



Curcumin therapy to treat vascular dysfunction in children and young adults with autosomal dominant polycystic kidney disease: Design and baseline characteristics of participants

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ARTICLE INFO

Keywords:

ADPKD
Clinical trial
Flow-mediated dilation
Pediatric
Pulse-wave velocity
Total kidney volume

ABSTRACT

Although often considered to be a disease of adults, complications of autosomal dominant polycystic kidney disease (ADPKD) begin in childhood. While the hallmark of ADPKD is the development and continued growth of multiple renal cysts that ultimately result in loss of kidney function, cardiovascular complications are the leading cause of death among affected patients. Vascular dysfunction (endothelial dysfunction and large elastic artery stiffness) is evident very early in the course of the disease and appears to involve increased oxidative stress and inflammation. Treatment options to prevent cardiovascular disease in adults with ADPKD are limited, thus childhood may represent a key therapeutic window. Curcumin is a safe, naturally occurring polyphenol found in the Indian spice turmeric. This spice has a unique ability to activate transcription of key antioxidants, suppress inflammation, and reduce proliferation. Here we describe our ongoing randomized, placebo-controlled, double-blind clinical trial to assess the effect of curcumin therapy on vascular function and kidney growth in 68 children and young adults age 6–25 years with ADPKD. Baseline demographic, vascular, and kidney volume data are provided. This study has the potential to establish a novel, safe, and facile therapy for the treatment of arterial dysfunction, and possibly renal cystic disease, in an understudied population of children and young adults with ADPKD.

1. Introduction

Autosomal dominant polycystic kidney disease (ADPKD) is the most common genetic kidney disorder leading to renal failure [1,2]. While the hallmark of ADPKD is the development and continued growth of multiple renal cysts that ultimately result in loss of kidney function [3], the leading cause of death among affected patients is cardiovascular disease (CVD) [1,2]. The proteins encoded by the *PKD1* and *PKD2* genes, polycystin-1 and -2, are expressed in vascular endothelial cells and smooth muscle cells of all major vessels, resulting in extrarenal manifestations of the disease [4]. Presently, there are limited treatment options for the prevention of CVD in patients with ADPKD, particularly in children.

Although often considered to be a disease of adults, clinical manifestation of ADPKD can begin in childhood [5] and it can in fact be diagnosed *in utero*. [6] Evidence of cardiovascular abnormalities, including elevated blood pressure and increased left ventricular mass index, begin to manifest in childhood [7–9]. Currently, adults with ADPKD have very limited treatment options for the prevention of CVD. Thus, childhood/early adulthood may represent the optimal therapeutic window to prevent future cardiovascular complications.

Two of the best established contributors to arterial dysfunction are the development of vascular endothelial dysfunction, most commonly assessed as impaired endothelium-dependent dilation (EDD), and stiffening of the large elastic arteries [10], both of which independently predict future cardiovascular events and mortality [11–14]. We have

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<https://doi.org/10.1016/j.conctc.2020.100635>

Received 27 April 2020; Received in revised form 29 July 2020; Accepted 9 August 2020

Available online 12 August 2020

2451-8654/© 2020 The Author(s).

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shown that vascular dysfunction develops very early in the course of ADPKD, as evidenced by impaired brachial artery flow-mediated dilation (FMD_{BA}; a measure of EDD) and increased aortic pulse-wave velocity (aPWV; a measure of large elastic artery stiffness) compared to matched healthy controls [15]. Targeting vascular dysfunction early in ADPKD is more likely to alter the long-term course of CVD compared to later intervention; thus, strategies to reduce vascular dysfunction in children and young adults with ADPKD have the potential for significant clinical impact.

Curcumin, from the plant *curcuma longa*, is a safe, naturally occurring polyphenol found in the Indian spice turmeric that has long been used in traditional Indian medicine. Curcumin has been proposed as a safe and novel *nutraceutical* for intervention in several disease states [16]. Curcumin administration reduces vascular dysfunction (increases EDD and/or reduces arterial stiffness) in rodent models of hypertension [17], diabetes [18,19], and aging [20]. Additionally, curcumin reduces vascular dysfunction associated with aging in healthy humans [21,22].

Furthermore, curcumin reduces histological evidence of kidney damage in various rodent models of kidney injury of chronic renal impairment [23–26]. Notably, curcumin also slows cyst growth in vitro in a dose-response manner in both the Madin–Darby canine kidney cell cyst model and an embryonic kidney cyst model [27]. Similarly, curcumin improves renal histology and reduces proliferative index, cystic index, and kidney weight (normalized to total body weight) in the *Pkd1*-deletion mouse model (iKsp-*Pkd1*^{del}; a model that does not develop extra renal manifestations of ADPKD and is utilized to evaluate renal cyst growth) [28]. Thus, curcumin might also decrease the rate of total kidney volume (TKV) growth in children and young adults with ADPKD.

Accordingly, we designed a randomized, controlled trial to determine the efficacy of curcumin for improving FMD_{BA} and reducing aPWV (primary aim), as well as slowing kidney cyst growth (exploratory aim), in children and young adults aged 6–25 years of age with ADPKD.

2. Material and methods

2.1. Study design

This is a randomized, placebo-controlled, double-blind, parallel-group study involving 68 children and young adults age 6–25 years with ADPKD. There are two treatment arms: curcumin versus placebo. Curcumin is dosed according to body weight (25 mg/kg body weight/day). Individual subject participation is for one year.

2.1.1. Hypothesis

The primary hypothesis is that oral curcumin therapy will reduce vascular endothelial dysfunction (i.e., increase EDD) and arterial stiffness in children and young adults with ADPKD as compared to placebo. A secondary hypothesis is that the improvements in arterial function observed with curcumin treatment will be associated with reduced oxidative stress and inflammation. An exploratory hypothesis is that curcumin will reduce the rate of kidney cyst growth, as reflected by total kidney volume (TKV), in children and young adults with ADPKD as compared to placebo.

2.1.2. Specific aims

The first specific aim is to determine: a) FMD_{BA} and aPWV before and after 12 months of oral curcumin or placebo. The second specific aim is to measure circulating and urine markers of oxidative stress and inflammation, and in a sub-set of participants aged 18–25 years, to measure FMD_{BA} during normal vs. reduced (via acute ascorbic acid infusion) oxidative stress, before and after 12 months of curcumin or placebo. The third (exploratory) specific aim is to quantify height-adjusted TKV (htTKV) measured by magnetic resonance imaging (MRI) before and after 12 months of curcumin or placebo.

2.2. Design and procedures

2.2.1. Organization of trial

This trial is supported by the National Institutes of Health (K01 DK103678) and is being conducted at the University of Colorado Anschutz Medical Campus. Recruitment began in November 2016, and was completed in December 2019. Subjects were recruited nationally from our ADPKD research registry, from physician referrals, and from responses to information from clinicaltrials.gov (NCT02494141) and the PKD Foundation. The Colorado Multiple Institutional Review Board reviewed and approved the study protocol. All participants (or parents as appropriate) signed an informed consent. Informed assent was obtained from all children aged 6–17 years. A data safety monitoring board was established to review the progress of participants and any adverse events on an annual basis.

2.2.2. Inclusion and exclusion criteria

Eligible participants were children and young adults aged 6–25 years with a diagnosis of ADPKD, based on the presence of bilateral renal cysts in the setting of a family history of ADPKD [29], and fairly preserved renal function with an estimated glomerular filtration rate (eGFR) > 80 mL/min/1.73 m² (using CKiD Schwartz bedside equation for ages 6–17 [30] and the CKD-EPI equation for ages 18–25 [31]). Participants were excluded if they were currently taking a curcumin supplement, had a history of smoking within the past 12 months, or used cannabis within two weeks or antioxidants and/or omega-3 fatty acids within four weeks prior to baseline testing. In addition, those who suffered from alcohol dependence or abuse, had a history of hospitalization within the last three months, had active infection or were receiving antibiotic therapy, were pregnant, lactating, or unwilling to use adequate birth control were also excluded. As vascular measurements can be challenging and/or inaccurate if severely obese (based on over a decade of experience with assessments), individuals who had a body-mass index (BMI) ≥ 95th percentile in ages 6–17 or >40 kg/m² in ages 18–25 were excluded, as were those with an inability to cooperate with study personnel and/or clinical contraindication for MRI, including severe claustrophobia, implants, devices, or non-removable body piercings.

2.2.3. Procedures

Participants underwent screening and vascular testing at the University of Colorado Anschutz Medical Campus Division of Renal Diseases and Hypertension Clinical Vascular Physiology Laboratory, which is part of the Clinical Research Unit. A local contracted laboratory (Quest Diagnostics) and medical records were used for screening out of state participants, in conjunction with a pre-screening questionnaire. MRI scans were performed at the Brain Imaging Center.

2.2.3.1. Baseline assessment. At the baseline visit, participants underwent a medical history and physical exam, including growth parameters and Tanner staging when indicated [32–34], a urine pregnancy test for all female participants of possible childbearing potential (Tanner Stage 2 or higher), blood sampling for clinical labs (e.g., serum creatinine, aspartate aminotransferase [AST], alanine aminotransferase [ALT]) and subsequent batched analysis of circulating curcumin levels by liquid chromatography-tandem mass spectrometry and markers of inflammation (subset of participants 18–25 years), and urine sampling for later batched analysis of markers of oxidative stress (described below).

All vascular measurements were made following standard recommendations including an overnight fast [35]. FMD_{BA} was determined using duplex ultrasonography (Xario 200, Toshiba, Tustin, CA) with ECG-gated end-diastolic ultrasound images analyzed by a single blinded analyst using a commercially available software package (Vascular Analysis Tools 5.10.10, Medical Imaging Applications, Coralville, IA), as described in detail previously [15,36]. Endothelium-independent dilation (brachial artery dilation to 0.4 mg of sublingual nitroglycerin) was

assessed as a standard index of smooth muscle cell sensitivity to exogenous nitric oxide in participants ≥ 18 years of age without contraindications (systolic blood pressure < 100 mmHg, resting heart rate < 60 , or history of migraines) ($n = 35$) [36,37].

Carotid-femoral PWV (aPWV) was measured as described in detail previously [15,36]. Briefly, a transcutaneous custom tonometer (Noninvasive Hemodynamics Workstation [NIHem], Cardiovascular Engineering Inc., Norwood, MA) was positioned at the carotid, brachial, radial, and femoral arteries to non-invasively assess carotid-femoral PWV and carotid-radial PWV (an index of peripheral stiffness). Additionally, as secondary indices of arterial stiffness, the tonometry assessment in conjunction with ultrasound imaging of the carotid artery also provided blinded assessment of carotid artery compliance and carotid artery β -stiffness index, as described previously [15,36]. Carotid intimal medial thickness (cIMT) and carotid BP were also assessed [15, 36].

In a sub-set of participants ≥ 18 years of age ($n = 24$), the influence of oxidative stress on FMD_{BA} was assessed by infusing a supraphysiological dose of ascorbic acid that produces plasma concentrations known to inhibit superoxide production *in vitro* [38] as compared to isovolumic saline, and measuring FMD_{BA} during the “drip infusion” when peak plasma concentrations occur, as described previously [36,39]. A priming bolus of 0.075 g ascorbic acid/kg of fat free mass (maximal dosage set at 5.0 g) was dissolved in 150 mL of saline and infused intravenously at 5 mL/min for 20 min, followed immediately by a “drip-infusion” of 0.5 mL/min over the period where FMD_{BA} was measured. Fat free mass was estimated using a previously validated predictor equation considering age, sex, and body-mass index [40]. We have previously shown that plasma ascorbic acid levels substantially increase in response to the infusion [41].

MRIs to quantify TKV were acquired using a 3-T Siemens Skyra magnet (Siemens Healthcare USA, Tarrytown, NY). Renal images were acquired in similar manner and volumetric measurements determined as described for the CRISP study [42], by stereology using Analyze software (Analyze 11.0, Mayo Foundation, Rochester, MN). No contrast agents were utilized. Individuals involved in the acquisition and analysis of images were blinded regarding group assignment. For analysis, DICOM images were de-identified and evaluated by a single analyst using the Analyze software. To account for normal growth in children, TKV and was adjusted for height (htTKV) [43]. In addition, height-adjusted cyst volume, fractional cyst volume (percent of TKV measured as cystic), and renal parenchyma (TKV – cyst volume) were measured. Mayo classification was determined for those participants ≥ 15 years of age [44].

2.2.3.2. Randomization. After baseline measurements, randomization (curcumin or placebo) was performed by the statistician, using a computer-generated blocked randomization sequence, with stratification by age group (6–13 or 14–25 yrs).

2.3.5. Description of intervention

To enhance the bioavailability of curcumin, manufacturers have developed formulations that may prevent glucuronidation, as unprocessed curcumin is insoluble in water and quickly metabolized via glucuronidation. The compound used in this study was a solid lipid curcumin particle (SLCP, Longvida™, Verdure Sciences) that increases plasma levels of curcumin significantly compared to traditional formulations in healthy volunteers [45] and is safe in both this group and in osteosarcoma patients, including children [45]. Curcumin was delivered as a dose of 25 mg/kg/day rounded to the nearest 100 mg, delivered as powder measured in 100 mg and 500 mg scoops (Longvida™, Verdure Sciences). The placebo was selected to be carrot powder, based on a similar texture and color to active powder and was also prepared by Verdure Sciences. The powder delivery was selected to improve adherence, particularly among young children who may have difficulty

swallowing capsules. Participants were instructed to take the powder mixed with food (e.g., mixed with an applesauce or yogurt, sprinkled on top of toast). Absorption of Longvida™ is the same when taken with a small amount of food compared with water or milk.

Curcumin is the active ingredient in the Indian spice turmeric and gives it a yellowish color. It is found naturally in the root of the plant *Curcuma longa* and has traditionally been used for the prevention and/or treatment of ailments related to the skin, liver, gastrointestinal tract, and the common cold [46]. Curcumin has been investigated extensively for safety and toxicity because of its use in foods. Because of its color, flavor, and antioxidant-stabilizing abilities, curcumin is commonly used in yellow mustard, pickles and sauces. Curcumin has been used to flavor foods and treat ailments for over 5,000 years and is generally recognized as safe (GRAS) by the FDA (this is also true specifically of Longvida™). Studies conducted in rats and humans using standard toxicology protocols have shown no toxic effects of curcumin *in vivo* at doses as high as 8 g/day (8,000 mg/day, which is several-fold higher than the dose administered in the current study [45,47–49]. Few side-effects of curcumin have been reported with doses up to 8–12 g/day (8000–12,000 mg/day) of supplementation in adults [47,48,50]. and children [45,51, 52]. The primary side-effect is gastrointestinal discomfort [47,50,52].

2.3.6. Follow-up study visits

A safety questionnaire is administered over the phone at months 1, 3, 6, and 9. Serum creatinine, ALT, and AST are measured after 1, 6, and 12 months, and for non-local participants, measurements are performed at a local contracted laboratory (Quest Diagnostics). Home pregnancy tests are required (at monthly intervals from the baseline visit) for all females of possible childbearing potential (\geq Tanner Stage 2). All subjects are given a digital blood pressure monitor (A&D Medical, UA767) with an appropriately sized cuff and the subject/parent is instructed in its use. Home blood pressures are taken monthly (average of 3 readings with 3 min between each reading, taken in the seated position following 5 min of rest) and reported during phone safety checks. At twelve months, participants return to the University of Colorado and all baseline measurements are repeated. A sub-group of participants (those local) also returned at 6 months for assessment of vascular measurements.

2.3.7. Outcome measures

The co-primary outcomes are changes at 12 months in FMD_{BA} and aPWV. Secondary outcomes are changes in oxidative stress-associated suppression of EDD (ΔFMD_{BA} with an acute infusion of ascorbic acid), urinary markers of oxidative stress (8-iso-prostaglandin F₂ α [8-iso-prostane] and 8-hydroxy 2 deoxyguanosine [8-OHdG]), and circulating inflammatory markers (C-reactive protein and interleukin-6). C-reactive protein and interleukin-6 were selected as markers of general systemic inflammation based on epidemiological literature supporting an independent association with cardiovascular risk [53] and because both are elevated in ADPKD [54]. An exploratory outcome is change in htTKV at 12 months.

2.3.8. Adherence

Study progress is reviewed with regular phone inquiry of medication supply and an adherence questionnaire. If non-compliance is noted or suspected during the phone inquiry, suggestions are made to help increase study compliance (e.g., alternated foods to try mixing the powder with). At the final study visit, all bottles are returned and the remaining powder is weighed. Percent compliance is calculated based on the total powder consumed relative to the expected total consumption. Additionally, circulating levels of curcumin will be measured using liquid chromatography-mass spectrometry in batched analyses upon completion of all study participants. All participants completing the study will be included in the final analysis based on a pre-specified intent to treat analysis, regardless with level of study compliance.

2.3.9. Statistical considerations

2.3.9.1. Sample size justification. Power calculations were based on clinically meaningful differences in the primary outcomes (FMD_{BA} and aPWV), limited published literature using curcumin in a healthy older population [21], and experience of the study team regarding the variability of the proposed outcomes [37,55,56]. A sample size of 27 in each group will have 90% power to detect a mean increase in FMD_{BA} of 1.5%, given a standard deviation of 2.0 [37], with $\alpha = 0.025$, adjusted for two primary endpoints. Akazawa et al. [21] demonstrated in healthy post-menopausal women a Δ FMD_{BA} of +1.5% with curcumin compared to a decrease in FMD_{BA} of -0.2% in controls. Similarly, a sample size of 27/group will have 90% power to detect a mean decrease of aPWV of 100 cm/s given a standard deviation of 125 [55] with $\alpha = 0.025$, adjusted for two primary endpoints. To account for a potential dropout of 20%, 34 subjects/group were enrolled. A sub-sample size of 10 in each group (of 18–25 year olds) was selected to provide 89% power to detect a 50% reduction in Δ FMD_{BA} with ascorbic acid (given Δ FMD = $+3.0 \pm 1.0\%$ with ascorbic acid in the placebo group [37]), with a two-sided $\alpha = 0.05$. Aim 3 was considered exploratory, thus power was not calculated.

2.3.9.2. Statistical analyses. Descriptive statistics of all data will be provided and tables and figures will be used to present the results. For example, mean and standard deviation will be calculated for continuous variables and frequency and proportion will be provided for categorical variables. Median and interquartile range will be calculated if appropriate for a continuous variable. The 95% confidence interval will be calculated as well. Data transformation will be performed if appropriate before further analysis.

Differences between groups at baseline will be assessed by an independent *t*-test and chi-squared test. A linear regression model will be fit to assess the curcumin effect vs. placebo (independent variable) on the outcomes by regressing each outcome variable at 12 months (dependent variable) on study group, with adjustment for the baseline measurement of the outcome variable and potential confounders (such as puberty). For the subset of those with additional measure of an outcome at 6 months, exploratory analysis with a linear mixed effects model with random intercept and random slope will be performed to assess the curcumin effect and all data from the three measurements will be included in one model. This approach is also flexible to missing (at random) data. We will test for any interactions based on age, sex, and race/ethnicity. The intent-to-treat analysis will apply. For the co-primary outcomes (i.e., FMD_{BA} and aPWV) in aim 1, a two-sided significant level of 0.025 is applicable to each. For other outcomes, a two-sided significant level of 0.05 will apply.

In the current paper, the difference in FMD_{BA} following an acute ascorbic acid infusion vs. saline infusion was evaluated using a paired *t*-test. We additionally performed an analysis to examine the association of baseline vascular function (FMD_{BA} and aPWV) with htTKV using multivariable linear regression. The initial model was unadjusted, then adjusted models were performed to include age, sex, race (model 1), and model 1 plus systolic blood pressure (SBP), body mass index category (normal weight, overweight, or obese), and eGFR (model 2). Analyses were performed using SAS version 9.4 (SAS Institute, Cary, NC) and statistical significance was set at $p < 0.05$.

3. Results

3.1. Baseline data

3.1.1. Baseline participant characteristics

Of the 68 participants who were screened for participation, all 68 were randomized to receive either the curcumin or placebo. An additional 84 participants were pre-screened for enrollment but determined

to be ineligible or not interested. Baseline characteristics of all enrolled participants are shown in Table 1. Participants ranged in age from 6 to 25 years of age, and $n = 25$ participants (37%) were children less than 18 years of age. BMI is presented categorically, as classification is based on percentiles for age, sex, and height in children. Of note, no participants were using tolvaptan. Baseline vascular function data are presented in Table 2. Neither CFPWV nor FMD_{BA} differed according to use of an angiotensin converting enzyme inhibitor/angiotensin receptor blocker, any anti-hypertensive agent, or a statin ($p \geq 0.31$ for all). FMD_{BA} was significantly improved in response to acute infusion of ascorbic acid, as compared to isovolumetric saline infusion, as shown in Fig. 1. Height-corrected TKV, cyst volume, renal parenchyma, and Mayo Class (applicable only to age 15 and older), are shown in Table 3.

3.1.2. Baseline associations

Greater baseline aPWV was independently associated with increased baseline htTKV; however, there was no association of baseline FMD_{BA} with htTKV (Table 4).

4. Discussion

This study has the potential to establish a novel, safe, and facile therapy for the treatment of arterial dysfunction, and possibly renal cystic disease, in an understudied population of children and young adults with ADPKD. Targeted enrollment was achieved within an approximately four-year recruitment window, and the final participant will complete the intervention in December of 2020. All participants who qualified based on pre-screening met screening criteria and were randomized to receive either curcumin or placebo. While there was no specific goal for the number of enrolled children, we were able to enroll 25 pediatric participants (37%), spanning the full inclusion criteria range of 6–25 years. Enrollment of males and females was approximately equal. Although enrollment of minorities was somewhat limited, this is consistent with other NIH-funded clinical trials in ADPKD populations [57,58].

Table 1
Baseline characteristics of study participants.

Variable	All Participants (n = 68)
Age, y	18 ± 5
Sex, n (%) Female	38 (55%)
Race/Ethnicity, n (%)	
Non-Hispanic White	58 (85%)
Hispanic	7 (10%)
Asian or other	3 (4%)
eGFR, ml/min/1.73m²	117 ± 16
BMI Category	
Normal weight or underweight	49 (72%)
Overweight	12 (18%)
Obese	7 (10%)
SBP, mmHg	116 ± 13
DBP, mmHg	73 ± 10
Medications	
ACEi/ARB	24 (35%)
Any antihypertensive	26 (38%)
Statin	11 (16%)
Oral contraceptive or IUD	11 (29%) ^a
Psychiatric medications	8 (12%)

Data are mean ± S.D. or n (%). eGFR, estimated glomerular filtration rate by the CKiD Schwartz bedside equation for ages 6–17 and the Chronic Kidney Disease Epidemiology Collaboration equation for adults ≥18 years of age; BMI, body-mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; ACEi, angiotensin converting enzyme inhibitor; ARB, angiotensin receptor blocker; IUD, intrauterine device. Blood pressures are the mean office reading and were taken in the seated position.

^a % of females.

Table 2
Vascular parameters at baseline.

Vascular Parameters	All Participants (n = 68)
Brachial artery FMD, % Δ	9.3 \pm 0.5
Brachial artery FMD, mm Δ	0.28 \pm 0.12
Baseline brachial artery diameter, mm	3.0 \pm 0.4
Peak shear rate, sec ⁻¹	1073 \pm 38
Brachial artery dilation to NTG, % Δ	32.8 \pm 8.2
Brachial artery dilation to NTG, mm Δ	0.99 \pm 0.22
Brachial SBP, mmHg	115 \pm 13
Brachial DBP, mmHg	65 \pm 9
Carotid intimal medial thickness, mm	0.40 \pm 0.05
Carotid-femoral PWV, cm/sec	510 \pm 95
Carotid-radial PWV, cm/sec	806 \pm 142
Carotid augmentation index, %	-6.4 \pm 15.7
Carotid artery compliance, mm/mm Hg * 10 ⁻¹	0.16 \pm 0.06
Carotid β -stiffness index, A.U.	4.4 \pm 1.1
Carotid SBP, mmHg	106 \pm 2
Carotid DBP, mmHg	65 \pm 1

Data are mean \pm S.D. FMD, flow-mediated dilation; NTG, nitroglycerin; SBP, systolic blood pressure; DBP, diastolic blood pressure; PWV, pulse-wave velocity. Blood pressures were taken in the supine position. NTG was administered to n = 35 participants \geq 18 years of age.

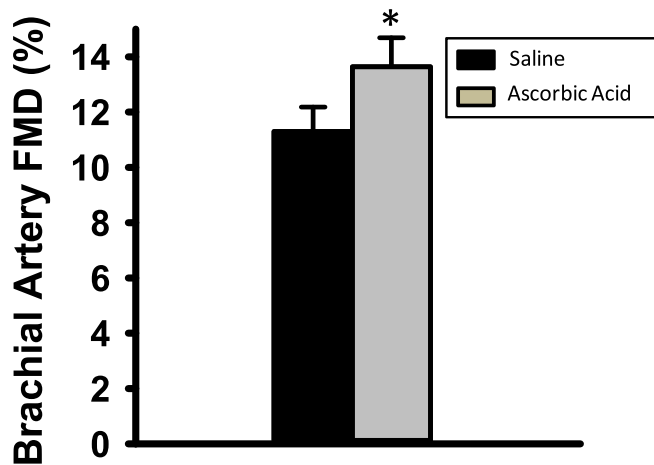


Fig. 1. Change in brachial artery flow-mediated dilation (FMD) in response to acute infusion of saline (black bar) and a supraphysiological infusion of ascorbic acid to inhibit vascular oxidative stress (gray bar) in a sub-group (n = 24) at baseline. *p < 0.01 by a paired *t*-test. Data are presented as mean \pm s.e.m.

The baseline vascular function data indicate slightly better baseline FMD_{BA} and slightly lower aPWV than our previous reports of vascular function in children/young adults [15], as well as in slightly older young adults with ADPKD [41]. This may be due to study of a younger population and a larger number of children under 18 years of age. Available data on changes in vascular function across childhood development suggest that FMD_{BA} and aPWV can change across puberty with post-puberty benefits observed in females [59–62]. We do not anticipate such changes to be a major limitation in the interpretation of the study results as randomization will ensure similar changes across the active and control groups, and we can account for puberty in statistical analyses. However, we do recognize that the small number of participants across stages of puberty may limit our statistical power. Consistent with our previously reported observation in a young adult ADPKD population, an acute ascorbic acid infusion known to inhibit vascular oxidative stress improved FMD_{BA} in the sub-group of participants \geq 18 years of age that participated in this procedure. If FMD_{BA} is improved in the curcumin group at one year as compared to placebo group, and the acute

Table 3
Kidney parameters at baseline.

Kidney Parameters	All Participants (n = 68)
Total kidney volume	534 (392, 797)
Right kidney volume	258 (171, 379)
Left Kidney Volume	307 (215, 507)
Height-corrected TKV, ml/m	333 (234, 475)
Height-corrected cyst volume, ml/m	143 (74, 261)
Fractional cyst volume, %	39 \pm 16
Renal parenchyma, ml	358 (275–446)
Mayo Classification, n, (%)^a	
1A	2 (3%)
1B	8 (12%)
1C	9 (13%)
1D	15 (22%)
1E	17 (25%)
Not applicable	17 (25%)

Data are mean \pm S.D, median (IQR), or n (%). TKV, total kidney volume.

^a Mayo classification is only applicable in participants \geq 15 years of age (n = 51). Those in the not applicable category were <15 years of age.

Table 4

Associations (β -estimate [95% confidence interval]) of vascular function (FMD_{BA} and aPWV) with htTKV at baseline.

Model	β -estimate	P-value
FMD_{BA}		
Unadjusted	7.11 [-9.8, 24.0]	0.40
Model 1	5.75 [-11.0, 22.5]	0.49
Model 2	5.82 [-10.5, 22.1]	0.78
aPWV		
Unadjusted	1.54 [0.91, 2.16]	<0.0001
Model 1	1.14 [0.28, 2.00]	0.01
Model 2	0.96 [0.08, 1.83]	0.03

β -estimates are per 1-unit increase in vascular function.

FMD_{BA}, brachial artery flow-mediated dilation; aPWV, aortic pulse-wave velocity; htTKV, height-corrected total kidney volume.

Model 1 is adjusted for age, race, and sex.

Model 2 is further adjusted for systolic blood pressure, body-mass index category, and estimated glomerular filtration rate.

ascorbic acid infusion no longer improves FMD_{BA} in this group, this would support the hypothesis that curcumin improved vascular function in part by reducing vascular oxidative stress. Systemic markers of oxidative stress and inflammation, which will be batch analyzed at the end of study, will provide additional mechanistic insight.

Baseline total kidney volume was, on average, fairly large in this population. In those individuals \geq 15 years of age, whom could be categorized by the Mayo Classification [44], the majority of participants were classified as 1D or 1E. Previous trials in children and young adults observed an annual kidney growth of approximately 7–10% in this age group [43,63]. While we recognize that the proposed study duration and sample size will only provide a trend for any changes in TKV with curcumin, if encouraging, this exploratory evidence will provide preliminary data for a power calculation to form the basis for a subsequent clinical trial with a larger sample size and longer treatment duration.

Quite interestingly, we observed an independent association between baseline aPWV and htTKV, although no association of FMD_{BA} with htTKV. Increased arterial stiffness reduces peripheral impedance to the forward component of the arterial pulse-wave and increases pulsatile energy transmission to the microcirculation [64]. This increased blood flow and pressure pulsatility can lead to damage of high flow, low impedance organs, including the kidney [64]. It is possible that such damage to kidney microcirculation could exacerbate cyst growth in ADPKD. Likewise, greater htTKV may reflect worsening kidney function not yet accounted for by eGFR, which could contribute to increased arterial stiffness.

We recognize that baseline medications, such as an angiotensin converting enzyme inhibitor or statin may independently influence vascular function. However, we allowed participants to be included who were using these medications because: 1) the medications were held constant throughout the study; 2) inclusion increases generalizability of the results (i.e., whether curcumin has an additive benefit beyond these medications), as it is reflective of the typical ADPKD population; 3) study randomization should ensure an equal number of participants using these medications in both the curcumin and placebo group. We acknowledge that a sub-group analysis based on baseline medication use would be challenging given the small sample size, although note that baseline vascular function did not differ according to usage. The final results of this clinical trial will provide important insight regarding any reductions in vascular function and changes in kidney growth with curcumin therapy in children and young adults with ADPKD. Mechanistic insight will also be ascertained. Given the slowly progressive, lifelong nature of ADPKD, and the favorable safety profile of curcumin, this trial has the potential for high clinical impact.

Disclosures

Verdure Sciences (Noblesville, IN) kindly provided the curcumin (Longvida™) and placebo for this investigator-initiated study.

Funding

This work was supported by the National Institutes of Health (NIH) National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK; grant numbers K01 DK103678 and R03 DK118215), NIH National Center for Advancing Translational Sciences, Colorado Clinical and Translational Science Awards Grant Number UL1 TR002535, NIH High-End Instrumentation Grant S10OD018435, the Baltimore PKD Research and Clinical Core Center Pilot and Feasibility Program (grant number P30DK090868), and by funding from the Zell Family Foundation.

Declaration of competing interest

We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

Acknowledgements

We thank the families who participated in this study.

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