The RENAL LIFECYCLE trial

A Randomized Controlled Clinical Trial to Assess the Effect of Dapagliflozin on Renal and Cardiovascular Outcomes in Patients with Severe Chronic Kidney Disease

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DOCUMENT HISTORY

Document	Date of version	Summary of Changes
Protocol version no. 10	11-Mar-2024	Updated to CTR protocol format; Additional safety laboratory visit for participants if participant has stopped IMP longer than 28 days after receiving kidney transplantation; Clarification of urine sample collection (morning void vs 24u urine); Clarifying timing of re-start IMP for participants whom have received a kidney transplant; Removal of exploratory cost-effectiveness endpoint.
Original protocol version no. 9	19-Oct-2023	Not applicable

CONFIDENTIALITY STATEMENT

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1. ABBREVIATIONS

AE	Adverse Event
AEoSI	Adverse event of specific interest
AR	Adverse Reaction
ATMP	Advanced Therapy Medicinal Product
AxMP	Auxiliary Medicinal Product
AZ	AstraZeneca
CA	Competent Authority
CCMO	Centrale Commissie Mensgebonden Onderzoek (Central Committee on Research Involving Human Subjects)
CKD	Chronic Kidney Disease
CRA	Clinical Research Associate
CRF	Case Report Form
CRO	Contract Research Organisation
CRP	C-Reactive Protein
CT	Clinical Trial
CTA	Clinical Trial Authorisation
CV	Curriculum Vitae
DEA	if the AE is the reason for discontinuation from IMP
DSMB	Data Safety Monitoring Board
EC	Ethics Committee
e-CRF	Electronic Case Report Form
eGFR	Estimated Glomerular filtration rate
EQ-5D	The Euroqol study group quality of life questionnaire
EU	European Union
EMA	European Medicines Agency
GCP	Good Clinical Practice
GDPR	General Data Protection Regulation
IB	Investigator's Brochure
IC	Informed Consent
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
IMP	Investigational Medicinal Product
IMPD	Investigational Medicinal Product Dossier
MS	Member State
NSAIDs	Nonsteroidal anti-inflammatory drugs
NT-proBNP	N-Terminal Prohormone of Brain Natriuretic Peptide
PI	Principal Investigator
QoL	Quality of Life
RSI	Reference Safety Information
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SGLT2	Sodium Glucose Transport Inhibitor 2
SmPC	Summary of Product Characteristics
SOP	Standard Operating Procedure

Sponsor	The sponsor is the party that commissions the organization or performance of the research, for example a pharmaceutical company, academic hospital, scientific organization or investigator. A party that provides funding for a study but does not commission it is not regarded as the sponsor but referred to as a subsiding party.
SUSAR	Suspected Unexpected Serious Adverse Reaction
UACR	Urine Albumin-to-Creatinine ratio
UAVG	Dutch Act on Implementation of the General Data Protection Regulation; in Dutch: Uitvoeringswet AVG
WMO	Medical Research Involving Human Subjects Act; in Dutch: Wet Medisch-wetenschappelijk Onderzoek met mensen
WOCPB	Women of Childbearing Potential

2. SYNOPSIS

Short title:	The RENAL LIFECYCLE trial
Full protocol title:	A Randomized Controlled Clinical Trial to Assess the Effect of Dapagliflozin on Renal and Cardiovascular Outcomes in Patients with Severe Chronic Kidney Disease
EudraCT number:	2021-005446-15
EU trial number:	2023-508389-13

Rationale

Sodium glucose co transporter 2 (SGLT2) inhibitors are a relatively new class of agents, originally developed as oral antihyperglycemic drugs. SGLT2 inhibitors are clinically available since 2012 for the treatment of patients with diabetes mellitus type 2. Later, SGLT2 inhibitors appeared to have also specific reno- and cardioprotective effects. Remarkably, the trials that have been performed thus far excluded patients with an eGFR below 25 mL/min/1.73m2 at inclusion, prevalent dialysis patients, and kidney transplant recipients. This is unfortunate, because especially these patients are at high risk of reaching kidney failure requiring dialysis, cardiovascular complications and mortality, whereas there are only few proven effective therapies. There is emerging evidence from experimental studies and post hoc-analyses of randomized clinical trials that SGLT2 inhibitors may also be effective in preventing cardiovascular and mortality outcomes in these patients with severe CKD, including patients receiving dialysis or living with a kidney transplant. For instance, subgroup analysis of the DAPA-CKD trial comparing 624 patients with an eGFR<30 to the remainder of the trial population with better kidney function, demonstrated that the efficacy of the SGLT2 inhibitor dapagliflozin in reducing cardiovascular, heart failure and renal outcomes persisted in the population with impaired kidney function. Furthermore, in the DAPA-CKD trial patients continued to use dapagliflozin or placebo when dialysis was initiated. In the subgroup of patients who initiated dialysis, dapagliflozin was associated with a relative risk reduction for mortality of 21%. Finally, in kidney transplant recipients, SGLT2 inhibitors have been shown to be effective in lowering HbA1c, body weight, blood pressure and stabilize kidney function, and these agents were well tolerated and safe. Taken these findings together there is a sound rationale to study the long-term reno- and cardioprotective efficacy and safety of SGLT2 inhibitors in patients with severe CKD.

Novel aspects

A unique patient population with severe chronic kidney disease at very high risk of adverse outcomes for whom very few proven effective therapies exist. The initiation and efficacy of SGLT2 inhibitors, including dapagliflozin, has not been studied before in this population. However, they have the potential to be very effective and well tolerated which would imply a major advance in the pharmacotherapy of these patients.

When patients reach a renal endpoint, e.g. start of chronic dialysis or receiving a kidney transplantation, study medication will not be stopped as customary in many other trials in nephrology, but continued in the new phase of their “renal lifecycle”, hence the name of the trial.

Objective

To establish the reno- and cardioprotective efficacy and safety of dapagliflozin in patients with severe CKD

Main trial endpoint

Combined endpoint of all-cause mortality, kidney failure[1], and hospitalization for heart failure in the overall study population[2]

Secondary trial endpoints

Incidence in the overall study population of:

- All-cause mortality
- Kidney failure requiring kidney replacement therapy (chronic dialysis, kidney transplantation, or death due to kidney failure)
- Hospitalization for heart failure

Incidence of the composite outcome in:

- Patients with eGFR ≤25 mL/min/1.73m^2 (not on dialysis and not living with a kidney transplant)
- Patients on dialysis[3] with a residual diuresis ≥500 mL/24h
- Patients with a kidney transplant and an eGFR ≤45 mL/min/1.73m^2

Safety outcomes:

SAEs, AEs of specific interest and (S)AEs leading to drug discontinuation

Exploratory outcomes:

These will include among others quality of life as measured with the EQ-5D and SF-12 of SGLT2-inhibition in the overall study population as well as in the three subpopulations.

Trial design

This is a multicenter, randomized, controlled, double blinded, pragmatic, interventional trial

Study duration:

The study consists of a 30 month recruitment phase and an 18 month follow-up after enrollment of the last patient. Total study duration intended to last 48 months. It should be noted that the trial is event driven and will be terminated when 468 primary composite outcomes have occurred. The exact trial duration may therefore be shorter or longer than the intended 48 months.

Study visits:

Screening, baseline, week 2, month 3, month 6 and every 6 months thereafter. Information needed for the trial will be obtained as much as possible from visits taking place as part of routine clinical care.

[1] Kidney failure defined as start of chronic dialysis or kidney transplantation, or death due to kidney failure. Death due to kidney failure is defined as death due to kidney failure because dialysis treatment / a kidney transplant was deliberately withheld, not started or discontinued for any reason. Death due to kidney failure will be adjudicated by the Clinical Endpoint Adjudication Committee

[2] For dialysis patients the primary outcome is restricted to the combined endpoint of all-cause mortality and hospitalization for heart failure

[3] Dialysis includes hemodialysis as well as peritoneal dialysis

Trial population

- Patients with advanced CKD, i.e. an eGFR ≤25 mL/min/1.73m^2
- Patients on dialysis with a residual diuresis ≥500 mL/24h (at least 3 months after start of dialysis)
- Patients with a kidney transplant and an eGFR ≤45 mL/min/1.73m^2 (at least 6 months after transplantation)
- Participants aged ≥18 years and willing to sign informed consent

Interventions

Dapagliflozin 10 mg/day or matching placebo

Ethical considerations relating to the clinical trial including the expected benefit to the individual subject or group of patients represented by the trial subjects as well as the nature and extent of burden and risks

The efficacy and safety of dapagliflozin has been established in multiple studies including more than 25,000 patients with type 2 diabetes mellitus and in patients with CKD with and without type 2 diabetes mellitus. Overall dapagliflozin is very well tolerated, with few side effects besides urinary tract and genital infections. Two independent clinical trials have demonstrated that dapagliflozin does not change HbA1c or induce hypoglycemia in participants without diabetes mellitus. In addition, in patients with CKD dapagliflozin did not cause diabetic keto-acidosis.

The trial is a pragmatic, randomized, controlled, clinical trial. Patients have to visit the clinic for a screening visit. Eligible patients will then visit the clinic at the randomization visit, week 2, month 3, month 6, and every 6 months thereafter. This frequency of clinic visits aims to avoid visits beyond those needed for routine clinical care, and also data collection for the trial will be done as much as part of routine clinical care to minimize burden for patients.

The expected time investment for patients is 1 hour or less per patient visit. Patients receive no priority in treatment of other diseases in the clinic during this study. Participation in this study is on a free-will base.

3. INTRODUCTION AND RATIONALE

3.1 Therapeutic condition and current treatment status

Chronic kidney disease (CKD) affects approximately 10% of the adult population worldwide. The most common causes of CKD are diabetes, hypertension, and chronic glomerulonephritis. Subjects with CKD are at high risk for various complications, among others cardiovascular morbidity and mortality, heart failure, and end-stage kidney disease requiring kidney replacement therapy.[1,2] Treatment for CKD encompasses tight blood pressure control, preferably with angiotensin converting enzyme inhibitor (ACE-I) or angiotensin II receptor blockers (ARBs) as well as a tight glucose control in diabetic patients to prevent or delay progression of CKD and CVD. These interventions have been proven efficacious, but still the residual risk to develop cardiovascular complications and to reach kidney failure remains high.[3] There is therefore a need for additional interventions.[4]

3.2 Mechanism of action, drug class

The sodium glucose co-transporter 2 is located in the first segment of the proximal tubule and is major transporter responsible for glucose reabsorption in the kidney.[5,6] SGLT2 inhibitors are highly selective and reversible inhibitors of this transporter. Various SGLT2 inhibitors such as dapagliflozin, empagliflozin, and canagliflozin are now available for clinical use. The mechanism of action of SGLT2 inhibitors is that these drugs cause direct and insulin independent elimination of glucose by the kidneys, which results in reduced blood glucose levels in type 2 diabetes patients. In addition, dapagliflozin has a mild diuretic and natriuretic effect which results in a reduction in body weight and blood pressure, and increase in haematocrit.[7] Phase 2/3 clinical studies have also shown that dapagliflozin reduces albuminuria, an important risk marker of renal and cardiovascular disease progression.[8,9]

3.3 Clinical trial rationale

The cardiovascular efficacy and safety of SGLT2 inhibitors has been established in two dedicated kidney outcome trials recruiting more than 4000 patients in each trial. The first trial demonstrated that in adult patients with type 2 diabetes and CKD (eGFR 30-90 mL/min/1.73m^2), the SGLT2 inhibitor canagliflozin reduced the risk of renal and cardiovascular outcomes.[10] The second trial, DAPA-CKD, was designed to assess the efficacy and safety of dapagliflozin in patients with CKD with and *without* type 2 diabetes.[11] The trial enrolled adult patients with an estimated glomerular filtration rate between 25 and 75 mL/min/1.73m^2 and a urinary albumin:creatinine ratio between 200 and 5000 mg/g. After a median of 2.4 years follow-up, the trial was terminated early by the independent data monitoring committee for overwhelming efficacy. Dapagliflozin compared to placebo treatment reduced the primary outcome of 50% eGFR decline, end-stage kidney disease, renal or cardiovascular death by 39%. In addition, dapagliflozin reduced the risk of the composite endpoint of heart failure hospitalizations or cardiovascular death by 29% and it reduced the risk of all-cause mortality by 31%. These effects were present both in patients with and without type 2 diabetes. Interestingly, dapagliflozin, although developed as a glucose lowering agent, only reduced HbA1c compared to placebo by 0.1% in patients with type 2 diabetes, whereas it did not reduce HbA1c in patients without type 2 diabetes. These data highlight that other mechanisms account for the beneficial effects of dapagliflozin on clinical outcomes. Other proposed mechanisms include reduced intra-glomerular pressure through an enhanced tubuloglomerular feedback mechanism,[12] reduced glucose and sodium transport by proximal tubular cells,[13,14] increased natriuresis,[7] and reduced systemic blood pressure. Interestingly, there is also a theoretical rationale for an improved oxygen availability in the impaired kidney during dapagliflozin treatment. This could potentially be achieved via two mechanisms.

Firstly, an improved oxygen delivery due to a small but consistently observed haemo-concentration and secondly through a reduced renal oxygen consumption due to a reduced energy requirement from the potassium sodium pump situated on the basolateral membrane striving to retain the sodium gradient over the tubular cells.[15,16] Finally, there are emerging theories around metabolic benefits connected to a mild ketosis induced by SGLT2 inhibition.[17] Based on the CREDENCE and DAPA-CKD trials, canagliflozin and dapagliflozin have been approved for the treatment of chronic kidney disease (canagliflozin in patients with type 2 diabetes) by regulatory agencies. The recent international KDIGO clinical guidelines recommend the use of SGLT2 inhibitors for the treatment of chronic kidney disease due to type 2 diabetes in patients with eGFR ≥ 30 mL/min/1.73m^2. Start of SGLT2 inhibitors is, however, not indicated in patients with lower kidney function, patients receiving dialysis, or kidney transplant patients since these patients were not included in most trials and the long-term efficacy and safety of SGLT2 inhibitors including dapagliflozin, are not established in these patients.

There is emerging evidence that SGLT2 inhibitors may be effective in preventing cardiovascular and mortality outcomes in patients with severe CKD including patients receiving dialysis and kidney transplantation. In these patients with a reduced number of functioning nephrons, it may be expected that SGLT2 inhibitors have less efficacy, because the number of proximal tubules containing SGLT2 transporters is reduced. There is, however, evidence that SGLT2 inhibitors may exert other extra-renal benefits. An experimental study using serum of patients with severe CKD showed that the uremic milieu impairs endothelial cell biology and alters cardiomyocyte function. This deleterious effect was blunted with the SGLT2 inhibitor empagliflozin.[18] Since this study was performed in an in-vitro system, it suggests that empagliflozin exerts direct effects on endothelial-cardiac tissues which are unlikely mediated by the SGLT2 transporter in the proximal tubule of the kidney. Of note, cardiomyocytes express SGLT1 but not SGLT2. Direct effects on cardiac tissue should then be mediated via non-SGLT2 mechanisms. Interestingly, in vitro data have suggested that SGLT2 inhibitors inhibit the sodium-hydrogen exchange transporter (NHE1) in cardiac tissues through binding to the extracellular Na^+ binding site of NHE1 resulting in a reduction in cytosolic Na^+ levels and decreased Ca^{2+} concentration Reduced cytosolic Ca^{2+} can lead to improved cardiac function. Moreover, in constant infused Langendorff-perfused mouse hearts, empagliflozin and canagliflozin were reported to induce coronary vasodilation, an effect independent of SGLT2 transporter inhibition.[19] These SGLT2 transporter independent benefits are supported by an artificial intelligence study to evaluate cellular mechanisms of SGLT2 inhibition.[20] The conclusion of this study was that inhibition of NHE1, potentially through restoration of the antiapoptotic activity of X-linked inhibitor of apoptosis (XIAP) and baculoviral IAP repeat-containing protein 5 (BIRC5) may explain the heart failure protective effects of SGLT2 inhibitors. Finally, in a porcine model of heart failure SGLT2 inhibitors have been shown to improve cardiac energetics, possibly through extra-renal direct mechanisms on the heart.[21] These studies collectively suggest that even in the setting of minimal diuresis (and thus little disposition of SGLT2 inhibitors to the transporter as expected in dialysis patients) SGLT2 inhibitors may still exert favorable effects on cardiac function.

There is also clinical evidence that supports the hypothesis that SGLT2 inhibitors reduce dialysis, heart failure and mortality in patients with eGFR below 30 mL/min/1.73m^2. A subgroup analysis of the DAPA-CKD trial among 624 patients with eGFR<30 (the majority having an eGFR between 25 and 30 mL/min/1.73m^2) demonstrated that the efficacy of the SGLT2 inhibitor dapagliflozin in reducing clinical outcomes persisted in this population.[22] Furthermore, in the DAPA-CKD trial patients continued to use dapagliflozin or placebo when dialysis was initiated. In the subgroup of patients who initiated dialysis, dapagliflozin was associated with a relative risk reduction for mortality of 21%.[*data on file]*

In addition, in kidney transplant recipients, SGLT2 inhibitors have been shown to be effective in lowering HbA1c, body weight, blood pressure and stabilize kidney function, and are well tolerated as shown in a meta-analysis involving 8 clinical studies and 132 patients. However, eGFR was above 60 in the majority of kidney transplant recipients and the efficacy and safety of SGLT2 inhibitors has yet to be established in kidney transplant recipients with more severe CKD.[23]

3.4 Hypothesis

We hypothesize that dapagliflozin reduces clinical outcomes (all-cause mortality, kidney failure, and heart failure hospitalizations) in patients with stage 4 or 5 CKD, dialysis patients, and kidney transplant recipients with and without type 2 diabetes.

3.5 Rationale for study population

This is a randomized, double-blind, parallel-group study. Randomization and double blinding will minimize potential bias. A parallel group design was chosen because a crossover study cannot assess major clinical outcomes. The study will be multicenter in various geographic regions (Netherlands, Belgium, Australia and Germany) to provide a wide applicability of results.

The study population chosen for this study is a broad population of patients with severe CKD. Three strata of patients will be included: Patients with an eGFR≤25 mL/min/1.73m^2 (not on dialysis or living with a kidney transplant); prevalent dialysis patients with residual diuresis ≥500 mL/24-hour (including hemo- and peritoneal dialysis), and kidney transplant recipients. These patients are nearly always excluded from clinical trials while they are at very high risk of adverse outcomes and few effective therapies are available for these patients. However, patients with type 1 diabetes or patients with a life-expectancy less than 6 months in the opinion of the treating clinician or a scheduled start of dialysis within 3 months or kidney transplantation within 6 months will be excluded.

3.6 Rationale for primary outcome measure

The primary outcome measure is the incidence of a composite of all-cause mortality, kidney failure, and heart failure hospitalization. This is a clinically relevant outcome measure and previous trials with dapagliflozin in patients with earlier stages of CKD than enrolled in the current trial have shown that dapagliflozin reduces the incidence of each of these outcomes.

3.7 Rationale for dose regimen/dose justification

The marketed dose, 10 mg of dapagliflozin, has been demonstrated to be well tolerated and effective for the treatment of CKD in patients with and without type 2 diabetes. In a trial of patients with heart failure and reduced ejection fraction, dapagliflozin 10 mg was also effective and safe. Dapagliflozin is metabolized by the liver to inactive 3-O-glucuronide, and this inactive metabolite is eliminated by the kidneys into urine, with a small part via feces. From a pharmacokinetics (PK) and pharmacodynamics perspective, dapagliflozin 10 mg is therefore appropriate for use in patients with kidney function impairment and dose adjustment in case of (severely) impaired kidney function is therefore not needed.

4. STRUCTURED RISK ANALYSIS

4.1 Potential issues of concern

4.1.1 a. Level of knowledge about mechanism of action

The sodium glucose co-transporter 2 (SGLT2), located in the proximal tubule of the kidney, is an effective transporter system which is responsible for the nearly complete reabsorption of glucose in order to maintain appropriate glucose levels. Each glucose molecule that is reabsorbed is accompanied by reabsorption of a sodium molecule in a 1:1 ratio. SGLT2 inhibitors reversely inhibit the SGLT2 transporter which leads to enhanced glucose and sodium excretion and a reduction in plasma glucose and HbA1c. The effects of dapagliflozin on the SGLT2 transporter are well characterized and sufficient knowledge is available about the mechanisms of action.

4.1.2 b. Previous exposure of human beings

Previously, twelve phase 3 randomized controlled clinical trials were conducted involving more than 6000 patients with type 2 diabetes mellitus of whom ~4000 were treated with dapagliflozin. Eleven studies were 24-weeks in duration with extension in 6 studies up to 78 weeks. One study was 52 weeks in duration with extension of another 52 weeks.

More recently, the DAPA-CKD trial reported the efficacy and safety of dapagliflozin in 4,304 patients with chronic kidney disease with or without type 2 diabetes. The trial demonstrated that dapagliflozin significantly reduced the risk of kidney and cardiovascular endpoints and prolonged survival. Dapagliflozin was well tolerated in keeping with the established safety profile. Specifically, in this population there was no diabetic ketoacidosis with dapagliflozin. In patients with CKD without type 2 diabetes no major hypoglycemic events were reported.

4.1.3 c. Induction of the mechanism in animals and/or *ex-vivo*

Various animal models and cell lines are available to study the effect of SFGLT2 inhibitors in more detail at a tissue/cell level.

4.1.4 d. Selectivity of the mechanism

SGLT2 is highly localized to the kidney and major effects are expected to be related to glycosuria and natriuresis.

4.1.5 e. Analysis of potential effect

In placebo controlled clinical trials the following adverse reactions have been identified:

- Infections and infestations: vulvovaginitis, balanitis and related genital infections, urinary tract infections (common); fungal infection (uncommon)
- Metabolism and nutrition disorders: hypoglycemia (when used with SU or insulin) (very common); volume depletion (uncommon)
- Nervous system disorders: dizziness (common)
- Gastrointestinal disorders: constipation, dry mouth (uncommon)
- Skin and subcutaneous tissue disorders: rash (common)
- Musculoskeletal and connective tissue disorders: back pain (common)
- Renal and urinary disorders: dysuria, polyuria (common); nocturia (uncommon)
- Reproductive system and breast disorders: vulvovaginal pruritis, pruritus genital (uncommon)
- Investigations: hematocrit increased, creatinine renal function decreased during initial treatment, dyslipidemia (common); blood creatinine increased during initial treatment, blood urea increased, weight decreased (uncommon).

Definitions are: common (>1/100 to <1/10) and uncommon (>1/1000 to <1/100).

Few adverse events led to discontinuation of treatment and adverse events were balanced across study groups. The most commonly reported events leading to discontinuation in patients treated with dapagliflozin 10 mg/day were increased blood creatinine (0.4%), urinary tract infections (0.3%), nausea (0.2%), and rash (0.2%). It should be noted that the transient rise in serum creatinine may reflect a reduction in intra-glomerular pressure which may be associated with long-term structural renal protection.

A pooled analysis of 3152 patients who received dapagliflozin in doses between 2.5 and 10 mg/day showed that the incidence of urinary tract infection was increased with dapagliflozin dosage. Most diagnosed infections were mild to moderate and responded to standard antimicrobial treatment.[17]

Overdose:

Dapagliflozin did not show any toxicity in healthy subjects at single oral doses up to 500 mg (50 times the dose aimed to be used in the present study). These subjects had detectable glucose in the urine for a dose-related period of time (at least 5 days for the 500 mg dose) with no reports of dehydration, hypotension, or electrolyte imbalance and with no clinically meaningful effect on QT_c interval. The incidence of hypoglycemia was similar to placebo. In clinical studies where once-daily doses of up to 100 mg dapagliflozin were administered for 2 weeks in healthy subjects and type 2 diabetes mellitus subjects, the incidence of hypoglycemia was slightly higher than placebo and was not dose-related. Rates of adverse events including dehydration or hypotension were similar to placebo, and there were no clinically meaningful dose-related changes in laboratory parameters, including serum electrolytes and biomarkers of renal function. In the event of an overdose, appropriate supportive treatment should be initiated as dictated by the patient's clinical status. The removal of dapagliflozin by hemodialysis has not been studied.

4.1.6 f. Pharmacokinetic considerations

Dapagliflozin is filtered in glomeruli and reabsorbed via the SGLT2 transporter. Dapagliflozin is metabolized in the liver and excreted with feces. Only a very small part (0.2%) is renal excreted. Dose adjustment in patients with impaired kidney function is therefore not needed.

In in-vitro studies, dapagliflozin neither inhibited cytochrome P450 (CYP) 1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP3A4 nor induced CYP1A2, CYP2B6, CYP3A4. Therefore, dapagliflozin is not expected to alter the metabolic clearance of co-administered medicinal products that are metabolized by these enzymes.

Interaction studies conducted in healthy subjects, using mainly a single dose design, suggest that the pharmacokinetics of dapagliflozin are not altered by metformin, pioglitazone, sitagliptin, glimepiride, voglibose, hydrochlorothiazide, bumetanide, valsartan, or simvastatin.

4.1.7 g. Predictability of effect

Efficacy is monitored by measuring the urine albumin-to-creatinine ratio / blood pressure / body weight / eGFR / plasma glucose, which are accepted and accurate surrogates to evaluate efficacy.

4.1.8 h. Interaction with other products

See F

4.1.9 i. Managing of effects

Patients are regularly monitored and asked about adverse effects of urinary tract infections or infestations. As reported above, dapagliflozin administration is associated with an increased risk of urinary tract and genital infections which can be managed with standard antimicrobial treatment.[16]

4.1.10j. Study population

The enrolled population is in a stable condition and no unexpected serious adverse events are foreseen.

4.2 Overall synthesis of the direct risks for the research subjects

The available data show that dapagliflozin decreases glucose, blood pressure and body weight in patients with type 2 diabetes. The drug received marketing approval from the European Medicines Agency (EMA) and is registered in various countries in the European Union (EU). Dapagliflozin increases incidence of urinary tract infections. This adverse effect led in rare instances to treatment discontinuation and is manageable with standard antimicrobial treatment.

5. OBJECTIVES AND ENDPOINTS

Objective(s)	**Endpoint(s)**
Primary objective(s)	**Endpoint for the primary objective(s)**
To determine whether dapagliflozin is superior to placebo in reducing the incidence of the primary composite endpoint.	Composite endpoint for kidney failure[4], hospitalization for heart failure[5], and all-cause mortality in the overall patient group, consisting of patients with eGFR ≤25mL/min/1.73m^2, dialysis patients with residual diuresis ≥500 mL/24hr , and kidney transplant recipients with eGFR ≤45mL/min/1.73m^2.
Secondary objective(s), if applicable	**Endpoint(s) for secondary objective(s), if applicable**
To determine if dapagliflozin is superior to placebo in reducing the incidence of each of the components of the primary composite endpoint in the overall patient group	• All-cause mortality • Kidney failure (chronic dialysis, kidney transplantation or mortality due to kidney failure) • Hospitalization for heart failure
To determine whether dapagliflozin is superior to placebo in reducing the incidence of the primary composite endpoint of all-cause mortality, kidney failure, or heart failure hospitalization in each of the three subgroups of patients	• Patients with advanced CKD i.e. an eGFR ≤25 mL/min/1.73m^2 • Dialysis patients with residual diuresis ≥500 mL/24h • Transplant patients with an eGFR ≤45 mL/min/1.73m^2

[4] Kidney failure is defined in the pre-dialysis and in the kidney transplant subgroups as start of chronic dialysis or receiving a kidney (re)transplant. In the dialysis subgroup there will be no kidney failure end-point. Death due to kidney failure events will be adjudicated by an independent clinical end-point adjudication committee that is blinded to treatment assignment. Endpoint definition and details about the adjudication process will be provided in the endpoint charter.

[5] Hospitalization for heart failure is defined as admission to hospital for a diagnosis of, or a diagnosis compatible with heart failure. Potential hospitalization for heart failure events will be adjudicated by an independent clinical endpoint adjudication committee that is blinded to treatment assignment. Endpoint definition and details about the adjudication process are provided in the endpoint charter.

Exploratory objective(s)	**Endpoint(s) for exploratory objective(s)**
To determine whether dapagliflozin is superior to placebo in reducing the incidence of all-cause mortality in pre-dialysis patients, dialysis patients, kidney transplant recipients separately	All-cause mortality in pre-dialysis patients, dialysis patients and kidney transplant recipients
To determine whether dapagliflozin is superior to placebo in reducing the incidence of kidney failure in pre-dialysis and kidney transplant recipients	Incidence of kidney failure in pre-dialysis and kidney transplant recipients
To determine whether dapagliflozin is superior to placebo in reducing the incidence of hospitalization for heart failure in pre-dialysis patients, dialysis patients, kidney transplant recipients	Incidence of hospitalization for heart failure in pre-dialysis patients, dialysis patients, kidney transplant recipients
To determine whether dapagliflozin is superior to placebo in reducing the incidence of new onset type 2 diabetes mellitus6 in patients without DM	Incidence of new onset type 2 diabetes mellitus in patients without DM
To determine whether dapagliflozin is superior to placebo in reducing the incidence of Diuresis <200 ml/24hr in the dialysis subgroup	Incidence of Diuresis <200 ml/24hr in the dialysis subgroup (for this purpose 24hr urine samples will be collected ≥2 times per year)
To determine whether dapagliflozin is superior to placebo in reducing the incidence of eGFR slope	Incidence of eGFR slope (for the dialysis subgroup slope residual kidney function will be calculated using the average of 24hr urinary creatinine and urea clearance values over time)
To determine whether dapagliflozin is superior to placebo in reducing the incidence of the primary outcome in patients with and without type 2 diabetes separately	Primary outcome in patients with and without type 2 diabetes separately
To determine quality of life	• EQ-5D • SF-12
Safety objectives	**Endpoints for safety objectives**
To evaluate the safety of dapagliflozin	• Serious adverse events • Adverse events leading to drug discontinuation • Adverse events of special interest • clinically significant hypoglycemia (as defined as glucose concentration <3.0 mmol/L, i.e. 54 mg/dL) • diabetic ketoacidosis • urinary tract infections • genital infections

[6] De novo diabetes mellitus type 2 is defined as repeating (twice) HbA1C>6.5% and/or start of glucose lowering drugs

6. STUDY PLAN AND DESIGN

6.1 Trial Design

The RENAL LIFECYCLE trial consists of a screening period and a double blind treatment period. An overview of the study design and key study assessments is shown in figure 1. Potential eligible participants will be recruited via both academic and non-academic hospitals.

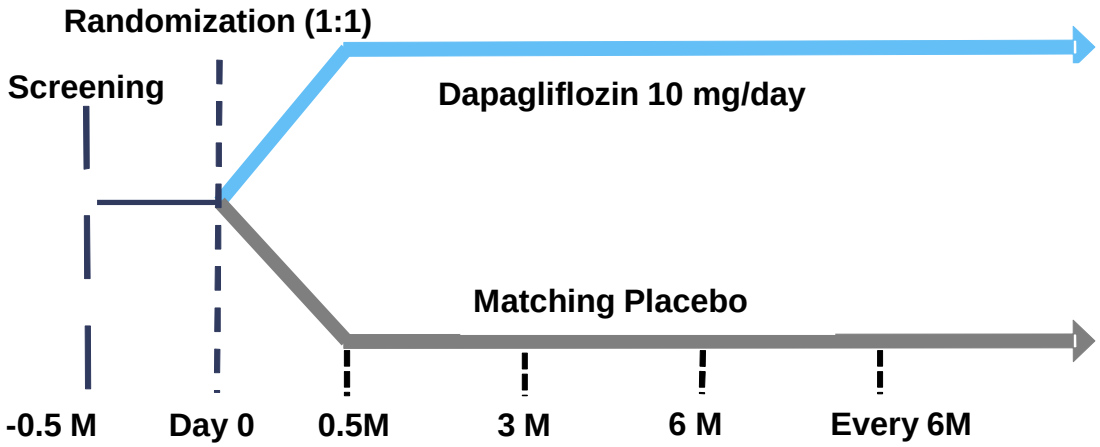

Figure 1: Study design

6.2 Number of Patients

The total number of patients to be included in this study is 1500. Recruitment will be monitored to ensure that in each stratum of patients (pre-dialysis, dialysis, kidney transplant recipients) at least 400 and no more than 600 subjects will be enrolled.

6.3 Overall study duration and follow-up

6.3.1 Screening and study eligibility (visit 1)

The procedures to be performed at the initial Screening visit are outlined in the Study Activities Table. The investigator will register the patient through the Interactive Web Response System (IWRS) and obtain a subject number. The procedures to be performed at the screening visit are outlined in Table 1 Study visits and procedures. Potential eligible pre-dialysis patients with eGFR ≤25 mL/min/1.73m^2 have to be on a stable dose (no changes in dose or type of drug) of ACEis or ARBs for at least 4 weeks prior to the screening visit to be eligible to proceed to the randomization visit unless there is documented evidence that the patient does not tolerate an ACEi or ARB. These subjects will maintain their stable doses of ACEis or ARBs throughout the trial (when possible and tolerated by the patient). ACEi or ARBs are not required for patients on maintenance dialysis or kidney transplant recipients. Dietary advices will be provided according national or local clinical practice guidelines.

While the subject is in screening, tests for eligibility may be repeated twice if there is historical clinical evidence that the subject qualifies for the trial. Subjects who fail the screening test will be allowed to be rescreened per investigator's judgment after 1 month. These subjects have to sign a new informed consent and will receive a new subject number. Subjects who meet all entry criteria can proceed to the randomization visit.

6.3.2 Randomization visit (visit 2: start of double blind treatment phase)

The randomization visit will occur approximately 2 weeks after the subject's screening visit (with a window of ±2 weeks). Procedures to be performed at this visit are outlined in Table 1 Study visits and procedures. Randomization will be performed via web-based ALEA software. Participants will be randomly assigned in a 1:1 ratio to double blind treatment with dapagliflozin 10 mg/day or matching placebo. At the randomization visit, one early morning void urine sample will be collected in pre-dialysis and kidney transplant recipients for assessment of sodium, albumin, protein, and creatinine concentration. In dialysis patients, 24-hour diuresis, residual renal kidney function (average of 24-hour urinary urea and creatinine clearances) and Kt/V[7] will be recorded. Blood samples[8] will also be taken for clinical chemistry measurements, vital signs will be recorded and a physical examination performed. Patients will be instructed to take their study medication in the morning and adhere to treatment. Randomized subjects will receive a sufficient drug supply until the month 3 visit, including the maximal time window (±0.5 month).

6.3.3 Visits during double blind treatment phase (visit 3 and beyond)

Per protocol in-patient follow-up visits are scheduled after 2 weeks, 3 months, 6 months and every 6 months thereafter. The 3 month and 6 months visits can be scheduled such that they coincide with the visits taking place as part of routine clinical care. At each visit vital signs (heart rate, blood pressure and body weight) and eGFR should be recorded at a minimum as part of routine clinical care. At the 3-month visit and all following visits until EoS/EET, one early morning void urine sample should be collected for assessment of sodium, protein, albumin, and creatinine concentration.

In dialysis patients, 24-hour diuresis, residual renal function (average of 24-hour urinary urea and creatinine clearances) and Kt/V[9] will be recorded every 6 months. In addition, adverse events will be recorded at each visit. A study visit schedule with all study procedures is shown in table 1. Randomized subjects will receive a sufficient drug supply until the next visit, that will occur 6 months later, including the maximal time window (plus minus 1 month).

6.3.4 End of Study (EoS) visit and Early End of Treatment (EET) visit

At the end of the study, when the estimated 468 events have occurred, an EoS visit will be scheduled. When patients opt to withdraw early from the study, an EET visit will be scheduled 14 days (+-3) after discontinuation of study medication. Procedures at these visits include physical examination (only on indication), blood sampling, endpoint and adverse event recording. Pre-dialysis patients and kidney transplant recipients are also asked to collect one early morning void urine sample for sodium, albumin, protein, and creatinine assessment.

[7] Most recent Kt/V will be recorded with a time window of ± 6 months

[8] for hemodialysis patients, blood samples will be taken at start of dialysis during any dialysis session, but preferably the first dialysis session in a week

[9] Most recent Kt/V will be recorded with a time window of ± 6 months

6.3.5 Table 1 Study visits and procedures

		Double blind treatment						Study close-out visit
	Screening	Baseline						
Visits	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	**V7, 8, 9, 10, 11, 12 …**	**EoS or EET[e]**
Month	*-0.5*	*0*	*0.5*	*3*	*6*	*12*	*Every 6 months*	
Time window	*- 48 days*		± 14 days	± 14 days	± 28 days	± 28 days	± 28 days	
Informed Consent[a]	x							
Randomization		x						
Significant Medical History	x							
Physical examination	x	x[h]	x[h]	x[h]	x[h]	x[h]	x[h]	x[h]
Serum or Urine Pregnancy test[b]	x	x[b]	x[b]	x[b]	x[b]	x[b]	x[b]	x[b]
Blood sampling[f]	x	x	x	x	x	x	x	x
Early morning void urine sample[g]	x	x		x	x	x	x	x
24-hour urine collection[d+g]		x[p]		x	x	x	x	x
Residual renal function[d]		x			x	x	x	x
Kt/V[d]		x			x	x	x	x
Vital signs[j]	x	x	x	x	x	x	x	x
EQ5D-5L and SF12 questionnaires		x			x	x	x[i]	x
Biobanking (plasma and urine)		x		x				x
Endpoint assessment [c]			x	x	x	x	x	x
Dispense study medication		x		x	x	x	x	
Drug accountability (pill count)			x	x	x	x	x	x
Serious adverse events and AEoSI		x	x	x	x	x	x	x
Review medications	x	x	x	x	x	x	x	x
Cardiac-MRI[K]		x				x		
Cardiac echocardiography[L]		x			x	x[n]		
Body Composition Measurement[L+m]		x			x	x[n]		

Additional biobanking (blood, urine and peritoneal effluent)[L]		x			x	x[n]		
Peritoneal dialysis modality, average ultrafiltration 4 weeks prior to study visit and PET-data[L+o]		x			x	x[n]	x[n]	x[n]
6-Minute Walking Test[L+m]		x			x	x[n]		
Kansas City Cardiomyopathy Questionnaire[L]		x			x	x[n]		
Symbol digit modalities test		x			x	x	x[i]	x

a Informed consent is obtained before any study specific procedure is done.

b WOCBP must have a negative serum or urine pregnancy test result (minimum sensitivity 25 IU/L or equivalent units of HCG) at screening or if an pregnancy is suspected

c At each visit and throughout the study sites will collect information about primary, secondary and exploratory endpoints

d 24-hour diuresis, residual renal function and Kt/V only recorded in dialysis patients. The most recent Kt/V values for each visit will be recorded with a time window of ± 6 months

e EoS, end of study; EET, early end of treatment. EET follow-up visit to be scheduled 14 (+- 3) days after discontinuation of study medication.

f V1 (screening) and all other visits: sodium, potassium, creatinine and urea; V2 (baseline) and EoS/EET: Sodium, Potassium, Creatinine, Urea, Hb, HbA1c, Cholesterol, HDL-cholesterol, LDL-cholesterol, Calcium, Phosphate and PTH;

g assessment of sodium, creatinine and albumin or protein (whichever is available). For urine collection, please see section 11.3.4 for more information

h only on indication

i EQ5D-5L and SF 12 questionnaires and Symbol Digit Modalities Test to be completed once every year after visit 6 until EoS/EET

j vital signs: heart rate, blood pressure and body weight

k Cardiac MRI sub study only. The MRI-scan will be performed within a time window of ± 4 weeks

L Cardiac echocardiography sub study only. The echocardiography, body composition measurement, Kansas City Cardiomyopathy Questionnaire and 6-Minute Walking Test will be performed within a time window of ± 4 weeks

m if available on site

n only participants who still receive PD-treatment at that time-point and participate in the cardiac echography sub study.

O collection of the most recent PET-data if performed during routine medical care

p results of 24-hours urine collection at baseline should be determined within 3 months before the baseline visit . The other 24-hours urine collections should be taken within the time window of the visit.

6.4 Patient participation

The Dutch Kidney Patient Society (Nierpatienten Vereniging Nederland, NVN) is involved in the organisation and execution of the trial. This society has appointed a Patient Advisory Board. This Board will help the Steering Committee with the design of the trial (among others safeguarding patient friendliness and incorporation of relevant patient centered outcomes), optimizing patient inclusion (among others by advertising it on the national kidney patient website, and by raising attention for it during their annual national and regional meetings) and implementation of the results (by presentations during their annual national and regional meetings, and by editorials on relevant websites). To ensure optimal patient involvement, a member of the Patient Advisory Board will be member of the Steering Committee, and during the trial the Patient Advisory Board may provide the Steering Committee with solicited and unsolicited advice. For other participating countries similar cooperation with national and/or regional patient societies will be sought.

7. STUDY POPULATION

7.1 Population

Adult male and female participants with severe CKD are eligible to participate defined as an eGFR ≤25 ml/min/1.73m^2, on dialysis (hemo- as well as peritoneal dialysis) or living with a kidney transplant. Recruitment will be monitored to ensure that in each stratum of patients (pre-dialysis, dialysis, kidney transplant recipients) at least 400 and no more than 600 subjects will be enrolled.

7.2 Inclusion criteria

In order to be eligible to participate in the randomized controlled double blind trial subject must meet the criteria for one of the three strata:

- Patients with advanced CKD i.e. an eGFR ≤25 mL/min/1.73m^2
- Hemo- and peritoneal dialysis patients with a residual diuresis ≥500 mL/24h (at least 3 months after start of dialysis)
- Transplant patients with an eGFR ≤45 mL/min/1.73m^2 (at least 6 months after transplantation)

In addition, to be eligible all subjects must meet all criteria below

- Age >18 years
- Willing to sign informed consent
- Pre-dialysis patients with eGFR ≤25 mL/min/1.73m^2 have to be on a stable dose (no changes in dose or type of drug) of ACEis or ARBs for at least 4 weeks prior to the screening visit to be eligible to proceed to the randomization visit unless there is documented evidence that the patient does not tolerate an ACEi or ARB. These subjects will maintain their stable doses of ACEis or ARBs throughout the trial (when possible and tolerated by the patient). ACEi or ARBs are not required for patients on maintenance dialysis or kidney transplant recipients.

7.3 Exclusion criteria

- Mentally incapacitated subjects (i.e. not able to sign informed consent)
- Diagnosis of type 1 diabetes mellitus
- Concurrent treatment with SGLT2 inhibitor
- History of ≥2 urinary tract / genital infections during the last six months
- Life expectancy <6 months in the opinion of the treating physician.
- Scheduled start of dialysis within 3 months or scheduled kidney transplantation within 6 months
- patients treated for a renal indication during the last 6 months with a course of systemic immunosuppressive agents or intensification of treatment with systemic immunosuppressive agents, such as patients with a kidney transplant and acute rejection or patients with GPA (Morbus Wegener) and a recent flare.

  Of note, this implies that patients receiving non-systemic immunosuppression (e.g. topical, ophthalmic, rectal, intra-articular or inhaled corticosteroids) are allowed to participate, as well as patients having received a short course of oral/IV steroids within 6 months prior to screening for non-renal indications (e.g. for gout or an asthma flare) as well as patients receiving during the last 6 months stable low-dose immunosuppression for whatever reason (e.g. kidney transplant recipients, patients with GPA, patients with gout).

- Active malignancy aside from treated squamous cell or basal cell carcinoma of the skin.
- History of severe hypersensitivity or known severe hepatic impairment (Child-Pugh class C)
- History of severe noncompliance to medical regimens or unwillingness to comply with the study protocol.
- Current pregnancy, lactation or women of child-bearing potential (WOCBP) unless using highly-effective contraceptive measurements[10] until 4 weeks after last intake of the study medication
- Presence of other transplanted organ besides a kidney transplant
- Severe lactose intolerance[11]
- Autosomal Dominant Polycystic Kidney Disease (ADPKD) treated with tolvaptan

[10] Acceptable methods of birth control include consistent and correct use of: oral contraception; implantable or injectable contraceptive; intrauterine device/intrauterine system (IUDs/IUSs); transdermal patch; dual barrier method (i.e. two methods used at the same time, such as a male condom in combination with a female diaphragm/cervical cap plus spermicide foam/gel/film/cream/suppository)

[11] The study medication contains so little lactose that most people with lactose intolerance do not suffer from this.

8. STUDY TREATMENTS

8.1 Investigational Medicinal Product (IMP)

8.1.1 Investigational product/treatment

Dapagliflozin tablets and matching placebos will be purchased and provided by AstraZeneca (AZ). Patients take 10 mg dapagliflozin or matching placebo once daily in the morning according to a randomized treatment scheme.

8.1.2 Name and description of the IMP

Drug name: dapagliflozin (Forxiga, AstraZeneca, EU/1/12/795/009); Chemical structure: (2S,3R,4R,5S,6R)-2-[4-chloro-3-(4-ethoxybenzyl)phenyl]-6-(hydroxymethyl)tetrahydro-2H-pyran-3,4,5-triol;

8.1.3 Summary of findings from non-clinical studies

This is not applicable as dapagliflozin is already registered for the treatment of cardiovascular disease in patients with and without type 2 diabetes, including subjects with CKD.

8.1.4 Summary of findings from clinical studies

Dapagliflozin

SGLT2, located in the proximal tubule of the kidney, is an effective transporter system that is responsible for reabsorption of glucose and sodium. Dapagliflozin is an SGLT2 inhibitor that reversely inhibits the SGLT2 transporter. This leads to enhanced glucose and sodium excretion and reductions in HbA1c, plasma volume, body weight and blood pressure. When given as mono-therapy or as add on to other glucose lowering agents dapagliflozin reduces HbA1c by 0.5 to 0.9% and reduced body weight and systolic blood pressure by approximately 2 kg and 3 mmHg (Dapagliflozin Investigator Brochure).
A cardiovascular outcomes study (DECLARE) was conducted to determine the effect of 10 mg dapagliflozin compared with placebo in 17,160 patients with type 2 diabetes mellitus with or without established cardiovascular disease on cardiovascular and renal events.[11] 8,582 patients were randomized to dapagliflozin 10 mg and 8,578 to placebo and were followed for a median of 4.2 years. The co-primary outcome was CV death or heart failure hospitalization and 2) nonfatal myocardial infarction, nonfatal stroke, or CV death. Dapagliflozin was superior to placebo in preventing the primary composite endpoint of heart failure hospitalization and CV death (hazard ratio 0.83 (95%CI 0.73, 0.95); p=0.005). There were also numerically fewer MACE events in the dapagliflozin group compared with the placebo group but the treatment effect did not show superiority (hazard ratio 0,93 (95%CI 0.84, 1.03); p=0.172).

The DAPA-CKD trial was conducted in 4,304 patients with CKD, with or without type 2 diabetes.[8] Participants were randomly assigned to receive dapagliflozin (10 mg once daily) or placebo. The trial was recommended to stop by the independent data monitoring committee because of efficacy of dapagliflozin in patients with CKD. The primary outcome was a composite of a sustained decline in the eGFR of at least 50%, end-stage kidney disease, or death from renal or cardiovascular causes. Over a median of 2.4 years, a primary outcome event occurred in 197 of 2152 participants (9.2%) in the dapagliflozin group and 312 of 2152 participants (14.5%) in the placebo group (HR 0.61, 95% CI 0.51 – 0.72; p <0.001). The number needed to treat to prevent one primary outcome event was 19 (95% CI 15 – 27). The hazard ratio for the composite of a sustained decline in the eGFR of at least 50%, end-stage kidney disease, or death from renal causes was 0.56 (95% CI 0.45 – 0.68; p <0.001). Dapagliflozin also reduced mortality. Both cardiovascular and non-cardiovascular deaths occurred less frequently with dapagliflozin compared to placebo.[24]

No major hypoglycemic events were reported in patients without type 2 diabetes and in these patients the difference in Hba1c between dapagliflozin and placebo was 0.0%. The effects of dapagliflozin on efficacy and safety were similar in participants with type 2 diabetes and in those without type 2 diabetes.[25] The known safety profile of dapagliflozin in patients with CKD, regardless of the presence or absence of diabetes, was confirmed.

In the DAPA-CKD trial patients with eGFR between 25 and 75 mL/min/1.73m^2 and substantial albuminuria (proteinuric range) were recruited. Whether the results of the DAPA-CKD trial can be extrapolated to a broad population of patients with an eGFR ≤25 mL/min/1.73m^2, dialysis patients, or kidney transplant recipients is unknown and it is the objective of the present study to answer this question.

8.1.5 Summary of known and potential risks and benefits

Type 2 diabetes mellitus

The primary assessment of safety and tolerability of dapagliflozin was conducted in a pre-specified pooled analysis of 13 short-term (up to 24 weeks) placebo-controlled studies with 2,360 subjects with diabetes mellitus type 2 treated with dapagliflozin 10 mg and 2,295 with placebo.

In the 13 short-term placebo controlled studies, the overall incidence of adverse events (short-term treatment) in subjects treated with dapagliflozin 10 mg was similar to placebo. Few adverse events led to discontinuation of treatment and were balanced across study groups. The most commonly reported events leading to discontinuation in patients treated with dapagliflozin 10 mg or placebo were increased blood creatinine (0.4%), urinary tract infections (0.3%), nausea (0.2%), dizziness (0.2%), and rash (0.2%).

The frequency of minor episodes of hypoglycemia was similar between treatment groups, including placebo, with the exceptions of studies with add-on sulphonylurea and add-on insulin therapies. Combination therapies with sulphonylurea and add-on insulin had higher rates of hypoglycemia.

In the 13-study safety pool, vulvovaginitis, balanitis and related genital infections were reported in 5.5% and 0.6% of subjects who received dapagliflozin 10 mg and placebo, respectively. Most infections were mild to moderate, and subjects responded to an initial course of standard treatment and rarely resulted in discontinuation from dapagliflozin treatment. These infections were more frequent in females (8.4% and 1.2% for dapagliflozin and placebo, respectively), and subjects with a prior history were more likely to have a recurrent infection. In the dapagliflozin cardiovascular outcomes study (DECLARE), the number of patients with serious adverse events of genital infections were few and balanced: 2 patients in each of the dapagliflozin and placebo groups.

In the large scale clinical studies in type 2 diabetes, more than 15,000 patients have been treated with dapagliflozin. The most frequently reported adverse reactions across the clinical studies were urinary tract and genital infections. No additional safety signals were found.

Heart failure

In the dapagliflozin cardiovascular outcome study in patients with heart failure with reduced ejection fraction (DAPA-HF study), 2,368 patients were treated with dapagliflozin 10 mg and 2,368 patients with placebo for a median exposure time of 18 months. The patient population included patients with type 2 diabetes mellitus and without diabetes, and patients with eGFR ≥ 30 mL/min/1.73 m2. The overall safety profile of dapagliflozin in patients with heart failure was consistent with the known safety profile of dapagliflozin.

Chronic kidney disease

In the dapagliflozin renal outcome study in patients with chronic kidney disease (DAPA-CKD), 2,149 patients were treated with dapagliflozin 10 mg and 2,149 patients with placebo for a median exposure time of 27 months. The patient population included patients with type 2 diabetes mellitus and without diabetes, with eGFR ≥ 25 to ≤ 75 mL/min/1.73 m2, and albuminuria (urine albumin creatinine ratio [UACR] ≥ 200 and ≤ 5000 mg/g). Treatment was continued if eGFR fell to levels below 25 mL/min/1.73 m2.

In the DAPA-CKD trial, the proportion of patients with serious adverse events was similar in the dapagliflozin and placebo group. This was consistent for patients with and without type 2 diabetes. Adverse events of interest in the DAPA-CKD trial, including diabetic keto-acidosis, severe hypoglycemia, fractures, amputations, AKI were similar between the dapagliflozin and placebo groups. Notably, there were no hypoglycemia events reported in patients without diabetes. In addition, diabetic ketoacidosis did not occur in patients receiving dapagliflozin.

According to the label no dose adjustment is needed for dapagliflozine with mild, moderate or severe hepatic impairment. In subjects with severe hepatic impairment (Child-Pugh class C), however, it was reported that mean Cmax and AUC of dapagliflozin were 40% and 67% higher than matched healthy controls, respectively. In this study, known severe hepatic impairment will therefore be an exclusion criterion.

8.1.6 Description and justification of dosage and route of administration

Dapagliflozin is absorbed from the digestive tract and can therefore be orally administered.

Dapagliflozin will be administered in a dose of 10 mg/day. Dapagliflozin is excreted by the kidney. Dose adjustment in case if impaired kidney function is therefore not needed.

8.2 Placebo

Study medication (dapagliflozin 10 mg OD or matching placebo) will be given on top of standard care. Patients will thus not be withheld any treatment, neither in the actively treated, nor in the placebo treated patients. The efficacy and safety of SGLT-2 inhibition has yet not been studied in the three subgroups of patients that are included in the trial. Theoretically, there are reasons to assume that this class of drugs may have less efficacy when compared to the patients with chronic kidney disease in whom the efficacy of this class of drugs has been shown. The use of a placebo is therefore justified, because this will be necessary to determine the efficacy to safety ratio of this intervention.

8.3 Preparation and labelling of the study treatment(s)

8.3.1 Preparation and labelling of Investigational Medicinal Product

AZ supply chain provides the Investigational Medicinal Product Dapagliflozin 10 mg and matching placebo in unlabeled bottles. Design of labels, labeling, QP release and distribution of finished packs is the responsibility of the Pharmacy of the University Medical Center Groningen.

9. OTHER TREATMENTS AND RESTRICTIONS

9.1 Concomitant therapy

9.1.1 Prohibited medication(s)

Open label SGLT-2 inhibitors are not allowed to be used during the trial.

9.1.2 Permitted medication(s)

No other medication, than mentioned under 9.1.1., is disallowed. Of note, the glucose lowering efficacy of dapagliflozin is dependent on kidney function, and is reduced in patients with an eGFR ≤45 mL/min/1.73m^2. Therefore, additional glucose-lowering treatment should be considered in patients with diabetes mellitus when deemed necessary.

9.2 Lifestyle restrictions

9.2.1 Contraception measures

Since there is no clinical experience regarding the safe use of dapagliflozin in pregnant or lactating women, contraceptive measures for these women are incorporated in the protocol. Women who are currently pregnant or lactating will be excluded from participation. Women of child-bearing potential will be required to use a form of highly effective contraception for the duration of the trial until 4 weeks after last intake of study medication.

Highly effective methods of contraception consist of consistent and correct use of: oral contraception; implantable or injectable contraceptive; intrauterine device/intrauterine system (IUDs/IUSs); transdermal patch; dual barrier method (i.e. two methods used at the same time, such as a male condom in combination with a female diaphragm/cervical cap plus spermicide foam/gel/film/cream/suppository)

10. TRACEABLILITY, STORAGE, ACCOUNTABLILITY AND COMPLIANCE

10.1 Traceability and storage of the study treatment

10.1.1 Handling and dispensing

Study medication is received at the study site by a designated and qualified person, handled and stored safely and properly according to the instructions specified on the drug labels. Study medication is kept in a secured location. Storage conditions are adequately monitored.

The local principal investigator is responsible for ensuring that it is dispensed only to study subjects and only from official study sites by authorized personnel, as dictated by local regulations. The local principal investigator is responsible for ensuring that the investigational product is stored under the appropriate environmental conditions (temperature, light, and humidity).

Subjects are asked to return all unused study drug and packaging at each visit (including at the end of the study or at the time of study drug discontinuation). Unused drugs are destroyed by the local pharmacy department at the end of the study.

If concerns regarding the quality or appearance of the investigational product arise, the investigational product will not be dispensed and Astra Zeneca will be contacted immediately.

10.1.2 Drug ordering

Initial Orders

Contact the protocol manager at the University Medical Center Groningen for information.

Re-Supply

Contact the protocol manager at the University Medical Center Groningen (trials@apoth.umcg.nl) for information.

10.2 Accountability of the study treatment(s) and compliance

All study medications will be stored at room temperature at the pharmacy department of participating centers. Study medication will be collected from the pharmacy department by the local principle investigator. Non-used medication will be returned to the pharmacy where it will be destroyed. Adherence to study drug will be monitored by pill count.

11. STUDY ASSESSMENTS AND PROCEDURES

11.1 Screening procedure

Informed consent is obtained before any study specific procedure is done. Thereafter, study procedures as listed in table will be completed accordingly.

11.2 Randomisation, blinding and treatment allocation

Treatment assignment of dapagliflozin or placebo will be randomized. A minimization randomization method will be used. The randomisation procedure including the drug supply management tool will be designed and implemented by IM Onderzoek UMCG (IM-O) using a web based system (ALEA). Randomization will be done with stratification for four subgroups (predialysis, peritoneal dialysis, hemodialysis, kidney transplant recipient),), centre and type 2 diabetes mellitus status (yes/no). The randomization system will register if a participants participates in either the MRI-substudy and/or echo substudy. Every user will receive an individual login code with which they can randomize their patients.

The investigational drugs will be dispensed by the principal investigator or delegate based on the randomization number generated in the central web based system. The pharmacy of each participating center will store the randomization code.

11.3 Study procedures and assessments

11.3.1 Physical examination

Patients will be subjects to a physical examination at the screening visit. This examination entails a routine investigation of at least heart, lungs and abdomen (and other body parts when indicated). At the study visits thereafter, a physical examination will only be performed on indication. At each examination it will be noted whether a patient has a functioning arteriovenous shunt (upper or lower arm).

11.3.2 Blood pressure and body weight measurements

Blood pressure will be measured as office blood pressure by an automated device during the study visits. Patients will be in a semi-supine position during the blood pressure measurement. The average of three readings will be used. Body weight will be recorded at each in-patient clinical visit. Patients will be asked to take off their shoes and jacket. Patient's height will be recorded at the randomization visit to calculate BMI.

11.3.3 Venipuncture

At each of the study visits a venipuncture will be performed for routine blood tests in accordance with local practice (for hemodialysis patients' blood can be drawn from the inlet blood line or the access cannulation line during a dialysis session).

11.3.4 Urine collection

At baseline and during the study, pre-dialysis patients and kidney transplant recipients are asked to collect an early morning void urine sample. The urine sample will be collected in the morning of the study visit. Sodium, creatinine and albumin or protein (whichever is available) will be determined in urine samples. In patients on dialysis, a 24-hour urine sample instead of an early morning sample will be collected for assessment of protein, albumin, creatinine, sodium and urea.

- Pre-dialysis (CKD) and transplant participants: the preferred urine sample collection is early morning void urine. If urine parameters are measured as part of standard care in 24-hour urine instead of early morning void, then these results can be used instead. When 24-hour urine collection is performed, the volume of the 24-hour urine collection should be entered into the eCRF. The type of urine collection (morning void or 24-hour urine) should be entered into the eCRF and should remain the same for the duration of the trial (except when participants move to another sub-group e.g. from CKD to dialysis).
- For dialysis participants: the preferred urine sample collection is 24-hour urine collection. If urine parameters are measured as part of standard care in early morning void instead of 24-hour urine collection, then these results can be used instead. The type of urine collection (morning void or 24-hour urine) should be entered into the eCRF and should remain the same for the duration of the trial (except when participants move to another sub-group e.g. from dialysis to transplant).

11.3.5 Laboratory measurements

All routine laboratory measurements (table 2) of this study will be assessed in local laboratories of participating centers. The laboratory measurements will be part of standard care.

Table 2: Blood and urine laboratory assessments

Laboratory Assessments	**Parameters**	
	Baseline and EoS/EET visit	**Screening and all other visits**
Plasma[1]	Sodium[5]	Sodium[5]
	Creatinine	Creatinine
	Urea	Urea
	Potassium	Potassium
	Hb	
	HbA1c	
	Cholesterol	
	HDL-cholesterol	
	LDL-cholesterol	
	Calcium	
	Phosphate	
	PTH	
	Baseline and EoS/EET visit	**Screening and all other visits[3]**
Urine	Sodium	Sodium
	Creatinine	Creatinine
	Albumin or protein[4]	Albumin or protein[4]
	Pregnancy Test[2]	

1 For blood sampling of hemodialysis patients, samples are to be taken from inlet blood line or the access cannulation line at the start of the first dialysis session in a week
2 Pregnancy test only done at the screening visit for WOCBP or if a pregnancy is suspected
3 except for visit V3
4 whichever is available

11.3.6 Biobank samples

Blood and urine (and in the cardiac echography substudy also peritoneal effluent) will be collected and stored for potential future analysis for exploratory biomarkers to assess correlations with disease activity, effects of dapagliflozin, clinical outcomes, and toxicity. Biomarker samples will be collected, handled and shipped as detailed in the laboratory manual . It is mandatory to obtain the patient's consent to the donation and use of biological samples. The consent date will be recorded in the eCRF. Patient not consenting to donate biological samples for future biomarker analysis are still able to participate in the study, but without providing samples for biomarker analysis.

Samples will be stored in the freezer facility of the University Medical Center Groningen (or in the Amsterdam UMC for the blood, urine and peritoneal effluent samples collected in the cardiac echography substudy) for a maximum of 25 years from the date of the last patient's last visit, after which they will be destroyed. The results of this biomarker research will be reported either in the clinical study report or separately in a scientific report or publication. The results of this biomarker research may be pooled with biomarker data from other studies with dapagliflozin to generate hypotheses to be tested in future research.

The Principal Investigator (PI) ensures that samples are labeled and shipped in accordance with the Laboratory Manual. Samples can be shipped to specialist labs around the world and analysed by academic collaborators or commercial partners.

A full chain of custody is maintained for all samples throughout their lifecycle. The PI at each site keeps full traceability of collected biological samples from the patients while in storage at the study site until shipment or disposal (where appropriate) and keeps documentation of receipt of arrival. The sample receiver keeps full traceability of the samples while in storage and during use until used or disposed of or until further shipment and keeps documentation of receipt of arrival.

12. STUDY DISCONTINUATION AND COMPLETION

12.1 Definition End of Trial

The study will have an 30 month recruitment phase and a 18 months follow-up after enrollment of the last patient. Total study duration is therefore intended to last 48 months. It should be noted that for individual patients the duration of the trial can differ. In principle, the first patient will be in the trial for 48 months, and the last patient for 30 months. In addition, the trial is powered to be event driven and will be terminated when 468 primary composite outcomes have occurred (see paragraph 4.4 Power Analysis). The exact trial duration may therefore be shorter or longer than the intended 48 months.

12.2 Criteria for temporary halt and early termination of the clinical trial

There are no predefined criteria for premature termination of the study. If, however, during the conductance of the study information becomes available showing that continuation of the study would result in a significant safety risk for the patients, the principal investigator and project leader will decide to terminate the study.

12.3 Withdrawal of individual subjects

Patients are free to withdraw from the study at any time, without consequences for further treatment. Reasons for withdrawal of individual subjects can be at the request of the subject, occurrence of adverse events (such as allergic reactions to study drug, pregnancy, laboratory abnormalities) leading to substantial changes in the individual risk-benefit considerations that suggest a discontinuation of study drug or new medical conditions not allowing for continuation of the protocol conform the treatment. In case a subject withdraws from the study, he/she will be asked to have an Early End of Treatment (EET) visit 14 days (+-3 days) after discontinuation of the study medication (see Table 1). Study medication can be stopped temporarily for medical reasons for a period of 28 days. A longer period of study drug discontinuation may be allowed, but only after written permission by the study coordinator or study chairs. The reason why medication is stopped temporarily will be noted in the eCRF. In case study medication is stopped definitively, the reason for this decision will be noted in the eCRF. Withdrawal of consent should only occur if the patient does not agree to any kind of further assessments or contact whatsoever. Discontinuation of study medication in itself is not considered withdrawal of consent.

Withdrawal of consent must be ascertained and documented in writing by the investigator who must inform the study central coordinator and document the withdrawal of consent in the eCRF and medical records. A patient who withdraws consent will always be asked about the reason(s) and the presence of any AE. The investigator will follow up AEs reported outside of the clinical study. If a patient withdraws from participation in the study, then his/her enrolment and randomization codes cannot be reused. Withdrawn patients will not be replaced. Data generated to the time of complete withdrawal from the study will not be destroyed.

Participants that received a kidney transplantation after randomization may require longer temporarily stop of the study medication. Per protocol participants are asked to restart the study medication within 3 months after receiving a kidney transplant. After restart of the study medication, it is advised to perform an unscheduled additional safety visit after 14 days to ensure subject safety.

12.4 Guidance for treatment of participants for fasting, procedure or surgical intervention

To minimize the risk of diabetic ketoacidosis, if the participant requires a scheduled surgery, the study medication should be withheld temporarily for at least 3 days prior to the surgery.[27] Specialist endocrinology advice is recommended for participants requiring an unscheduled or emergency surgery.[28] These participants should be admitted until eating and drinking normally. Regular monitoring of blood glucose and ketone levels might be required. Restart the study medication post-operatively only when the patient is eating and drinking normally or close to discharge from hospital.

The same process should be followed if participants require an iodine-based intravenous contrast. If surgery is urgent or needed in an emergency, study drug should be withheld at the time of the surgery. Unblinding of study allocation is usually not indicated. However, unblinding may be considered but should only be performed if knowledge of randomized treatment allocation will influence clinical management. If study drug is permanently ceased, the date and reason of permanent study drug discontinuation should be documented in the database.

12.5 Guidance for treatment of participants with gastrointestinal symptoms and/or fever

In case of threating dehydration, for instance due to vomiting (>3 times), diarrhea (>3 times a day during at least 2 days) or fever (>38.5 C during at least 2 days), study drug can be ceased as is routinely often done with diuretics and RAAS inhibition. If study drug is ceased, it should be re-started if clinically appropriate, as soon as deemed feasible. If study drug is permanently ceased, the date and reason of permanent study drug discontinuation should be documented in the eCRF.

12.6 Guidance for treatment of participants with symptomatic hypotension

If the participant experiences symptomatic hypotension, consider down-titration of anti-hypertensives and/or diuretics, at the discretion of the treating physician, prior to consideration of discontinuation of study drug. If study drug is ceased, it should be re-started if clinically appropriate, as soon as deemed feasible. If study drug is permanently ceased, the date and reason of permanent study drug discontinuation should be documented in the eCRF.

12.7 Discontinuation of study medication

In case of pregnancy, hypersensitivity to study drug or known severe hepatic impairment (Child Pugh C), study drug will be immediately discontinued upon the investigator receiving first knowledge of the event. Any reason for study drug discontinuation will be noted in the eCRF, and the patient involved will be asked to continue all trial assessments as planned at beforehand.

When patients reach a renal endpoint, e.g. start or chronic dialysis or receiving a kidney transplantation, study medication will not be stopped as customary in many other trials in nephrology, but continued in the new phase of their “renal lifecycle”, hence the name of the trial.

12.8 Replacement of individual subjects after withdrawal

Randomized subjects who wish to withdraw from the study will not be replaced.

12.9 Follow-up subjects withdrawn from treatment

To ensure validity of study data, it is very important to collect as much data as possible throughout the study and especially vital status (dead or alive) at study closure (also for patients who have withdrawn their informed consent). The investigator will therefore attempt to collect information on all patients' vital status from publicly available sources at study closure, even if informed consent has been withdrawn completely, in compliance with local privacy laws/practices.

12.10 End of study

When the study is stopped (either as planned or when discontinued prematurely) all patients will be invited to perform an End-of-Study visit (see Table 1).

12.11 Arrangements for subjects after their participation in the clinical trial ended

After participation patients will return to their regular standard of care in consultation with their treating physician.

13. SAFETY REPORTING

13.1 Definitions

13.1.1 Adverse events (AEs)

Adverse events are defined as any untoward medical occurrence in a subject to whom a medicinal product is administered and which does not necessarily have a causal relationship with this treatment.

13.1.2 Serious adverse events (SAEs)

Serious adverse event is any untoward medical occurrence in a patient or trial subject that at any dose:

- results in death,
- is life-threatening,
- requires inpatient hospitalization or prolongation of existing hospitalization,
- results in persistent or significant disability/incapacity,
- is a congenital anomaly/birth defect

An elective hospital admission will not be considered as a serious adverse event.

13.1.3 Suspected unexpected serious adverse reactions (SUSARs)

Unexpected adverse reactions are SUSARs if the following three conditions are met:

1. The event must be serious;
2. There must be a certain degree of probability that the event is a harmful and an undesirable reaction to the medicinal product under investigation, regardless of the administered dose;
3. The adverse reaction must be unexpected, that is to say, the nature and severity of the adverse reaction are not in agreement with the product information as recorded in the reference safety information (RSI).

13.2 Recording of AEs/SAEs/SUSARS

In this study AEs will not be collected, unless they meet the criteria for potential endpoints, SAE, if the AE is the reason for discontinuation from IMP (DAE), or Adverse Event of Special Interest (AEoSI). The term AE in this document refers only to the above categories.

An AE reported by the patient should be recorded in the eCRF only if it qualifies as:

- An SAE (as defined in Section 9.2.2 Serious adverse events (SAEs))
- If the AE is the reason for discontinuation from IMP (DAE)
- An adverse event of specific interest (AeoSI):
  - diabetic ketoacidosis
  - urinary tract infections
  - genital infection
  - significant hypoglycaemia (i.e. glucose concentration <3.0 mmol/L or 54 mg/dL)

The causal relationship between IP and each AE (for those collected above) will be assessed, and investigators are asked to answer ‘yes’ or ‘no’ to the question ‘Do you consider that there is a reasonable possibility that the event may have been caused by the IP?’

Given the reversible renal hemodynamic effect of SGLT2-inhibitors, a decline in eGFR of 20-25% from baseline upon start could be acceptable and will not considered an AE. A decline of ≥25% for CKD subgroup and transplant subgroup will be considered an adverse event. A sudden decline in eGFR may result in a temporary stop of study medication, but first a stop or down-titration of concomitant diuretics (if used) should be considered. It is up to the investigator whether the study medication should be stopped. If so, study medication may be stopped for a maximum of 2 weeks. The eGFR should then be checked for reversibility, and study medication should preferably be restarted within 2 weeks.

13.3 Reporting of AEs and SAEs

13.3.1 Reporting of SAEs by the investigator to the sponsor

AEs or laboratory abnormalities that are critical to the safety evaluation are reported to the Sponsor upon judgement by the investigator. SAEs are reported by the investigator to the Sponsor without undue delay but not later than within 24 hours of obtaining knowledge of the events and will be submitted to CTIS in the annual safety report. Where relevant, a follow-up report is send to the sponsor to allow the sponsor to assess whether the SAE has an impact on the benefit-risk balance of the clinical trial.

13.4 Follow-up of adverse events

All SAEs and AeoSIs will be followed until they have abated, or until a stable situation has been reached. Depending on the event, follow-up may require additional tests or medical procedures as indicated, and/or referral to the general physician or a medical specialist. SAEs and AeoSIs need to be reported till end of study, as defined in the protocol.

13.5 Reporting of SUSARs by the sponsor to EudraVigilance

The sponsor will keep detailed records of all AEs which are reported to him/her by the investigator or investigators (CTR: Article 41(3)). The sponsor will report electronically and without delay to EudraVigilance all relevant information about any SUSAR (CTR: Article 42). The period for the reporting of SUSARs by the sponsor to EudraVigilance will take account of the seriousness of the reaction and will be as follows:

- In the case of fatal or life-threatening SUSARs, as soon as possible and in any event not later than **7 days** after the sponsor became aware of the reaction (CTR: Article 42(2(a)));
- In the case of non-fatal or non-life-threatening SUSARs, not later than **15 days** after the sponsor became aware of the reaction (CTR: Article 42(2(b)));
- In the case of a SUSARs which was initially considered to be non-fatal or nonlife threatening but which turns out to be fatal or life-threatening, as soon as possible and in any event not later than **7 days** after the sponsor became aware of the reaction being fatal or life-threatening (CTR: Article 42(2(c))).

Where necessary to ensure timely reporting, the sponsor may, in accordance with section 2.4 of Annex III, submit an initial incomplete report followed up by a complete report (CTR: Article 42(2)).

13.6 Annual safety report

Regarding investigational medicinal products other than placebo, the sponsor shall submit annually through CTIS to all Member States concerned a report on the safety of each investigational medicinal product used in a clinical trial (CTR: Article 43).

13.7 Unblinding procedures for safety reporting

In case of an emergency code break, the online ALEA system has an emergency procedure in place to unblind the subject after approval by the PI. In case ALEA is down the UMCG Pharmacy has a paper backup list to follow the procedure described above. Even though the code is broken for an individual patient, any blood samples for safety or pharmacodynamic assessments will continue to be drawn, for at least 24hr following the last dose as long as doing so will not compromise subject welfare.

It is the responsibility of the Investigator to ensure that there is a procedure in place to allow access to the code break procedures in case of emergency. Study drug must be discontinued after unblinding, but the subject will be followed until resolution of the adverse event. At the conclusion of the study, the occurrence of any emergency code breaks will be determined after return of all code break reports.

The investigator will only unblind the treatment allocation of a subject in the course of a clinical trial if unblinding is relevant to the safety of the subject (CTR: Annex III 2.5(17)).

When reporting a SUSAR to the EMA, the sponsor will only unblind the treatment allocation of the affected subject to whom the SUSAR relates (CTR: Annex III 2.5(18)).

Unblinded information will be accessible only to persons who need to be involved in the safety reporting to the EMA, to Data Safety Monitoring Boards (DSMB), or to persons performing ongoing safety evaluations during the clinical trial (CTR: Annex III 2.5(20)).

13.8 Temporary halt for reasons of subject safety

The sponsor will suspend the study if there is sufficient ground that continuation of the study will jeopardise subject health or safety. The sponsor will submit the notification through CTIS without undue delay of a temporary halt but not later than in 15 days of the date of the temporary halt. It shall include the reasons for such action and specify follow-up measures. The study will be suspended pending a further positive decision by the concerned member state (CTR: Article 38). The investigator will take care that all subjects are kept informed.

13.9 Urgent safety measures and other relevant safety reporting

Where an unexpected event is likely to seriously affect the benefit-risk balance, the sponsor and the investigator will take appropriate urgent safety measures to protect the subjects. In addition the sponsor will notify the Member States concerned, through CTIS, of the event and the measures taken. That notification will be made without undue delay but no later than **7 days** from the date the measures have been taken (CTR: Article 54).

13.10 Data Safety Monitoring Board (DSMB)/Data Monitoring Committee (DMC)

An independent DSMC will be appointed and will report to the Steering Committee. The DSMC will be responsible for safeguarding the interests of the patients in the study by assessing the safety of the IMP during the study, and for reviewing the overall conduct of the study. The DSMC will receive from IM-O the collected study data merged with the coded individual treatment codes before the DSMC meeting. On request IM-O can provide a code list of the randomization outcome. A DSMC charter will be prepared to detail precise roles and responsibilities and procedures to ensure maintenance of the blinding and integrity of the study in the review of accumulating data and interactions with the Steering Committee.

13.11 Safety reporting to AstraZeneca

The Sponsor is responsible for informing AstraZeneca (AZ). SAEs (both SUSAR and SSAR) related to the IP must be provided to AZ on an ongoing basis (initial and follow-up information) as well as individual case reports (ICSRs). SUSARs must be provided unblinded and at the same time these events are notified to the Regulatory Authority.

During the study, the Sponsor shall:

- Send SAE reports (ICSRs and quarterly line listings) including accompanying cover page via secure mail to AEmailboxclinicaltrialTCS@astrazeneca.com
- Provide the Company with a copy of their DSUR (Development Safety Update Report) in those cases the safety reference information used by the Sponsor, is inconsistent with the AZ IB and Local Label.
- Emerging Safety Events: provide AZ with any emerging safety issues, unanticipated problems or actions as a result of a safety signal with the IMP within 24 hours of knowledge.
- Provide the Company yearly with a line listing of all SAEs notified to regulatory authority and Company during the study, for reconciliation purpose. Send via secure mail to AEmailboxclinicaltrialTCS@astrazeneca.com.

At end of Study, provide the Company with a final unblinded summary line listing of all Safety events notified to the regulatory authority and/or IRB/EC and Company during the Study, for reconciliation purposes. Send via secure mail to AEmailboxclinicaltrialTCS@astrazeneca.com.

14. STATISTICAL ANALYSIS

14.1 Description of statistical methods

The Renal Life Cycle trial is an endpoint driven clinical outcome trial. Once the pre-specified primary endpoints have accrued the study will be completed and the primary and secondary endpoints will be analysed as described in paragraph 14.6. No interim analysis is planned for this trial

14.2 Analysis sets

14.2.1 Efficacy analysis set

All patients who have been randomised to study treatment will be included in the full analysis set (FAS) irrespective of their protocol adherence and continued participation in the study. Patients will be analysed according to their randomised IP assignment, irrespective of the treatment actually received. The FAS will be considered the primary analysis set for the primary and secondary variables and for the exploratory efficacy variables.

14.2.2 Safety analysis set

All patients who receive at least one dose of randomised treatment will be included in the safety population. Patients will be analysed according to the treatment actually received. The Safety analysis set will be considered the primary analysis set for all safety variables.

14.3 Participant demographics and other baseline characteristics

A description of the study population will be given using the study subjects characteristics at base-line. Baseline characteristics include but are not limited to age, sex, blood pressure, kidney function (eGFR and UACR), Hba1c, concomitant medications and medical disease history.

Baseline characteristics will be presented using descriptive statistics, i.e. for quantitative parameters the mean and standard deviation (or SEM) or median and minimum/maximum with number of valid observations, depending on normality of data. Mean and median will be reported to a precision of one decimal place more than the individual measurements; standard deviation will be reported to a precision of two decimal places more than the individual measurements; and min/max will have the same precision as the individual measurements. For qualitative parameters (categorical or ordered), frequency counts and percentages of each category will be calculated by treatment group. Percentages will be reported up to 1 decimal place. Baseline characteristics will be summarized overall, and by treatment when applicable.

14.4 Randomisation and blinding

A minimization randomization method will be used. Randomization will be done with stratification for four subgroups (predialysis, peritoneal dialysis, hemodialysis, kidney transplant recipient), centre and type 2 diabetes mellitus status (yes/no). Patients will either receive IMP, Dapagliflozin 10 mg, or matching placebo.

14.5 Sample size, trail power and level of significance used

Assuming 25% risk reduction in the 12.5% annual incidence rate of the primary outcome in a trial of 4 year duration (1.5 year inclusion and 2,5 year follow-up after last patient in) with a power of 80% and an alpha of 0.05, it follows that 1442 patients have to be included, which is rounded to 1500[12].

Event Rate	Risk reduction				
	20%	**22.5%**	**25%**	**30%**	**33%**
7.5%	3716	3186	2305	1549	1254
10%	2849	2441	1765	1184	958
12.5%	2330	1996	1442	966	781
15%	1985	1700	1227	822	664

Previous studies with SGLT2 inhibitors in patients with CKD have shown that SGLT2 inhibitors reduce the risk of renal failure, heart failure and all-cause mortality by 30 to 40%. These effects were consistent regardless of whether participants had/did not have type 2 diabetes, and regardless of renal function or degree of albuminuria[10, 22, 25,26]. Based on these studies, we expect that in the Renal Life Cycle Trial dapagliflozin will reduce the risk of the composite endpoint of renal failure, heart failure and death by at least 25%. The study is powered for an effect measure that is lower than observed in previous studies with patients with chronic kidney damage. It is important to note that the trial will reach statistical significance if the observed effect shows a risk reduction of 22%.

The overall 12.5% annual event rate for the primary composite outcome of incident kidney failure, hospitalization for heart failure and all-cause mortality is based on data from previous trials and observational studies. Details are provided in Appendix A. Given that the incidence rates of the composite endpoint in each of the three subgroups, we assumed a 12.5% annual event rate for the overall study population. Because the incidence rates in these subgroups differ slightly, inclusion will be weighed to allow a better estimate of the treatment effect within each subgroup.

[12] Calculations made with PASS Power Analysis and Sample Size Software.

We will therefore include 450 to 525 patients with CKD stages G4/5, 400 to 475 patients on dialysis, and 550 to 650 patients with a kidney transplant. In case the maximum patient number of a subgroup is reached, inclusion of patients will be stopped for this subgroup, with a total number of patients to be included being 1500. An annual loss-to follow up of 1% is accounted for in the power analysis (a conservative estimate and higher than those observed in other randomized clinical trials with SGLT2 inhibitors in CKD). Of note, when pre-dialysis patients start dialysis or are transplanted, they will continue in the trial, and this will not be a reason for study discontinuation. Likewise when dialysis patients are transplanted, they will also continue in the trial. A 5% annual study drug discontinuation is also incorporated.

With the above assumptions (25% risk reduction / 12.5% annual event rate / 1500 participants) we expect to accrue 468 events which provide 82% power. It should be noted that the trial is event driven and will be terminated when 468 primary composite outcomes have occurred. The exact trial duration may therefore be shorter or longer than the intended 48 months.
A sample size of 1500 patients leads to a minimal detectable effect size at alpha 0.05 of 18% (Hazard ratio 0.82) assuming a primary outcome event rate of 12.5%.

An interim analysis of the sample size will determine if the sample size should be adjusted based on the actual event rate observed during the trial. Moreover, when financial resources allow it, it may be considered to increase sample size to allow more solid conclusions on the efficacy of the SGLT2 inhibitor versus placebo on the primary endpoint in each of the three subgroups separately. In case it will be deemed appropriate to increase the sample size METc approval will be sought before doing so.

14.6 Planned analysis

14.6.1 Analysis primary endpoint

The primary variable is the time to first event included in the primary composite endpoint. The primary analysis will be based on the intention to treat (ITT) principle using the FAS.

In the analysis of the primary composite endpoint, treatments (dapagliflozin versus placebo) will be compared using a Cox proportional hazards model with a factor for treatment group, stratified by randomization stratification factors (subgroup (advanced CKD/dialysis/kidney transplant), centre, and type 2 diabetes mellitus status (yes/no). In general, the analysis will use each patient's last contact as the censoring date for patients without any primary events. The p value, hazard ratio (HR), and 95% confidence interval will be reported.

The contribution of each component of the primary composite endpoint to the overall treatment effect will be examined. Last contact will be treated as the censoring date for patients without the endpoint of interest. Hazard ratios (HR) and 95% confidence intervals will be reported.

Kaplan-Meier estimates of the cumulative incidence to the first occurrence of any event in the primary endpoint will be calculated and plotted, for overall analysis and for the individual components.
Methods similar to those described for the primary analysis will be used to separately analyze the time from randomization to the first occurrence of each component of the primary composite endpoint.

14.6.2 Analysis secondary endpoint(s)

The secondary variables will be analysed in the similar manner as the primary variable.

Subgroup variables for the primary efficacy endpoint and secondary efficacy endpoints include demography, baseline disease characteristics, baseline concomitant medications and others. Cox proportional hazards models will be performed to examine treatment effects within relevant subgroups separately. The p-values for the subgroup analyses will not be adjusted for multiple comparisons as the tests are exploratory and will be interpreted descriptively. Event rates by treatment and HRs with 95% confidence intervals will be reported for each subgroup. HRs and Cis for overall analysis and subgroups will be presented with forest plots. Subgroup analysis by baseline age, sex, treatment modality, diabetes status, and other relevant subgroups will be performed by testing for interaction.

The Fine-Gray model will be used to estimate the cumulative incidence to take into account competing risks. Further details of the subgroup analysis, including the list of subgroup variables, will be provided in the Statistical Analysis Plan (SAP).

14.6.3 Safety analysis:

The number and percent of patients with SAEs, AEs leading to (temporary) study drug interruptions, AEs of special interest, will be summarized by treatment group. Changes in relevant clinical chemistry/haematology parameters will be summarized over time by treatment group. In addition, the number and percent of patients with a marked abnormality in clinical laboratory tests will be summarized over time by treatment group. For safety analyses, summaries will be provided using both on treatment observations and using all observations regardless of whether patients are on or off study treatment**.**

14.7 Interim analysis

No efficacy interim analysis is planned in the study. Safety will be monitored on an ongoing basis by the DSMC.

14.8 (Statistical) criteria for termination of the trial

In case the DSMC notices clinically relevant differences in safety between the two groups during the execution of the trial, it can advise to terminate the trial early, i.e. before the last study visit. Whether or not to accept this advice will be upon the responsibility of the Steering Committee. In case the Steering Committee will not follow the advice of the DSMC, the Steering Committee will inform the IRB about the advice of the DSMC and why the Steering Committee has decided not to follow this advice.

14.9 Procedure for accounting for missing, unused and spurious data

For continuous measures with missing data the pattern of missingness will be investigated a and appropriate imputation techniques will be used.

14.10 Procedure for reporting any deviation(s) from the original statistical plan.

Any deviation(s) from the original statistical plan will be described and justified in amendment to the protocol and/or in the final study report, as appropriate.

15. ETHICAL CONSIDERATIONS

15.1 Declaration of Helsinki

The study will be conducted according to the principles of the Declaration of Helsinki (Fortaleza, 2013) and in accordance with the Medical Research Involving Human Subjects Act (WMO). The Medical Ethical Committee of the University Medical Center in Groningen has to approve the study before any study specific procedure commences. For participating sites outside the Netherlands ethical approval should be sought according to prevailing local and national regulations.

15.2 Recruitment and informed consent procedures

Patients will be recruited from academic and non-academic hospitals in Europe and Australia. Prior to their visit to the outpatient clinic, patients will be invited to participate in the study verbally or by sending a letter via the treating physician. In this letter, patients will find a full explanation of the study, advantages and disadvantages of participating, and contact information of the research team members working on this study. Moreover, the letter contains contact information of an independent physician, to whom subjects can address questions about the research before, during and after a study. The patients will be given 2 weeks to consider their decision, any questions will be answered and thereafter patients will be asked to sign their written informed consent before they take part in the study.

15.3 Benefits and risks assessment, group relatedness

There are no direct benefits for the patients to be included. Participation in the study is on a free-will base. Patients will receive restitution of all costs of transportation that is necessary beyond transportation normally made for patient care. Patients will not receive priority for treatment of other diseases in the clinic during this study. Participation in the proposed study is accompanied with only minor risks. The blood samples will be drawn by means of venipuncture that will be performed during the visit to the outpatient clinic as much as possible as part of routine clinical care. All further performed measurements are non-invasive and therefore only minor risks are associated with participation.

15.4 Compensation for injury

The sponsor University Medical Center Groningen (UMCG) has a liability and subject insurance which is in accordance with the legal requirements in the Netherlands (Article 7, subsection 6 of the WMO). This insurance provides cover for damage to research subjects through injury or death caused by the study that becomes apparent during the study or within 4 years after the end of the study.

Participating centers within the Netherlands fall into the scope of the subject insurance. All patients will receive written information about this insurance. For participating sites outside the Netherlands, liability and/or subject insurance should be organized by these sites according to prevailing local and national regulations.

15.5 Compensation for subjects

Participation of patients in the study is a free-will decision. Patients will receive restitution of costs for transportation which fall outside the standard care visits. Patients do not receive priority for treatment of other diseases in the clinic during this trial

15.6 Compensation for investigators

Agreements on compensation for each participating centre are documented in the clinical trial agreements (CTAs) for this specific study.

16. ADMINISTRATIVE ASPECTS, MONITORING AND CONFIDENTIALITY

The study will be conducted in compliance with the protocol, Directive 2001/20/EC and with the principles of GCP. As of February 2024 the study will be conducted in compliance with CTR No 536/2014 (CTR: Annex I D17a).

16.1 Approval initial application and substantial modifications

The trial protocol, informed consent form, subject information leaflet, investigational medicinal product dossier, investigators brochure and any other documents required by the Regulation will be submitted for the regulatory approval before the clinical trial is started via CTIS.

The sponsor will also submit and obtain approval for substantial modifications to the original approved documents via CTIS.

A 'substantial modification' is defined in the CTR as any change to any aspect of the clinical trial which is made after notification of a decision referred to in Articles 8, 14, 19, 20 or 23 and which is likely to have a substantial impact on the safety or rights of the subjects or on the reliability and robustness of the data generated in the clinical trial.

16.2 Monitoring

Independent clinical site monitoring and quality assurance is conducted at all hospitals to ensure that the rights and well-being of human subjects are projected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial is in compliance with the currently approved protocol, with GCP, and with applicable regulatory requirement(s). For the Netherlands monitoring will be based on basis of the NFU (Nederlandse Federatie van Universiteiten) Guideline 2020 for moderate risk classification.

16.3 Recording, handling and storage of information

16.3.1 Handling of data and data protection

A subject identification code list will be made to link the data to the subject in order to be able to trace data to an individual subject. This code will not be based on the patient initials and birthdate. The key to the code will be safeguarded by the investigator since the data will be kept for a period of 25 years. The handling of personal data will comply for Dutch sites with the General Data Protection Regulation (GDPR) (in Dutch: De Algemene verordening gegevensbescherming, AVG) and for other sites with their respective local and national regulations.

16.3.2 Source documents and case report forms (CRF)

Source documents for this study will include hospital records and procedure reports and data collection forms. These documents will be used to enter data on the CRFs. Data reported on the CRF that are derived from source documents must be consistent with the source documents or the discrepancies must be explained.

All documents will be stored safely in confidential conditions. On all study-specific documents other than the signed consent, the subject will be referred to by the study subject identification code.

16.3.3 Clinical trial master file and data archiving

The sponsor and the investigator shall keep a clinical trial master file. The clinical trial master file shall at all times contain the essential documents relating to the clinical trial which allow verification of the conduct of a clinical trial and the quality of the data generated (CTR: Article 57).

The sponsor and the investigator shall archive the content of the clinical trial master file for at least 25 years after the end of the clinical trial, unless other EU law requires archiving for a longer period. The medical files of subjects shall be archived in accordance with national law (CTR: Article 58).

The content of the clinical trial master file shall be archived in a way that ensures that it is readily available and accessible, upon request (CTR: Article 57).

16.3.4 Collection and storage of biological samples

Blood will be collected and processed for storage at 3 study visits. Table 1 gives an overview of the timing and nature of these samples. Samples will be stored under the unique study number of the participant, for which the key is kept in a secured place in the hospital. Only the investigators of subjects in the specific centres will have access to this key. Samples will initially be stored locally. Samples will be stored for 25 years for future analyses that as defined in the protocol section, 11.3.6. For storage of this material, separate informed consent will be asked. Samples will be destroyed when storage is no longer necessary.

16.4 Audits and inspections and direct access to source data/documents

This trial may be subject to internal or external monitoring, auditing or inspections procedure to ensure adherence to GCP. Access to all trial-related documents including direct access to source data will be given at that time.

16.5 Reporting of serious breaches

The sponsor will notify the Member States concerned about a serious breach of the Regulation or of the version of the protocol applicable at the time of the breach through CTIS without undue delay but not later than **seven days** of becoming aware of that breach (CTR: Article 52).

16.6 Notification of the start and the end of the recruitment

The sponsor will notify within 15 days each Member State concerned of the start of a clinical trial in relation to that Member State through CTIS (CTR: Article 36(1)).

The sponsor will notify within 15 days each Member State concerned of the first visit of the first subject in relation to that Member State through CTIS (CTR: Article 36(2)).

The sponsor will notify within 15 days each Member State concerned of the end of the recruitment of subjects for a clinical trial in that Member State through the EU (CTR: Article 36(3)).

16.7 Temporary halt/(early) termination

The sponsor will notify within 15 days each Member State concerned of the end of a clinical trial in relation to that Member State through CTIS (CTR: Article 37(1)).

The sponsor will notify within 15 days each Member State concerned of the end of a clinical trial in all Member States concerned and in all third countries in which the clinical trial has been conducted through CTIS (CTR: Article 37(3)).

16.7.1 Temporary halt/early termination for reasons not affecting the benefit-risk balance

The sponsor will notify with 15 days each Member State concerned of a temporary halt of a clinical trial in all Member States concerned for reasons not affecting the benefit-risk balance through CTIS (CTR: Article 37(5)).

When a temporarily halted clinical trial for reasons not affecting the benefit-risk balance is resumed the sponsor will notify each Member State concerned through CTIS (CTR: Article 37(6)).

The sponsor will notify to the EU portal CTIS of early termination of the clinical trial for reasons not affecting the benefit-risk balance through CTIS. The reasons for such action and, when appropriate, follow-up measures for the subjects will be provided as well (CTR: Article 37(7)).

16.7.2 Temporary halt/early termination for reasons of subject safety

In accordance to article 38 of the CTR, the sponsor will suspend the study if there is sufficient ground that continuation of the study will jeopardise subject health or safety. The temporary halt or early termination of a clinical trial for reasons of a change of the benefit-risk balance will be notified to the Member States concerned through the EU portal CTIS without undue delay but not later than in 15 days of the date of the temporary halt or early termination. It shall include the reasons for such action and specify follow-up measures. The restart of the clinical trial following a temporary halt as referred to in paragraph 1 shall be deemed to be a substantial modification subject to the authorisation procedure laid down in Chapter III of the CTR (CTR: Article 38).

16.8 Summary of the results

Within one year from the end of a clinical trial in all Member States concerned, the sponsor will submit to the EU database CTIS a summary of the results of the clinical trial. The content of the summary of the results is set out in CTR Annex IV. It shall be accompanied by a summary written in a manner that is understandable to laypersons. The content of the summary is set out in CTR Annex V (CTR: Article 37(4)).

16.9 Public disclosure and publication policy

Publication policy is in agreement with the Dutch CCMO publication statement. Nor the sponsors, nor the principal investigator has a right of veto regarding the way of publishing the results.

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