly below the previously recorded stable pressure.
6. The subject has stable blood pressure and is maintained on a stable anti-hypertensive medication regimen, if treatment for hypertension is necessary. If treatment includes an angiotensin-converting-enzyme inhibitor (ACEI) or an angiotensin receptor blocker (ARB), that treatment must have been initiated at least 8 weeks prior to renal biopsy. Treatment must be stable during the 6-week period immediately prior to REACT injection. Stable treatment is defined as dose adjustment to no less than one half of the current dosage or to no more than 2 times the current dosage. Dose interruptions up to 7 days due to medical necessity are allowed.
7. A minimum of 3 measurements of eGFR or sCr should be obtained at least 3 months apart prior to the Screening Visit or within the previous 24 months to define the rate of progression of CKD. The subject should have adequate, historical clinical data to provide a reasonable estimate of the rate of progression of CKD. The Medical Monitor may be consulted to ensure there is sufficient data.
8. The subject is willing and able to refrain from NSAID consumption (including aspirin) as well as clopidogrel, prasugrel, or other platelet inhibitors during the period beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).
9. The subject is willing and able to refrain from consumption of fish oil and platelet aggregation inhibitors, such as dipryridamole (i.e., Persantine®), during the period beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).
10. The subject is willing and able to cooperate with all aspects of the protocol.
11. The subject is willing and able to provide signed informed consent.

5.2. Subject Exclusion Criteria

Subjects who satisfy any exclusion criterion listed below are not eligible to participate in the study. Exclusion criteria will be assessed at the Screening Visit, before renal biopsy, and before each REACT injection unless otherwise noted.

1. The subject has a history of type 1 diabetes mellitus.
2. The subject has a history of renal transplantation.
3. The subject has a serum HbA_{1c} level greater than 10% at the Screening Visit.
4. The subject has uncontrolled diabetes (defined as metabolically unstable by the Investigator).
5. The subject has hemoglobin levels less than 9 g/dL prior to each REACT injection.
6. The subject has abnormal coagulation status as measured by activated partial thromboplastin time (APTT), prothrombin time international normalized ratio (PT INR), and/or platelet count at the Screening Visit.
7. The subject has a bleeding disorder(s) or is taking anticoagulants, such as Coumadin® (warfarin) or direct thrombin inhibitors that, in the judgment of the Investigator, would interfere with the performance of study procedures.
8. The subject has small kidneys (average size less than 9 cm) or has only one kidney, as assessed by ultrasound and/or MRI prior to renal biopsy, unless earlier radiology reports (generated within 1 year of the Screening Visit) are made available to confirm kidney size and number.
9. The subject has a known allergy or contraindication(s), or has experienced severe systemic reaction(s) to kanamycin or structurally similar aminoglycoside antibiotic(s)
10. The subject has a history of anaphylactic or severe systemic reaction(s) or contraindication(s) to human blood products or materials of animal origin (e.g., bovine, porcine).
11. The subject is not a good candidate to undergo percutaneous REACT injection, in the judgment of the surgeon or physician who will perform the procedure. This includes individuals who are morbidly obese (defined as BMI greater than 45 kg/m^2), have excessive fat surrounding the kidney, or who are otherwise at excessive risk for serious complications.
12. The subject has a history of severe systemic reaction(s) or any contraindication to local anesthetics or sedatives.
13. The subject has a clinically significant infection requiring parenteral antibiotics within 6 weeks of REACT injection.
14. The subject has acute kidney injury or has experienced a rapid decline in renal function during the last 3 months prior to REACT injection.
15. The subject has any of the following conditions prior to REACT injection: renal tumors, polycystic kidney disease, anatomic abnormalities that would interfere with the REACT injection procedure or evidence of a urinary tract infection.

    Note: anatomic abnormalities are not exclusionary if the kidney remains accessible and meets the criteria to receive the REACT injection.
16. The subject has incapacitating cardiac and/or pulmonary disorders.

17. The subject has a history of cancer within the past 3 years (excluding non-melanoma skin cancer and carcinoma in situ of the cervix).
18. The subject has clinically significant hepatic disease (ALT or AST greater than 3 times the upper limit of normal) as assessed at the Screening Visit.
19. The subject is positive for active infection with Hepatitis B Virus (HBV), or Hepatitis C Virus (HCV), and/or Human Immunodeficiency Virus (HIV) as assessed at the Screening Visit.
20. The subject has a history of active tuberculosis (TB) requiring treatment within the past 3 years.
21. The subject is immunocompromised or is receiving immunosuppressive agents, including individuals treated for chronic glomerulonephritis within 3 months of REACT injection.

    Note: inhaled corticosteroids and chronic low-dose corticosteroids (less than or equal to 7.5 mg per day) are permitted as are brief pulsed corticosteroids for intermittent symptoms (e.g., asthma).
22. The subject has a life expectancy less than 2 years.
23. The female subject is pregnant, lactating (breast feeding), or planning a pregnancy during the course of the study. Or, the female subject is of child-bearing potential and is not using a highly effective method(s) of birth control, including sexual abstinence. Or, the female subject is unwilling to continue using a highly effective method of birth control throughout the duration of the study.

    Note: A highly effective method of birth control is defined as one that results in a low failure rate (i.e., less than one percent per year) when used consistently and correctly, such as implants, injectables, combined oral contraceptives, some intrauterine devices IUDs, sexual abstinence, or a vasectomized partner.
24. The subject has a history of active alcohol and/or drug abuse that, in the judgment of the Investigator, would impair the subject's ability to comply with the protocol.
25. The subject's health status would, in the judgment of the Investigator, be jeopardized by participating in the study.
26. The subject has used an investigational product within 3 months prior to REACT injection without receiving written consent from the Medical Monitor.

5.3. Prohibited and Concomitant Medications

- The consumption of NSAIDs (including aspirin) as well as clopidogrel, prasugrel, or other platelet inhibitors is prohibited during the study beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).
- Aspirin, up to a dose of 100 mg/day, may be continued if doing so is the standard of care at the investigational site, for subjects with diabetes who are greater than 40 years of age or have additional risk factors for cardiovascular disease or stroke, and for whom the perceived benefits of aspirin therapy outweigh the risks associated with treatment.
- Intake of fish oil and platelet aggregation inhibitors, such as dipryridamole (i.e., Persantine), is prohibited during the study beginning 7 days before through 7 days after both the renal biopsy and REACT injection(s).

- Subjects who are undergoing treatment with an ACEI or an ARB must have initiated therapy at least 8 weeks prior to renal biopsy. Treatment must be stable during the 6-week period immediately prior to REACT injection. Stable treatment is defined as dose adjustment no less than one-half of the current dosage and no more than 2 times the current dosage. In addition, except where medically necessary, no changes should be made to the ACEI or ARB dosing regimen from Screening through the 24-month EOS Visit. Dose interruptions up to 7 days due to medical necessity are allowed.
- Medications that interfere with measurements of SCr should be avoided during the study, such as trimethoprim, dronedarone, and cimetidine. If such medications are required based on medical necessity, then the circumstance should be discussed with the Medical Monitor and documented within the CRF.
- Use of investigational drugs is prohibited during the course of the study, unless preapproved by the Medical Monitor. Investigational drugs are defined as drugs that have not been approved for use by the FDA.

5.4. Subject Withdrawal Criteria

Subjects may withdraw from the study at any time and for any reason without penalty or prejudice, and without jeopardizing access to future medical care. If the Investigator determines that continuing in the study is no longer in the best interest of the subject, then the Investigator should withdraw the subject from the study. If a subject voluntarily withdraws from the study, the reason(s) should be documented. Subjects who withdraw from the study or are withdrawn by the Investigator should undergo procedures specified at the EOS Visit.

If a subject withdraws from the study before having the renal biopsy, the subject will be considered a screen failure. If a subject withdraws from the study following the renal biopsy but before the first REACT injection, the subject will not be a screen failure, but will not be considered enrolled and may be replaced. If a subject withdraws from the study after REACT injection but before the end of the follow-up period, the subject cannot be replaced.

Patients that do not meet I/E criteria on the RMCL-002 protocol but had a biopsy may be enrolled into REGEN-003. Patients that have completed a biopsy in RMCL-002 will not undergo another biopsy if they enroll in REGEN-003.

6. STUDY VISITS

The schedules of clinical assessments and procedures to be performed during the study are displayed in the time and events tables, i.e., Table 1. Similarly, the schedules of sample collection and clinical laboratory evaluations planned for the study are displayed in the laboratory time and events tables, i.e., Table 2. Before conducting any study specific assessments or procedures (including screening), the subject must provide written informed consent in accordance with ICH GCP guidelines and 21 CFR Part 50.

Due to the COVID-19 pandemic that began in March 2020, subject follow-up visits may be conducted remotely or via home health services to ensure the safety of the subject. Any procedures listed in the schedule of events that are not completed due to COVID-19 pandemic restrictions will be documented as protocol deviations. Clinical laboratory evaluations are permitted to be completed by a local lab convenient for the subject. Results of all evaluations performed by local laboratories will be collected by ProKidney.

6.1. Screening

All screening assessments should take place in a timeframe that will allow for scheduling of the renal biopsy within 60 days of the Screening Visit.

Renal ultrasound will be performed at the Screening Visit to verify subject eligibility (i.e., no evidence of renal tumors, polycystic kidney disease, renal cysts or other anatomic abnormalities that would interfere with the REACT injection procedure) along with obtaining a baseline echogenicity reading. Additionally, a MRI study without contrast will be performed from the time of Screening Visit through Day -1 before renal biopsy to determine kidney size and volume.

To qualify for study enrollment, the subject's eGFR must be between 14 and 20 mL/min/1.73m^2 inclusive at the Screening Visit. To define the eGFR for entry criteria, the site should use the eGFR assessed during screening and calculate using the CKD-EPI equation.[63]

If a subject does not meet a specific eligibility criterion, but the Investigator believes that the subject would be an excellent candidate for the clinical trial, then that criterion may be reassessed one time. In general, a subject who does not qualify may be rescreened once as long as the Investigator has sufficient clinical justification. If the Investigator has any questions concerning the appropriateness of rescreening a subject, then the Investigator should contact the Medical Monitor.

If, for whatever reason, the biopsy cannot be conducted within 60 days of the Screening Visit, then the Investigator and Medical Monitor should discuss and agree upon the need for repeating the screening assessments on a case-by-case basis. For example, laboratory assessments performed between 1 and 3 days before the renal biopsy might be used in place of the screening assessments to satisfy final eligibility criteria. In other cases, it may not be appropriate or necessary to repeat the clinical diagnostic procedures, including ECG or MRI studies.

6.2. Biopsy

The biopsy should be scheduled within 60 days of the Screening Visit. The biopsy should take place in a time frame that will allow for scheduling of the first REACT injection approximately one month later. Due to shipping and manufacturing schedules, the preferred day for collecting the renal biopsy cores is on a Wednesday or Thursday. Biopsies should not be collected on a Friday.

Subjects will report to the hospital or clinical research center one to three days before the biopsy for pre-biopsy assessments. As much as possible, pre-biopsy laboratory samples may be collected and

assessed to verify continued eligibility. After admission and final verification of inclusion and exclusion criteria, the biopsy should be performed as described in Section 7.5.1.

A minimum of 2 biopsy cores measuring 1.5cm in length a piece and collected using a 16-gauge biopsy needle under sterile conditions from each enrolled subject should be sent to ProKidney using the refrigerated shipping container provided by ProKidney and according to procedures detailed in the Study Reference Manual. ProKidney will contact the site to confirm receipt of sufficient biopsy material to manufacture REACT. If the biopsy cannot be used to manufacture REACT, the subject should be discontinued from the study.

Subjects who do not experience complications from the biopsy may be discharged the same day consistent with site standard practice. Otherwise, the subject should remain in the hospital overnight for observation. The subject may be discharged on the day after the biopsy so long as any biopsy-related AEs have resolved, stabilized, or returned to baseline.

6.3. REACT Injection

Due to shipping and manufacturing schedules, REACT injection should be scheduled on a Wednesday, Thursday, or Friday. If it is necessary to schedule the REACT injection on a Tuesday, the site should contact ProKidney in advance to make certain that a Tuesday delivery can be accommodated. The REACT injection should not be scheduled on a Monday.

Subjects will report to the clinic 10 to 14 days before the scheduled REACT injection for pre-treatment assessments. If the subject remains eligible, based on verification of inclusion and exclusion criteria, the site will perform a renal scintigraphy study and contact ProKidney indicating that the REACT product should be produced for that particular subject.

It is anticipated that all subjects will receive two planned REACT injections to allow dose-finding and to evaluate the duration of effects. As indicated on the time and events tables (i.e., Table 1), the series of 2 REACT injections will be administered 6 months apart with a study visit window of 4 weeks. Regardless, **every attempt should be made to ensure that the second REACT injection is administered 6 months after the first injection**. In the event that the subject cannot schedule his/her second REACT injection 6 months after receiving the first injection or cannot keep his/her scheduled 6-month visit, ProKidney and the Medical Monitor must be notified immediately. The expectation is that the subject and the clinical center will accommodate the Sponsor's preference that the series of 2 REACT injections should be administered no more than 6 months apart.

In some cases, a subject or the Investigator may decide to postpone or withhold the second REACT injection. For example, if there appears to be any untoward safety risk, or rapid deterioration of renal function, or the development of uncontrolled diabetes or uncontrolled hypertension, or the development a malignancy or an intercurrent infection, then the second REACT injection should not be administered.

If continued eligibility cannot be verified for a particular subject, the site should contact ProKidney who will alert the manufacturing staff not to prepare the REACT product. The Investigator and ProKidney will discuss the reason(s) that the subject did not qualify and determine the best course of action. For example, if the subject was excluded for a temporary condition (e.g., active infection) then it may be possible to manufacture REACT at a later date, assuming future eligibility can be established. If the results of pre-treatment assessments indicate that it is highly unlikely for that subject to become eligible for REACT injection(s), then the subject should be discontinued from the study. If REACT injection is delayed (e.g., due to an intercurrent illness), the renal scintigraphy study, if conducted, does not need to be repeated prior to injection.

Eligible subjects will arrive at the hospital or clinical research center on the morning of REACT treatment. The subject will be injected with autologous REACT using a percutaneous approach as discussed in Section 7.5.2.

6.4. Discharge After REACT Injection

On the day after REACT injection and prior to discharge, an ultrasound will be performed to detect possible, subclinical adverse effects (e.g., swelling, fluid accumulation). If product- or procedure-related AE's occurred following REACT injection, the subject should not be discharged until the AE's have resolved, stabilized, or returned to baseline. If consistent with the site's standard practice, the subject may be discharged the same day as the REACT injection after no less than 2 hours of observation and monitoring.

6.5. Follow-up Visits

The subject will return to the clinic for follow-up visits on Days 1 (if discharged day of REACT injection), 7, 14, and 28 (± 3 days) and Months 2, 3, 4 and 5 (± 7 days) after the first REACT injection, and on Days 1 (if discharged day of REACT injection), 7, 14, and 28 (± 3 days) and Months 2 and 3 (± 7 days) after the second REACT injection (Table 1 and Table 2).The subject will also report to the clinical center 10 to 14 days before the planned, final REACT injection to undergo pre-treatment assessments. **Every attempt should be made to ensure that the second REACT injection is administered 6 months after the first injection.**

Following the final REACT injection, subjects will complete long-term, follow-up assessments of safety and efficacy through Months 6, 9, 12, 15, 18, 21, and 24 (± 7 days) post-treatment.

6.6. End-of-Study Visit

This section describes situations in which a subject will undergo the EOS visit; for example, due to premature discontinuation from the study or completion of all protocol-specified follow-up visits.

- If a subject discontinues from the study after undergoing the renal biopsy but before REACT injection, then that subject should complete all EOS assessments except for the MRI and/or renal scintigraphy studies. These imaging studies should only be completed if, in the judgment of the Investigator, one and/or the other would provide critical safety information needed for patient care. If the subject is experiencing an investigational product- or study procedure-related SAE, then the subject will not be discontinued until the SAE has resolved, stabilized, or returned to baseline.
- If a subject discontinues from the study after undergoing one or two REACT injections but before completing all of the protocol-specified follow-up visits, then he/she should have the EOS Visit at the time of discontinuation. For these subjects, the Investigator will determine whether or not it is clinically prudent to perform one or both of the imaging studies (i.e., MRI and renal scintigraphy). If the subject is experiencing an investigational product- or study procedure-related SAE, then the subject will not be discontinued until the SAE has resolved, stabilized, or returned to baseline.
- If a subject completes all of the protocol-specified follow-up visits, he/she will undergo all EOS assessments 24 months after the final REACT injection at the EOS Visit. If the subject is experiencing an investigational product- or study procedure-related SAE, then the subject will not be discontinued until the SAE has resolved, stabilized, or returned to baseline.

6.7. Study Completion

Completion of the study is defined as the time when the last subject completes the EOS Visit, or when the last subject is considered lost to follow-up, withdraws consent, or dies.

7. STUDY ASSESSMENTS AND PROCEDURES

7.1. Demography and Medical History

Demographics characteristics will be obtained for each subject at the Screening Visit.

All CKD-related medical history and all other significant medical history will be recorded in the CRF beginning at the Screening Visit. Throughout the study, medical conditions that are still ongoing will be regularly updated in the CRF.

7.2. Clinical Evaluations

7.2.1. Vital Signs

Vital signs to be measured include systolic/diastolic blood pressure, heart rate, respiration rate, and temperature. Blood pressure will be measured after the subject has rested in a sitting position for a minimum of 5 minutes. At the Pre-Biopsy Visit (Day -3 to Day-1) and Pre-Injection Visit (Day -14 to Day -10 Visit), three BP measurements will be taken and the average of the 3 measurements (for systolic and diastolic pressure) used to satisfy entry criteria and entered into the CRF.

7.2.2. Physical Examination

The Investigator or designee will perform the PE. The comprehensive examination will assess all pertinent body systems while the interim examination will include specific assessments of those body systems deemed appropriate for that subject by the Investigator. As a general rule for the interim examination, the Investigator will review the subject's AEs prior to, or in conjunction with, the examination and include assessment of related body systems as appropriate. Only clinically significant abnormalities will be recorded in the CRF.

The subject's weight will be measured at every visit that includes a complete or interim physical examination. The subject's height will be taken at the Screening Visit. Body Mass Index (BMI) will be calculated as kg/m^2.

7.2.3. ECG

A 12-lead ECG will be obtained after the subject has been resting on their back for 5 minutes.

7.2.4. Concomitant Medications

Concomitant medications will be recorded in the CRF as follows:

- **Screening Visit until first REACT injection:** Record any CKD-specific medications as well as medications that may affect renal hemodynamics and/or serum creatinine measurements. In addition, record any medications used to treat an AE that is documented in the CRF. Surgical medications used during the biopsy procedure do not need to be captured in the CRF unless their use falls outside of expected dosages and/or frequencies of administration.
- **First REACT injection until 6 to 7 months of follow-up:** Record any medications taken until 6 to 7 months after treatment, depending on when the second REACT injection will be administered. Surgical medications used during the REACT injection procedure do not need to be captured in the CRF unless their use falls outside of expected dosages and/or frequencies of administration.

- **Second (Final) REACT injection until 6 months of follow-up:** Record any medications taken until 6 months after the last REACT treatment. Surgical medications used during the REACT injection procedure do not need to be captured in the CRF unless their use falls outside of expected dosages and/or frequencies of administration.
- **6 months of follow-up through the EOS Visit:** Record CKD-specific medications, that may affect renal hemodynamics and medications that may affect serum creatinine measurements. Record medications used to treat any AE documented in the CRF.

7.2.5. Funduscopic Exam of Retina

This exam should be completed at screening, month 12, and month 24/EOS to monitor for diabetic retinopathy of both eyes. Retinal photography should be collected and kept within the patient records and any worsening from baseline should be reported as an AE if not already documented in medical history.

7.3. Laboratory Assessments

Planned clinical laboratory evaluations are listed in Table 12. Analyses will be conducted by a central laboratory, except as noted. The schedule for collecting biological samples during the study are shown in Table 2.

Table 12: Clinical Laboratory Evaluations

Clinical Chemistry	Hematology	Urinalysis (24hr and Urine Chemistry)
Alkaline phosphatase ALT AST ß2-Microglobulin Bilirubin Creatinine kinase FSH (females only) GGT HbA_{1c} LDH PTH (intact)	Hematocrit Hemoglobin RBC count & indices WBC count & differential	Albumin ß2-Microglobulin Creatinine Protein Protein & Albumin:Creatinine Ratio NGAL
	Pregnancy	
	hCG (serum) – confirmatory	
	Serology	**Standard Panel**
	HBV HCV HIV	pH Ketones Protein Blood Glucose Microscopic analysis
Renal Analytes	**Coagulation Status**	
Albumin BUN Calcium CO_2, total Creatinine Cystatin-C CRP eGFR (calculated) Glucose Phosphorus Potassium Sodium	APTT PT-INR Platelet count	
	Lipid Panel	**Drug Screen**
	Cholesterol LDL HDL LDL:HDL ratio Triglycerides	Amphetamine Barbiturates Benzodiazepines Cocaine Opiates Tetrahydrocannabinol Phencyclidine

Abbreviations: ALT (Alanine Aminotransferase); APTT (Activated Partial Thromboplastin Time);AST (Aspartate Aminotransferase); BUN (Blood Urea Nitrogen); CRP (C-Reactive Protein); FSH (Follicle-Stimulating Hormone); GGT (gamma-glutamyl transferase); HbA_{1c} (glycosylated hemoglobin); HBV (Hepatitis B Virus); hCG (Human Chorionic Gonadotropin); HCV (Hepatitis C Virus); HDL (High Density Lipoprotein); HIV (Human Immuno-deficiency Virus); LDL (Low Density Lipoprotein); NGAL (Neutrophil Gelatinase-Associated Lipocalin); iPTH (Parathyroid Hormone, intact); PT-INR (Prothrombin Time- International Normalized Ratio).

7.3.1. eGFR

GFR will be estimated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation that incorporates both serum creatinine and Cystatin C.[63] The specific assay for measuring creatinine will be defined by ProKidney, and the samples will be analyzed by the central laboratory. For comparison to each subject's historical values, it may be necessary to perform a second analysis at the site laboratory used to generate the historical data. The need for any additional assays conducted locally by the site laboratory will be defined at the Site Initiation Visit.

7.3.2. Urine Collection

Urine will be collected and analyzed via standard panel and urine chemistry. The schedules for collecting each type of urine sample are shown in Table 2.

Urine will be collected over two different time periods: 24-hour collection and "spot" urine. Spot urine collections will be used for dipstick urinalysis (test stick) assessments. The schedules for collecting each type of urine sample are shown in Table 2. To provide a comprehensive picture of protein and albumin excretion, both total protein and albumin will be assessed in all samples.

7.3.3. Hematology

Hemorrhage following biopsy and REACT injection is a known and foreseeable risk to subjects participating in this study. Therefore, hemoglobin and hematocrit will be measured by the site's local laboratory before and after each procedure and compared to baseline levels. Other bleeding parameters (e.g., APTT, PTT-INR, platelets) also will be measured throughout the study.

7.3.4. Virus Serology

The biopsy cores obtained from each subject will be used for the expansion and selection of SRC. Contamination with HIV, HBV, and/or HCV would prevent ProKidney from manufacturing REACT product for that subject. Therefore, each subject will undergo testing for viral blood-borne pathogens, including HIV, HBV, and HCV.

7.3.5. Drug Screen

Consistent with Exclusion Criterion #24 (See Section 5.2), subjects are not eligible to participate in the study if they have an "…active history of … drug abuse that, in the judgment of the Investigator, would impair the subject's ability to comply with the protocol." Therefore, subjects will undergo testing for drugs of abuse.

7.3.6. Pregnancy Screen

A qualitative urine pregnancy test will be performed at the site using a test-strip. If the test is positive, then a confirmatory test will be performed by the clinical laboratory. If site practices do not accept the results of a test-strip, then a urine sample should be sent to the central laboratory for analysis. Post-menopausal women with a confirmatory FSH test do not have to undergo pregnancy testing throughout the study.

7.3.7. Research Samples

Research samples (serum/plasma and urine) will be collected, frozen, and stored for the evaluation of novel biomarkers. Participating subjects can opt out of long-term storage of their unused specimens during the informed consent process or at any time during the study (Section 3.5.2.2).

7.4. Renal Imaging

7.4.1. Ultrasound

Renal ultrasound will be performed at the Screening Visit to verify subject eligibility (i.e., no evidence of renal tumors, polycystic kidney disease, renal cysts or other anatomic abnormalities that would interfere with the REACT injection procedure) along with obtaining a baseline echogenicity reading. Ultrasound will also be performed following the in-patient renal biopsy on Day 0 and Day 1 and following the in-patient REACT injection(s) on Day 0 and Day 1 with the aim of monitoring possible, subclinical AEs. Findings from the ultrasound (e.g., resistance index, length, etc.) will be recorded on the CRF. An ultrasound may be conducted at other times, in the judgment of the Investigator, if needed for additional safety evaluation(s).

7.4.2. Computerized Tomography

Computerized tomography (CT) must be used in conjunction with ultrasound during the REACT injection procedure, according to the usual standards of care at the investigative site. Use of fluoroscopic CT may be used in conjunction with ultrasound.

7.4.3. Magnetic Resonance Imaging

An MRI study without contrast will be performed from the Screening Visit through Day -1 before renal biopsy to determine kidney size and volume. During the site initiation visit, the MRI process will be defined for each site, depending on the MRI equipment available. Generally, a 1.5-T unit should be used. MRI imaging studies will help determine kidney volume (for dosing calculations). MRI will be performed using standard sequences *without injection of contrast agents*. Renal volume measurements may be calculated, for example, using a fast 3D gradient- echo sequence, VIBE, with an acquisition time of 22 seconds and spatial resolution of 2 x 1.4 x 1.2 mm. Imaging parameters will be recorded in the source documents and CRF. A total of two MRIs will be performed on patients.

7.4.4. Renal Scintigraphy

Renal scintigraphy will be used to assess left and right kidney function using the radioactive tracer ^{99m}Tc- dimercaptosuccinic acid (DMSA) or Tc99m MAG3 (Mercaptoacetyl triglycine). This method is considered as the most reliable for measuring renal cortical function and is the preferred agent for this clinical trial. If the site standard practice is to use a radiopharmaceutical other than ^{99m}Tc-DMSA or Tc99m MAG3 then the site must discuss their procedure with ProKidney or the Medical Monitor prior to site initiation. If the site's standard practice is considered sufficiently equivalent to the procedure using ^{99m}Tc-DMSA or Tc99m MAG3, then the site will be allowed to follow their procedure. In this case, a copy of the site's procedure will be signed/dated by the Project Leader from ProKidney and maintained in the site's regulatory binder. All patients in this study will receive five renal scintigraphy studies. Renal scintigraphy will be performed before the first REACT injection, before the last REACT injection, at the 6-Month Visit after the last REACT injection, at the 12-Month Visit after the last REACT injection, and at the EOS Visit for all patients.

7.5. Surgical Procedures

7.5.1. Biopsy

Renal biopsy will be performed under sterile conditions using an ultrasound- or CT-guided approach consistent with site practices. Two biopsy cores will be needed to provide sufficient material for the selection of SRC and manufacture of REACT. Likewise, a 16-gauge needle should be used to insure adequate cortical material is obtained. If required, a 15-gauge needle may be used following consultation with the Medical Monitor and Manufacturing Facility. Bedside examination of the biopsy cores may be performed, if available, to ensure sufficient cortical material has been obtained.

Since the biopsy tissue will be used to manufacture REACT, the site must ensure that the tissue cores are harvested using sterile conditions so that the risk of contamination during subsequent cell expansion and selection is minimized. Contamination of the tissue cores could significantly jeopardize the ability of ProKidney to manufacture REACT product for that subject.

Guidance on wound care and pain management following the biopsy procedure will be provided in the Interventional Radiologist Manual. Briefly, the subject will remain supine for 4 hours with monitoring of hemoglobin, blood pressure, gross hematuria, abdominal/flank pain, and flank ecchymosis. As long as any biopsy-related AEs have resolved, stabilized, or returned to baseline, the subject can be discharged from the hospital on the day after the biopsy consistent with site standard practice. Importantly, any pain medication administered after the renal biopsy should be selected carefully, avoiding medications with nephrotoxic potential.

If a subject experiences significant AEs following the biopsy that, in the opinion of the Investigator, would put the subject at increased risk for significant adverse effects following REACT injection, then he/she will not be treated with REACT but will be followed until resolution of the event(s) and then discontinued from the study.

7.5.2. REACT Injection

Before performing the REACT injection, the operating physician will evaluate the subject as follows:

- Perform a physical examination to determine the feasibility of the procedure.
- Evaluate bleeding parameters, including coagulation panel, PTT-INR, platelets, hemoglobin, hematocrit, and other pertinent laboratory studies.
  - *Note: Hemorrhage following REACT injection is a known and foreseeable risk to subjects participating in this study. Therefore, hemoglobin and hematocrit will be measured before and after each REACT injection per the site's standard practice and compared to baseline levels.*
- Review imaging studies, including ultrasound, MRI, and CT or fluoroscopic CT, to determine route of access, depth of kidney, and appearance of cortical-medullary junction.
- Map potential REACT cell deposition sites.
- Determine classification and associated perioperative /post-operative risk according to the American Society of Anesthesiologists (ASA) with respect to airway assessment, medical history, allergies, and medications.
- Interview the subject and the subject's family/supporters to discuss the procedure, its risks and possible complications. Answer questions and obtain written informed consent.

Prophylactic antibiotics will be given intravenously according to site standard practice. An initial CT scan may be ordered, if necessary, to evaluate adjacent viscera, renal location, and the presence of renal cysts. A CT scan must be used in conjunction with ultrasound to locate the cortical-medullary junction.

REACT is targeted for injection into the kidney cortex via a needle/cannula and syringe compatible with cell delivery. The intent is to introduce REACT via penetration of the kidney capsule and deposit REACT into multiple sites of the kidney cortex. Initially, the kidney capsule will be pierced using a 15- to 20-gauge trocar /access cannula inserted approximately 1 cm into the kidney cortex.

In the Phase 1 clinical study, REACT was administered via an 18-gauge needle. The proposed Phase 2 study will utilize an 18-gauge or smaller needle for REACT delivery. The needle will be threaded inside the access cannula and advanced into the kidney, from which the REACT will be administered. Injection of the REACT will be at a rate of 1 to 2 mL/min. After each 1-to-2-minute injection, the inner needle will be retracted along the needle course within the cortex to the second site of injection, and so forth, until the needle tip reaches the end of the access cannula or until the entire REACT product has been injected. Using a percutaneous delivery approach, placement of the access cannula /trocar and delivery needle will be performed using direct, real-time imaging. A CT scan must be used in conjunction with ultrasound for the REACT injection procedure.

During the procedure, moderate conscious sedation will be employed; vital signs will be measured continuously. REACT injection will cease if there is imaging evidence of cell extravasation into

central or peripheral renal blood vessels, the medullary portion of the kidney, or through the renal cortex and into the retroperitoneal soft tissues, or evidence of active bleeding.

Following completion of the REACT injection, the inner needle will be withdrawn, and the outer cannula will remain in place for track embolization. During removal of the outer cannula (trocar), the site of the renal cortex puncture and needle track through the retroperitoneum will be embolized with absorbable gelatin particle/pledgets (e.g., Gelfoam® [Pfizer]) or fibrin sealant (e.g., TISSEEL [Baxter]) or other suitable agent to prevent excessive renal bleeding.

Upon completion of the procedure, non-contrast CT scan or ultrasound with color Doppler evaluation will be performed to image puncture site cell injection and any hematoma or bleeding events. The subject will be monitored for 2 to 3 hours post-procedure in a recovery-room environment with nursing assessment and measurement of vital signs. Subjects who do not experience complications may be discharged the same day as REACT injection, consistent with site standard practice.

8. SAFETY ASSESSMENTS AND MANAGEMENT

8.1. Adverse and Serious Adverse Events

8.1.1. Definition of Adverse Events

An AE is the development of an undesirable medical condition (including abnormal laboratory findings) or the deterioration of a pre-existing medical condition following or during exposure to a study treatment, whether or not considered to have a causal relationship with study procedures or the investigational product. A pre-existing condition is a clinical condition (including a condition being treated) that is diagnosed before the subject signs the Informed Consent Form and is documented as part of the subject's medical history. Pre-existing conditions that are stable or unchanged should not be considered AEs.

The Investigator is responsible for ensuring that all AEs observed by the Investigator or reported by the subject that occur from the day of the biopsy procedure through 24 months after the final injection of REACT are monitored and recorded in the subject's medical record as well as the CRF provided by the Sponsor or its designee.

Treatment-emergent adverse events (TEAEs) are defined as any AE that started after the first injection of REACT or started prior to the first injection but increased in severity or frequency after the first injection of REACT.

Unscheduled visits may be performed at any time during the study as judged necessary by the Investigator to assess and conduct follow-up on AEs. Evaluations and procedures to be performed at unscheduled visits will be at the Investigator's discretion in consultation with the Sponsor and may be based on those listed in the Schedule of Events.

The Investigator's clinical judgment is used to determine whether a subject is to be removed from treatment due to an AE.

An AE is considered unexpected if it is not consistent in nature or severity with information contained in the current version of the Investigator's Brochure provided by the Sponsor.

8.1.1.1. Definition of Serious Adverse Events

An SAE is an AE that occurs during any phase of the study (i.e., baseline, treatment, washout, or follow-up) at any dose of the investigational product, comparator, or placebo, and fulfils one or more of the following:

- Results in death
- It is immediately life-threatening
- It requires in-patient hospitalization or prolongation of existing hospitalization
- It results in persistent or significant disability or incapacity
- Results in a congenital abnormality or birth defect
- It is an important medical event that may jeopardize the subject or may require medical intervention to prevent one of the outcomes listed above.

All SAEs that occur from the day of the biopsy procedure, during treatment, or within 24 months following the final REACT injection, whether or not they are related to study procedures or the

investigational product, must be recorded in the subject's medical record as well as the CRF provided by the Sponsor or its designee.

8.1.1.2. Other Significant Adverse Events

Significant events of particular clinical importance include SAEs and AEs leading to premature discontinuation of subjects from the study. These events will be recorded in the subjects' medical records as well as the CRF provided by the Sponsor or its designee. Narratives of these events may be prepared for inclusion in the Clinical Study Report.

The following sections describe "adverse events of special interest" concerning procedure- and product-related events. Subjects should be carefully monitored for the occurrence of these potential AEs.

8.1.1.3. Procedure-Related Events

Post-procedure pain: If the subject experiences pain following the biopsy or REACT injection, administration of paracetamol or paracetamol-codeine combinations is recommended. More severe pain in the loin or abdomen requires ultrasonography to exclude significant perirenal hemorrhage. If severe pain occurs, administration of opiates may be necessary. If analgesic doses higher than the maximum authorized doses are required to alleviate pain, then the Investigator must perform additional clinical evaluations to ascertain the probable cause(s) of excessive pain.

Hemorrhage: Following renal biopsy and REACT injection procedures, subjects undergo regular hemoglobin and blood pressure monitoring. Subjects will be confined to bed and monitored for maintenance of normal coagulation indices. If bleeding occurs and the subject is hypotensive despite bed rest, a blood transfusion may be considered. If the bleeding is still not controlled, surgery may be considered. In rare cases, renal angiography may be performed to identify the source of bleeding. Coil embolization can be performed during the same procedure.

Other complications: In very rare cases, other organs (such as liver, gallbladder and lungs) may be penetrated during the biopsy procedure. In these cases, appropriate treatment and follow- up may be discussed with consulting surgeons.

Death: Deaths resulting from renal biopsies occur in <0.01% of patients.[55,56,57] Adherence to strict inclusion /exclusion criteria will ensure that subjects who may be predisposed to uncontrolled or excessive bleeding will not be enrolled in this trial.

8.1.1.4. Product-Related Events

No REACT product-related events are expected to occur. This assumption is based on the autologous nature of REACT (i.e., REACT is manufactured from renal cells isolated from the same kidney to which they are being returned), the lack of product-related events observed in pre- clinical animal studies, and the lack of specific REACT- related events seen during and following implant of the first 5 subjects in the Phase 1 clinical trial. However, subjects will be extensively monitored throughout the course of the study for any unexpected events.

If an event occurs which is assessed as related to the investigational product, REACT, then the event will be immediately reviewed by the Investigator and Medical Monitor.

8.2. Adverse Event Intensity and Relationship Assessment

8.2.1. Intensity Scale

Intensity will be assessed by the Investigator using the "Common Terminology Criteria for Adverse Events" (CTCAE) version 4.03, from the US National Cancer Institute (refer to http://evs.nci.nih.gov/ftp1/CTCAE/CTCAE_4.03_2010-06-14_QuickReference_8.5x11.pdf).

If the AE is not included in the CTCAE, then the Investigator will determine the intensity of the AE according to the following criteria:

- **Mild (Grade 1)**: The AE is noticeable to the subject but does not interfere with routine activity.
- **Moderate (Grade 2)**: The AE interferes with routine activity but responds to symptomatic therapy or rest.
- **Severe (Grade 3)**: The AE significantly limits the subject's ability to perform routine activities despite symptomatic therapy. Severe events are usually incapacitating.
- **Life-Threatening (Grade 4)**: The subject is at immediate risk of death.
- **Death (Grade 5)**

If the intensity (grade) changes within a day, the maximum intensity (grade) should be recorded. If the intensity (grade) changes over a longer period of time, the changes should be recorded as separate events (having separate onset and stop dates for each grade).

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria under Definition of Serious Adverse Events (See Section 8.1.1.1). Therefore, an AE of severe intensity may not necessarily meet the criteria for seriousness.

8.2.2. Relationship Assessment

An Investigator listed on Form FDA 1572 may make the determination of relationship to the study procedure or investigational product for each AE. The Investigator should judge whether there is a reasonable possibility that the AE may have been caused by the study procedure or investigational product. If no valid reason exists for suggesting a relationship, then the AE should be classified as "not related." If there is any valid reason, even if undetermined, for suspecting a possible cause-and-effect relationship, then the AE should be considered "possibly related" or "related" to the study procedure or investigational product.

Definitions of relatedness categories are:

- **Not Related**: Exposure to the study treatment did not occur, or the occurrence of the AE is not reasonably related in time, or the AE is considered unlikely to be related to the study treatment.
- **Unlikely Related**: The study treatment and the AE were not closely related in time, and/or the AE could be explained more consistently by causes other than exposure to the study treatment product.
- **Possibly Related**: The study treatment and the AE were reasonably related in time, and the AE could be explained equally well by causes other than exposure to the study treatment product.

- **Related**: The study treatment and the AE were reasonably related in time, and the AE was more likely explained by exposure to the study product than by other causes, or the study treatment was the most likely cause of the AE.

For the purpose of safety analyses, all AEs judged by the Investigator to be "possibly related" or "related" will be considered treatment-related adverse events.

8.3. Recording and Reporting Adverse Events

Adverse events spontaneously reported by the subject and/or in response to an open question from study personnel, or revealed by observation, or documented via laboratory reports, imaging reports, consult notes, survey instruments and other data collection tools, will be recorded in the subject's medical records and CRF provided by the Sponsor or its designee.

Adverse events will be reported using standard medical terminology, whenever possible. A clinically significant change in laboratory values or vital signs need not be reported as an AE unless the abnormal change constitutes an SAE and/or leads to discontinuation of treatment or withdrawal from the study.

For each AE, the Investigator will record the start date, the stop date, the intensity of each reportable event, the Investigator's judgment of the relationship to the study procedure or investigational product, the action taken, severity (if applicable), and whether the event resulted in discontinuation of treatment or withdrawal from the study. Follow-up information on any SAE may be requested by ProKidney or its designee.

All SAEs must be promptly reported by the Investigator to ProKidney or its designee within 24 hours from the time when the Investigator first becomes aware of the event. All SAEs must be reported whether or not they are considered causally related to the study procedure or investigational product. SAE forms will be provided to each clinical site.

The information collected will include subject number, a narrative description of the event, and an assessment by the Investigator as to the intensity of the event and relatedness to the investigational product. The Investigator must complete, sign and date the SAE pages, and verify the accuracy of the information recorded against the corresponding source documents. Follow-up information on the SAE may be requested by ProKidney or its designee.

Contact information for reporting SAEs appears below:

Email: CTISafety@ctifacts.com
SAE Hotline: 1877-755-0742
eFax: 1-800-541-1501

ProKidney or its designee is responsible for notifying the relevant regulatory authorities of certain AEs. It is the Principal Investigator's responsibility to notify the IRB /EC of all SAEs that occur at the site.

8.3.1. Serious Unexpected Drug Reactions

If there are serious, unexpected adverse drug reactions (SUSARs) associated with the use of the investigational product, ProKidney or its designee will notify the FDA and all participating Investigators on an expedited basis and in accordance with applicable regulations. It is the responsibility of the Investigator to promptly notify the IRB /EC and other appropriate institutional regulatory bodies of all unexpected serious adverse drug reactions involving risk to human subjects.

8.3.2. Pregnancy

Pregnancy is neither an AE nor an SAE, unless a complication relating to the pregnancy occurs. All reports of congenital abnormalities /birth defects are SAEs. Spontaneous miscarriages should be reported and handled as SAEs. However, elective abortions without complications should not be handled as AEs.

All pregnancies experienced by female subjects enrolled in this study are to be reported in the same time frame as SAEs using the Pregnancy Form of the CRF. The course of all pregnancies, including perinatal and neonatal outcome, regardless of whether the subject has discontinued participation in the study, will be followed until resolution, including follow-up of the health status of the newborn to 6 weeks of age.

The effects of administration of the investigational product on the pregnant female or the developing fetus are unknown. Therefore, female subjects of child-bearing potential who are planning a pregnancy during the course of the study, or who are not using a highly effective method(s) of birth control, or who are unwilling to continue using a highly effective method of birth control throughout the duration of the study are not eligible to participate in the study. (Refer to Exclusion Criterion # 23; Section 5.2).

8.4. Data Safety Monitoring Board

A Data Safety Monitoring Board (DSMB) will be chartered to oversee subject safety, especially as it relates to unexpected investigational product-related events. The DSMB will minimally consist of 3 members who have expertise directly related to protocol-specified activities. It will function independently, and its members will have no other engagement with ProKidney. The DSMB will meet by teleconference at regular intervals, depending on the speed of subject enrollment and the amount of new data generated. The DSMB will advise ProKidney on aspects concerning the safety of subjects participating in the clinical trial. Apart from reviewing study data, the DSMB will consider feedback from the Sponsor and Investigators. The DSMB will share its recommendations with the study centers, Institutional Review Boards /Ethics Committees, and regulatory authorities, as appropriate. Other specific activities and responsibilities of the DSMB will be detailed in the DSMB charter.

8.5. Dose Adjustment Criteria

The dates for the first 3 subjects who are scheduled to receive a second REACT injection under IND 16482 will be staggered at a minimum of 3-week intervals. The DSMB will sequentially review the status of each of these subjects prior to the next subject undergoing his/her second REACT injection. After the third subject has received his/her second REACT injection, the DSMB will meet to assess AEs and other safety parameters obtained to date for these 3 subjects described in the protocol. The DSMB may recommend changes concerning the second REACT injection as well as protocol-specified study evaluations, and/or follow-up procedures.

Apart from reviewing study data, the DSMB will evaluate relevant feedback from the Sponsor and Investigators.

8.6. Stopping Rules for an Individual Subject

The Investigator, Medical Monitor, DSMB, and Sponsor may remove any subject from the study for:

- Any clinical AE, laboratory abnormality, intercurrent illness, other medical condition or situation whereby continued participation in the study would not be in the best interest of the subject.
- Development of any exclusion criterion.

If a subject is terminated from the study, EOS assessments should be conducted at the last visit.

If any of the following events occur, no additional subjects can receive REACT injections until review by the DSMB has been completed:

- An SAE that is rated as severe or life-threatening and, in the judgment of the Investigator, is related to REACT or study procedures
- Death of an enrolled subject
- Similar SAE's in more than one subject that are related to REACT, in the judgment of the Investigator
- Inability to deliver a minimum of 50% of the dose of REACT in more than one subject due to surgical or other issues

8.7. Study Suspension or Study Termination

As long as subjects are enrolled and participating in the study, including follow-up, the Medical Monitor, DSMB, IRB /EC, Sponsor, FDA and other regulatory agencies will review serious, unexpected, procedure- and product-related AEs.

Enrollment in the study will be suspended if any patient develops an apparent tumor, whether in the kidney or at a remote anatomic site. A DSMB review will then determine whether or not study amendment or termination is needed. The study will also be suspended in the event that two or more patients develop a similar unexpected SAE or AEs that are more severe than expected with the study drug or study procedures. A DSMB review will then determine whether or not study amendment or termination is needed. If a decision is made to suspend or terminate the study, the ProKidney will notify the Investigators /sites and ProKidney as well as the IRB /EC and regulatory authorities, as required.

The DSMB, IRB /EC, Sponsor, Investigator(s), FDA and other regulatory authorities, as part of their duties to ensure human subject protection, *may suspend the study at any time* due to concerns for the safety of study subjects. Once the clinical trial is halted (i.e., no new enrollments and no further REACT injections), a report will be submitted to the IRB /EC.

9. STATISTICAL METHODS AND PLANNED ANALYSES

9.1. Sample Size

Up to 10 subjects who complete screening procedures and satisfy all inclusion and exclusion criteria will be enrolled.

Statistical analyses will be primarily descriptive in nature and no statistical hypothesis testing is planned for the study. Unless otherwise specified, continuous variables will be summarized by presenting the number of non-missing observations (n), mean, standard deviation, median, minimum, and maximum. Categorical variables will be summarized by presenting frequency count and percentage for each category.

9.2. Criteria for Evaluation

9.2.1. Analysis Objectives and Endpoints

9.2.1.1. Primary

To assess the safety of REACT injected in one recipient kidney.

- ***Primary Endpoint:*** Procedure and/or product related AEs through 24 months post-injection.

9.2.1.2. Secondary

To assess the safety and tolerability of REACT administration by assessing renal-specific AEs over a 24-month period following injection.

- ***Secondary Endpoints:*** Renal-specific laboratory assessments through 24 months post injection.

9.2.1.3. Exploratory

To assess the impact of REACT on renal function over a 24-month period following injection and on the Quality of Life.

- ***Exploratory Endpoints:*** 1) Clinical diagnostic and laboratory assessments of renal structure and function (including eGFR, serum creatinine, and proteinuria) to assess changes in the rate of progression of renal disease, 2) patient-reported outcomes from the Kidney Disease Quality of Life (KDQOL) and EQ-5D-5L surveys obtained at baseline (i.e., after randomization, but before REACT injection) through 24 months after the last REACT injection.

9.3. Demographic and Baseline Characteristics

Demographic data and baseline characteristics will be summarized via sample size, mean, standard deviation, median, minimum, and maximum for the continuous variables as well as the frequency and proportion for categorical variables. These summaries will be produced for both the full analysis set and the injection analysis set. Demographic and baseline characteristic information will be presented as descriptive statistics; generating inferential statistics is not planned. These data will be provided in a tabular listing.

9.4. Efficacy Analysis

The primary efficacy endpoint is serial measurements of eGFR obtained at 1, 3, 6, 12, 15, 18, 21, and 24 months after the last REACT injection. GFR will be estimated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation that incorporates both serum creatinine and Cystatin C. [1]

Estimated GFR measured at each time point will be summarized by presenting descriptive statistics of raw data and change from baseline values for each treatment group.

9.5. Exploratory Analysis

An exploratory analysis will be conducted to examine potential changes in health-related Quality of Life (HR-QoL).

9.6. Statistical Methods

Subjects will complete the KDQOL-SF™ survey (i.e., Kidney Disease and Quality of Life Short Form). The KDQOL-SF is a 36-item, validated, HR-QoL instrument relevant to patients with kidney disease.[64] This disease-specific, HR-QoL instrument consists of the following subscales:

- The "SF-12 measure of physical (PCS) and mental (MCS) functioning" contains items about general health, activity limits, ability to accomplish desired tasks, depression and anxiety, energy level, and social activities.
- The "Burden of Kidney Disease subscale" contains items about how kidney disease interferes with daily life, takes up time, causes frustration, or makes the respondent feel like a burden.
- The "Symptoms and Problems subscale" contains items about how bothered a respondent feels by sore muscles, chest pain, cramps, itchy or dry skin, shortness of breath, faintness/dizziness, lack of appetite, feeling washed out or drained, numbness in the hands or feet, nausea, or problems with dialysis access.
- The "Effects of Kidney Disease on Daily Life subscale" contains items about how bothered the respondent feels by fluid limits, diet restrictions, ability to work around the house or travel, feeling dependent on doctors and other medical staff, stress or worries, sex life, and personal appearance.

9.7. Safety Analysis

9.7.1. Laboratory Evaluations

Baseline values will be collected immediately prior to REACT injection. Observed and change from baseline laboratory data will be summarized via sample size, mean, standard deviation, median, minimum, and maximum for the continuous variables as well as frequency and proportion for the categorical variables. Laboratory abnormalities will be defined using the NCI CTCAE grading scheme. Abnormal laboratory values will be flagged as above or below the normal range. The results of laboratory testing for renal function, specifically sCr, BUN, and urinary albumin, are of particular interest for this study.

The incidence of treatment-emergent laboratory abnormalities, defined as values that increase at least one toxicity grade between baseline and any time post-baseline up to six months following REACT injection, will be summarized. If baseline data are missing, then the latest value between

biopsy and injection will be used as the baseline value. If baseline and pre-injection data are missing, then any graded abnormality (i.e., at least a Grade 1) will be considered as treatment-emergent. These values will be summarized for both the full analysis set and the injection analysis set. Observed and change from baseline values will be presented as descriptive statistics; generating inferential statistics is not planned. These data will be provided in a tabular listing.

9.7.2. Adverse Events

Clinical and laboratory AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) version 18.1 by System Organ Class (SOC) and Preferred Term (PT). Adverse events will be graded using the CTCAE version 4.03 from the US National Cancer Institute.

A treatment-emergent AE will be defined as any AE that started after the first injection of REACT or started prior to the first injection but increased in severity or frequency after the first injection of REACT. Summaries (frequency and proportion) of treatment-emergent AE's will be presented by SOC and PT. Additional summaries will include, but are not limited to, treatment- emergent AE judged to be related to the procedure and/or investigational product, intensity, reason for subject withdrawal, SAEs, and deaths. The numbers of events (occurrence) and the number of subjects (incidence) who experienced treatment-emergent AEs will be reported by treatment group. Adverse event data will be provided in a data listing.

9.7.3. Other Safety Evaluations

Change from baseline for vital signs will be calculated for each subject and provided in a data listing. The number and percent of subjects who exhibit change(s) in their physical examinations (such as from normal to abnormal) will be summarized via a data listing. The number and percent of subjects who develop abnormal heart rhythms or QT-interval prolongation during the study will be provided in a data listing. Data from medical history, concomitant medications, ultrasound, renal scintigraphy, and MRI assessments will be provided in a data listing.

Descriptive statistics for these evaluations will be generated as warranted.

9.8. Biopsy and REACT Injection(s)

Biopsy and REACT injection data will be provided in a data listing.

10. ETHICAL AND REGULATORY CONSIDERATIONS

10.1. Good Clinical Practice

This study will be conducted in compliance with the protocol, in accordance with ICH E6 Harmonized Tripartite Guideline (ICH-GCP), in general agreement with the most recent version of the Declaration of Helsinki, and in accordance with all applicable United States and European regulations.

10.2. Delegation of Principal Investigator Responsibilities

The Investigator will ensure that all persons assisting with the trial are adequately informed about GCP requirements, the protocol, any amendments to the protocol, the study treatments as well as their trial-related duties and functions. The Investigator will maintain a list providing the names, positions, signatures, and initials of sub-investigators as well as other appropriately qualified personnel, including those authorized to make entries and corrections on the CRF.

10.3. Institutional Review Board / Ethics Committee

The final study protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and patient-reported questionnaires/surveys (if applicable), including respective version dates, must be approved or given a favorable opinion in writing by an IRB /EC as appropriate. A copy of the written approval must be provided to the Sponsor. This documentation should clearly mention the approval/favorable opinion of the protocol, the subject Informed Consent Form, and subject recruitment materials (if applicable), and patient-reported questionnaires/surveys (if applicable) along with the respective version dates. The written approval and a list of current membership, or Department of Health and Human Services (DHHS) Assurance Number, or letter from the IRB /EC stating that the membership list is on file, must be provided to the Sponsor prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB /EC has direct participation in this clinical trial, written notification regarding his or her abstinence from voting must also be obtained.

The Investigator must inform the IRB /EC of any amendment to the protocol, provide updates about the ongoing trial at intervals (at least annually) specified by the respective IRB /EC, and submit final study reports to the IRB /EC. In addition, the IRB /IEC must approve all advertising used to recruit subjects for the study along with any written information to be provided to subjects (e.g., diaries, calendars, patient-reported surveys) and updates to the Informed Consent Form.

It is the Investigator's responsibility to notify the IRB /EC of all SAEs that occur at his or her site. The Sponsor is responsible for notifying the relevant regulatory authorities of certain safety events, including unexpected, serious, drug-related adverse reactions that occur during the clinical study. The Investigator is responsible for notifying its IRB /EC of these unanticipated SAEs.

Initial IRB approval, and all materials approved by the IRB for this study including the subject consent form and recruitment materials must be maintained by the Investigator and made available for inspection.

10.3.1. Subject Informed Consent

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continuing throughout the individual's study participation. The Investigator will ensure that potential participants (and their families) are given full and adequate oral and written

information about the nature, purpose, possible risk(s) and benefit(s) of the study. IRB-approved consent forms explaining study procedures and the investigational product /intervention(s) will be provided. The Investigator will explain the research study and answer any questions that may arise. Potential participants should have sufficient time to discuss the study, ask questions, and process the information during the consent process before deciding whether or not to participate.

The rights and welfare of potential subjects will be protected by emphasizing that the quality of their medical care will not be adversely affected should they decide not to participate in the research study. Those individuals who agree to enroll must sign and date the current version of the Informed Consent Form prior to starting any study-related procedures, including screening. Subjects may withdraw consent at any time throughout the course of the clinical trial without penalty or prejudice toward future medical care.

The acquisition of informed consent will be documented in the participant's medical records, as required by 21 CFR 312.62. The Informed Consent Form will be signed and personally dated by the participant and the person who conducted the informed consent discussion. A copy of the signed Informed Consent Form must be given to the subject while the original, signed Informed Consent Form must be retained by the Investigator.

10.4. Subject Confidentiality

Individual subject medical information obtained as a result of this study is considered confidential and disclosure to third parties is prohibited. Such medical information may be given only after approval of the subject to his/her physician or to other appropriate medical personnel responsible for the subject's well-being. The Sponsor affirms the subject's right to protection against invasion of privacy. Only a subject identification number and/or initials (where allowed by local or national regulations) will identify subject data retrieved by the Sponsor. However, ProKidney requires the Investigator to permit the Sponsor, its designated representative(s), the IRB /EC, and when necessary, representatives of the regulatory health authorities to review and/or to copy any medical records relevant to the study.

10.5. Substantial Amendment to the Protocol

A substantial amendment to the protocol must be agreed upon in writing by the Sponsor, then submitted to and approved by the respective regulatory authority before the amendment can be implemented. It is the responsibility of ProKidney or its designee to ensure compliance with the appropriate regulatory requirements.

Written approval of a protocol amendment is not required prior to implementation of changes that eliminate an immediate hazard to the study subject; however, approval must be obtained as soon as possible thereafter. Any protocol amendment also must be signed by the Investigator, who will provide a copy to the IRB /EC.

10.6. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be attributed to the subject, the Investigator, or the site staff. All protocol deviations will be documented and reported by the monitor during the course of the study as described in the Study Monitoring Plan. ProKidney will work with the site to develop and implement any corrective actions to address protocol deviations, as appropriate.

Serious breaches of the protocol that are likely to significantly affect the safety of a subject or the integrity of the data generated must be reported to the Sponsor, IRB /EC, and regulatory authorities.

Although, in principle, no deviations from or changes to the protocol are permitted, under emergency circumstances protocol deviations may proceed without prior approval from ProKidney, IRB /EC and regulatory authorities to protect the rights, safety, and well-being of study subjects.

11. DATA HANDLING AND RECORDKEEPING

11.1. Data Collection and Review

11.1.1. Data Collection

ProKidney or its designee will provide the clinical sites with access to CRFs for each subject. A CRF must be completed for every subject who provides written, informed consent and has undergone at least one protocol-specified, study-specific assessment. CRF completion instructions will be provided to the site in the Study Reference Manual. The Investigator or designated representative should complete the CRFs as soon as possible after the information has been collected. The Investigator is responsible for ensuring the accuracy, completeness, and timeliness of the data reported in a subject's CRF. Source documentation supporting the CRF data should indicate the subject's participation in the study and document the dates and details of study procedures, AEs, and subject status.

When a subject completes the study, the Investigator must review and sign the CRF indicating that he/she has reviewed the completed CRF and pertinent clinical data for that subject and that, to the best of his/her knowledge, all data recorded in the CRF accurately reflect the subject's clinical performance in the study.

11.1.2. Study Monitoring

Before an investigational site can enter a subject into the study, a representative of ProKidney or its designee will visit the investigational study site to:

- Determine the adequacy of the facilities.
- Discuss with the Investigator(s) and other personnel their responsibilities under the protocol as well as the responsibilities of ProKidney. This will be documented in a Clinical Study Agreement between ProKidney and the Investigator.

During the study, a study monitor appointed by ProKidney or its designee will have regular contacts with the site and conduct on-site monitoring visits as described in the Study Monitoring Plan. Monitoring and auditing procedures, approved by the Sponsor, will ensure that the study is conducted in accordance with the protocol and regulatory requirements while ensuring the safety of all study subjects. Apart from on-site visits, the monitor will maintain frequent communication with the Investigator and site personnel via letters, e-mails, telephone, and FAX.

Duties of the monitor include, but are not limited to:

- on-site review of the CRFs for completeness and clarity
- confirmation that the facilities remain acceptable
- confirmation that investigational product accountability checks are being performed
- confirmation that AEs and SAEs are properly documented in the CRFs, and that any SAEs have been forwarded to ProKidney or its designee and the IRB /EC
- recording and reporting of any protocol deviations not previously sent to ProKidney, the IRB /EC, and regulatory authorities, as required
- clarification of administrative matters

The monitor will perform source data verification, including a comparison of the data in the CRF with the subject's medical records at the hospital or clinic, and other records relevant to the study. Source data verification requires direct access to all original records for each subject. The review of medical records will be performed in a manner that ensures subject confidentiality.

Regulatory authorities and the IRB /EC may request access to all source documents for on-site inspection or an audit. Likewise, the Sponsor or its designee may conduct a quality assurance to ensure compliance with GCP and all applicable regulatory requirements. Direct access to source documents must be guaranteed by the Investigator, who will provide support at all times for these activities.

11.2. Audits and Inspections

The purpose of an audit or inspection by the Sponsor, regulatory authorities, the IRB /EC, or other appropriate institutional bodies is to systematically and independently examine all study-related activities and documents to determine whether these activities were conducted, and the data were recorded, analyzed, and accurately reported, in accordance with the protocol, ICH-GCP, and any applicable regulatory requirements. The Investigator and the site /institution must provide support at all times for on-site audits or inspections by providing direct access to all source documents, CRFs, and other study documentation. The Investigator must notify ProKidney or its designee immediately if contacted by a regulatory agency about an inspection.

Additionally, the Sponsor or its designee will be allowed to conduct site visits to the investigational facilities for the purpose of monitoring any aspect of the study. The Investigator agrees to allow the monitor to inspect the drug storage area, study drug stocks, drug accountability records, subject charts, study source documents, and other records relative to study conduct.

11.3. Retention of Records

The Investigator agrees to keep records and those documents that include (but are not limited to): the identification of all participating subjects; medical records; study-specific source documents; source worksheets; all original signed and dated Informed Consent Forms; records of any body fluids or tissue samples retained; query responses; and, detailed records of investigational product accountability to enable evaluations or audits from regulatory authorities, the Sponsor, or its designees.

These documents are to be retained until at least two years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region, or at least two years have elapsed since the formal discontinuation of clinical development of the investigational therapy. If the Investigator cannot meet this obligation, he/she must ask the Sponsor for permission to make alternative arrangements; details of these arrangements should be documented. If the Investigator withdraws from the responsibility of retaining the study records, custody must be transferred to a person willing to accept the responsibility. ProKidney must be notified in writing if a custodial change occurs.

12. QUALITY CONTROL AND QUALITY ASSURANCE

Independent quality assurance (QA) and clinical quality control (QC) systems are implemented and maintained using written standard operating procedures (SOPs) to ensure that the trial is conducted, and data are generated, documented (recorded), and reported in compliance with the protocol, ICH-GCP, and the applicable regulatory requirements.

To ensure compliance with Good Clinical Practices and all applicable regulatory requirements, ProKidney may conduct a quality assurance audit. Refer to Audits and Inspections (Section 11.2) for more details regarding the audit process.

13. PUBLICATION POLICY

All information regarding the investigational therapy is the confidential property of ProKidney. The Investigator agrees to use this information to conduct the study and will not use it for other purposes without written approval from the Sponsor. It is understood that there is an obligation to provide ProKidney with complete data obtained during the study. ProKidney retains full rights over any invention, discovery, or innovation, patentable or not, that may occur during the conduct of the clinical trial.

It is anticipated that the results of this study will be presented at scientific meeting(s) and/or published in a peer-reviewed journal(s). A Publications Committee, composed of Investigators participating in the study and representatives from the Sponsor, will oversee the publication and presentation of study results, which will reflect the experience of all participating clinical sites.

The International Committee of Medical Journal Editors has adopted a trials-registration policy requiring that all clinical trials be registered in a public database (such as ClinicalTrials.gov and clinicaltrialsregister.eu) as a condition for publication in member journals. It is the responsibility of ProKidney to register the present study in an acceptable clinical trial registry on or before the onset of subject enrollment.

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A PHASE II, OPEN-LABEL, SAFETY AND TOLERABILITY STUDY OF A RENAL AUTOLOGOUS CELL THERAPY (REACT) IN PATIENTS WITH TYPE 2 DIABETES AND CHRONIC KIDNEY DISEASE (REGEN-003)

Summary of Changes from Protocol Version 1.6 (05MAR2019) to Version 1.7 (12JAN2021)

Administrative and formatting changes are not highlighted below but can be found in the "track-changes" version of the protocol.

Protocol Section	Original Text	Updated Text	Reason for Change
Throughout	V 1.6 05MAR2019	V 1.7 12JAN2021	Updated protocol version and date as needed.
Throughout	10 Market Street, #688 Camana Bay Grand Cayman, Cayman Islands KY1-9006	8020 Arco Corporate Drive, Suite 118 Raleigh, NC 27617	Updated Sponsor address as needed.
Throughout	inRegen	ProKidney	Updated Sponsor Name as needed.
Throughout	Twin City Bio LLC or Twin City Bio	ProKidney, LLC or ProKidney	Updated Manufacturer Name as needed.
Throughout	NKA	REACT	Updated product name as needed
Throughout	Autologous Neo-Kidney Augment	Renal Autologous Cell Therapy	Updated product name as needed
Section 6 Study Visits		Due to the COVID-19 pandemic that began in March 2020, subject follow-up visits may be conducted remotely or via home health services to ensure the safety of the subject. Any procedures listed in the schedule of events that are not completed due to COVID-19 pandemic restrictions will be documented as protocol deviations. Clinical laboratory evaluations are permitted to be completed by a local lab convenient for the subject. Results of all evaluations performed by local laboratories will be collected by ProKidney.	Updated collection of laboratory assessments to allow for more flexibility due to the pandemic.