Supplemental Information ALTERED STUDY

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Supplemental Methods

1. Study Population

Amendments to inclusion criteria

At the study outset participants were restricted to those under 80 years of age and patients with echocardiographic evidence of LVH. Following full regulatory and ethical review an amendment to protocol was made to allow recruitment of patients over 80 years of age and those who did not have LVH as recruitment was more challenging than originally anticipated and it was considered that these changes could be made without affecting the integrity of the study.

Amendments to exclusion criteria

At the study outset participants who had any history of gout, those who were taking warfarin and patients with above ankle amputations were excluded from participation. These exclusion criteria were removed in an amendment following regulatory and ethical review in an effort to improve study recruitment without impeding trial integrity.

2. CMR Methods

An ECG gated fast imaging steady-state free precession (SSFP) sequence was used to acquire cine images in both long axis and short axis planes. Each short axis slice was 6mm thick with an 4mm gap between each slice. Typical cine acquisition parameters included: repetition time 47.88ms, flip angle=45°, echo time = 1.5ms, matrix = 216 x 256 pixels and voxel size = 1.4 x 1.4 x 6mm, slice thickness = 6mm, band-with= 930Hz/pixel.

A single blinded observer analyzed acquired anonymized images in a random order using dedicated software (Siemens Argus, Erlangen, Germany) to determine cardiac indices including LVM and function.

The reproducibility of LVM assessment was derived by the same observer from the above repeated blinded measurements at both time points and an intra-class correlation of 0.998 (95% confidence interval 0.996-0.999) was achieved. The LVMI was calculated using the mean of the two blinded CMR LVMI readings at baseline to determine baseline LVMI and the two blinded CMR LVMI readings at follow up (month 12) to determine follow up LVMI.

3. Flow Mediated Dilation Methods

A secondary outcome of this study was to determine if there is a difference in endothelial function with allopurinol compared with placebo, measured by flow mediated dilation (FMD).

Place and Timing of FMD

FMD was performed at baseline, month 9 and month 12 of the study. Reactivity Task Force guidelines and was in line with FMD practices performed at both Dundee and Glasgow Universities.[3,4]

FMD Acquisition

FMD was measured using a Siemens Accuson Sequoia 512 ultrasound system with an 8 MHz linear array probe. FMD was performed at randomisation, month 9 and month 12. FMD was an optional part of the study and not all participants consented to this aspect of the study. Patients with previous bilateral arteriovenous vascular access surgeries were unable to participate. FMD was either performed immediately prior to a haemodialysis session or on a post-dialysis day. For each individual participant, the timing of FMD in relation to their haemodialysis session was kept consistent throughout the study.

FMD was performed in a dark and quiet room. After arrival patients lay supine for ten minutes. Their non-fistula upper limb was held supine at a 90° angle level with the heart. The brachial artery was located using the ultrasound probe which was then held securely in place using a pneumatic probe holder. A blood pressure cuff was placed around the forearm 10cm distal to the brachial artery recording site. To

minimise interference of the naturally occurring variability of diameter of the brachial artery throughout the cardiac cycle, all ultrasound recordings were ECG-gated. A baseline recording of the brachial artery was taken for 5 minutes. Following this, the blood pressure cuff was then rapidly inflated to either 200mmHg or 30mmHg above the participants' systolic blood pressure (if that was higher than 200mmHg). The cuff was then held inflated for 5 minutes before the pressure was rapidly released. Following this, a 5-minute recording was taken. After this recording of endothelial dependent vasodilation, there were 5 minutes of recovery time. Thereafter, a further 2-minute baseline recording of the artery was taken prior to the administration of 400mcg of sublingual glyceryl trinitrate (GTN). One minute after administration of GTN a further 5-minute recording of the brachial artery was made to record endothelial independent vasodilation.

FMD Analysis

FMD analysis was performed offline using dedicated Vascular Research Tools software (Medical Imaging Applications LLC, Coralville, IA, USA). FMD was recorded as the percentage change in maximal diameter achieved after the cuff was deflated relative to the baseline average brachial artery diameter. As well as the percentage change, the absolute change in brachial artery diameter in millimetres from the baseline average to maximal diameter post cuff deflation was recorded. The same recordings were made for the pre- and post- GTN images with an average of the baseline being compared to the maximal post GTN brachial artery diameter. All FMD acquisition and analysis was performed by a single trained observer (ER) who was blinded to study allocation.

4. Pulse Wave Analysis and Pulse Wave Velocity

A secondary outcome of this study was to determine if there was a difference in endothelial function with allopurinol compared with placebo, measured by pulse wave analysis. Pulse wave velocity and augmentation indices were undertaken at baseline, month 9 and month 12 of the study. These tests were an optional component of this study and were only undertaken in participants who chose also to undergo FMD analysis. The pulse wave tests took place in the same room as FMD at each centre immediately prior to FMD being performed. Participants with a history of bilateral upper limb vascular access procedures were not able to participate. All pulse wave acquisition was performed by a single observer (ER) who was blinded to treatment allocation.

Augmentation Indices

Analysis was undertaken using the SphygmoCor® Vx machine (AtCor Medical, Sydney, Australia) using a highly sensitive micro-manometer. Participants lay supine for 5 minutes prior to the analysis being performed. The radial artery of the non-fistula arm was used to acquire peripheral pressure waveforms by applanation tonometry. The SphygmoCor software then automatically calculated the central aortic pressure. The difference between the first and second systolic peaks from the calculated aortic pressure wave were then used to calculate the augmentation index. The adjusted augmentation index was automatically normalised to a heart rate of 75 beats per minute.

Pulse Wave Velocity

Radial and carotid waveforms were obtained using ECG gating. The distance between the radial artery and the carotid was measured and inputted. The SphygmoCor system calculates the time from the R wave of the ECG to the arterial wave forms. Using these times and the distance between the two measurement sites allows calculation of the pulse wave velocity.

5. Safety and Trial withdrawals

At each study visit participants were monitored for any potential side effects of allopurinol and pre-dialysis safety blood tests were taken (full blood count, renal function, liver function, random blood glucose, hemoglobin A1C, lipids, calcium and phosphate). If there was any significant deterioration in safety blood tests, then the trial medication was stopped and no further medication issued. Urate was also measured at study visits 2,4,5,6 and 7 but to ensure blinding, the results were not made available to investigators until after unblinding at the end of the trial.

Rash was considered the main potential side effect of allopurinol and the protocol specified that participants would stop trial medication if a rash was marked as persistent. If other concerns arose, then at the discretion of the trial team, the trial medication dose could be reduced to the dose last tolerated and the participant remain in the trial. Unless patients underwent kidney transplantation, participants who stopped trial medication during the study were invited to continue with all trial protocol visits for intention to treat analysis.

Supplemental Table 1. Breakdown of medication use at study baseline

Baseline Medication (%)	**Allopurinol (n=39)**	**Placebo (n=40)**	**p value**
Vitamin D	89.8	82.5	0.352
Beta Blocker	43.6	47.5	0.727
Statin	59.0	45.0	0.214
Non-calcium containing phosphate binders	71.8	70.0	0.861
Intravenous Iron	94.9	80.0	0.087
CCBs	30.8	35.0	0.689
ESAs	87.2	85.0	0.780
Loop diuretics	23.1	32.5	0.350
ARBs	20.5	5.0	0.048
Calcium containing phosphate binders	30.8	32.5	0.869
ACE Inhibitors	10.3	17.5	0.352

Abbreviations: ACE – angiotensin converting enzyme, ARBs – angiotensin receptor blockers, CCBs – calcium channel blockers, ESAs erythropoietin stimulating agents.

Supplemental Table 2 – Breakdown of baseline characteristics for completed cases

Baseline Characteristics	**Allopurinol (n=28)**	**Placebo (n=25)**	**p valu**
Age (years)	56.5 ± 11.3	58.8 ± 13.4	0.51
Gender (% male)	44.0	60.7	0.28
Ethnicity White Caucasian (%) Other (%)	 96.4 3.6	 100 0	 0.53
Pre-Systolic BP (mmHg)	141 ± 23	142 ± 23	0.93
Pre-Diastolic BP (mmHg)	68 ± 12	74 ± 13	0.09
Ultrafiltration volume (l)	1.8 ± 0.9	1.7 ± 0.9	0.44
Weight (kg)	72.5 (60.7 – 85.4)	78.0 (66.0 – 94.0)	0.41
BMI (kg/m^2)	25.6 (21.3 - 29.6)	28.9 (22.6 – 33.5)	0.25
RRT (months)	32 (16 – 115)	39 (23 – 82)	0.81
Duration hemodialysis (months)	30 (13 – 69)	36 (17 – 52)	0.90
Dialysis Access (%) Fistula or Graft Line	 92.9 7.1	 84.0 16.0	 0.44
Primary renal disease (%) Diabetic nephropathy ADPKD Glomerulonephritis Renovascular disease Chronic pyelonephritis Other/Unknown Hypertension	 28.6 10.7 25.0 3.6 0.0 28.5 3.6	 8.0 20.0 24.0 4.0 16.0 28.0 0.0	 0.06 0.29 0.59 0.73 0.04 0.59 0.53
Past medical history (%) Diabetes Hypertension Cerebrovascular disease Peripheral vascular disease Ischemic heart disease Dyslipidemia	 32.1 82.1 32.1 25.0 17.9 53.6	 16.0 64.0 8.0 4.0 24.0 32.0	 0.38 0.12 0.03 0.04 0.42 0.10
Smoking History (%) Ex/Current Never	 57.2 42.8	 44.0 56.0	 0.48
Hemoglobin(g/dL)	11.6 (11.3 – 12.6)	111 (10.5 – 11.8)	0.27
URR (%)[a]	76.6 ± 5.5	74.0 ± 8.4	0.22
Albumin (g/L)	35 (32 – 37)	33 (31 – 36)	0.36
Urate[b] (mmol/L)	342 ± 68	360 ± 90	0.44
Phosphate[a] (mmol/L)	1.53 ± 0.38	1.68 ± 0.44	0.20

LVM (g)	124.3 ± 34.4	115.4 ± 39.9	0.39
LVMI (g/m^2)	67.0 ± 16.2	60.8 ± 19.3	0.22
End Diastolic Volume (ml)	154.2 ± 32.9	160.7 ± 44.8	0.56
End Systolic Volume (ml)	62.1 ± 22.9	68.0 ± 28.3	0.41
Ejection Fraction (%)	60.6 ± 8.9	58.4 ± 10.0	0.41
Post systolic BP (mmHg)	126(116 – 146)	118 (109 – 139)	0.33
Post-diastolic BP (mmHg)	65 ± 13	67 ± 15	0.65
24h systolic BP[c] (mmHg)	129 ± 14	133 ± 20	0.64
24h diastolic BP[c] (mmHg)	73 ± 15	78 ± 12	0.47
FMD – baseline cuff (% change)[d]	3.8 ± 2.2	3.7 ± 4.4	0.95
FMD – baseline GTN (% change)[d]	11.3 ± 6.0	14.1 ± 7.3	0.30
PWV (m/s)[e]	7.4 ± 1.5	7.5 ± 2.2	0.91
Aix (%)[e]	25.2 ± 10.1	22.6 ± 19.5	0.70

Data presented as mean ± standard deviation, or median (inter quartile range) if non-parametric
Data available for [a]50, [b]52,[c]16, [d]28, [e]25 participants

BMI - body mass index, RRT – renal replacement therapy, ADPKD – autosomal dominant polycycstic kidney disease, URR – urea reduction ratio, LVM – left ventricular mass, LVMI – left ventricular mass indexed to body surface area, BP – blood pressure, FMD – flow mediated dilation, PWV – pulse wave velocity, Aix – augmentation index

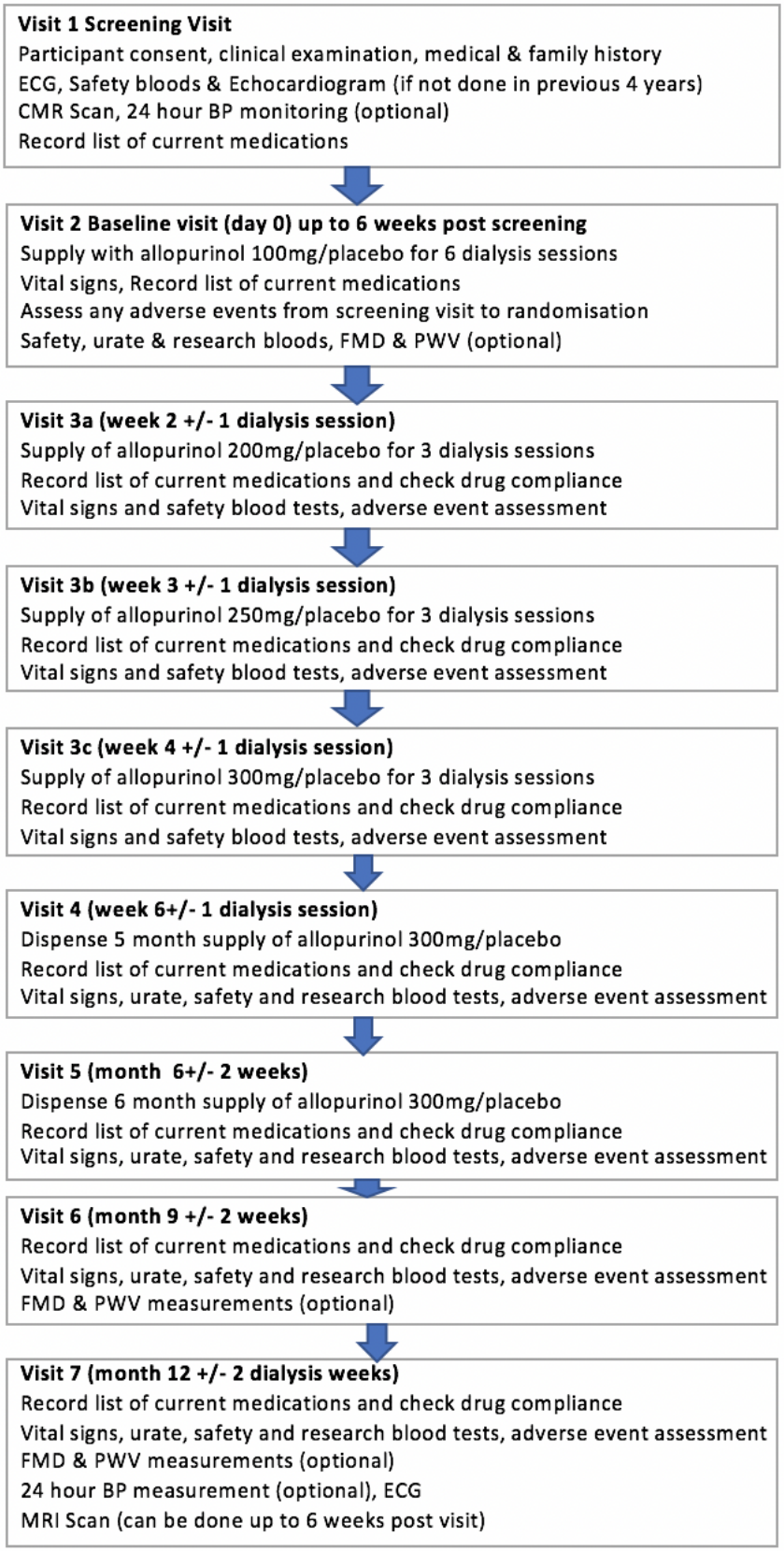
Supplemental Table 3. Breakdown of all Adverse Events in the Study

MedDRA CODING	Allopurinol	Placebo	Grand Total
1. Adverse Events	**81**	**74**	**155**
Blood and lymphatic system disorders		1	1
Cardiac disorders	9	1	10
Endocrine disorders	1		1
Eye disorders	1		1
Gastrointestinal disorders	5	3	8
General disorders and administration site conditions	8	5	13
Hepatobiliary disorders	1		1
Infections and infestations	21	21	42
Injury, poisoning and procedural complications	9	10	19
Investigations	4	4	8
Metabolism and nutrition disorders	2	3	5
Musculoskeletal and connective tissue disorders	2	5	7
Nervous system disorders	1	2	3
Product issues	3	5	8
Renal and urinary disorders	1	1	2
Respiratory, thoracic and mediastinal disorders	5	2	7
Skin and subcutaneous tissue disorders		3	3
Surgical and medical procedures	6	7	13
Vascular disorders	2	1	3
2. Adverse Reactions	**15**	**15**	**30**
Gastrointestinal disorders	5	2	7
General disorders and administration site conditions	1		1
Infections and infestations	2		2
Injury, poisoning and procedural complications		1	1
Investigations		1	1
Metabolism and nutrition disorders	1	1	2
Musculoskeletal and connective tissue disorders	2	1	3
Nervous system disorders	2	5	7
Psychiatric disorders		1	1
Skin and subcutaneous tissue disorders	2	3	5
Serious Adverse Events	**16**	**28**	**44**
Blood and lymphatic system disorders	1		1
Cardiac disorders	1	1	2

Gastrointestinal disorders	2	4	6
Hepatobiliary disorders		1	1
Immune system disorders		1	1
Infections and infestations	3	3	6
Injury, poisoning and procedural complications	1	5	6
Metabolism and nutrition disorders		1	1
Musculoskeletal and connective tissue disorders	1	2	3
Neoplasms benign, malignant and unspecified	1	1	2
Nervous system disorders	3	1	4
Respiratory, thoracic and mediastinal disorders		2	2
Skin and subcutaneous tissue disorders	1		1
Surgical and medical procedures	1	4	5
Vascular disorders	1	2	3
Serious Adverse Reaction		**1**	**1**
Skin and subcutaneous tissue disorders		1	1
GRAND TOTAL	**112**	**118**	**230**

Repetitions of the same adverse event for any given participant have been excluded from this table.

MedDRA – Medical dictionary for regulatory activities



Supplemental Figure 1. Flow chart of patient visits