REDUX: A Multicenter, Open-label Study of DM199 (recombinant human tissue kallikrein-1) in Patients with Stage II or III Chronic Kidney Disease

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Introduction

DM199 is a recombinant form of the endogenous human tissue kallikrein-1 protein (KLK1) and is being studied as a subcutaneously injectable protein drug to treat chronic kidney disease (CKD) and ischemic diseases like acute ischemic stroke (AIS). Tissue kallikrein (KLK1), a serine protease that cleaves kinin peptides from the substrate low molecular weight kininogen thereby leading to nitric oxide and prostaglandin production and vasodilation, is expressed in different organs, including the kidney, pancreas, and the vasculature.

KLK1, isolated from porcine pancreas, has been approved in Japan, China and Korea as a treatment of arterial hypertension, chronic kidney disease (CKD), and numerous vascular diseases including hypertension, retinopathy, and acute ischemic stroke (AIS).[1-5]

DM199 is a recombinant, injectable form of KLK1 that has been developed recently by DiaMedica for the above clinical indications. DM199 therapy is intended to restore normal KLK1 levels, which are often reduced in patients with salt sensitive hypertension, renal and ischemic disease, and thereby enable a return to physiological production of bradykinin (BK). Restoration of downstream mediators of kinin action, nitric oxide and prostaglandin, may ultimately result in normalization of blood pressure, protection of renal function documented by improvement in the estimated glomerular filtration rate (eGFR) and urine albumin-to-creatinine ratio (UACR).

Seminal evidence suggests that DM199 may also have immunomodulatory functions, regulating both innate and adaptive immune responses, which are regarded as important pathogenic mechanisms in vascular and renal diseases. This action of DM199 could be therapeutically relevant in renal diseases that have an autoimmune substrate such as IgA nephropathy (IgAN).

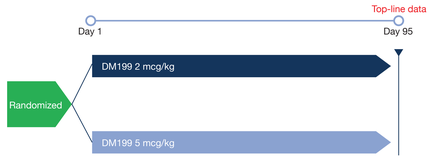
Here, we report the latest results of a pilot study (REDUX) conducted to determine the feasibility of DM199 in patients with nephropathies, associated with arterial hypertension in African-Americans (AA) and IgAN.

Methods

Study Design

- REDUX is an open-label, Phase 2 study of the safety, efficacy, pharmacokinetics, and pharmacodynamics of DM199 as a treatment for CKD. Three cohorts targeting 30 subjects each:
  - AA – African American, non-diabetic with Stage II or III CKD and hypertension
  - IgAN – IgA nephropathy subjects with Stage II or III CKD
  - DKD – diabetic kidney disease – topline results reported at an earlier date
- Subjects who qualified for a cohort were assigned to receive DM199, at a dose level of either 2 µg/kg or 5 µg/kg subcutaneously (SC) twice weekly for 95 days.
- Primary endpoints are safety and tolerability, kidney function (eGFR, UACR), systolic and diastolic blood pressure, and plasma and urine pharmacokinetics of DM199 (KLK1).

Figure 1. Study Design



Study endpoints at day 95:
- Proteinuria (UACR), eGFR and blood pressure
- Safety, tolerability and pharmacodynamics (IgA1, April)

Subject Selection

- Overall eligibility criteria:
  - Stage II and III: eGFR 30 to 90 mL/min/1.73 m^2
  - Albuminuria (UACR): 150-5,000 mg/g
- AA Cohort
  - Non-diabetic (hemoglobin A1c <7%)
  - AHA Stage I Hypertension: systolic BP ≥130 mmHg or diastolic BP ≥80 mmHg or on medication for treating hypertension
- IgAN Cohort
  - confirmed by medical history with biopsy

Treatment

- Randomized to one of two subcutaneous (SC) doses of DM199 twice weekly for 95 days:
  - 2 µg/kg or
  - 5 µg/kg

Study Endpoints

- Safety and tolerability
- Change from baseline to Day 104 for:
  - UACR
  - eGFR
- Change from baseline to Day 95 for:
  - Systolic and diastolic blood pressure
- Change from baseline to Day 95 for biomarkers indicative of IgA nephropathy
  - APRIL (proliferation-inducing ligand)
  - Anti-GD IgA1 (galactose-deficient IgA1)

Results

- The study is being conducted at 13 clinical sites in the U.S.
- Subjects currently are being enrolled
- Approximately two-thirds of patients are male
- Mean age range is 29-87 years
- Baseline eGFR and UACR are characteristic of patients with CKD (Table 1).

Table 1. Baseline Characteristics

	AA Cohort N=22	IgAN Cohort N=23
Age, years	56.0 ± 14.0	49.2 ± 11.5
Age range, years	29 – 87	30 – 73
Male, n (%)	13 (59.1)	15 (65.2)
Race, n (%)		
Asian	0	2 (8.7)
Black or African American[a]	22 (100)	2 (8.7)
White	0	18 (78.3)
Other	0	1 (4.3)
Hispanic or Latino	0	9 (39.1)
Estimated GFR, mL/min/1.73m^2[b]	47.5 ± 19.8	45.2 ± 14.7
Urine albumin to creatinine ratio, µg/mg	737.5 ± 868.5	963.6 ± 730.5
UACR > 500 µg/mg, n (%)	10 (45.5)	11 (57.9)
Sitting systolic blood pressure, mm Hg	145.9 ± 25.0	128.0 ± 12.7
Sitting diastolic blood pressure, mm Hg	89.0 ± 13.8	81.3 ± 9.7

[a]AA or black self-identified
[b]eGFR using CKD EPI Stage II 60 to <90 mL/min/1.73m^2 or Stage III 30 to < 60 mL/min/1.73m^2
[c]Mean ± standard deviation

Efficacy

- DM199 produced improvement of elevated systolic and diastolic BP, UACR for A.A. and IgAN cohorts and eGFR remained stable for each cohort, also showing reduction for APRIL and IgA1 in the IgAN patients. (Figures 2, 3, and 4).

Figure 2. Mean Change From Baseline to Day 95 by Dose for Sitting Systolic and Diastolic BP Among Each Cohort

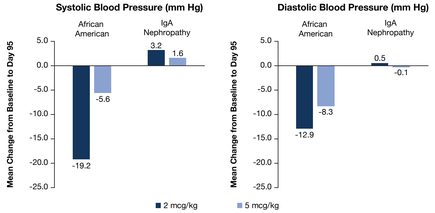
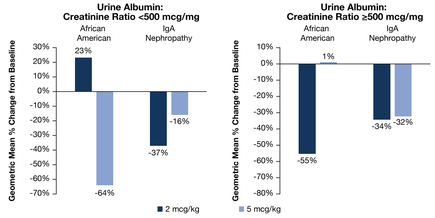
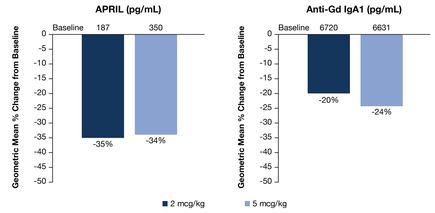


Figure 3. Geometric Mean Change From Baseline to Day 104 for UACR Among Each Cohort by UACR Levels Above or Below 500 mcg/mg (First Morning Void Collection)



- DM199 reduced levels of APRIL and Anti-GD IgA1 in the plasma of patients in the IgAN cohort (Figure 4).

Figure 4. Mean Baseline and Change From Baseline to Day 95 for Biomarkers in the Anti-Gd IgA1 Nephropathy Cohort



Tolerability

- At least 1 TEAE was reported by 24 (31.2%) patients across all 3 cohorts
- The most common TEAEs were of the gastrointestinal system (Table 2).
- The only TEAEs occurring in >1 patient were injection site events (11 events)
- No treatment-related serious AEs were reported
- 2 (2.6%) patients discontinued treatment for diarrhea and death, not related to DM199

Table 2. Incidence of TEAEs by Organ System for Each Cohort

System Organ Class	Number (%) of Patients: AA Cohort (N=22)	IgAN Cohort (N=23)
Number of patients with any TEAE	11 (50.0)	8 (34.8)
Cardiac disorders	0	1 (4.3)
Gastrointestinal disorders	3 (13.6)	0
General disorders and administration site conditions	3 (13.6)	6 (26.1)
Infections and infestations	2 (9.1)	3 (13.0)
Injury, poisoning and procedural complications	0	0
Metabolism and nutrition disorders	0	1 (4.3)
Musculoskeletal and connective tissue disorders	1 (4.5)	1 (4.3)
Psychiatric disorders	1 (4.5)	1 (4.3)
Renal and urinary disorders	0	0
Respiratory, thoracic, and mediastinal disorders	0	0
Skin and subcutaneous tissue disorders	3 (13.6)	0
Vascular disorders	1 (4.5)	2 (8.7)

Summary

The data shown here suggest that the treatment with DM199 may benefit the outcomes of IgAN and AA patients.

IgA nephropathy. IgAN is an auto-immune disease for which no effective treatment is currently available. APRIL promotes plasma cell survival and increases antibody class switching, which are involved in lowering Gd-IgA1 levels. Gd-IgA1 is the core of the immune complexes that cause disease progression and mortality in IgAN patients. Gd-IgA1 is the core of the immune complexes that cause disease progression and mortality in IgAN patients. High Gd-1gA1 levels are highly associated with poor renal outcomes in IgAN patients. Here, we explored the change in two biomarkers, Proliferation-Inducing Ligand (APRIL), and anti-GD IgA1. Anti-Gd-IgA1 autoantibodies form complexes with IgA1 that cannot be cleared because of the large size. The complexes are deposited in the glomerular mesangium, subsequently activating mesangial cells to proliferate and overproduce extracellular matrix proteins and cytokines, thereby inciting injury of the glomerulus.

In addition to native immunity, an alteration in the control of adaptive immune response can contribute to IgAN., T cells play an important role in IgAN pathogenesis and are correlated with its clinical severity[13]. In an earlier in vivo study, DM199/KLK1 reportedly influenced the CTLA4 dependent modulation of Tregs (T cell activation) through nitric oxide and TGF-β1 mediating mechanisms. This led to resetting the balance between regulatory Tregs and Th17 cells in favor of CD4+ CD25+ Foxp3+ T suppressor Tregs, thus dampening the autoimmune response.[11,12] This mechanism warrants further investigation for its possible link to the outcomes of IgAN following DM199 treatment.

KLK1 may act at the very core of the native immune response by cleaving immunocomplexes, a mechanism previously described for bacteria. These microorganisms can evade the host immune response through the production of serine proteases similar to KLK1.[8] Bacteria-derived proteases are capable of degrading pathogenic IgA1 and derived immune complexes in vitro and also in vivo, as demonstrated by pioneer research from Lamm and colleagues[9] in a passive mouse model of IgAN. These initial findings on the therapeutic value of bacterial serine proteases in blunting renal damage at the glomerulus level were confirmed by others in a murine model.[10]

Treatment with DM199 lowered levels of the two key IgAN biomarkers, APRIL and IgA1. In particular, at the 2 ug/kg dose, APRIL was reduced 35% reduction in anti-GD IgA1 was reduced by 20%, which may be early indication of disease modifying.

Hypertensive African Americans: DM199 demonstrated to be associated with encouraging decreases in blood pressure in hypertensive AA patients while blood pressure remained stable in normotensive patients. In particular, in the group given the 2 ug/kg dose, systolic and diastolic blood pressure decreased by 19.2/12.9 mm Hg, respectively.

The AA cohort was selected as a potentially preferrable target for treatment because AA subjects exhibit salt sensitive hypertension and exhibit reduced endogenous KLK1 levels and renal blood flow compared with Caucasians[14]. KLK1 is also involved in regulating the EnAC channel and salt regulation. Therefore, DM199 treatment could be more advantageous to AA patients. One caveat is that AA patients have a high risk for angioedema, a life-threatening condition where exceptionally high kinin levels can play a pathogenic role. However, no angioedema was reported in our study, and this is in line with the fact the DM199 induces physiological increase in kinin.

Our report is the first to demonstrate administration of KLK1 with DM199, is associated with reduction of the circulating immunocomplexes, APRIL and IgA1, in patients with IgAN. These results encourage further controlled studies in this pathology. In addition, DM199 may offer a potential treatment option for Hypertensive AA by improving kidney function and controlling blood pressure. Being a recombinant human protein, DM199 could be a more safe and effective way to cleave pathogenic immunocomplexes that bacterial proteases and for a potential treatment for AA patients.

Conclusions

- Interim results from this Phase II CKD study demonstrating positive signals on interim analysis of a 3 month study:
  - Hypertensive African Americans
    - Reductions in systolic and diastolic BP
    - Reduction in UACR
    - Stable eGFR
  - IgAN
    - Reductions in IgAN biomarkers (APRIL and IaA1) levels as potential signals which are associated with the pathogenesis of nephropathy
    - Reduction in UACR
    - Stable eGFR
    - No change in BP in cohort of normotensive patients
- No Serious Adverse events associated with DM199
  - No angioedema
- DM199 was well tolerated with injection site AEs the only events occurring in >1 patient
- Treatment for patients with CKD is primarily targeted for the underlying disease state, (i.e., hypertension and IgAN).
- DM199 is being studied as a treatment with the potential to stabilize and/or improve kidney function.
- Results from this interim analysis provide proof-of-concept for the effectiveness, safety, and tolerability of DM199 in these specific subgroups of subjects with Stage II or III CKD.
- Future clinical studies in subjects with CKD will provide additional information on the efficacy and tolerability of DM199 for the long-term management of CKD.
- Results from Phase 1 studies with DM199 demonstrated a consistent PK profile with a signal for improvement in renal function in subjects with moderate or severe CKD.[6,7]
- BP was reduced in hypertensive patients with no effect in normotensives

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Presented at Kidney Week, November 4–7, 2021