A Phase II, randomized, double-blinded, placebo-controlled clinical trial to evaluate the efficacy of BASIS™ (Nicotinamide Riboside and Pterostilbene) treatment for kidney protection in patients treated by complex aortic aneurysm repair and aortic arch reconstruction

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A Phase II, randomized, double-blinded, placebo-controlled clinical trial to evaluate the efficacy of BASIS™ (Nicotinamide Riboside and Pterostilbene) treatment for kidney protection in patients treated by complex aortic aneurysm repair and aortic arch reconstruction

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List of Abbreviations

AE	Adverse Event
AKI	Acute Kidney Injury
BASIS™,	Nicotinamide Riboside and Pterostilbene
CKD-EPI	Chronic Kidney Disease Epidemiology Collaboration
CNS	Central Nervous System
CrCl	Creatinine Clearance
CRF	Case Report Form
DNA	Deoxyribonucleic acid
eGFR	Estimated Glomerular Filtration Rate
EVAR	Endovascular Aortic Repair
FDA	Food and Drug Administration
GFR	Glomerular Filtration Rate.
GRAS	Generally Recognized as Safe
HIPAA	Health Insurance Portability and Accountability Act
IND	Investigational New Drug Application
IP	Investigational Product
IRB	Institutional Review Board
KDIGO	Kidney Disease: Improving Global Outcomes
LOAEL	Lowest-Observed-Adverse-Effect-Level
NAAD	Nicotinic Acid Adenine Dinucleotide
NAD	Nicotinamide Adenine Dinucleotide
NMN	Nicotinamide Mononucleotide
NOAEL	No-Observed-Adverse-Effect-Level
NR	Nicotinamide Riboside
PHI	Protected Health Information
PI	Principal Investigator
Pter	Pterostilbene
RIFLE	Risk, Injury, Failure; Loss, End-Stage Renal
SAE	Serious Adverse Event/Serious Adverse Experience
SCr	Serum Creatinine
SIRT1, 2 or 3	Sirtuin

Study Summary

Title	A Phase II, randomized, double-blinded, placebo-controlled clinical trial to evaluate the efficacy of BASIS™ (Nicotinamide Riboside and Pterostilbene) treatment for kidney protection in patients treated by complex aortic aneurysm repair and aortic arch reconstruction
Running Title	Protection from Acute Kidney Injury (AKI) with Basis™ treatment
Protocol Number	18-000162
Phase	Phase II
Methodology	Single-center, prospective, randomized, double-blinded, placebo-controlled clinical trial
Overall Study Duration	2 – 3 years
Subject Participation Duration	Six months
Single or Multi-Site	Single site
Objectives	Determine the efficacy of Basis™ therapy in preventing AKI among patients undergoing complex aortic aneurysm repair and aortic arch reconstruction
Number of Subjects	238
Diagnosis and Main Inclusion Criteria	Patients undergoing open and endovascular repair for complex aortic aneurysm and open aortic arch reconstruction
Study Product, Dose, Route, Regimen	Basis™ - mixture of Nicotinamide Riboside (NR) (250 mg) and Pterostilbene (Pter) (50 mg) - orally 1000 mg of NR and 200mg of Pter per day for minimum of 2 weeks before surgery and 6 weeks after surgery
Duration of Administration	8 weeks
Reference therapy	Placebo
Statistical Methodology	The current investigation utilizes a randomized design with 1:1 allocation to treatment group or placebo. All randomized subjects who completed open surgical or endovascular repair of complex abdominal aortic aneurysms and open aortic arch reconstruction will be included in the analysis based on their randomized treatment assignment using an intention to treat model. No covariate adjustment will be included in the primary analysis. Descriptive statistics will be used to summarize baseline demographics, cardiovascular risk factors, anatomical characteristics and procedure details. Clinical data will be summarized for the entire sample and by specific treatment group. In addition to the primary analyses which will include all randomized subjects, secondary analyses will be performed which include only subjects who are found to be ≥50% compliant with medication at the admission appointment for surgery. We hypothesize that this incidence will be reduced by >50% (i.e. AKI <15%) in patients receiving Basis™. Based on these assumptions we have determined that an effective sample-size of N=208 (N=104 per group) will provide statistical power (two-tailed, alpha=0.05) to detect a difference between groups. In order to accommodate attrition of approximately 15%, a total sample-size of N=238 (N=119 per group) is proposed.

1 Introduction

This document is a protocol for a human research study. This study will be carried out in accordance with the applicable United States government regulations and Mayo Clinic research policies and procedures.

1.1 Background

1.1.1 Disease

Acute kidney injury (AKI) is a frequent complication in patients who undergo open surgical or endovascular repair of complex aortic aneurysms, and is an independent predictor of early and late mortality[1]. Among the perioperative causes for this complication are renal ischemia associated with aortic clamping, reperfusion injury, embolization of atherosclerotic plaque and renal arterial injury due to surgical and/or catheter-related trauma. Aortic wall thrombus has been shown to be the one of the most important predictors of acute kidney injury and late renal function deterioration, likely due to embolization during implantation of aortic stent-grafts or open surgical manipulation of the aorta and its branches. The use of iodinated contrast during implantation of aortic devices and for surveillance of endoleaks has also been implicated in the etiology of renal function deterioration.

Complex aortic aneurysms are defined based on their extent of aortic and side branch involvement. Aneurysms encroaching the renal and mesenteric arteries are defined as complex due to need for more extensive dissection, aortic clamping above the renal or mesenteric arteries, and possible reconstruction of aortic branches. The potential deleterious effects of renal and visceral ischemia include renal insufficiency, mesenteric ischemia, ischemic colitis, distal embolization, coagulation disorder, multisystem organ failure, and death.

Acute kidney injury continues to be the most significant complication associated with the extent of repair. Several groups have used surgical adjuncts to decrease the risk of renal injury, including meticulous and expeditious surgical techniques using renal cooling, ice packing and shorter aortic cross-clamping time, minimizing the use of supra-celiac clamp whenever possible.

Mechanisms of cardiac surgery-associated AKI include perioperative renal ischemia, reperfusion injury, cardio-pulmonary bypass (CPB)-induced hemolysis and pigment nephropathy[2], oxidative stress[3], and inflammation (Figure 1). Renal perfusion is complex and highly regulated. Although 20% of cardiac output perfuses the kidneys, the majority of blood filtered by cortex glomeruli is shunted away from the vasa recta. This shunt may help maintain the electrolyte and water concentration gradients in the renal medulla required for tubule and collecting system reabsorption, but renders the renal medulla and cortico-medullary junction hypoxic relative to other tissues (PO_2 10-20 mmHg)[4]. This may be a protective mechanism for oxidative injury but increases susceptibility to ischemia. During surgery many factors alter renal perfusion, and tubules at the corticomedullary junction and in the medulla are often damaged.

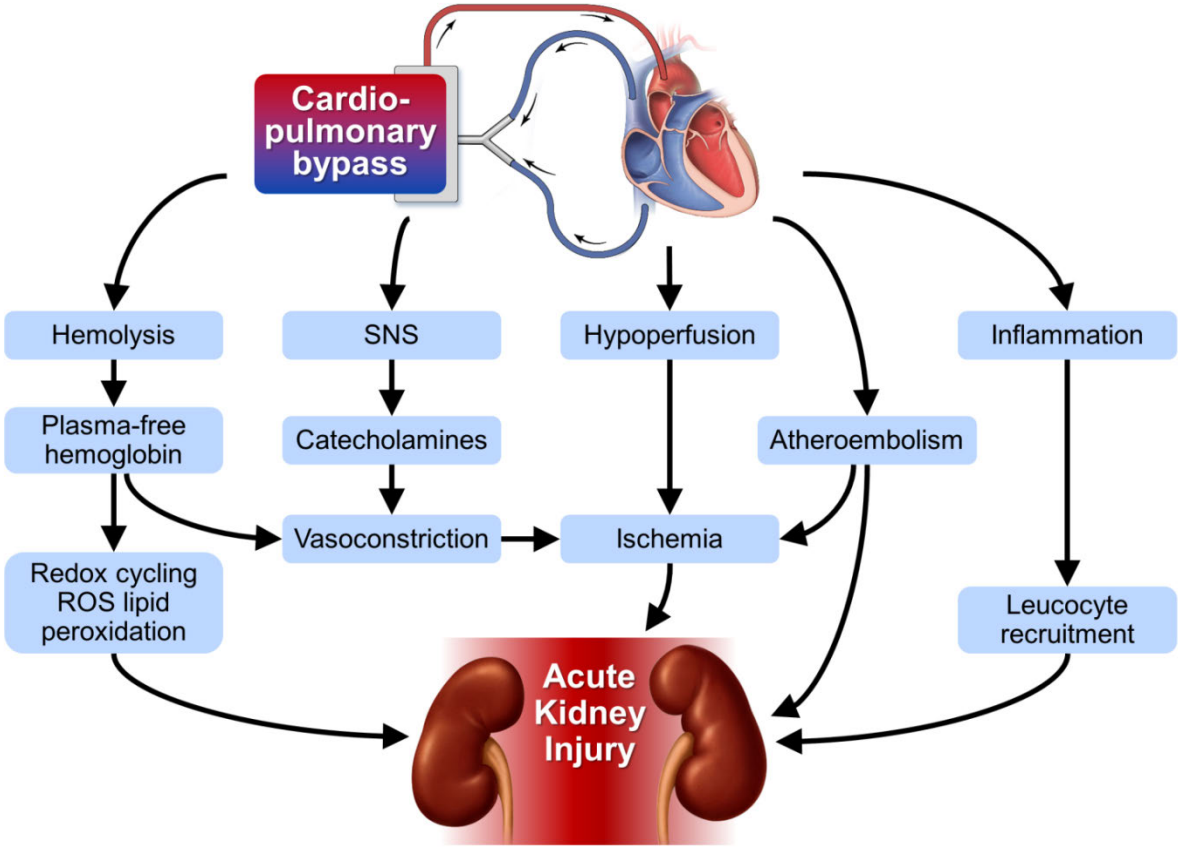


Figure 1 - Mechanisms of cardio-pulmonary bypass and the association with AKI.

CPB provides nonpulsatile blood flow and may dysregulate the balance between cortical and medullary perfusion. Paradoxically, increased cortical perfusion may precipitate corticomedullary ischemia due to increased medullary oxygen consumption from increased solute transport[5]. Aorta cannulation and cross-clamping increase atheroemboli to the kidneys, further exacerbating ischemia and inducing inflammation[6]. Other factors including sympathetic nervous system activation, the endogenous release of circulating cathecolamines, and induction of the renin-angiotensin-aldosterone cascade may further impair renal oxygenation during surgery[7-9].

Cardiac surgery also induces renal and systemic inflammation. Elevated postoperative plasma concentrations of inflammatory cytokines are associated with the subsequent diagnosis of AKI and increased mortality[10]. The mechanisms that increase inflammation during cardiac surgery are not fully understood, but contact activation from the exposure of blood to the CPB circuit, ischemia reperfusion injury, and oxidative damage all contribute[9,11].

1.1.2 Defining Acute Kidney Injury (AKI)

AKI, formerly called acute renal failure (ARF), is commonly defined as an abrupt decline in renal function, clinically manifesting as a reversible acute increase in nitrogen waste products – measured by blood urea nitrogen (BUN) and serum creatinine levels – over the course of hours to weeks. The vague nature of this definition has historically made it difficult to compare between scholarly works and to generalize findings on epidemiologic studies of AKI to patient populations. Several classification systems have been developed to streamline research and

clinical practice with respect to AKI[12-15]. Despite a broad consensus of support, however, the nephrology community continues to point out the shortcomings of these classifications systems[16].

1.1.2.1 RIFLE Classification

In 2002, the Acute Dialysis Quality Initiative (ADQI) was created with the primary goal of developing consensus and evidence-based guidelines for the treatment and prevention of AKI. The first order of business was to create a uniform, accepted definition of AKI; hence, the Rifle criteria were born (Table 1). RIFLE is an acronym of Risk, Injury, and Failure; and Loss; and End-stage kidney disease[17].

Table 1 - RIFLE classification system for AKI

Class	GFR Criteria	UO Criteria
Risk	SCr increased 1.5-2 times baseline or GFR decreased > 25%	UO < 0.5 mL/kg/h < 6h
Injury	SCr increased 2-3 times baseline or GFR decreased > 50%	UO < 0.5 mL/kg/h > 12h
Failure	SCr increased > 3 times baseline or GFR decreased 75% or SCr ≥ 4mg/dL; acute rise ≥ 0.5mg/dL	UO < 0.3 mL/kg/h 24h (oliguria) or anuria 12 h
Loss	Persistent acute renal failure: complete loss of kidney function > 4 weeks (requiring dialysis)	
ESRD	Complete loss of kidney function > 3 months (requiring dialysis)	

GFR = Glomerular filtration rate; UO = urine output; SCr = Serum Creatinine; ESRD = end-stage renal disease.
Note: Patients can be classified either by GFR criteria or by UO criteria. The criteria that support the most severe classification should be used. The superimposition of acute in chronic failure is indicated with the designation RIFLE-FC; failure is present in such cases even if the increase in SCr is less than 3-fold, provided that the new SCr is greater than 4 mg/dL (350 μmol/L) and results from an acute increase of at least 0.5 mg/dL (44 μmol/L). When the failure classification is achieved by UO criteria, the designation of RIFLE-FO is used to denote oliguria. The initial stage, “risk”, has high sensitivity; more patients are classified in this mild category, including some who do not actually have renal failure. Progression through the increasingly severe stages of RIFLE is marked by decreasing sensitivity and increasing specificity.

A recent study to assess RIFLE urine output criteria based on creatinine concentrations for the development of contrast-induced acute kidney injury (CA-AKI) in intensive care (ICU) patients found low predictive value. It was concluded that there was limited use for assessing the effects of therapeutic interventions on the development and progression of AKI in this population[18].

1.1.2.2 Acute Kidney Injury Network (AKIN)

In September 2004, the AKIN was formed. AKIN advised that the term AKI be used to represent the full spectrum of renal injury, from mild to severe, with the latter having increased likelihood for unfavorable outcomes (e.g., loss of function and end-stage renal disease (ESRD)[19].

A report by the AKIN proposed the following criteria for AKI[19]:

- Abrupt (within 48 hours) reduction in kidney function currently defined as an absolute increase in serum creatinine of 0.3 mg/dL or more (≥ 26.4 μmol/l) or
- A percentage increase in serum creatinine of 50% or more (1.5-fold from baseline) or
- A reduction in urine output (documented oliguria of < 0.5 ml/kg/h for > 6h)

The AKIN criteria differ from the RIFLE criteria in several ways. The RIFLE criteria are defined as changes within 7 days, while the AKIN criteria suggest using 48 hours. The AKIN classification includes less severe injury in the criteria and AKIN also avoids using the glomerular filtration rate as a marker in AKI, as there is no dependable way to measure glomerular filtration rate and estimated glomerular filtration rate are unreliable in AKI.

AKIN notes that the diagnostic criteria proposed only after volume status has been optimized and urinary tract obstructions must be excluded when using oliguria as diagnostic criteria.

Fujii and colleagues assessed the three systems discriminative ability based on serum creatinine for the prediction of hospital mortality and found the AKIN classification system to be inferior to the RIFLE and KDIGO systems[20].

1.1.2.3 KDIGO Clinical Practice Guidelines

In 2012 the Kidney Disease Improving Global Outcomes (KDIGO) released their clinical practice guidelines for AKI[21]. KDIGO defines AKI as any of the following:

- Increase in SCr by ≥ 0.3 mg/dl within 48 hours or
- Increase in SCr to ≥ 1.5 times baseline within the last 7 days or
- Urine output < 0.5 ml/kg/h for 6 hours.

Table 2 – KDIGO Staging for AKI severity

Stage	Serum creatinine	Urine output
1	1.5-1.9 times baseline or ≥ 0.3 mg/dL increase	< 0.5 mL/kg/h for 6 hours
2	2.0-2.9 times baseline	< 0.5 mL/kg/h for 12 hours
3	3.0 times baseline or Increase in SCr to ≥ 4.0 mg/dL or Initiation of renal replacement therapy	< 0.3 mL/kg/h for 24 hours or Anuria for ≥ 12 hours

1.1.3 Aneurysms Classification

Complex abdominal aortic aneurysms can be classified using the recommended reporting standard for aortic aneurysm repair of the Society for Vascular Surgery [22]. Aneurysms were categorized (Figure 2) into those involving renal and mesenteric arteries (suprarenal aneurysm), one or both renal arteries (pararenal aneurysm), or encroaching one or both renal arteries without an aneurysmatic dilatation of these vessels (juxtarenal aneurysm) [22]. In addition, aneurysms extending above the celiac axis and up to the diaphragmatic muscle were considered as type IV thoracoabdominal aneurysms.

Thoracoabdominal aortic aneurysms involve the visceral arteries and variable extent of thoracic aorta, which renders any type of repair more complex due to the need to incorporate these vessels into the repair, while minimizing the risk of end-organ ischemia. While the infra-renal aorta is felt to be the most common site for aneurysmal disease to develop, it can involve any region of the aorta and its major branches.

Type IV thoracoabdominal aortic aneurysms are also classified under the category of complex abdominal aortic aneurysms, which is justified because of its similarities with paravisceral aortic aneurysms. These similarities include the operative approach and frequent need to incorporate the celiac axis and superior mesenteric artery into the repair, as well as similar clinical outcomes reported in the literature. Open conventional repair for both types of aneurysms often requires a lower left thoracoabdominal incision with medial visceral rotation. Endovascular repair with fenestrated endografts is similar for both types of aneurysms, in that the stent graft design has most often four fenestrations and the proximal landing zone is approximately two to four centimeters higher with type IV thoracoabdominal aneurysms [23-28]. In addition, results of open and endovascular repair of type IV thoracoabdominal aortic aneurysms are markedly different (and better) than other types of thoracoabdominal aortic aneurysms (I, II and III). These results include substantially higher rates of spinal cord injury, renal insufficiency, morbidity and mortality with more extensive types I to III thoracoabdominal aortic aneurysms [23,29-35].

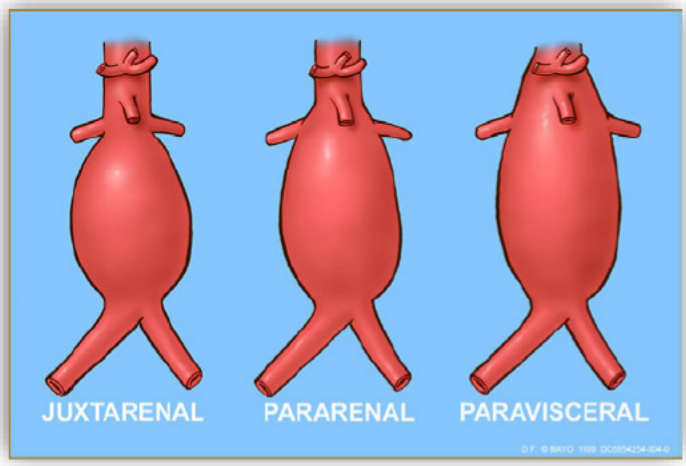


Figure 2 - Classification of complex abdominal aortic aneurysms based on the recommended reporting standard for aortic aneurysm repair of the Society for Vascular Surgery

1.1.4 Types of Complex Aortic Aneurysm Repair

Complex aortic aneurysms requiring surgical intervention can be managed with open surgical repair or Endovascular aortic repair (EVAR).Several studies, have demonstrated immediate clinical benefit in reduction of operative morbidity for EVAR when compared with open surgical repair for the treatment of that disease. Nonetheless, open surgical repair of complex aortic aneurysm remains a viable treatment option, especially in patients with anatomic presentation unsuitable for EVAR.

1.1.4.1 Open Conventional Repair

Open surgical repair has been considered the gold standard for repair of aortic aneurysms against which novel technology should be compared [36]. The simplest form of aortic aneurysm affects the infra-renal aorta, where the operation is performed using midline trans-peritoneal or a left retroperitoneal incision. The operation does not require opening the chest cavity, and aortic cross clamping is typically located below the renal arteries, allowing the reconstruction to be performed with no visceral ischemia. After the aorta is clamped and opened, the organized thrombus is evacuated and multiple pairs of lumbar arteries are suture ligated. The aorta is reconstructed using either a polyester or PTFE graft with straight or bifurcated configuration.

The technical difficulty of the operation increases with aortic aneurysms that involve the visceral segment. The degree of complexity in these cases correlates with the need to expose and dissect more proximal aorta above the renal or mesenteric arteries, the more cranial position the aortic clamp and consequently longer visceral ischemia time, which is required to reconstruct the visceral arteries. Therefore, it is logical that patients with more complex abdominal aortic aneurysm have higher mortality and morbidity rates as compared to patients undergoing repair of infra-renal abdominal aortic aneurysms.

Open repair of thoracoabdominal aortic aneurysms has been associated with high mortality and morbidity in most centers [23-35]. Type IV thoracoabdominal aortic aneurysm represents the simplest and less extensive form, with outcomes similar to suprarenal aortic aneurysms, yet with higher mortality rates in most centers. Only a few aortic centers have shown

that these operations can also be performed with satisfactory results. Coselli and associates [23]reported 2,286 patients treated by open TAAA repair, with an operative mortality of 7%. Other aortic centers have shown mortality rates in the range of 5% to 15%. However, 'real world' data using national and regional datasets have demonstrated more ominous results. In the study by Rigberg *et al.* [37] on 797 Medicare beneficiaries who underwent elective open TAAA repair in the State of California the mortality was 19% at 30-days and 31% in 1 year.

Most importantly, not only are mortality and morbidity high, a large number of patients require prolonged hospitalization and additional rehabilitation. Oderich *et al.* [38] reported the outcomes in 5,776 Medicare beneficiaries treated by open repair for TAAAs. In that study, only 31% of the patients were dismissed home from the hospital, whereas 16% died from complications and 53% required to be transferred to another hospital or to assisted care or rehabilitation centers.

1.1.4.2 Endovascular Repair

Endovascular aortic aneurysm repair (EVAR) has gained widespread acceptance and is currently considered the first treatment option for most patients with abdominal aortic aneurysms. Randomized trials[39-41] have demonstrated several short-term advantages over open conventional repair, including less blood loss, operative time, hospital stay, mortality and morbidity. Several anatomical constraints limit the use of endovascular stent grafts in patients with complex aortic aneurysms. The presence of short infra-renal aortic neck, severe angulation or involvement of the visceral arteries continues to limit the application of endovascular approaches. In these patients, open conventional repair remains the standard treatment, but technical complexity increases with more extensive dissection, higher clamp site, prolonged visceral ischemia and more extensive reconstruction. It is logical to speculate that the advantages achieved with endovascular repair of infra-renal aneurysms will pale in comparison to the potential for reduction in morbidity and mortality for treatment of more complex aneurysms that involve the visceral and thoracoabdominal aortic segment.

Table 3. Comparison between early outcomes of open surgery repair and endovascular repair

Early outcomes	*OSR n = 127*	*F-BEVAR n = 160*	*P value*
30-day mortality	6.3%	3.1%	0.26
In-Hospital mortality	7.1%	3.8%	0.21
Major Adverse Event	48%	16%	<.001
Myocardial infarction	12%	4.4%	.02
Respiratory failure	38%	3.1%	<.001
Acute kidney injury (>50% ↓ eGFR)	28%	13%	<.001
New-onset dialysis	19%	1.3%	<.001
Paraplegia (3a-3c SCI)	5.5%	3.1%	.38
Stroke	3.2%	1.9%	.70

Bowel ischemia (+ resection)	3.2%	1.9%	.70
Early re-interventions	17%	13%	.33
Length of hospital stay (days)	19±19	8.5±6.5	<.001

OSR = Open Surgery Repair; F-BEVAR = Fenestrated-branched endovascular aortic repair; eGFR = Estimated Glomerular Filtration Rate; SCI = Spinal Cord Injury.

1.1.5 Open Aortic Arch Repair

Aortic arch replacement remains one of the most formidable operations in cardiovascular surgery. The aortic arch is defined anatomically as that segment of the aorta from which the innominate artery, left common carotid artery (LCCA), and subclavian artery (LSCA) arise. Open replacement of the arch segment entails interrupting circulation to these brachiocephalic vessels during the repair, necessitating the use of strategies to protect the target organs, mainly the brain.

Approach to open repair of the aortic arch features right axillary artery CPB inflow and moderate hypothermic circulation arrest (HCA) with antegrade cerebral perfusion (ACP). This approach is usually used for all open arch replacement procedures, including hemiarch replacement, total arch replacements (Figure 3), and elephant trunk procedures (Figure 4). When performing total arch replacement, the Y-graft technique can also be used (Figura 4).

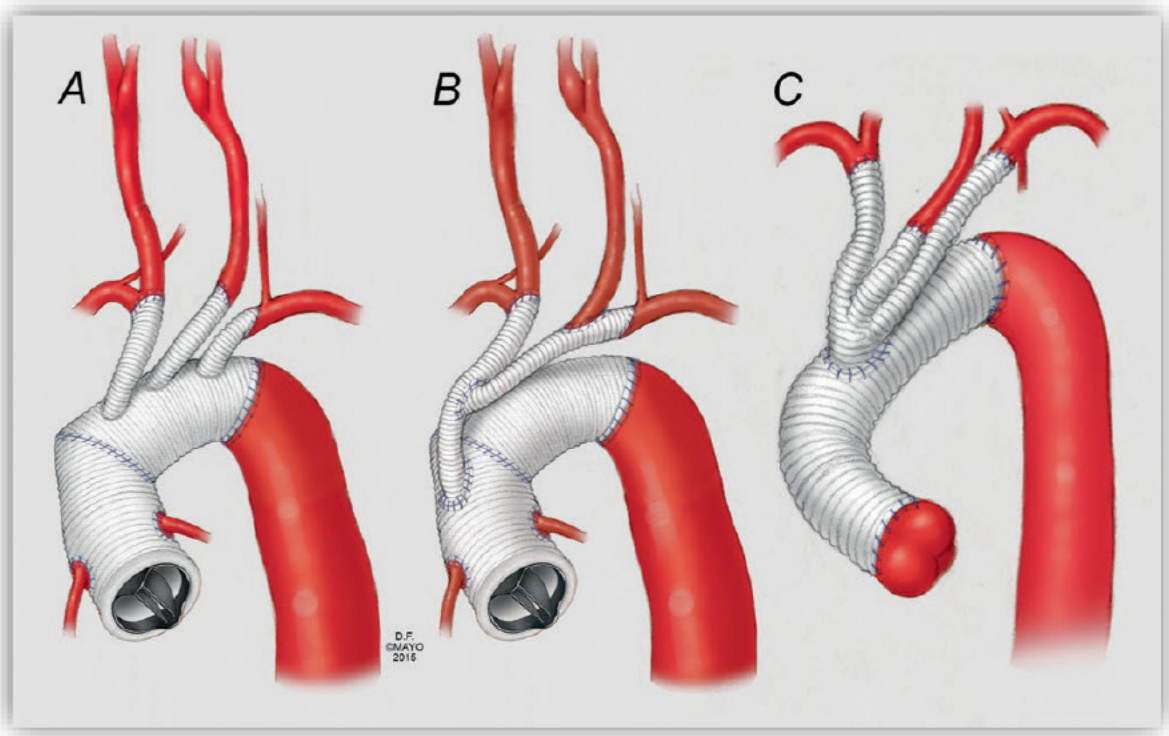


Figure 3 - Patient with extensive arch and ascending aortic disease, total arch replacement performed. (A) Graft configuration in a patient who need aortic valve replacement and separate grafts into all three supra-aortic trunks or (B) use of a bifurcated graft. (C) A trifurcated graft is less favored due to risk of compression.

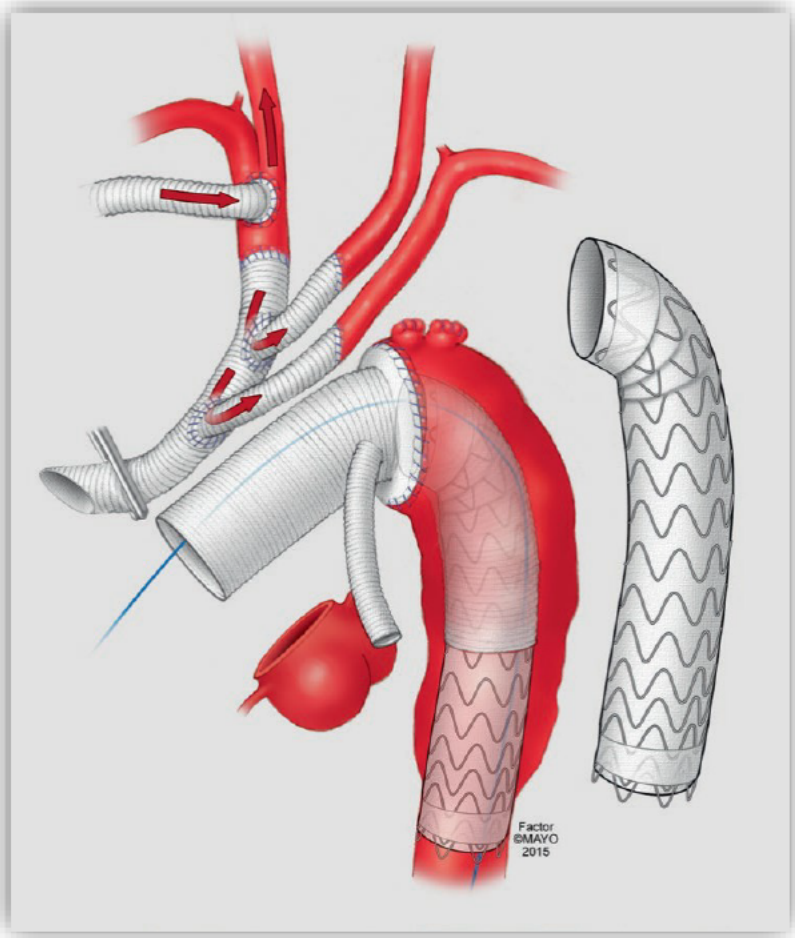


Figure 4 - Endograft deployment inside the elephant trunk graft with double Y- graft

1.1.6 Description of Study Population

Patients with aortic arch disease in need of a surgical replacement or repair who are eligible for open reconstruction, undergoing cardio-pulmonary bypass will be considered as potential subjects for enrollment in this study. For this study, it will be also considered patients with complex aortic aneurysms who are being evaluated for open surgical reconstruction. Those patients undergoing endovascular repair for complex aortic aneurysm will only be considered as potential subjects for this study if they are Crawford extent I to IV (Figure 5) and have known stage III (including IIIA and IIIB) chronic kidney disease (CKD) or if the patient has solitary kidney and known stage III (including IIIA and IIIB) CKD.

The study is designed with standard eligibility criteria to address any known or foreseeable factors that may compromise the outcome of the clinical investigation or the interpretation of results. Only patients who meet all of the inclusion criteria and none of the exclusion criteria listed in sections 5.1 and 5.2 will be enrolled. Subject eligibility will be initially assessed by the research team. Eligibility will be confirmed by the PI or Cardio and Vascular Surgery team based on review of anatomical extend of aneurysm disease and type of open surgical or endovascular repair. All patients will undergo a computed tomography angiography

of the chest, abdomen, and pelvis to determine the extent of aortic involvement. All patients will sign an informed consent form approved by the Mayo Clinic Institutional Review Board.

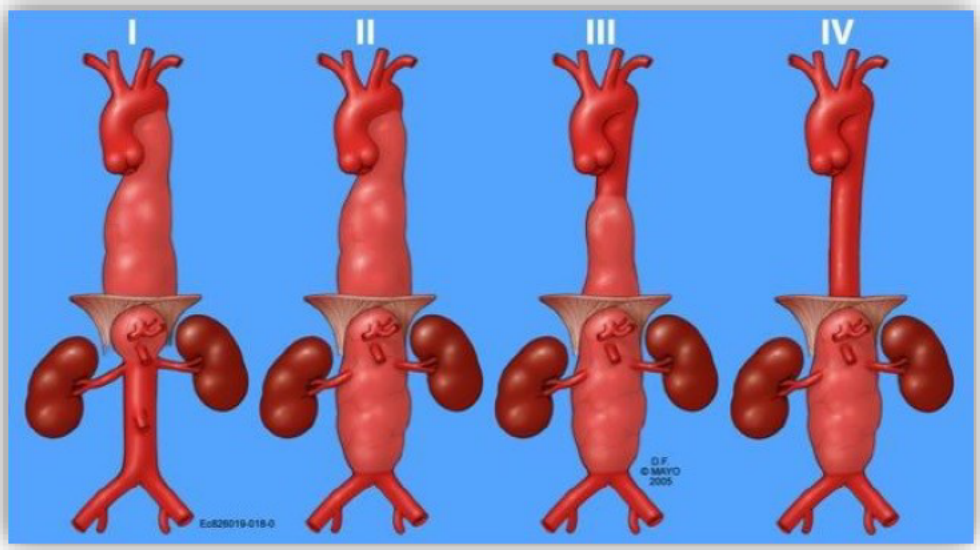


Figure 5 – Crawford classification of thoracoabdominal aortic aneurysms.

The decision of type of aortic aneurysm repair (e.g. open surgical or endovascular aortic repair) will be left at the discretion of the Vascular operating team. In general, endovascular repair is considered in patients who are anatomical candidates and have suitable vascular access and renal-mesenteric targets. Open surgical repair is considered in younger, fit patients or in those who do not meet minimum anatomical criteria for endovascular repair due to lack of sealing zones or unsuitable target vessels. Subjects who are offered either open or endovascular repair will be considered for enrollment in the study. The patient pre-operative clinical risk will be evaluated using cardiac, pulmonary, and renal non-invasive testing and standardized clinical scores (e.g. American Society of Anesthesiology Classification and Society for Vascular Surgery clinical comorbidity scores).

1.1.7 Rationale for Drug Use

As Tran MT *et al.*[42] have shown the correlation between oxidative metabolism through NAD biosynthesis and renal protection.

The energetic burden of continuously concentrating solutes against gradients along the tubule may render the kidney especially vulnerable to ischemia. Tran MT *et al.* demonstrated that the mitochondrial biogenesis regulator, PGC1α$^{-/-}$, is a pivotal determinant of renal recovery form injury by regulating NAD biosynthesis. Following renal ischemia, PGC1α$^{-/-}$, (also known as *Ppargc1a*$^{-/-}$) mice develop local deficiency of the NAD precursor niacinamide (NAM, also known as nicotinamide), marked fat accumulation, and failure to reestablish normal function.

Exogenous NAM improves local NAD levels, fat accumulation, and renal function in post-ischemic PGC1α$^{-/-}$mice. Inducible tubular transgenic mice (iNephPGC1α) recapitulate the effects of NAM supplementation, including more local NAD and less fat accumulation with better renal function after ischemia. PGC1α $^{-/-}$ coordinately upregulates the enzymes that synthesize NAD *de novo* from amino acids whereas PGC1α$^{-/-}$ deficiency or AKI attenuates the *de novo* pathway. NAM enhances NAD via the enzyme NAMPT and augments production of

the fat breakdown product β-hydroxybutyrate, leading to increased production of prostaglandin PGE2[43], a secreted autacoid that maintains renal function. NAM treatment reverses established ischemic AKI and also prevented AKI in an unrelated toxic model. Inhibition of β-hydroxybutyrate signaling or prostaglandin production similarly abolishes PGC1α-dependent renoprotection. Given the importance of mitochondrial health in ageing and the function of metabolically active organs, the results implicate NAM and NAD as key effectors for achieving PGC1α-dependent stress resistance [42].

Nicotinamide Riboside (NR) has been investigated as an NAD-boosting molecule because of its ability to cross the plasma membrane and, different than NAM, it does not inhibit NAD-hydrolyzing enzymes such as PARP-1 and SIRTUINS[44]. Recently, diet supplementation with NR was shown to increase NAD levels in mammalian cells and mice tissue and activate SIRT1 and SIRT3. This supplementation ameliorated metabolic defects of obese mice, boosted their energy expenditure and oxidative performance of skeletal muscle and brown adipose tissue, and, overall, reverted metabolic impairment[45]. NR supplementation also was able to increase mitochondrial NAD levels and improve mitochondria function of old mice[46].

Dellinger *et al.* have validated the safety and efficacy of repeated short-term dose BASIS™[47] in increases of NAD^+. Given the relatively benign safety profiles of NR and Pter the bioavailability of these substances and the animal experimentation to date, it would seem appropriate to explore possible benefits in humans for a variety of conditions, including cognitive impairment, NAFLD and kidney disease.

The beneficial effects of Pter in the prevention and treatment of disease has been largely attributed to its antioxidant and anti-carcinogenic actions[48]. The antioxidant activity of Pter was first characterized by Rimando *et al*., who demonstrated that Pter was similar to resveratrol in suppressing free radical formation caused by herbicide-induced oxidative stress in plants[49]. Later, it was shown that oral Pter treatment (40 mg/kg) for 6 weeks was able to enhance the levels of antioxidants (superoxide dismutase, catalase, and GSH peroxidase) in the liver and kidneys of Wistar diabetic rats[50].

Gao *et al.* suggested in your study that Pter may protect against acute renal ischemia reperfusion injury in rats potentially via inhibition of oxidative stress, iNOS expression and inflammation. In addition, the protective effects of Pter may be associated with inhibition of the TLR4/NF-κB signaling pathway[51].

Riche *et al.* have demonstrated in a trial the safety of long-term pterostilbene administration in humans. There were no statistically significant self-reported or major adverse drug reactions, concluding that pterostilbene is generally safe for use in humans up to 250 mg/day[52].

1.2 Investigational Agent

The investigational product, Basis™, contains a mixture of Nicotinamide Riboside (NR) (250 mg) and Pterostilbene (Pter) (50 mg) in capsular form. It will be a white substance contained within an opaque capsule. Non-medicinal ingredients contained within the capsule include microcrystalline cellulose, silicon dioxide and magnesium stearate.

1.3 Preclinical Data

1.3.1 Nicotinamide Riboside (NR)

In vitro studies have demonstrated that NR supplementation resulted in increased NAD^+ levels in yeast and cultured human cells [53,54]. NR also extended the lifespan of yeast cells in a SIRT2-dependent manner, suggesting that NAD^+-mediated sirtuin activation is the potential mechanism of action[53].

More recently, two *in vivo* studies evaluated the effects of prolonged NR-supplementation in mice[45,55]. Canto *et al.*[45] fed wild-type mice a high fat diet (35% fat, 26% carbohydrate, and 26% protein) with and without NR for 12 weeks. The mice receiving NR gained weight at a slower rate, had improved insulin sensitivity, increased endurance, enhanced mitochondrial oxidative capacity in brown adipose tissue, were more able to maintain body temperature during cold exposure, and increased muscle and liver NAD^+ concentrations, compared to the mice receiving the high fat diet alone[45]. Khan *et al.*[55] fed wild-type (control) and Deletor mice, which are prone to developing mitochondrial myopathy, a normal diet (11% fat, 65% carbohydrate, and 24% protein), supplemented with and without NR, for 16 weeks[55]. Although NR supplementation did not significantly increase skeletal muscle NAD^+ concentrations in either wild-type or Deletor mice, the NR containing diet increased mitochondrial biogenesis in skeletal muscle and enhanced lipid oxidation in brown adipose tissue of both wild-type and Deletor mice[55].

The effects of NR supplementation have also been assessed in several animal models of neurodegeneration[56,57]. Gong *et al.* [56] fed Tg2576 transgenic mice that normally develop symptoms resembling Alzheimer's disease a diet containing NR for 4 months, and found that NR supplementation attenuated the effects of beta-amyloid toxicity at the behavioral and cellular levels[56]. Shindler *et al.*[57] injected NR intra-ocularly to mice with experimental autoimmune encephalomyelitis. Compared to mice injected with phosphate buffered saline, injections of NR delayed retinal ganglion cell death. Sasaki *et al*.[58] treated mouse dorsal root ganglion neurons with NAD^+ precursors including NR, nicotinamide and NA, and found that all three increased the activity of the NAD^+ pathway and delayed axonal degeneration. In this study, it was demonstrated that NR is as effective as other NAD^+ precursors (nicotinamide and NA) in increasing NAD^+ levels. These results suggest that increased NAD^+ availability resulting from NR supplementation may delay axonal degeneration in some degenerative diseases.

Brown *et al.*[59] reported that mice injected twice daily with NR, at a dose of 1,000mg/kg for five days, and subjected to specific noise levels, did not exhibit expected noise-induced hearing loss. Subsequent examination of harvested cochlea of the ear showed that that NR prevented spiral ganglia neurite degeneration, and the authors suggested that these beneficial effects were mediated by NAD^+-dependent SIRT3. Middle-aged mice of a phenotype susceptible to non-alcoholic fatty liver disease (NAFLD) were found to have a reduced systemic NAMPT and NAD^+ level, in addition to many of the characteristics of NAFLD, including lipid accumulation, inflammation and insulin insensitivity]. When a high-fat diet was consumed, NAMPT and NAD^+ level decreased further and many NAFLD characteristics were exacerbated. NR given orally improved NAD^+ levels and attenuated many NAFLD characteristics, even in the presence of a high fat diet. For example, body weight decreased, and Kupffer cell accumulation of lipid was decreased, as were levels of the inflammatory cytokines TNFα and IL-1β in the liver [60].

Mukherjee *et al.*[61] also reported beneficial effect on the livers of mice subjected to partial hepatectomy. Following the addition of NR to drinking water, deoxyribonucleic acid (DNA) synthesis and restoration of regenerating livers was observed, and there was a lessening of liver regeneration-induced steatosis. Mills *et al.*[62] utilized the physiological product of NR metabolism, NMN, in the diets of aging mice, to mitigate many typical aging processes such as increased body weight, decreased insulin sensitivity, deteriorating eye function, and reduction in bone density. In a mouse model of muscular dystrophy, treatment with NR was reported to prevent the muscle stem cell senescence that is normally exhibited in this disease. In addition, NR supplementation induced a mitochondrial protein effect that rejuvenated muscle stem cells in aging mice, thus ameliorating the decrease in muscle mass often seen in aging[63].

Overall, these studies have demonstrated that in animal models, supplementation with NR increases the availability of NAD^+ *in vivo*, resulting in several beneficial outcomes in cell mitochondria. Chi and Sauve[64] have proposed that NR is a third form of Vitamin B3 and plays a role in ameliorating metabolic and age-related deficiencies of mitochondrial function.

1.3.2 Pterostilbene (Pter)

The numerous beneficial effects of Pter in the prevention and treatment of human disease has been largely attributed to its antioxidant and anti-carcinogenic actions [48]. The antioxidant activity of Pter was first characterized by Rimando and colleagues, who demonstrated that Pter was similar to resveratrol in suppressing free radical formation caused by herbicide-induced oxidative stress in plants[49]. Later, it was shown that oral Pter treatment (40 mg/kg) for 6 weeks was able to enhance the levels of antioxidants (superoxide dismutase, catalase, and GSH peroxidase) in the liver and kidneys of Wistar diabetic rats[50].

Post myocardial infarction, restoration of blood supply often causes further damage to cardiac muscle cells, possibly with the involvement of SIRT-1. In an in vitro model of ischemia reperfusion injury, Guo *et al.* (2016) demonstrated that adding Pter at concentrations of 0.1 to 0.3 μMol to culture media was cardioprotective, and was specifically related to both the enhancement of SIRT-1 expression and the stimulation of SIRT-1 activity[65].

1.4 Clinical Data to Date

In a recent randomized, double-blind, three-arm crossover, 24-hour, single-dose pharmacokinetic study was conducted to evaluate the pharmacokinetics of NR and its metabolites in humans[66]. Twelve healthy subjects were enrolled (6 male, 6 female) and randomized to a three-treatment sequence after screening and passing eligibility criteria. Subjects served as their own control and received a single dose of 100mg, 300mg, or 1,000mg of NR on three test days, separated by a 7-day washout period. The highest dose (1,000 mg/day) was equivalent to 16.7 mg/kg bw/day in a 60-kg individual. Additionally, these values were at least fifty-six times less than those shown to be safe in single oral acute dose and 14-day toxicity studies in rats (5,000 mg/kg/day). NR supplementation increased blood concentrations of NAD^+, NMN and nicotinic acid adenine dinucleotide (NAAD) in a dose-dependent manner[66]. At each dose of NR, both NAAD at 8 hours and NAD^+ at 24 hours were elevated. NAAD is considered to be an appropriate biomarker of NAD^+ absorption because it is lacking in blood at baseline, thus reducing background interference. No serious adverse events were reported. Several clinical trials are still being performed in humans (Table 4).

A recent randomized, double-blind, placebo-controlled trial found that treatment with daily Pter (250 mg) for 6-8 weeks reduced blood pressure and promoted minor weight loss in

hypercholesterolemic subjects[67]. However, LDL levels increased in subjects taking Pter, and this is believed to be due to its ability to act as peroxisome proliferator-activated receptors (PPAR) agonist, which is a transcription factor that regulates lipid metabolism[67].

Table 4. Clinical Trial with NR/NMN

Author	Research Institution	Population	n	NR/NMN	Endpoint	Dose/day Duration	Trial ID #	Status
Samuel A.J. Trammell *et al.* [66]	KGK Synergize Inc.	Healthy Subject	12	NR	Safety and efficacy	1000mg 7 days	NCT0219 1462	Published
Ryan W. Dellinger *et al.* [47]	Elysium Health	Healthy Subjects (60 to 80 years of age)	120	NR (Basis®)	Dose-dependent effect	250-500mg 60 days	NCT0267 8611	Published
Kevin O'Brien *et al.* [68]	University of Washington Seattle, US	Healthy Subjects	8	NR	Pharmaco kinetic (Percent change average/st eady state)	250-2000mg 9 days	NCT0268 9882	Published
Christopher R Martens *et al.* [69]	Clinical Translational Research Center Boulder, Colorado, US	Healthy middle-aged and older adults	24	NR (Niagen®)	Adverse event	1000mg 6 weeks	NCT0292 1659	Published
Ole Dollerup *et al.* [70]	Medical Research Laboratories, Aarhus University Hospital Aarhus, Denmark	Obese subjects	40	NR (Niagen®)	Insulin sensitivity	2000mg 3 months	NCT0230 3483	Published
Kevin O'Brien *et al.*	University of Washington Seattle, US	Heart Failure	30	NR	Adverse event	500-2000mg 12 weeks	NCT0342 3342	Recruiting
Patrick Chinnery *et al.*	Cambridge University, UK	Mitochondrial Diseases	10	NR (Basis®)	Safety/Bioavailability	20 mg/kg 4 weeks	NCT0343 2871	Recruiting
Patrick Schrauwen *et al.*	Maastricht University, Netherlands	Obesity/Insulin resistance	15	NR (Niagen®)	Insulin sensitivity	1000mg 6 weeks	NCT0283 5664	Recruiting

Michael N Sack *et al.*	National Institutes of Health Clinical Center Bethesda, Maryland, US	Healthy Subjects	42	NR	Refeeding IL-1 secretion	1000mg 7 days	NCT02812238	Recruiting
Dost Ongur *et al.*	Mclean Hospital	Healthy Subjects	60	NR	Brain NAD/NADH ratio	1000mg 2 weeks	NCT03151707	Recruiting
Britt Christensen *et al.*	University Hospital Aarhus, Denmark	Healthy male	12	NR (Niagen®)	Serum NR	500-1000mg 8 hours	NCT02300740	Completed (unpublished)
Gordon Schacter *et al.*	KGK Synergize Inc. London, Ontario, Canada	Healthy Subjects	140	NR (Niagen®)	Blood, urinary, and muscle NR metabolites	100-1000mg 8 weeks	NCT02712593	Completed (unpublished)
Brent A. Bauer *et al.*	University of Minnesota Minneapolis/MN,US Mayo Clinic Rochester/MN, US	Mild concussion	48	NR	Changes in brain NAD levels	750mg 12 weeks	NCT02721537	Recruiting
Eugene Rhee *et al.*	Massachusetts General Hospital Boston, Massachusetts, US	Acute Kidney Injury	24	NR (Basis®)	NR levels and adverse event	500-2000mg 2-8 days	NCT03176628	Recruiting
Dale Wilson *et al.*	KGK Synergize Inc. London, Ontario, Canada	Healthy Subjects	12	NR (Niagen®)	Terminal half-life (t ½)	100-1000mg 24 h (3 times)	NCT02191462	Completed (unpublished)
Gareth Lavery *et al.*	University Hospitals Birmingham NHS Foundation Trust Birmingham, West Midlands, UK	Aging	12	NR	Mitochondrial function	1000mg 3 weeks	NCT02950441	Recruiting
Becky Powers *et al.*	University of Texas Health San Antonio	Mild Cognitive Impairment	26	NR	Change in cognitive assessment	250-1000mg	NCT02942888	Recruiting

	San Antonio, Texas, US					10 weeks		
Christopher R Martens *et al.*	Neurovascular Aging Laboratory Newark, Delaware, United States	"Amnestic" mild cognitive impairment	58	NR (Niagen®)	Cognitive scores at 12 weeks	1000mg 12 weeks	NCT03482167	Not yet recruiting
Rudy Valentine *et al*.	Iowa State University, United States	Aging Lipemia	40	NR (Niagen®)	Blood and peripheral blood mononuclear cell (PBMC) levels	500mg 7 days	NCT03501433	Recruiting
Mary Anne Buggia *et al.*	Boca Raton, Florida, United States	Older Adults	40	NR (Niagen®)	Cognitive Function, Mood and Sleep	300mg-1000mg 8 weeks	NCT03562468	Recruiting
Michael N Sack *et al.*	(NHLBI)Bethesda, Maryland, United States	Heart failure		NR (Niagen®)	Mitochondrial function in skeletal muscle	500mg-2000mg 12 weeks	NCT03565328	Not yet recruiting
Charalampos Tzoulis *et al.*	Haukeland University Hospital	Parkinson's Disease	200	NR	Clinical rating scale approved by movement disorder society.	1000mg 52 weeks	NCT03568968	Not yet recruiting
Bryan Kestenbaum *et al.*	University of Washington Seattle, US	Chronic kidney disease	30	NR or Coenzyme Q10 (Cross-over trial)	Maximal aerobic capacity during cycle ergometry	1200mg 6 weeks	NCT03579693	Not yet recruiting
Samuel Klein *et al.*	Washington University School of Medicine Saint Louis, Missouri, US	Glucose Metabolism Disorders	50	NMN	Change in insulin sensitivity	250mg 8 weeks	NCT03151239	Recruiting

1.5 Dose Rationale

The choice of dose, route of administration, dosage regimen and dosage period were based on animal and human studies that demonstrated safety.

The safety profile of NR has now been established in rodent species, i.e., the no-observed-adverse-effect-level (NOAEL) and lowest-observed-adverse-effect-level (LOAEL) of 300 and 1,000 mg/kg bw/day in mice, and the safety of a single dose of up to 5,000 mg/kg bw in rats.

The conversion ratio between human adult and mouse dose of NR is a factor of 12.3 [66]. Preliminary research in humans has resulted in the observation of no serious adverse events (AE) in subjects given up to 1,000 mg NR per day.

Limited animal model and human data exist on the pharmacokinetics and safety of orally administered NR. However, in the aforementioned randomized, double-blind, cross-over, pharmacokinetic study, NR was well tolerated and presented no toxicity following a single dose up to 1,000 mg. Moreover, in an acute toxicology study, rats that were given a single oral dose (5,000 mg/kg) of NR did not show clinical signs of toxicity or mortality (unpublished results).

1.6 Risks and Benefits

Given that both NR and Pter are present in small amounts in dietary sources, humans are continuously exposed to this compound. NR has been granted generally recognized as safe (GRAS Notice No 635) status from the FDA. Based on the structural similarities between NR and nicotinamide, on the fact that nicotinamide is a downstream metabolite of NR digestion, and on safety findings in a recently-published study, it is known that NR is very similar to nicotinamide in its toxicological profile. The safety profile of NR has been established in rodent species, i.e. the NOAEL and LOAEL of 300 and 1,000 mg/kg bw/day in mice, and the safety of a single dose of up to 5,000 mg/kg bw in rats. The conversion ratio between human adult and mouse dose of NR is a factor of 12.3. Preliminary research in humans has resulted in the observation of no serious AEs in subjects given up to 1,000 mg NR per day. Similarly, subjects consuming up to 250 mg Pter reported no significant adverse events. Similar to NR, Pter is present in the human diet in sources such as blueberries, grapes, and legumes[71]. In a comprehensive review of Pter, Kosuru *et al.* examined the evidence for the protective effects of Pter against heart disease, brain injuries and cancer, as well as its improved bioavailability, compared to resveratrol [72]. The authors concluded that Pter "represents an active phytonutrient with pleiotropic applications". In their safety review, which included several of the studies reviewed in this document, they suggested that Pter has no significant toxic effects.

There are no known interactions between NR and other medicinal products and other forms of interactions. NR has not been tested in pregnant or lactating women, therefore it is recommended for women of childbearing age to use effective contraception when using this product. NR is not for use in children under 18 years of age. Mild nausea, headaches, diarrhea and abdominal discomfort have been reported with the use of NR. Despite no serious adverse effects and no drug interactions being reported, we will monitor liver function, coagulation/bleeding time, muscle aches, blood pressure, and heart rate, as well as periodic consultations to ensure the product safety.

Animal research has suggested that the combination of NR and PT may prove to be beneficial in a variety of human conditions such as cognitive impairment, non-alcoholic fatty liver disease (NAFLD), and rosacea. Currently, human research is limited. The information gleaned from animal studies, together with the knowledge of the safety of specific dosages of NR and PT, will assist in determining an appropriate dosage of EH301, as well as suitable indications, for the application of EH301 for future clinical trials.

2 Study Objectives

2.1 Primary Objective

To assess the efficacy of Basis™ (Nicotinamide Riboside and Pterostilbene) in preventing acute kidney injury (AKI) in patients undergoing complex aortic aneurysm repair and open aortic arch reconstruction.

2.2 Secondary Objective

To assess the efficacy of Basis™ (Nicotinamide riboside and Pterostilbene) in reducing the incidence of heart attacks and central nervous system (CNS) complications (stroke and spinal cord injury) in patients undergoing complex aortic aneurysm repair and open aortic arch reconstruction.

3 Study Design

3.1 General Description

This study is a single-center, prospective, randomized, double-blinded, placebo-controlled phase II clinical trial to evaluate the efficacy of "NAD^+ supplementation" with Basis™ (Nicotinamide Riboside and Pterostilbene) in preventing acute kidney injury (AKI) in patients undergoing complex aortic aneurysm repair and open aortic arch reconstruction.

3.2 Number of Subjects

The sample size will be 119 patients in each group (placebo vs. Basis™), 238 patients total, considering a dropout rate of 15%.

3.3 Duration of Participation

The duration of patient's participation is expected to be concluded in 6 months post index procedure.

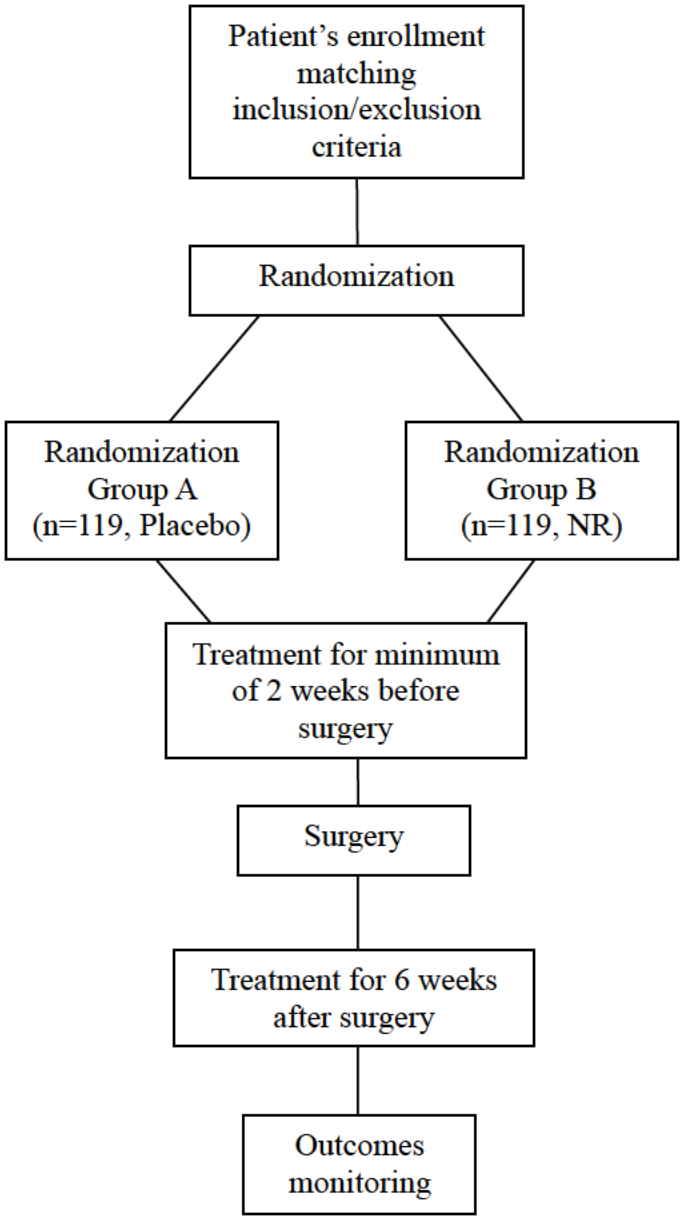


Figure 2 - Study flowchart

3.4 Primary Study Endpoints

The primary endpoint will be improvement in incidence of AKI measured by eGFR using the RIFLE criteria. The laboratory tests will be compared to the baseline and performed after surgery and the assessment of improvement/no change/worsening of AKI will be followed at 6-week, and 6-month period after surgery. The follow-up duration will last 6 months.

To define AKI, the RIFLE criteria will be used and the patients should be classified using the highest baseline eGFR value and the lowest post-op eGFR value within 30 days. Also the following criteria will be applied to define AKI: increase of 1.5x in SCr or decrease of 25% or more in GRF.

Levels of urea, creatinine clearance (CrCl), cystatin C, and N-gal will also be measured in blood. eGFR will be determined by using predictive MDRD[73]. In urine, levels of TIMP-2, KIM-1, and IGFBP7 will be measured.

$$eGFR = 175 \times (S_{Cr})^{-1.154} \times (age)^{-0.203} \times 0.742 \text{ [if female]} \times 1.212 \text{ [if Black]}$$

eGFR (estimated glomerular filtration rate) = mL/min/1.73 m^2
S_{cr} (standardized serum creatinine) = mg/dL
age = years

3.5 Secondary Study Endpoints

The secondary endpoint will be the decrease in the incidence of myocardial infarction, stroke, bowel ischemia, and spinal cord injury. However, due to the low incidence of these complications, the study may not have the power to detect potential small differences between the outcomes, but might provide proof of concept of protective effect from injuries in those organs. Also, other parameters such as NAD, NMN, NR levels, PCR for inflammatory markers, ELISA, renal biomarkers, and metabolomics levels of *de novo* pathway will also be measured.

As another secondary endpoint, we will survey the subject's quality of life using SF-36 Health Survey after the procedure. The comparison will be done at the following time points: pre-procedure, 6-week follow-up, and 6-month follow-up.

3.6 Identification of Source Data

Source data will be obtained from the electronic medical record and documented in study specific case report forms (paper and/or electronic data capture).

4 Study Drug

4.1 Description

4.1.1 Chemical Structure of Nicotinamide Riboside (NR)

NR is a single chemical moiety containing nicotinamide and ribose[43]. The investigational product, Basis™, contains a synthetic NR that is nature-identical to naturally-occurring NR, does not induce flushing or pruritus and has no effect on lipid levels.
Common name: Nicotinamide riboside chloride
Chemical name: 3-(Aminocarbonyl)-1-β-D-ribofuranosyl-pyridinium chloride (1:1)
Chemical formula: C11H15N2O5.Cl
Molecular weight: 290.70 g/mol

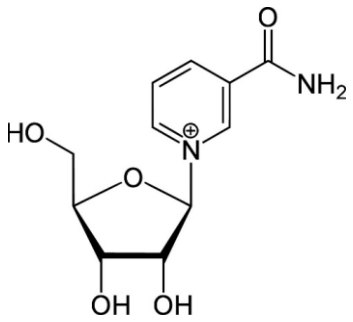
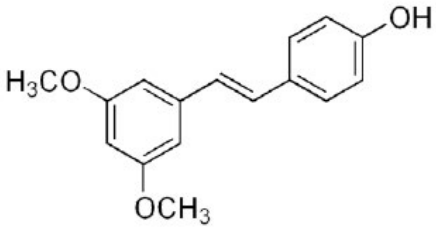


Figure 3 - NR Structural formula

4.1.2 Chemical Structure of Pterstilbene (Pter)

Pterostilbene (Pter) is a stilbenoid compound, characterized by two aromatic rings connected by a methylene bridge backbone, and it has two methoxy groups and one hydroxyl group extending from the aromatic rings.

Common name: Pterostilbene
Chemical name: 3,5-dimethoxy-4'-hydroxystilbene
Chemical formula: C16H16O3
Molecular weight: 256.296 g/mol



Pterostilbene

Figure 3 - Pter Structural formula

4.2 Treatment Regimen

Study medication will be administered a total of 1000 mg/day of NR and 200 mg/day of Ptero in a regimen of 2 capsules of Basis™ (each capsule 250 mg of NR and 50 mg of Ptero) 2 times a day by mouth for a minimum of 2 weeks before surgery and 6 weeks after surgery. Study medication is to be taken every 12 hours. In the event of a missed dose, the study medication can be taken as soon as a missed dose is noted, making sure to allow at least 6 hours interval period between doses. Missed doses (not taken) will be recorded in the medication diary.

→If surgery is rescheduled, the patient will receive instructions based on how long the period of delay will be (see section "rescheduling surgery").

→ Basis™ or placebo will be held while patients are in a NPO status. The administration of Basis™ or placebo will be restarted when clinically indicated.

→ Patients unable to take Basis™ or placebo orally for more than 5 days post procedure will be withdrawn from the study (see "when and how to withdraw subjects")

4.3 Method for Assigning Subjects to Treatment Groups

The total number of 238 patients will be randomly assigned into one of two groups. The treatment group will be treated with Basis™ (NR + Pter) and the control group, with correspondent placebo. As the study is designed to be double-blinded, neither the patient nor the investigator and the research team will know what is being given, the patient order list will be under awareness and ownership of Research Pharmacy team only. Research Pharmacy will be responsible for dispensing, according to the randomization.

4.4 Preparation and Administration of Study Drug

Both compounds, Basis™ and placebo, will be provided by Elysium Health Company. Basis™ and placebo will be provided ready for use, discarding the need of drug manipulation by the investigator or any study team member.

The storage and control of Basis™ and placebo will be the responsibility of Mayo Clinic Outpatient Research Pharmacy [redacted] The awareness of randomization and what the patient is receiving will be exclusively the responsibility of the Research Pharmacy team, as the study is double blinded (patients and investigator's team).

After all the patient enrollment process is fully disclosed, and the baseline blood tests are collected, treatment allocation will be established by randomization and the patient will receive the appropriate compound by the Research Pharmacy, unknown by the patient and by the investigator and his team.

Via of administration will be by mouth, 2 capsules (each capsule containing 250 mg NR and 50 mg Ptero) every 12 hours for a minimum of 2 weeks before surgery and 6 weeks after surgery.

4.5 Subject Compliance Monitoring

Subjects will be provided with a medication diary (see "Attachment" Section) in which they will track their administration of the Basis™ or placebo. At Visit 4 (procedure day), patients will be asked to present the medication diary to check compliance. A new medication diary will be provided to the patient for the remainder of the treatment period. The study team will also assess subject's compliance at Visit 7 (see table 6).

Subjects, who are found to be less than 90% compliant, excluding NPO cases, will be counseled in the importance of taking the study medication daily. Patients who meet less than 50% of compliance will still be asked to come back for remaining follow-up visits and data collection according to intention to treat model. However, notifications and documentations will be appropriately filed by study team for further data analysis. See details in section "Data Collection and Follow-up for Non-Complaint (protocol and/or medication) and Withdrawn Subjects".

4.6 Prior and Concomitant Therapy

Medications taken routinely by patients will be allowed. There will be no rescue therapy - open-label administration of active drug when a patient's condition is deteriorating.
Patients taking multivitamins containing vitamin B3 derivatives in a dose > 200 mg/day will be excluded from the study.

4.7 Packaging

The investigational product, Basis™, is a white substance contained within an opaque capsule of 300 mg. Non-medicinal ingredients contained within the capsule includes microcrystalline cellulose, silicon dioxide and magnesium stearate. The placebo will appear identical.

Study drug/placebo will be packaged and shipped in separate boxes clearly labeled and factory sealed. All the contents of any kits or boxes will have associated labeling.

4.8 Randomization / Blinding

Participants will be randomized in a 1:1 fashion to one of two arms (Nicotinamide Riboside and Pterostilbene versus Placebo), using a Pocock-Simon dynamic allocation procedure[74]. This allocation procedure utilizes a minimization algorithm balances the marginal distributions of the stratifications factors across the intervention groups to balance the marginal distributions of the stratifications factors across the intervention groups. For this study the randomization will be stratified according to the type of surgery using the following strata:

1. Open total aortic arch replacement or repair;
2. Open non-total aortic arch replacement or repair;
3. Open reconstruction of complex thoracoabdominal aortic aneurysm Crawford extent I to IV;
4. Open reconstruction of complex abdominal aortic aneurysm of juxtarenal, pararenal, and paravisceral type;
5. Endovascular repair of complex pararenal or thoracoabdominal Crawford extent I to IV aortic aneurysms and known stage III and IV CKD;
6. Endovascular repair of complex pararenal or thoracoabdominal Crawford extent I to IV aortic aneurysms and solitary or single functioning kidney and known stage III and IV CKD.

After study personnel confirm that a subject meets all study entry criteria, they will be randomized. All study medication will be labeled, per pharmacy guidelines/policy, in a blinded fashion and dispensed by the pharmacy according to subject name.

4.9 Receiving, Storage, Dispensing and Return

4.9.1 Receipt of Drug Supplies

Investigational Products (IP) from Elysium Health Company are received by pharmacy personnel. Research pharmacy staff verifies information on the packing slip matches the contents of the shipment. The following are verified by receiving personnel upon receipt of shipment: product identification, sponsor product label, quantity of items received, package size(s), lot numbers, expiration dates if available, product condition, and maintenance of storage conditions. Any discrepancies in shipment are communicated immediately to study PI. Investigational products that do not contain an expiration date are deemed in date and acceptable to use until notice of expiration is received from sponsor or supplier. Research staff contacts research team and/or suppliers quarterly to determine if retest date or expiration is available. After shipment has been checked and verified, IP is immediately placed in appropriate storage conditions as indicated in study documentation. For product delivered with a temperature recording device which may be electronically interrogated, data will be downloaded and saved in PDF format in

the Investigational Drug Services (IDS) electronic management application if the device supports such function.

If the data is not downloadable, guidance documents provided with shipment are followed for receipt of product. If temperature recording device indicates an alarm condition was present during shipment, IP is quarantined and sponsor is contacted for further instructions. Unless specified by sponsor, all temperature logging devices received with IP shipment are discarded post inspection / interrogation. Invoices are dated, signed, scanned, and uploaded electronically into the IDS electronic management application at time of receipt. Signed/dated hard copies of invoices are double checked per page against scanned uploaded images to ensure scanned image represents a true copy of the original document. Once accuracy of the scanned image has been verified, hard copies of invoices/shipping documents are discarded. If required by protocol, IVRS/IWRS receipt verification is completed.

4.9.2 Storage

The Investigational Pharmacy is a secure, badge access area accessible to authorized pharmacy personnel only. Investigational products are stored as per study documentation. Either medication or placebo will be stored in a secure location in room temperature and light protected conditions by Outpatient Research Pharmacy. Temperature monitoring is recorded via a continuous electronic monitoring system. All storage areas and devices that contain investigational products are monitored by this system. Temperature sensor devices are calibrated per local policy and calibration certificates are available upon request. Sponsor specific temperature monitoring devices are not utilized. Temperature data from the continuous electronic temperature monitoring system is stored electronically in a dedicated server and is readily retrievable upon request. Local procedures are followed in the event of an out of range condition reported by the continuous electronic monitoring system.

In the event of a temperature excursion during storage, investigational product(s) will be quarantined if required by protocol. IP may be relocated to backup storage sites per local policy if necessary. The study monitor, sponsor, principal investigator (PI), and clinical research coordinator are notified of the temperature excursion. Documentation of temperature excursion is described in a Note to File and saved in the electronic IDS management system. Additionally, the temperature excursion is documented in a report generated by the electronic IDS system and is submitted to sponsor(s) if required per protocol. The report generated by the electronic IDS system along with any correspondence regarding the excursion is saved within the electronic IDS management system. Sponsor specific temperature excursion reporting forms are not utilized. Instructions from sponsor regarding the disposition of IP involved in a temperature excursion are followed.

4.9.3 Dispensing of Study Drug

Pharmacists and/or pharmacy technicians under the supervision of a pharmacist will prepare and dispense investigational products per study protocol. Lot numbers and expiration dates of commercial supplies utilized in the preparation of investigational products such as syringes, needles, IV bags, tubing, diluents, and standard of care products are not tracked.

4.9.4 Return or Destruction of Study Drug

In general, IP and IP wastes generated on site as a consequence of participation in a investigator’s study are disposed of via placement in a designated, appropriate waste container

per local policy and regulation. Investigator specific destruction forms are not utilized to document medication destruction. Certificates of destruction for any individual container are not available.

Empty containers, vials, or empty injectable syringes of investigational products generated from preparation and subsequent administration of an IP to a patient are not retained for monitor inspection following dose preparation; instead these items are disposed of by placement in an appropriate waste container based on the hazard risk of the IP (e.g. hazardous, chemotherapeutic, biohazardous). Disposition is documented in the IDS electronic management application. Unless pharmacy is the only unblinded party in a study, IDS pharmacy staff will not document IP destruction or drug returns within sponsor electronic systems. In addition, all investigational products (i.e. tablets, capsules, injections, and/or containers) that are returned by subjects, as well as any returned empty boxes and cartons are disposed of in the manner described above immediately after appropriate documentation of such return has occurred in the IDS electronic management application. These items are not retained for monitor inspection.

Any expired or quarantined investigational products accumulated as a result of site participation in a sponsor's study will be segregated from regular IP inventory, sealed in a tamper evident bag and marked "DO NOT USE". Monitors will be contacted by the investigational pharmacy to schedule a monitoring visit within 90 days to reconcile these products so that these items may be shipped back to sponsor at sponsor's expense if required by sponsor, or disposed of on site. If sponsor extends expiration date it is the responsibility of the sponsor or their designee to relabel IP. At the end of the 90 day period, if monitor has not scheduled a visit or made other arrangements, all expired or quarantined products are disposed of on site and same is documented in the IDS electronic management application. Upon closure of a protocol, study monitors are requested to reconcile all remaining investigational products on hand within 90 days. At the conclusion of the 90 day period, unless sponsor has contacted the investigational pharmacy to make arrangements, remaining products are disposed of on site, and same will be documented in the IDS electronic management application.

5 Subject Selection Enrollment and Withdrawal

5.1 Inclusion Criteria

A patient may be included in the study if the following conditions are present:

1. Male or female;
2. Age ≥ 18 years old;
3. Patients who match the criteria for indication of elective open aortic arch replacement or repair:
    a. Total arch;
    b. Non-total arch;
4. Patients who match the criteria for indication of elective complex aortic aneurysm repair has to respect the following conditions:
    a. Patients undergoing complex aortic aneurysm open reconstruction with a suprarenal clamp;
        i. thoracoabdominal aortic aneurysms Crawford extent I to IV ;
        ii. abdominal aortic aneurysms of juxtarenal, pararenal, or paravisceral type;
    b. For endovascular approach:

i. patients with pararenal or thoracoabdominal aortic aneurysms Crawford extent I to IV and known stage III or IV CKD;
ii. patients with pararenal or thoracoabdominal aortic aneurysms Crawford extent I to IV and solitary or single functioning kidney and known stage III or IV CKD;

5. Patients will be allowed to participate in concomitant endovascular aortic stent-graft trials and prospective cohort studies as long as these do not involve another investigational study drug.

5.2 Exclusion Criteria

Patients must be excluded from the study if any of the following conditions are true:

5.2.1 General Exclusion Criteria

1. Unwilling to comply with the follow-up schedule;
2. Inability or refusal to give informed consent by the patient or a legally authorized representative;
3. Known pregnancy or breastfeeding;
4. Subject who takes multivitamins containing vitamin B3 derivatives in a dose > 200 mg/day;

5.2.2 Clinical / Laboratory Exclusion Criteria

5. Renal failure defined as eGFR< 15 mL/min/1.73m^2
6. Patients in permanent Renal Replacement Therapy;
7. Patients with chronic liver disease: Child-Pugh score class B and C;

5.2.3 Medication Exclusion Criteria

8. Patients in chemotherapy scheme;
9. Patients taking any immunosuppressant, except for corticosteroids;
10. Patients taking any of these well-known P-glycoprotein substrates: digoxin, fexofenadine, indinavir, sirolimus.

➔ For some types of P-glycoprotein substrates, subjects will be closely monitored (see section “Use with caution medications”).

5.3 Subject Recruitment, Enrollment and Screening

Patients will be recruited from cardiac and vascular surgery.

5.3.1 Informed Consent Process

All patients must provide informed consent prior to any study related procedures being performed. The Institutional Review Board (IRB) approved consent form will be signed and personally dated by the subject or legally authorized representative, and the person who conducted the informed consent discussion.

5.3.2 Pre-Screening/Screening

Subjects will be screened by the research team before enrollment into the study. Blood will be collected to assess baseline values. The research team will confirm subject's eligibility based upon review of required test results.

➔ Patients who do not qualify for participation in the study during the pre-screening assessment will be considered screening fail. Those cases will be documented and the patients will be informed.

5.3.3 Enrollment

Enrollment into this study will begin following Food and Drug Administration (FDA) and Investigational Site IRB approval. Subjects will be evaluated based upon their medical history, disease process suitability for the study.

If the research team determines that the patient is eligible for enrollment into the study, the patient will be contacted and the study will be discussed with the patient.

5.4 Rescheduling surgery

In case the surgery is needed to be rescheduled, for any reason, and the study medication has been already started, the patient will be instructed based on the period of delay:

1. If the delay period is up to 2 weeks (1 to 14 additional days), the patient will be instructed to continuously take the study medication until the day of surgery. In this case the patient will take the study medication before the surgery a minimum of 2 weeks + 1 day and a maximum of 4 weeks.
2. If the delay period is more than 14 days, the patient will be instructed to stop the study medication. A washout interval of 48-72 hours will be established. The study medication will be re-started beginning from day 0 for completion of 2 weeks period before surgery.

➔ Additional Basis™ or placebo will be provided to the patient as needed.

5.5 Follow-up

Subjects will be asked to return for follow-up visits at 6 weeks and 6 months after the procedure. At each follow-up visit, subjects will undergo an evaluation for adverse events, concurrent medication, and required laboratory testing (Table 6).

5.6 Follow-up Visit Windows

Follow-up visits will be scheduled at appointed times after the date of surgery. The investigator recognizes that subjects may not be able to return for follow-up visits on the exact date required. Thus, a period during which each visit is allowed is demonstrated below (Table 5).

Table 5. Follow-up and Associated Visit Windows

Follow-up Visit	Visit Window (days)
Treatment started	-14
Procedure day (reference)	0

6 - 8 weeks	42-56
6 months	150-210

*** Note: Discharge data may be collected up to 4 days prior to discharge date**

5.7 When and How to Withdraw Subjects - Early Withdrawal of Subjects

A patient may withdraw from the clinical study prior its completion at any time and should notify the research team of this event. The investigator may also withdraw the subject from the clinical study at any time based on their medical judgement, such as in the following cases:

1. Subject safety issues
2. Patients who had a major adverse event from the procedure (MAE) (Table 3), requiring long-term hospitalization and disabling the patient of following the treatment regimen.

5.8 Data Collection and Follow-up for Non-Compliant (protocol and/or medication) and Withdrawn Subjects

Subjects will be considered non-compliant if meet any of the following:

1. Less than 50% of compliance according to medication diary
2. Requirement of NPO for longer than 5 days after the procedure
3. Failure of subject to adhere to protocol requirements

In the aforementioned cases, patient will be asked to come back for remaining follow-up visits and data collection respecting intention to treat model. However, notifications and documentations will be appropriately filed by study team for further data analysis.

If subject refuses to return a follow-up visit, including “follow-up visit exception” with specimen collected somewhere else (see section “Follow-up Visits Exception”) the study staff will attempt to obtain at least vital status. A subject will be considered lost to follow-up and terminated from the study once they have missed two consecutive follow-up visits without contact and three documented attempts have been made by the research team to contact the subject. One of the three documented attempts must include a certified letter. In the event that the subject is lost to follow-up, the date of discontinuation of the study will be recorded as the last contact with the subject by the investigative site.

5.9 Subject Study Completion

A subject has completed the study when they have completed the 6-month follow-up visit. Any subject that does not complete this visit due to voluntary withdrawal, death, or any other reason will be considered a withdrawal.

6 Study Procedures

6.1 Schedule of events

6.1.1 Visit 1

This visit will be the initial consult before scheduling the surgery. In this visit:

1. Study will be explained to the patient
2. Informed Consent will be obtained

3. Inclusion/Exclusion Criteria questions will be applied
4. Blood will be collected to confirm patient eligibility and baseline labs.
5. Concurrent medications reviewed
6. Physical evaluation will be performed
7. Urine sample will be collected in order to have laboratories baseline values for metabolomics analysis

➔ In case the subject presents any reason to not be included in the study, he/she will be informed and no visits will be required for the study.
➔ In case the laboratory blood test value present some significant clinical abnormality listed in the exclusion criteria list, the patient will be considered screening fail and will be excluded from the study. In this case, the blood and urine samples collected for baseline values will be automatically discarded.

6.1.2 Visit 2

Prior to surgery, study medication or placebo will be mailed/shipped by Outpatient Research Pharmacy or delivered in person to patient by study team. In this visit:

1. Patient will be randomized in one of the two groups (Basis™ or placebo)
2. Basis™ or placebo compound will be provided to the patient
3. Medication diary will be provided to the patient

6.1.3 Visit 3 (see telephone script)

Prior to patients beginning study medication, study team will contact patient to review study medication instructions and diary completion. Also, study team will remind patient to bring the medication/placebo bottle and medication diary to the hospital at the surgery day. If the patient is on campus, an in-person visit is acceptable as well to complete this time point if the patient prefers that option.

6.1.4 Visit 4

This visit will be at the admission day for the surgical procedure. In this visit:

1. The patient will be admitted to the hospital
2. The medication diary will be reviewed and exchanged for a new one
3. Concomitant medication will be reviewed
4. Adverse event review
5. Blood sample (approximately 40 mL) will be collected
6. Urine sample will be collected
7. Retrieve medication/placebo bottle
8. Provide new bottle with medication/placebo sufficient for 6 weeks

- Basis™/placebo intake may be interrupted based on NPO status.

➔ If NPO duration is more than 5 days, patient will be withdrawn from the study.

6.1.5 Visit 5

This visit will be at post-procedure period within 24-48 hours after surgery (inpatient). In this visit:

1. Physical evaluation

2. Adverse event review
3. Blood sample (approximately 40 mL) will be collected
4. Urine sample will be collected

➔ As long as the patient stays in the hospital, blood sample will be daily collected as well as the adverse event review, until the discharge day. It may vary depending on the type of procedure and the length of hospital stay.

6.1.6 Visit 6

This visit is the pre-discharge visit. In this visit:

1. Physical evaluation
2. Adverse event review
3. Blood sample (approximately 40 mL) will be collected
4. Urine sample will be collected

➔ In the event that a patient is in the hospital 24 - 48 hours then discharged, Visit 5 and Visit 6 may be combined into a single visit.

6.1.7 Visit 7 (see telephone script)

Approximately three weeks post-op, study team will contact patient to review study medication and diary completion. Also, the study team will check on any adverse events. If the patient is on campus, an in-person visit is acceptable as well to complete this time point if the patient prefers that option.

6.1.8 Visit 8

This visit follow-up will ideally happen:

- Within 42 to 56 days after surgery (6-8 weeks)
- In this visit:
  1. Physical evaluation
  2. Blood sample (approximately 40 mL) will be collected
  3. Urine sample will be collected
  4. Concomitant medication reviewed
  5. Adverse event review
  6. Return medication diary
  7. Return medication/placebo bottle

- At this moment, patient will be finished the period of treatment.

6.1.9 Visit 9

This visit follow-up will happen 6 months after the surgical procedure. In this visit:

1. Physical evaluation
2. Blood sample (approximately 40 mL) will be collected
3. Urine sample will be collected
4. Concomitant medication reviewed
5. Adverse event review

➔ In case patients are not able to come for follow-up visits #8 and #9, they will be offered the possibility to obtain their serum creatinine and urea levels from their local provider. No research lab blood tests will be performed (see section "follow-up visits exception").

In case patients are not able to come for follow-up visits, within 42 to 56 days / 6-8 weeks (Visit 8) and at 6 months (Visit 9) the study team will call the patient to review any Adverse Event.

Table 6. Subject Schedule of Events

Schedule of Events										
Study Activity	Visit 1 (screening and baseline)	Visit 2 (randomization)	Visit 3 (telephone call #1)	Visit 4 (pre-op at surgery day)	Visit 5 (24-48h post-op)	Daily as long as patient is hospitalized	Visit 6 (discharge day)	Visit 7 (telephone call #2)	Visit 8* (6-8 weeks follow-up)	Visit 9* (6 months follow-up)
Study Agent(s)		X		X						
Pre-screening questions	X									
Informed consent	X									
History	X									
Reminder calls (take capsules and bring bottle)			X					X		
Concurrent meds	X			X					X	X
Vital signs	X				X		X		X	X
Height and weight	X									
Physical evaluation [a]	X				X	X	X		X	X
Serum Creatinine, urea, and cystatin C	X					X				
CBC w/diff, plts	X			X	X		X		X	X
Serum chemistry [b]	X			X	X		X		X	X
Prothrombin Time/INR	X			X**	X**	X**	X**		X**	X**
NAD, NMN, NR levels	X			X	X		X		X	X
Real-time PCR [c]	X			X	X		X		X	X
ELISA[d]	X			X	X	X	X		X	X
Metabolite measurement [e]	X			X	X		X		X	X
Renal biomarkers [f]	X			X	X		X		X	X
Adverse event evaluation				X	X	X	X	X	X	X
Check Medication Diary				X					X	

Return med/placebo bottle				X					X	
SF-36 survey	X								X	X

a: Heart, lungs, and abdomen evaluation
b: Albumin, alkaline phosphatase, total bilirubin, BUN, creatinine, calcium, chloride, glucose, LDH, phosphorus, potassium, total protein, SGOT [AST], SGPT [ALT], sodium, C-reactive protein, and cystatin C
c: in PBMCs: TNFα, IL-6, and PAI-1
d: in plasma: and N-gal
e: in urine: Quinolinate and tryptophan
f: in urine: TIMP-2, KIM-1, IGFBP7
* These tests may not apply for patients who are not able to come for follow-up visits. In this case, only blood for serum creatinine and blood urea nitrogen will be collected. No research lab blood tests will be performed. The study team will call the patient to review any Adverse Event.
**Tests will be ordered only in case the patient is currently taking anticoagulants.

6.2 Follow-up visits exception

Only for those patients who agree to enroll in the study but are not able to come back to Mayo Clinic for follow-up visits 8 and 9, for any reason, the follow-up visits exception will apply. They will commit to collect blood samples in their local clinical laboratory, so the tests results can be sent to the study team.

Orders for SCr and BUN will be sent to the local clinical laboratory determined by the patient. The tests will be collected at 6-8 weeks and at 6 months after surgery.

For those specific patients, research lab blood tests will not be performed.

- ➔ Patients who are not coming for follow-up visits will mail the drug/placebo bottle (empty or containing any remaining capsules) and their study medication diary after the 6-week period of treatment after surgery.
- ➔ For these patients only, the study team will contact patient by telephone call by end of 6-week period of treatment (Visit 8) to review study medication compliance, medication diary completion, and review any new or ongoing adverse event(s).
- ➔ For these patients only, the study team will contact patient by telephone call by 6-month follow-up (Visit 9) to review any adverse event(s).

6.3 Assessments

6.3.1 Blood collection

It will be collected approximately 40 mL of blood for the analysis of:

1. CBC, hematocrit, C-reactive protein, and serum glucose
2. Renal function tests: BUN, creatinine, and cystatin C
3. Liver function tests: albumin, total protein, alkaline phosphatase, SGOT [AST], SGPT [ALT], total bilirubin, and LDH
4. Electrolytes: calcium, chloride, sodium, phosphorus, and potassium
5. NAD levels in peripheral blood mononuclear cells (PBMCs)
6. NMN, NR levels in plasma
7. Real-time Polymerase Chain Reaction (PCR) in PBMCs:
    - TNFα

- IL-6
- PAI-1

8. ELISA in plasma:
    - N-gal
9. DNA methylation study

6.3.2 Urine Collection

It will be collected urine for the quantification of:

10. Quinolinate
11. Tryptophan
12. Renal biomarkers: TIMP-2, KIM-1, IGFBP7

➔ The samples will be sent by Clinical Research Trial Unit (CRTU) to different locations for the analysis as follows:

- Samples for analysis of items 1, 2, 3, and 4 will be sent to the clinical laboratory
- Samples for analysis of items 5, 6, 7, 8, 9, 10, 11, and 12 will be picked, processed and stored by the study team at Research laboratory.
- One mL of whole blood will be sent to Elysium for analyses of item 9.
- Specimens processed and stored for analysis of items 5, 6, 7, 8, 10, 11, and 12 will be sent in batches to [redacted] research laboratory located at Mayo Clinic Jacksonville campus in Florida.

7 Statistical Plan

7.1 Sample Size Determination

Based on preliminary data, we anticipate that the incidence of the primary endpoint (AKI) is approximately 30% in our target patient population. We hypothesize that this incidence will be reduced by >50% (i.e. AKI <15%) in patients receiving Nicotinamide Riboside and Pterostilbene. Based on these assumptions we have determined that an effective sample-size of N=208 (N=104 per group) will provide statistical power (two-tailed, alpha=0.05) to detect a difference between groups. In order to accommodate attrition of approximately 15%, a total sample-size of N=238 (N=119 per group) is proposed.

7.2 Statistical Methods

7.2.1 General Analysis Considerations

Unless otherwise specified, all randomized subjects will be included in the analysis based on their randomized treatment assignment (intention to treat). No covariate adjustment will be included in the primary analysis. We will use descriptive statistics to summarize baseline demographic and clinical risk factor characteristics. These summaries will be generated for the entire sample as well as according to treatment group. Given the randomized study design, we will not perform formal hypothesis testing of baseline characteristics to assess for significant baseline treatment differences. However, the investigators will review the baseline

characteristics prior to performing analyses for a given outcome and identify variables that should be included as covariates in secondary analyses. In addition to the primary analyses which will include all randomized subjects, secondary analyses will be performed which include only subjects who are found to be ≥ 50% compliant with medication at the admission appointment for surgery. Model assumptions for each analysis will be validated and transformations or nonparametric methods will be utilized as appropriate. In all cases, two-sided tests will be performed with $p < 0.05$ used to denote statistical significance.

7.2.2 Handling of Missing Data

The primary endpoints are collected immediately following surgery while patients are still hospitalized. For this reason, missing data are unlikely. However, for secondary endpoints collected at 6-week and 6-month following surgery it is anticipated that some patients will have missing data. In all cases, the primary analyses will be restricted to those patients with data available (complete case analyses) with supplemental analyses performed using multiple-imputation methods to account for missing data.

7.2.3 Primary Aim/Hypothesis

To assess the efficacy of Nicotinamide Riboside and Pterostilbene in preventing acute kidney injury (AKI) in patients undergoing complex aortic aneurysm repair and open aortic arch reconstruction.

The incidence of AKI, as defined in Primary Study Endpoints Section, will be summarized for each treatment group using a point estimate and corresponding 95% confidence interval. The incidence of AKI will be compared between groups using logistic regression with results summarized using the odds ratio and corresponding 95% confidence interval. To supplement the primary analysis, secondary analyses will be performed for levels of urea, CrCl, cystatin C and n-gal which are obtained immediately following surgery. These secondary endpoints will be analyzed as continuous variables using analysis of covariance with the post-surgical value of the given variable included as the dependent variable, treatment group as the independent variable, and the baseline value of the given endpoint will be included as the covariate. In addition, repeated measures analysis of covariance will be performed using measurements obtained immediately following surgery, at 6-week, and 6-monthafter surgery. In all cases, the estimated treatment effect will be summarized using a point-estimate and corresponding 95% confidence interval and two-tailed p-values < 0.05 will be considered statistically significant.

7.2.4 Secondary Aim/Hypothesis

To assess the efficacy of Nicotinamide Riboside and Pterostilbene in preventing perioperative MI and central nervous system complications (stroke and spinal cord injury) in patients undergoing complex aortic aneurysm repair and open aortic arch reconstruction.

For the primary analysis of this aim a composite binary endpoint will be created to indicate the occurrence either perioperative MI or central nervous system complication. This binary endpoint will be compared between groups using the chi-square test (or Fisher's exact test). In addition, the incidence of each specific complication will be compared between groups using Fisher's exact test.

7.2.5 Additional analyses

The percentage of subjects who discontinue study participation following randomization will be summarized and compared between groups using Fisher's exact test. Medication compliance will be assessed using a medication diary. Medication compliance will be summarized for each treatment group and the percentage of subjects who report taking less than 90% of the prescribed medication will be compared between groups using Fisher's exact test. The frequency of adverse events (See in "Safety and Adverse Events" Section) will be summarized and compared between groups using Fisher's exact test.

8 Safety and Adverse Events

To date, there are no known adverse effects associated with either compound, NR has been granted generally recognized as safe GRAS status from the FDA (GRAS Notice No 635). Animal and human studies indicate that oral administration of Basis™ is well tolerated. Despite no serious adverse effects were reported, we will be monitoring liver function as well as periodic consultations to ensure the product safety.

Below are the definitions of what will be considered as adverse events and how that information will be collected and reported.

8.1 Definitions

Unanticipated Problems Involving Risk to Subjects or Others (UPIRTSO)
Any unanticipated problem or adverse event that meets the following three criteria:

- Serious: Serious problems or events that results in significant harm, (which may be physical, psychological, financial, social, economic, or legal) or increased risk for the subject or others (including individuals who are not research subjects). These include: (1) death; (2) life threatening adverse experience; (3) hospitalization - inpatient, new, or prolonged; (4) disability/incapacity - persistent or significant; (5) birth defect/anomaly; (6) breach of confidentiality and (7) other problems, events, or new information (i.e. publications, DSMB reports, interim findings, product labeling change) that in the opinion of the local investigator may adversely affect the rights, safety, or welfare of the subjects or others, or substantially compromise the research data, **AND**
- Unanticipated: (i.e. unexpected) problems or events are those that are not already described as potential risks in the protocol, consent document, not listed in the Investigator's Brochure, or not part of an underlying disease. A problem or event is "unanticipated" when it was unforeseeable at the time of its occurrence. A problem or event is "unanticipated" when it occurs at an increased frequency or at an increased severity than expected, AND
- Related: A problem or event is "related" if it is possibly related to the research procedures.

Adverse Event

An untoward or undesirable experience associated with the use of a medical product (i.e. drug, device, biologic) in a patient or research subject.

Serious Adverse Event

Adverse events are classified as serious or non-serious. Serious problems/events can be well defined and include;

- death
- life threatening adverse experience
- hospitalization
- inpatient, new, or prolonged; disability/incapacity
- persistent or significant disability or incapacity
- birth defect/anomaly

and/or per protocol may be problems/events that in the opinion of the sponsor-investigator may have adversely affected the rights, safety, or welfare of the subjects or others, or substantially compromised the research data.
NR is potentially safe, well- tolerated and readily available.

All adverse events that do not meet any of the criteria for serious, should be regarded as **non-serious adverse events**.

Adverse Event Reporting Period

For this study, the treatment follow-up period will be defined as 30 days following the last administration of drug/placebo. The study period during which adverse events must be reported will be defined as the period from the initiation of any study procedures to the end of the study treatment follow-up.

Preexisting Condition

A preexisting condition is one that is present at the start of the study. A preexisting condition should be recorded as an adverse event if the frequency, intensity, or the character of the condition worsens during the study period.

General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a preexisting condition. At the end of the study, any new clinically significant findings/abnormalities that meet the definition of an adverse event must also be recorded and documented as an adverse event.

Post-study Adverse Event

All unresolved adverse events should be followed by the sponsor-investigator until the events are resolved, the subject is lost to follow-up, or the adverse event is otherwise explained. At the last scheduled visit, the sponsor-investigator should instruct each subject to report, to the sponsor-investigator, any subsequent event(s) that the subject, or the subject's personal physician, believes might reasonably be related to participation in this study.

Abnormal Laboratory Values

A clinical laboratory abnormality should be documented as an adverse event if they are considered clinically significant. Abnormal laboratory values induced by perioperative interventions and compatible and guided by specific clinical interventions are expected and not

considered an adverse event. Examples are increase in ACT after heparin treatment, increase in C-reactive protein after major vascular surgical intervention, and others.

Hospitalization, Prolonged Hospitalization or Surgery

Any adverse event that results in hospitalization or prolonged hospitalization should be documented and reported as a serious adverse event unless specifically instructed otherwise in this protocol.

Neither the condition, hospitalization, prolonged hospitalization, nor surgery are reported as an adverse event in the following circumstances:

- Hospitalization or prolonged hospitalization for diagnostic or elective surgical procedures for a preexisting condition. Surgery should not be reported as an outcome of an adverse event if the purpose of the surgery was elective or diagnostic and the outcome was uneventful.
- Hospitalization or prolonged hospitalization required to allow efficacy measurement for the study.
- Hospitalization or prolonged hospitalization for therapy of the target disease of the study, unless it is a worsening or increase in frequency of hospital admissions as judged by the clinical investigator.

8.2 Recording of Adverse Events

Information on all adverse events will be recorded immediately in the source document, and also in the appropriate adverse event section of the case report form (CRF). All clearly related signs, symptoms, and abnormal diagnostic, laboratory or procedure results will be recorded in the source document.

All adverse events occurring during the study period will be recorded. The clinical course of each event will be followed until resolution, stabilization, or until it has been ultimately determined that the study treatment or participation is not the probable cause. Serious adverse events that are still ongoing at the end of the study period will be followed up, to determine the final outcome. Any serious adverse event that occurs during the Adverse Event Reporting Period and is considered to be at least possibly related to the study treatment or study participation will be recorded and reported immediately.

8.3 Reporting of Serious Adverse Events and Unanticipated Problems

The sponsor-investigator is responsible to evaluate all adverse events to determine reporting requirements to the IRB and FDA.

When an adverse event has been identified, the study team will take appropriate action necessary to protect the study participant and then will complete the Study Adverse Event Worksheet and log. The sponsor-investigator will evaluate the event and determine the necessary follow-up and reporting required.

8.3.1 Sponsor-Investigator reporting: notifying the Mayo IRB

The following information will be collected on the adverse event worksheet (and entered in the research database):

•Disease/histology (if applicable):

•The date the adverse event occurred:

•Description of the adverse event:
•Relationship of the adverse event to the research (drug, procedure, or intervention*):
•If the adverse event was expected:
•The severity of the adverse event: (use a table to define severity scale 1-5**)
•If any intervention was necessary:
•Resolution: (was the incident resolved spontaneously, or after discontinuing treatment)
•Date of Resolution:
Quick Reference
The NCI (National Cancer Institute) Common Terminology Criteria for Adverse Events is a descriptive terminology which can be utilized for AE reporting. A grading (severity) scale is provided for each AE term.

Components and Organization
SOC
System Organ Class, the highest level of the MedDRA hierarchy, is identified by anatomical or physiological system, etiology, or purpose (e.g., SOC Investigations for laboratory test results). CTCAE terms are grouped by MedDRA Primary SOCs. Within each SOC, AEs are listed and accompanied by descriptions of severity (Grade).
CTCAE Terms
An Adverse Event (AE) is any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medical treatment or procedure that may or may not be considered related to the medical treatment or procedure. An AE is a term that is a unique representation of a specific event used for medical documentation and scientific analyses. Each CTCAE v5.0 term is a MedDRA LLT (Lowest Level Term).

Definitions
A brief definition is provided to clarify the meaning of each AE term.

Grades
Grade refers to the severity of the AE. The CTCAE displays Grades 1 through 5 with unique clinical descriptions of severity for each AE based on this general guideline:
Grade 1 Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
Grade 2 Moderate; minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental ADL*.
Grade 3 Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL**.
Grade 4 Life-threatening consequences; urgent intervention indicated.
Grade 5 Death related to AE.
A Semi-colon indicates ‘or’ within the description of the grade.

Activities of Daily Living (ADL)
*Instrumental ADL refer to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.

**Self-care ADL refer to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden.

The sponsor-investigator will report to the Mayo IRB any UPIRTSOs and Non-UPIRTSOs according to the Mayo IRB Policy and Procedures.

Serious adverse event (SAE) which the Principal Investigator has determined to be a UPIRTSO will be reported according to the Mayo IRB Policy and Procedures.

*** Relationship Index Example**
The relationship of an AE to the Investigational Drug is a clinical decision by the sponsor-investigator (PI) based on all available information and is graded as follows:

1. Not related: a reaction for which sufficient information exists to indicate that the etiology is unrelated to the study drug; the subject did not receive the study medication or the temporal sequence of the AE onset relative to administration of the study medication is not reasonable or the event is c1early related to other factors such as the subject's clinical state, therapeutic intervention or concomitant therapy.

2. Unlikely: a clinical event, including laboratory test abnormality, with a temporal relationship to drug administration which makes a causal relationship improbable and in which other drugs, chemicals, or underlying disease provide plausible explanations.

3. Possible: a clinical event, including laboratory test abnormality, with a reasonable time sequence to administration of the drug but which could also be explained by concurrent disease or other drugs or chemicals; information on drug withdrawals may be lacking are unclear.

4. Probable: a clinical event including laboratory test abnormality, with a reasonable time sequence to administration of the drug, unlikely to be attributed to concurrent disease or other drugs or chemicals and which follows a clinically reasonable response on withdrawal (de-challenge): re-challenge information is not required to fulfi1 this definition.

5. Definite: a reaction that follows a reasonable temporal sequence from administration of the drug, or in which the drug level has been established in body fluids or tissues, that follows a known or expected response pattern to the suspected drug, and that is confirmed by improvement on stopping or reducing the dosage of the drug, and reappearance of the reaction on repeated exposure (re-challenge).

8.3.2 Sponsor-Investigator reporting: Notifying the FDA

The sponsor-investigator will report to the FDA all unexpected, serious suspected adverse reactions according to the required IND Safety Reporting timelines, formats and requirements.

Unexpected fatal or life threatening suspected adverse reactions where there is evidence to suggest a causal relationship between the study drug/placebo and the adverse event, will be reported as a serious suspected adverse reaction. This will be reported to the FDA according to the FDA Policy and Procedures.

Other unexpected serious suspected adverse reactions where there is evidence to suggest a causal relationship between the study drug/placebo and the adverse event, will be reported as a serious suspected adverse reaction. This will be reported to the FDA according to the FDA Policy and Procedures.
Any clinically important increase in the rate of serious suspected adverse reactions over those listed in the protocol or product insert will be reported as a serious suspected adverse reaction. This will be reported to the FDA according to the FDA Policy and Procedures.
Findings from other studies in human or animals that suggest a significant risk in humans exposed to the drug will be reported. This will be reported to the FDA according to the FDA Policy and Procedures.

8.4 Unmasking/Unblinding Procedures

This protocol will employ a blinded design so that the participant, study personnel, and investigators will not know subject group assignment status. The unblinding would be done by contacting the research pharmacy. While the safety of the subject always comes first, it is still important to seriously consider if unmasking/unblinding the study therapy is necessary to ensure a subject's safety. The unmasking/unblinding will happen only when there is a serious adverse event; in this case this event will be logged in the specific CRF for SAE that is part of this protocol. However, in cases where unmasking/unblinding was not associated with an SAE, such actions will be reported in a same timeline requirements for reporting of SAEs.

8.5 Medical Monitoring

It is the responsibility of the Principal Investigator to oversee the safety of the study at his/her site. This safety monitoring will include careful assessment and appropriate reporting of adverse events as noted above, as well as the construction and implementation of a site data and safety-monitoring plan (see section "Study Monitoring, Auditing, and Inspecting"). Medical monitoring will include a regular assessment of the number and type of serious adverse events.

8.5.1 Use with caution medications

Given limited resources on Pterostilbene in clinical research and the fact that it is an inhibitor of P-glycoprotein, some subjects will be excluded from the study (see Exclusion Criteria) and some others will be monitored very closely depending on the medication they are already taking.

The medications and the actions taken are as follows:

- Anticoagulants: PT/INR will be closely monitored and it will be checked in every visit follow-up;
- Antiplatelets: Patients will be closely monitored being specifically asked about bleeding episodes in every Adverse Event Evaluation;
- Statins: Patients will be closely monitored for reported myopathies;
- Antiarrythimics / Antihypertensive: Patients will be closely monitored and blood pressure and heart rate will be checked in every visit follow-up.

8.6 Early Stopping Rule

The Independent Medical Monitor for this study is not associated with this research project and thus works independently of the PI. The Independent Medical Monitor is qualified to review

the patient safety generated by this study because of his unique expertise. When an adverse event has been identified, the study team will take appropriate action necessary to protect the study participant and then complete the Study Adverse Event Worksheet and log. The Independent Monitor will evaluate the events considered as serious or related to the study medication and determine the necessary follow-up and reporting required.

The study will be stopped if any patient is permanently disabled, seriously injured, or dies in a manner that could be consistent with the drug usage (see relationship index grade). The study will continue if the following criteria are determined by the Independent Medical Monitor: (1) the patient(s) were taking the placebo, or (2) the event is unrelated to the subject's participation in the investigational drug trial (e.g., caused by surgical complications).

8.6.1 Internal Data and Safety Monitoring Board

The internal data and safety monitoring is paramount to our study and to maintain the interest of patients as a top priority. Our study is monitored and evaluated by the Department of surgery (DoS), division of surgery research Data and Safety Monitoring Board (DSMB). The functions, goals and structure of the Mayo Clinic Department of surgery DSMB are described on the attached document (DSMB charter).

In brief, the DoS DSMB is structured to serve clinical intervention trials to provide data safety monitoring oversight. The function of a DSMB is to review data and endpoints on a timeline set forth by the Data Safety Monitoring Plan (DSMP) in the IRB approved protocol. Initial evaluation by the DSMB has been performed to our protocol. As described in our protocol the DSMB evaluated and approved based on the following criteria. The DSMB meet prior to enrollment of subjects to our study and evaluated the IRB protocol, informed consent and the Data Safety Monitoring Plan (DSMP) to determine:

- The study's risks and benefits and safety of research subjects
- Suggest improving the study design
- Identify what data will be required for review
- Identify early stopping rules.

All of these are described in the appropriate sections of our IRB document.

The DoS DSMB evaluates studies in a by-annual basis (every 6 months).

The Bi-Annual review includes:

- Interim data review to detect evidence efficacy or adverse events and determine if the study should continue, be modified, or be stopped.
- Progress evaluation to assess recruitment and retention of subjects, protocol adherence, data quality/completeness, and accrual.
- Multi-arm studies will require both clinical and statistical review

The meeting outcomes provide recommendation to the continuations, provisional continuation, suspension or closure.

The occurrence of events that prompt the activation of the protocol specific early stopping rules for pilot, phase II, and phase III studies require prompt review by the study team. The study team formulates an appropriate plan of action to ensure patient safety (as required by the IRB). This may include protocol changes and may also require immediate suspension of accrual, as deemed necessary to ensure patient safety.

The study team then forwards the action plan to the DSMB chair for review. A copy of the action plan is also provided to the DoS DMSB for review at the time it is submitted to the IRB. The DSMB provides a prompt review of the action plan and a summary of that review to the

IRB and to the study team. The review by the DSMB serves as an advisory statement to the IRB and either expresses approval of the action plan or provides recommendations for modifications to the plan.

9 Data Handling and Record Keeping

9.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (long term survival status that the subject is alive) at the end of their scheduled study period.

9.2 Source Documents

Source data is all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents. Examples of these original documents, and data records include: hospital records, clinical and office charts, laboratory notes, memoranda, subjects' diaries or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, microfiches, photographic negatives, microfilm or magnetic media, x-rays, subject files, and records kept at the pharmacy, at the laboratories, and at medico-technical departments involved in the clinical trial.

9.3 Case Report Forms

The study case report form (CRF) is the primary data collection instrument for the study. All data requested on the CRF must be recorded. All missing data must be explained. If a space on the CRF is left blank because the procedure was not done or the question was not asked, write "N/D". If the item is not applicable to the individual case, write "N/A". All entries should be printed legibly in ink. If any entry error has been made, to correct such an error, draw a single straight line through the incorrect entry and enter the correct data above it. All such changes must be initialed and dated. Do not erase or use "white-out" for errors. For clarification of illegible or uncertain entries, print the clarification above the item, then initial and date it. If the reason for the correction is not clear or needs additional explanation, neatly include the details to justify the correction.

9.3.1 Data Management

The Clinical Data Management System (CDMS) for this study will be stored in the format of eBinder.

9.3.2 Data Collection Methods – Quality, Security and Confidentiality

This study will report clinical data using the eBinder storage. The CDMS will be the database of record for the protocol and subject to regulatory inspections. All users will be trained to use the CDMS and will comply with study specific guidelines/instructions as well as applicable regulatory requirements.

9.3.3 Data Clarification Process

Once entered, data will be evaluated to ensure that it is complete, consistent and logically sound. If Changes to the data in the CDMS are required, all changes, reasons for changes, and persons making the changes will be captured in the CDMS's audit trail.

9.4 Records Retention

The study team will maintain complete, accurate and current study records as required by applicable regulatory requirements. Records will be maintained during the clinical study and for (1) a minimum of two years after the latter of the date on which the study is terminated or completed, or the date the records are no longer required to support regulatory approval of the drug (2) as outlined in the Mayo Clinic Research Policy Manual –"Retention of and Access to Research Data Policy", whichever is longer. In any event, clinical study records will not be disposed of, nor custody of the records transferred, without prior Investigator approval.

The study team shall not enroll any subjects prior to obtaining approval for the study from a properly constituted independent IRB. The study team will submit the protocol, informed consent forms; other information to be provided to subjects such as survey instruments or questionnaires, and any proposed advertising/recruitment materials, to the IRB for written approval.

All subject records will be kept confidential to the extent provided by applicable laws and regulations. The study monitors and other authorized representative of the investigator may inspect all documents and records required to be maintained by the investigator, including but not limited to medical records. Such records may also be reviewed by the Site's IRB. The investigator will inform the subjects that their records will be reviewed.

9.5 Specimen Samples Retention

Researchers at Mayo Clinic who aren't involved with this study may ask to use subject's data and/or samples for future research. Researchers at other institutions may also ask for data and/or samples for future studies. Samples collected for this study will be used or shared for future research, only in case the subject consents. Although, none of identifiable information such as your name, Mayo Clinic number, date of birth, address, phone number, social security number will be shared. The subject can still take part in the study even if he/she does not want information or samples used for future research.

Data and/or samples will only be shared consistent with consent from signed by the subject, and with all applicable laws and regulations.

Subject may withdraw consent for future use of data and/or samples at any time, by writing to the Principal Investigator at the address provided in the "Contact Information" section of the

consent form. Data and/or samples would be removed from any repository where they are stored, if possible. Data and/or samples already distributed for research use will not be retrieved.

In case of deceased subject samples will be considered a gift to Mayo Clinic. That means that Mayo Clinic can use it for research forever.

10 Study Monitoring, Auditing, and Inspecting

10.1 Study Monitoring Plan

The investigator will allocate adequate time for such monitoring activities. The Investigator will also ensure that the monitor or other compliance or quality assurance reviewer is given access to all the study-related documents and study related facilities (e.g. pharmacy, diagnostic laboratory, etc.), and has adequate space to conduct the monitoring visit.

This monitoring is conducted to assure data is secured. Mayo Clinic is committed to protecting the safety and confidentiality of information obtained about the subject in connection with this research study. Study records that identify the subject will be kept confidential. Data will be stored in locked office, or in password protected computers. Only research staff involved in the study will have access to the data.

During this research, information about the subject health will be collected under Federal law called the Privacy Rule, health information is private. However, there are exceptions to this rule, and the subjects should know who may be able to see, use and share their health information for research and why they may need to do so. Information about study subjects and their health will not be used in this research study without their written permission. When the subject signs the consent form, it will provide that permission.

As a service to the sponsor-investigator, this study may be monitored during the conduct of the trial by staff from the Mayo Clinic Office of Research Regulatory Support. Clinical trial monitoring may include review of the study documents and data generated throughout the duration of the study to help ensure the validity and integrity of the data along with the protection of human research subjects.

10.2 Auditing and Inspecting

The investigator will permit study-related monitoring, audits, and inspections by the IRB, the sponsor, and government regulatory agencies, of all study related documents (e.g. source documents, regulatory documents, data collection instruments, study data etc.). The investigator will ensure the capability for inspections of applicable study-related facilities (e.g. pharmacy, diagnostic laboratory, etc.).

Participation as an investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable compliance offices.

11 Ethical Considerations

This study is to be conducted according to United States government regulations and Institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted local Institutional Review Board (IRB), in agreement with local legal prescriptions, for formal

approval of the study. The decision of the IRB concerning the conduct of the study will be made in writing to the sponsor-investigator before commencement of this study.

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. This consent form will be submitted with the protocol for review and approval by the IRB for the study. The formal consent of a subject, using the Approved IRB consent form, must be obtained before that subject undergoes any study procedure. The consent form must be signed by the subject or the subject's legally authorized representative, and the individual obtaining the informed consent.

12 Study Finances

12.1 Funding Source

This study will be financed through Mayo funding.

12.2 Conflict of Interest

Dr. Chini has a license agreement with Elysium for CD38 inhibitors that is unrelated to this current protocol.

13 Publication Plan

This present clinical trial is intended for publication, even if terminated prematurely. The publication may include any or all of the following: posting of a synopsis online, abstract and/or presentation at a scientific conference, or publication of a full manuscript.

14 References

1 de Souza, L. R. *et al.* Editor's Choice - Comparison of Renal Outcomes in Patients Treated by Zenith((R)) Fenestrated and Zenith((R)) Abdominal Aortic Aneurysm Stent grafts in US Prospective Pivotal Trials. *Eur J Vasc Endovasc Surg* **53**, 648-655, doi:10.1016/j.ejvs.2017.02.005 (2017).

2 Huen, S. C. & Parikh, C. R. Predicting acute kidney injury after cardiac surgery: a systematic review. *Ann Thorac Surg* **93**, 337-347, doi:10.1016/j.athoracsur.2011.09.010 (2012).

3 Billings, F. T. t. *et al.* Obesity and oxidative stress predict AKI after cardiac surgery. *Journal of the American Society of Nephrology : JASN* **23**, 1221-1228, doi:10.1681/asn.2011090940 (2012).

4 Brezis, M. & Rosen, S. Hypoxia of the renal medulla--its implications for disease. *The New England journal of medicine* **332**, 647-655, doi:10.1056/nejm199503093321006 (1995).

5 Ricksten, S. E., Bragadottir, G. & Redfors, B. Renal oxygenation in clinical acute kidney injury. *Critical care (London, England)* **17**, 221, doi:10.1186/cc12530 (2013).

6 Granata, A. *et al.* [Atheroembolism renal disease: diagnosis and etiologic factors]. *Clin Ter* **163**, 313-322 (2012).

7 Fleming, G. A. *et al.* Angiotensin-converting enzyme inhibition alters the inflammatory and fibrinolytic response to cardiopulmonary bypass in children. *Pediatric critical care medicine : a journal of the Society of Critical Care Medicine and the World Federation of Pediatric Intensive and Critical Care Societies* **12**, 532-538, doi:10.1097/PCC.0b013e3181fe3925 (2011).

8 Fujii, T. *et al.* The role of renal sympathetic nervous system in the pathogenesis of ischemic acute renal failure. *Eur J Pharmacol* **481**, 241-248 (2003).

9 Sreedharan RDP, V. W. S. in *PediatricNephrology* 1579–1602 (Heidleberg: Springer-Verlag, 2009).
10 Zhang, W. R. *et al.* Plasma IL-6 and IL-10 Concentrations Predict AKI and Long-Term Mortality in Adults after Cardiac Surgery. *Journal of the American Society of Nephrology : JASN* **26**, 3123-3132, doi:10.1681/asn.2014080764 (2015).
11 CLEaRW, S. in *iseasesof the kidney and urinary tract.* 930–961 (Lippincott Williams &Wilkins, 2007).
12 Hui, W. F., Chan, W. K. & Miu, T. Y. Acute kidney injury in the paediatric intensive care unit: identification by modified RIFLE criteria. *Hong Kong medical journal = Xianggang yi xue za zhi* **19**, 13-19 (2013).
13 Ratanarat, R., Skulratanasak, P., Tangkawattanakul, N. & Hantaweepant, C. Clinical accuracy of RIFLE and Acute Kidney Injury Network (AKIN) criteria for predicting hospital mortality in critically ill patients with multi-organ dysfunction syndrome. *Journal of the Medical Association of Thailand = Chotmaihet thangphaet* **96 Suppl 2**, S224-231 (2013).
14 Ricci, Z. & Ronco, C. Neonatal RIFLE. *Nephrology, dialysis, transplantation : official publication of the European Dialysis and Transplant Association - European Renal Association* **28**, 2211-2214, doi:10.1093/ndt/gft074 (2013).
15 Roy, A. K. *et al.* A Comparison of Traditional and Novel Definitions (RIFLE, AKIN, and KDIGO) of Acute Kidney Injury for the Prediction of Outcomes in Acute Decompensated Heart Failure. *Cardiorenal medicine* **3**, 26-37, doi:10.1159/000347037 (2013).
16 Barasch, J., Zager, R. & Bonventre, J. V. Acute kidney injury: a problem of definition. *Lancet (London, England)* **389**, 779-781, doi:10.1016/s0140-6736(17)30543-3 (2017).
17 Bellomo, R., Ronco, C., Kellum, J. A., Mehta, R. L. & Palevsky, P. Acute renal failure - definition, outcome measures, animal models, fluid therapy and information technology needs: the Second International Consensus Conference of the Acute Dialysis Quality Initiative (ADQI) Group. *Critical care (London, England)* **8**, R204-212, doi:10.1186/cc2872 (2004).
18 Hocine, A. *et al.* Predictive value of the RIFLE urine output criteria on contrast-induced nephropathy in critically ill patients. *BMC nephrology* **17**, 36, doi:10.1186/s12882-016-0243-5 (2016).
19 Mehta, R. L. *et al.* Acute Kidney Injury Network: report of an initiative to improve outcomes in acute kidney injury. *Critical care (London, England)* **11**, R31, doi:10.1186/cc5713 (2007).
20 Fujii, T., Uchino, S., Takinami, M. & Bellomo, R. Validation of the Kidney Disease Improving Global Outcomes criteria for AKI and comparison of three criteria in hospitalized patients. *Clin J Am Soc Nephrol* **9**, 848-854, doi:10.2215/cjn.09530913 (2014).
21 KDIGO Board Members. *Kidney international supplements* **2**, 3, doi:10.1038/kisup.2012.3 (2012).
22 Chaikof, E. L. *et al.* Reporting standards for endovascular aortic aneurysm repair. *J Vasc Surg* **35**, 1048-1060 (2002).
23 Coselli, J. S., Bozinovski, J. & LeMaire, S. A. Open surgical repair of 2286 thoracoabdominal aortic aneurysms. *Ann Thorac Surg* **83**, S862-864; discussion S890-862, doi:10.1016/j.athoracsur.2006.10.088 (2007).
24 Kieffer, E. *et al.* Type IV thoracoabdominal aneurysm repair: predictors of postoperative mortality, spinal cord injury, and acute intestinal ischemia. *Ann Vasc Surg* **22**, 822-828, doi:10.1016/j.avsg.2008.07.002 (2008).
25 Richards, J. M. *et al.* Contemporary results for open repair of suprarenal and type IV thoracoabdominal aortic aneurysms. *Br J Surg* **97**, 45-49, doi:10.1002/bjs.6848 (2010).
26 Ockert, S. *et al.* Complex abdominal aortic pathologies: operative and midterm results after pararenal aortic aneurysm and type IV thoracoabdominal aneurysm repair. *Vascular* **17**, 121-128, doi:10.2310/6670.2009.00010 (2009).
27 Gustavo S. Oderich, T. T., Manju Kalra, Audra A. Duncan, Peter Gloviczki, Terri Vrtiska, Steve Cha, Thomas C. Bower. Contemporary Results of Open Complex Abdominal Aortic Aneurysm Repair Using a Standardized Classification for Comparison with Fenestrated Endografts. *JOURNAL OF VASCULAR SURGERY* **53**, 27S (2011).
28 Patel, V. I. *et al.* Continued favorable results with open surgical repair of type IV thoracoabdominal aortic aneurysms. *J Vasc Surg* **53**, 1492-1498, doi:10.1016/j.jvs.2011.01.070 (2011).

29 Schepens, M. A. *et al.* Long-term follow-up after thoracoabdominal aortic aneurysm repair. *Ann Thorac Surg* **83**, S851-855; discussion S890-852, doi:10.1016/j.athoracsur.2006.10.087 (2007).
30 Achneck, H. E., Rizzo, J. A., Tranquilli, M. & Elefteriades, J. A. Safety of thoracic aortic surgery in the present era. *Ann Thorac Surg* **84**, 1180-1185; discussion 1185, doi:10.1016/j.athoracsur.2007.05.038 (2007).
31 Conrad, M. F., Crawford, R. S., Davison, J. K. & Cambria, R. P. Thoracoabdominal aneurysm repair: a 20-year perspective. *Ann Thorac Surg* **83**, S856-861; discussion S890-852, doi:10.1016/j.athoracsur.2006.10.096 (2007).
32 Jacobs, M. J., van Eps, R. G., de Jong, D. S., Schurink, G. W. & Mochtar, B. Prevention of renal failure in patients undergoing thoracoabdominal aortic aneurysm repair. *J Vasc Surg* **40**, 1067-1073; discussion 1073, doi:10.1016/j.jvs.2004.08.055 (2004).
33 Safi, H. J. *et al.* Evolution of risk for neurologic deficit after descending and thoracoabdominal aortic repair. *Ann Thorac Surg* **80**, 2173-2179; discussion 2179, doi:10.1016/j.athoracsur.2005.05.060 (2005).
34 Grabitz, K. *et al.* The risk of ischemic spinal cord injury in patients undergoing graft replacement for thoracoabdominal aortic aneurysms. *J Vasc Surg* **23**, 230-240 (1996).
35 Svensson, L. G., Crawford, E. S., Hess, K. R., Coselli, J. S. & Safi, H. J. Experience with 1509 patients undergoing thoracoabdominal aortic operations. *J Vasc Surg* **17**, 357-368; discussion 368-370 (1993).
36 Clouse WD, C., R. P. *Complex Aortic Aneurysm: Pararenal, Suprarenal, and Thoracoabdominal* (2009).
37 Rigberg, D. A. *et al.* Thirty-day mortality statistics underestimate the risk of repair of thoracoabdominal aortic aneurysms: a statewide experience. *J Vasc Surg* **43**, 217-222; discussion 223, doi:10.1016/j.jvs.2005.10.070 (2006).
38 Gustavo S. Oderich, E. B. R., Joseph J. Ricotta, II, Peter Gloviczki, Thomas C. Bower, Audra A. Duncan, Manju Kalra, Carlos Timaran. In-Hospital Mortality of Open Thoracoabdominal Aortic Aneurysm Repair in the United States Prior to Widespread Use of Hybrid and Endovascular Branched Techniques. *JOURNAL OF VASCULAR SURGERY* **49**, 55S (2009).
39 De Bruin, J. L. *et al.* Long-term outcome of open or endovascular repair of abdominal aortic aneurysm. *The New England journal of medicine* **362**, 1881-1889, doi:10.1056/NEJMoa0909499 (2010).
40 United Kingdom, E. T. I. *et al.* Endovascular versus open repair of abdominal aortic aneurysm. *The New England journal of medicine* **362**, 1863-1871, doi:10.1056/NEJMoa0909305 (2010).
41 Schermerhorn, M. L. *et al.* Endovascular vs. open repair of abdominal aortic aneurysms in the Medicare population. *The New England journal of medicine* **358**, 464-474, doi:10.1056/NEJMoa0707348 (2008).
42 Tran, M. T. *et al.* PGC1alpha drives NAD biosynthesis linking oxidative metabolism to renal protection. *Nature* **531**, 528-532, doi:10.1038/nature17184 (2016).
43 Benyo, Z. *et al.* GPR109A (PUMA-G/HM74A) mediates nicotinic acid-induced flushing. *The Journal of clinical investigation* **115**, 3634-3640, doi:10.1172/JCI23626 (2005).
44 Chini, C. C. S., Tarrago, M. G. & Chini, E. N. NAD and the aging process: Role in life, death and everything in between. *Molecular and cellular endocrinology* **455**, 62-74, doi:10.1016/j.mce.2016.11.003 (2017).
45 Canto, C. *et al.* The NAD(+) precursor nicotinamide riboside enhances oxidative metabolism and protects against high-fat diet-induced obesity. *Cell Metab* **15**, 838-847, doi:10.1016/j.cmet.2012.04.022 (2012).
46 Gomes, A. P. *et al.* Declining NAD(+) induces a pseudohypoxic state disrupting nuclear-mitochondrial communication during aging. *Cell* **155**, 1624-1638, doi:10.1016/j.cell.2013.11.037 (2013).
47 Dellinger, R. W. *et al.* Repeat dose NRPT (nicotinamide riboside and pterostilbene) increases NAD(+) levels in humans safely and sustainably: a randomized, double-blind, placebo-controlled study. *NPJ Aging Mech Dis* **3**, 17, doi:10.1038/s41514-017-0016-9 (2017).
48 McCormack, D. & McFadden, D. A review of pterostilbene antioxidant activity and disease modification. *Oxid Med Cell Longev* **2013**, 575482, doi:10.1155/2013/575482 (2013).
49 Rimando, A. M. & Suh, N. Biological/chemopreventive activity of stilbenes and their effect on colon cancer. *Planta Med* **74**, 1635-1643, doi:10.1055/s-0028-1088301 (2008).

50 Amarnath Satheesh, M. & Pari, L. The antioxidant role of pterostilbene in streptozotocin-nicotinamide-induced type 2 diabetes mellitus in Wistar rats. *J Pharm Pharmacol* **58**, 1483-1490, doi:10.1211/jpp.58.11.0009 (2006).
51 Gao, D., Jing, S., Zhang, Q. & Wu, G. Pterostilbene protects against acute renal ischemia reperfusion injury and inhibits oxidative stress, inducible nitric oxide synthase expression and inflammation in rats via the Toll-like receptor 4/nuclear factor-kappaB signaling pathway. *Exp Ther Med* **15**, 1029-1035, doi:10.3892/etm.2017.5479 (2018).
52 Riche, D. M. *et al.* Analysis of safety from a human clinical trial with pterostilbene. *Journal of toxicology* **2013**, 463595, doi:10.1155/2013/463595 (2013).
53 Belenky, P. *et al.* Nicotinamide riboside promotes Sir2 silencing and extends lifespan via Nrk and Urh1/Pnp1/Meu1 pathways to NAD+. *Cell* **129**, 473-484, doi:10.1016/j.cell.2007.03.024 (2007).
54 Yang, T., Chan, N. Y. & Sauve, A. A. Syntheses of nicotinamide riboside and derivatives: effective agents for increasing nicotinamide adenine dinucleotide concentrations in mammalian cells. *Journal of medicinal chemistry* **50**, 6458-6461, doi:10.1021/jm701001c (2007).
55 Khan, N. A. *et al.* Effective treatment of mitochondrial myopathy by nicotinamide riboside, a vitamin B3. *EMBO molecular medicine* **6**, 721-731, doi:10.1002/emmm.201403943 (2014).
56 Gong, B. *et al.* Nicotinamide riboside restores cognition through an upregulation of proliferator-activated receptor-gamma coactivator 1alpha regulated beta-secretase 1 degradation and mitochondrial gene expression in Alzheimer's mouse models. *Neurobiol Aging* **34**, 1581-1588, doi:10.1016/j.neurobiolaging.2012.12.005 (2013).
57 Shindler, K. S., Ventura, E., Rex, T. S., Elliott, P. & Rostami, A. SIRT1 activation confers neuroprotection in experimental optic neuritis. *Investigative ophthalmology & visual science* **48**, 3602-3609, doi:10.1167/iovs.07-0131 (2007).
58 Sasaki, Y., Araki, T. & Milbrandt, J. Stimulation of nicotinamide adenine dinucleotide biosynthetic pathways delays axonal degeneration after axotomy. *J Neurosci* **26**, 8484-8491, doi:10.1523/JNEUROSCI.2320-06.2006 (2006).
59 Brown, K. D. *et al.* Activation of SIRT3 by the NAD(+) precursor nicotinamide riboside protects from noise-induced hearing loss. *Cell Metab* **20**, 1059-1068, doi:10.1016/j.cmet.2014.11.003 (2014).
60 Zhou, C. C. *et al.* Hepatic NAD(+) deficiency as a therapeutic target for non-alcoholic fatty liver disease in ageing. *British journal of pharmacology* **173**, 2352-2368, doi:10.1111/bph.13513 (2016).
61 Mukherjee, S. *et al.* Nicotinamide adenine dinucleotide biosynthesis promotes liver regeneration. *Hepatology (Baltimore, Md.)* **65**, 616-630, doi:10.1002/hep.28912 (2017).
62 Mills, K. F. *et al.* Long-Term Administration of Nicotinamide Mononucleotide Mitigates Age-Associated Physiological Decline in Mice. *Cell Metab* **24**, 795-806, doi:10.1016/j.cmet.2016.09.013 (2016).
63 Zhang, H. *et al.* NAD(+) repletion improves mitochondrial and stem cell function and enhances life span in mice. *Science (New York, N.Y.)* **352**, 1436-1443, doi:10.1126/science.aaf2693 (2016).
64 Chi, Y. & Sauve, A. A. Nicotinamide riboside, a trace nutrient in foods, is a vitamin B3 with effects on energy metabolism and neuroprotection. *Current opinion in clinical nutrition and metabolic care* **16**, 657-661, doi:10.1097/MCO.0b013e32836510c0 (2013).
65 Guo, Y., Zhang, L., Li, F., Hu, C. P. & Zhang, Z. Restoration of sirt1 function by pterostilbene attenuates hypoxia-reoxygenation injury in cardiomyocytes. *Eur J Pharmacol* **776**, 26-33, doi:10.1016/j.ejphar.2016.02.052 (2016).
66 Trammell, S. A. *et al.* Nicotinamide riboside is uniquely and orally bioavailable in mice and humans. *Nat Commun* **7**, 12948, doi:10.1038/ncomms12948 (2016).
67 Riche, D. M. *et al.* Pterostilbene on metabolic parameters: a randomized, double-blind, and placebo-controlled trial. *Evid Based Complement Alternat Med* **2014**, 459165, doi:10.1155/2014/459165 (2014).
68 Airhart, S. E. *et al.* An open-label, non-randomized study of the pharmacokinetics of the nutritional supplement nicotinamide riboside (NR) and its effects on blood NAD+ levels in healthy volunteers. *PLoS One* **12**, e0186459, doi:10.1371/journal.pone.0186459 (2017).

69 Martens, C. R. *et al.* Chronic nicotinamide riboside supplementation is well-tolerated and elevates NAD(+) in healthy middle-aged and older adults. *Nat Commun* **9**, 1286, doi:10.1038/s41467-018-03421-7 (2018).
70 Dollerup, O. L. *et al.* A randomized placebo-controlled clinical trial of nicotinamide riboside in obese men: safety, insulin-sensitivity, and lipid-mobilizing effects. *Am J Clin Nutr*, doi:10.1093/ajcn/nqy132 (2018).
71 McCormack, D. & McFadden, D. Pterostilbene and cancer: current review. *J Surg Res* **173**, e53-61, doi:10.1016/j.jss.2011.09.054 (2012).
72 Kosuru, R., Rai, U., Prakash, S., Singh, A. & Singh, S. Promising therapeutic potential of pterostilbene and its mechanistic insight based on preclinical evidence. *Eur J Pharmacol* **789**, 229-243, doi:10.1016/j.ejphar.2016.07.046 (2016).
73 Levey, A. S. *et al.* A new equation to estimate glomerular filtration rate. *Ann Intern Med* **150**, 604-612 (2009).
74 Pocock, S. J. & Simon, R. Sequential treatment assignment with balancing for prognostic factors in the controlled clinical trial. *Biometrics* **31**, 103-115 (1975).