Brain Small Chain Fatty Acid Metabolism in Parkinson Disease: Ketones

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CLINICAL PROTOCOL

STUDY TITLE

Brain Small Chain Fatty Acid Metabolism in Parkinson Disease: Ketones

Study Agents: Ketone Ester, [^{18}F]FDG

University of Michigan IRB HUM #00213035

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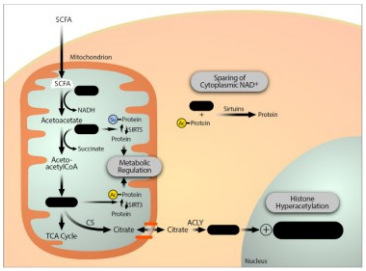
Brain Small Chain Fatty Acid Metabolism in Parkinson's Disease: Ketones

Cognitive decline, progressing usually to dementia, is a common disabling feature of Parkinson's disease (PD). Progression to dementia generally reflects progressive cortical pathologies. We and others showed that cognitive decline is accompanied and heralded by deficits in regional neocortical glucose metabolism, demonstrated with [^{18}F]fluorodeoxyglucose (FDG) PET imaging [1, 2]. These deficits are widely believed to reflect synaptic loss. Convergent imaging data, however, suggests relative preservation of neocortical synapses with abnormal neuronal glucose metabolism. Recent studies of PD and Alzheimer disease (AD) subjects with synaptic terminal SV2A PET ligands indicate preservation of cortical synaptic terminals at early to moderate disease stages. Ketone bodies, which are short chain fatty acids (SCFAs) are the major alternative brain energy substrates. Comparative studies of brain SCFA and glucose uptake in Mild Cognitive Impairment and early AD subjects indicates relative preservation of regional brain SFCA metabolism in subjects with abnormal regional glucose uptake, suggesting relative preservation of synapses.
This accumulated PET data is broadly consistent with the concept that mitochondrial dysfunction is a major feature of, and likely a pathogenetic mechanism in PD. Other data suggests relevant dysfunction of glucose uptake and metabolism mechanisms in PD. One implication is that enhancing neuronal SCFA metabolism will circumvent abnormalities in glucose uptake and initial metabolism, enhancing and restoring neuronal mitochondrial function. This would potentially provide symptomatic benefits and might be a disease-modifying therapy (**Figure 1**). There are other good reasons to pursue SCFA therapy in PD. SFCAs may be significant modulators of inflammatory, oxidative, and transcriptional regulatory processes linked to neurodegeneration in PD (**Figure 1**). Recent experimental work suggests important links between mitochondrial dysfunction and pathogenic inflammatory mechanisms in PD. SCFA therapy may address the brain-gut axis dysbiosis that seems to characterize PD.



Capitalizing on our experience with PD clinical research and development of novel PET methods, we propose an integrated series of experiments to measure brain SCFA metabolism and to assess target engagement of SCFA therapy in older adults and in persons with Parkinson disease (PwPD) as well as a subset of persons with Parkinson's-related dementias (Parkinson's disease dementia (PDD) or Lewy Body dementia (LBD)).

Aim 1: To explore glucose metabolism and clinical measures before and after open-label treatment with the ketone monoester (KE), ®-3-hydroxybut®(*R*)-3-hydroxy- butyrate or D-β-hydroxybutyrate in a small pilot study in patients with PD (n=16), LBD/PDD (n=10), and normal control older adults (n=4). To examine glucose metabolic changes including mean daily glucose for the first 7 days of wearing a continuous glucose monitor and fasting glucose that can be complemented by FDG PET in the subset undergoing PET imaging.

Hypothesis 1: Glucose metabolism (mean daily glucose and/or fasting glucose) may improve after approximately 30-day intervention with KE 25 g t.i.d. p.o. with meals in patients with PD and normal older adults.

Exploratory hypothesis: KE supplementation may affect cognition, autonomic, sleep, metabolic and other measures.

Impact: Positive findings in this small exploratory pilot trial may support future target engagement studies of ketone supplementation in PD and normal adults.

Background

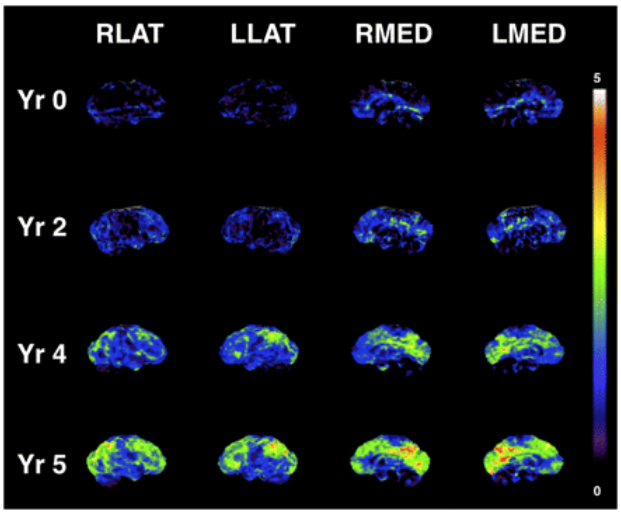
Dopamine replacement therapy (DRT) refractory features are major disabling aspects of PD. Health-related quality of life (QoL) studies and surveys of PwPD, caregivers, and clinicians involved in PD care identify DRT-refractory features as the most disabling aspect of PD and a high priority target for research aimed at improving the lives of PwPD [6]. One of the most important, and possibly most feared, DRT-refractory feature is dementia. Cross-sectional studies indicate high prevalence of dementia in PD, with increasing prevalence associated with disease progression [8]. In the prospective Sydney study of a clinical trial cohort, at least 80% of PwPD followed developed dementia after 15 years disease duration [10]. In a prospective follow-up of a Norwegian community-based cohort [14], 60% developed dementia at 12 years post diagnosis, and in the British CamPaiGN study of a community based inception cohort, approximately 50% met criteria for dementia at 10 years after study enrollment [17]. In non-demented PwPD, careful neuropsychological characterization demonstrates a high rate of cognitive impairments in the form of PD associated Mild Cognitive Impairment (PD-MCI) [19]. It is likely that all PwPD with disease of several years' duration will experience some form of significant cognitive impairment, if not overt dementia.

The pathophysiology underlying cognitive impairments and dementia is thought to largely reflect the progression of α-synucleinopathy in PD. The general evolution of cognitive impairments/dementia and their relationship to α-synucleinopathy is captured by the "Dual Syndrome" model of Kehagia et al [20]. This model, in turn, is based on the Braak et al. model of gradually advancing α-synucleinopathy in PD. Braak et al. hypothesized rostral spread of α-synucleinopathy from initial lower brainstem involvement to later invasion of forebrain and cortical structures [21]. In the Kehagia et al. correlative model, initial cognitive impairments reflect frontostriatal circuit dysfunctions consequent to nigrostriatal dopaminergic denervation with later cognitive dysfunctions and dementia [22] reflecting much broader cortical dysfunctions secondary to both loss of important subcortical afferent terminals [23], notably basal forebrain cholinergic corticopetal afferents, and increasing burden of α-synucleinopathy within cortical neurons.

While the Braak et al. and Kehagia et al. models are simplifications, they likely capture essential features of the pathogenesis and pathophysiology of cognitive impairments in PwPD. It is likely that overt neurodegeneration is proceeded by some period of neuronal dysfunction. Recent data suggest that neuronal dysfunction/neurodegeneration occur initially as "synaptopathy" with initial dysfunction and degeneration occurring at the level of neuronal terminals/synapses [24, 25]. This formulation suggests that there may be a period of terminal-synaptic dysfunction prior to loss of terminals-synapses in which appropriately targeted therapies might partially restore the function of dysfunctional synapses, producing symptomatic improvements.

Recent convergent data suggest that this concept of dysfunctional, but anatomically intact (sleeping or stunned) terminals-synapses may be particularly relevant to the cortical deficits underlying advancing cognitive impairments in PwPD. We and others showed that cognitive decline in PD is accompanied by and preceded by regional neocortical deficits of glucose uptake, demonstrated with [^{18}F]fluorodeoxyglucose positron emission

Figure 2: Serial FDG PET in PwPD and progressive cognitive decline. Progressive glucose metabolic reductions from within reference range at baseline (Yr 0), through stages of mild cognitive impairment (Yr 2 and Yr 4), and clinical diagnosis of dementia (Yr 5) in PwP converting to dementia over 5 years [1].



tomography (FDG-PET; **Figure 2**) [1, 2]. These FDG PET deficits are widely assumed to reflect terminal-synaptic degeneration, but recent *in vivo* human imaging data suggests that a significant component of neocortical FDG-PET deficits is metabolic dysfunction of anatomically intact terminals-synapses. The development of the synaptic terminal SV2A protein PET ligand [^{11}C]UCB-J allowed independent estimates of regional synaptic terminal densities. In mild Alzheimer disease (AD) spectrum, subjects expected to exhibit several regional neocortical FDG-PET deficits. [^{11}C[UCB-J PET revealed synaptic terminal deficits restricted to the hippocampal formation [26]. Similarly, initial studies with [^{11}C]UCB-J PET in mild PD subjects revealed synaptic terminal deficits restricted to the substantia nigra[27, 28].

The recent development of alternative methods for imaging brain metabolic processes also supports the concept of sleeping-stunned synapses in neurodegenerative disorders. Brain energy consumption is primarily devoted to maintaining the ionic gradients needed for neurotransmission-synaptic function. Under normal circumstances, glucose is the primary substrate for brain metabolism. Under fasting conditions, however, ketone bodies such as β-hydroxybutyrate (BHB) and other species of short chain fatty acids (SCFAs), substitute for glucose directly within mitochondria. Cunnane and colleagues developed [^{11}C]acetoacetate (AcAc) PET as a non-invasive method to study SCFA metabolism *in vivo*. In studies comparing [^{18}F]FDG PET and [^{11}C]AcAc PET in mild AD spectrum subjects, this group demonstrated normal regional [^{11}C]AcAc uptake in subjects with significant regional FDG PET deficits [29]. This result is consistent with SV2A/[^{11}C]UCB-J PET imaging results and supports the conclusion that a significant component of regional FDG PET deficits reflect dysfunctional (sleeping-stunned), as opposed to anatomically lost synapses. In turn, these results suggest that therapies circumventing regional glucose metabolism deficits might improve synaptic function in neurodegenerative disorders where comparative metabolic imaging studies demonstrate regional glucose uptake (FDG PET) deficits greater than SCFA metabolism (AcAc PET or analogous methods) deficits. SCFA supplementation (see below) may be such a therapy.

In PD, the concepts of synaptic dysfunction associated with metabolic deficits and potential remediation by circumventing metabolic dysfunction using a fuel repartitioning approach are consistent with a substantial body of data implicating metabolic defects in PD pathogenesis. Mitochondrial dysfunctions secondary to electron transport chain deficits are felt to be a major pathogenetic mechanism in sporadic PD with some monogenetic forms of PD resulting directly from mitochondrial dysfunctions [30-32]. Glucose transporters, essential for glucose transport across the blood-brain barrier and into CNS cells, may be down-regulated in PD [33]. Boosting neuronal energy metabolism by bypassing glycolysis and feeding directly into the Krebs cycle is a plausible approach to enhancing mitochondrial function. This is feasible via dietary ketone body-SCFA supplementation. These SCFAs are directly transported across the blood-brain barrier and into astrocytes and neurons via monocarboxylate transporters (MCTs). In addition, fasting or ketone body based (ketogenic) diets induce brain metabolism of ketone bodies-SCFAs, thereby reducing reliance on glucose as the primary energy substrate. This is amply demonstrated in experimental studies and is demonstrated more recently with effects of a ketogenic diet on AcAc PET in normal human volunteers [34]. Ketone body-SCFA supplementation may plausibly “awaken” sleeping-stunned synapses and produce symptomatic improvements in PD. Given the association between regional cortical metabolic deficits and cognitive decline in PD, we are targeting cognitive decline as the ultimate target for symptomatic

relief of this important feature of PD via SCFA supplementation.

Ketogenic diet and SCFA interventions in neurodegeneration:
The ketogenic diet based on preferential intake of predominant medium and long-chain fatty acids and minimization of carbohydrates has become a subject of recent research in metabolic and neurodegenerative disorders. Preliminary positive findings have been reported mainly from short-duration studies. Despite the benefits of this diet, health care professionals still question its safety due to the elevated serum ketones it induces and the limited dietary fiber intake [35]. To compound the controversy, patient compliance with the program is poor due to the restrictive nature of the diet, symptoms related to energy deficit, and gastrointestinal adversity during the introductory and energy substrate transition phase of the diet. A ketone ester, like the ketone monoester (KE), (*R*)-3-hydroxybutyl-(*R*)-3-hydroxy-butyrate, is a drink that, after ingestion, is cleaved by gut esterases into equimolar βHB and (*R*)-1,3-butanediol. Both molecules enter the portal circulation, and the latter is converted in the liver into βHB. Therefore, KE supplementation offers the advantage that no dietary restrictions are needed and can be taken together with regular meals. This approach of t.i.d. oral KE supplementation together with the study participant's regular diet has been successfully and safely used in 1-month trials in healthy human volunteers as well as in patients with type 2 diabetes mellitus [36, 37].

Overall strategy

Overview
The overarching goal of this small exploratory open-label pilot study is to explore [^{18}F]FDG PET glucose metabolism and clinical measures before and after open-label treatment with the ketone ester (R)-3-hydroxybutyl (R)-3-hydroxybutyrate (KE) 25 g t.i.d. po in a small open label phase I pilot study in PD and normal control older adults. Positive findings in this small exploratory pilot trial may support target engagement studies of ketone supplementation in normal adults, PD, and PDD/LBD. Additionally, the goal of the optional sub-study (see complete protocol for sub-study on page 19) is to assess the feasibility of an exercise and supplement intervention in a subset of participants.

Timetable: This study, to be conducted over a 2-year period, will include a net total of *n=30 subjects (NC, n=4; PD, n=16; LBD/PDD, n= 10; gross n=42 to allow for attrition).* Details of the enrollment and major test procedures are listed in **table 1** below. Additionally, 10 participants will be recruited to complete the optional sub-study.

Milestone of recruitment success are net completion of 9 persons for year 1.

Table 1: Study enrollment & main assessments	**NC**	**PD**	**PDD/LBD**	**TOTAL**
Baseline clinical assessment with (optional) [^{18}F]FDG PET-CT & brain MRI	NC=4	PD=16	PDD/LBD=10	n=30

30-day (± 7 days) KE 25 g tid po intervention with pre/post clinical assessments)	NC=4	PD=16	PDD/LBD=10	n=30
Post-intervention clinical assessment with (optional) [^{18}F]FDG PET-CT & brain MRI	NC=4	PD=16	PDD/LBD=10	n=30
Total subject net recruitment aims	NC=4	PD=16	PDD/LBD=10	n=30*

**Net* recruitment n=30; *gross* recruitment to account for attrition n=42. We will aim to complete 10 participants in the optional sub-study, all who will be recruited from the net recruitment pool.

Design: Exploratory open-label KE drink 25 g t.i.d. po (titrated up from 12.5g t.i.d) supplementation pilot study.

Methods: After screening for study eligibility and obtaining informed consent (including eConsent), study participants will undergo the baseline clinical (vital signs, respiration, and general neurological assessment and optional imaging protocol (separate days). For patients taking dopaminergic medication, the motor component of the test battery will be performed in the dopaminergic medication 'off' state in the morning after withholding their dopaminergic medications in the PD overnight, i.e., temporarily postponing the morning dose until the motor exam has completed. The remainder of the test battery will be performed in the medication 'on' state. Participants will also complete some testing in a fasted state. For participants who were not consented prior arriving for their baseline visit, study staff will obtain a waiver of consent during the screening call for fasting and withholding dopaminergic medications prior to their visit. Qualified personnel who have been trained in the use of the instruments and have undergone inter-rater reliability evaluations will perform the assessments.

There will be a total of 5 visits for this pilot study. The first two visits will consist of the initial clinical assessment and optional imaging split over 2 days (no specific sequence or preset time interval). At visit 1 or 2, participants will be instructed to wear physical activity and sleep trackers (ActivPal, which is a sensor that will be put on the upper leg and a ring called Oura ring, which will be worn at a finger) for approximately 1 week prior to visit 3. Participants will also wear a continuous glucose monitor during the same time. After the participants complete the approximate 1 week of physical activity, sleep and glucose monitoring they will return for visit 3, where a limited repeat clinical test battery will be administered. At visit 3 the participant will also be instructed to start the approximately 30-day supplementation of KE drink as well as to wear the sleep tracking ring throughout their time taking the supplement. Participants who are willing and eligible to take part in the optional single-day sub-study will complete the optional sub-study at visit 3, where they will still get the first dose of the KE drink in lab. See page 19 of protocol for more information on the optional sub-study. The physical activity tracker and the continuous glucose monitor will also be worn during the last week of the KE supplementation period. At the end of the approximately 30-day KE supplementation, the participants will return for visits 4 (repeat clinical assessment) and 5 (repeat optional imaging) again split over 2 days. There is no pre-set sequence which assessment (clinical or imaging) comes first. Blood tests will be collected for assessments of genetic variables (e.g. ApoE). Blood and/or urine assessments will also be used for metabolomics, inflammatory markers, and/or screening measures. Blood continuous glucose data will be collected blinded to the participant, except for the first week of taking the ketone ester as this

information is needed for safety monitoring purposes. Participants will also take readings of their glucose and ketone body levels using a Keto-Mojo device (see Table 2 for more information).

Labs to be performed:

- CBC w/ Diff
- CMP w/ eGFR
- High-sensitivity CRP
- Hemoglobin A1c
- RBC Magnesium
- Phosphorus
- Albumin
- TSH
- T4
- Reverse T3
- Lipid Panel
- Urinalysis
- Beta-2 Microglobulin
- Homocysteine
- Vit D (25-hydroxyvitamin D)
- Folate
- Uric acid – serum

Additionally, there will be an optional single-day sub-study involving combining the KE supplement with a coffee supplement and cardiovascular exercise. This optional sub-study will take place at Visit 3. Participants can still complete the rest of the study if they are not able or do not wish to participate in the sub-study. See page 20 of protocol for a complete sub-study protocol and schedule of events.

Details for the schedule of activities for the KE supplement intervention study are shown below (Table 2).

Table 2 Schedule of activities	Visit 1* / Visit 2*: Baseline assessment (clinical)	Visit 1* / Visit 2* Baseline assessment (imaging – OPTIONAL)	At-home wearable sensor use (7 days ± 3 days)	Visit 3 Pre-intervention assessment[4] (including optional sub-study if applicable)	At-home 30 (± 7 days) KE (25 g tid po) intervention[3]	Visit 4* / Visit 5* Post-intervention clinical assessment (while on KE)	Visit 4* / Visit 5* Post-intervention imaging assessment (while on KE, OPTIONAL)
Approximate time commitment	4-6 hours	3-6 hours	-	4-5 hours	-	4-6 hours	3- 6 hours
Test location	Dominos Farms	UM hospital	Home	Dominos Farms	Home	Dominos Farms	UM hospital
Informed Consent (or prior eConsent)	X						
Eligibility	X						
OPTIONAL [^{18}F]FDG PET-CT [1.2] (brain/body)*		X					X

DEXA	X					X	
OPTIONAL MRI brain*		X					X
Blood/urine sample collection[1]	X					X	
Ketone Ester Drink				X (first dose taken in lab; may be taken during optional sub-study)	X	X	X
Continuous glucose monitor for 7±3 days			X		X (first ~7 days of taking drink [unblinded] and last ~7 days of taking drink)	X	
Keto-Mojo glucose/ketone body monitoring***				X (before and after initial dose of KE in lab; optional)	X (first ~7 days of taking drink and weekly throughout intervention; optional)	X (prior to testing)	X (prior to imaging)
Sleep tracking ring			X	X	X (worn for duration of using ketone ester drink)	X	
Activity tracker for 7±3 days			X		X (last ~7 days of taking drink)		
Motor MDS UPDRS[2]	X			X (optional)		X	
Balance test MiniBESTest (sensored; optional for PDD/LBD/MCI patients)[2]	X			X (optional)		X	
Pegboard testing[2]	X			X (optional)		X	
Foot Tapping[2] (optional for PDD/LBD/MCI patients)	X			X (optional)		X	
Finger Tapping[2]	X			X (optional)		X	
Electronic Gait Mat/sensored walk[2] (optional for PDD/LBD/MCI patients)	X			X (optional)		X	
Beck Depression Inventory II scale	X			X		X	
Spielberger Trait Anxiety Scale	X			X		X	

Sleepiness scales	X					X	
MoCa, PDCRS (limited cognition)	X			X (optional)		X	
Cognitive test battery**	X			X (optional)		X	
PCFRS functional scale	X					X	
Clinical Dementia Rating (CDR) Scale****	X					X	
Quality of life scale PDQ-39	X					X	
Adverse event assessment	X			X		X	
Phone Call / email drug monitoring					X		
Drug accountability				X		X	
Pregnancy test in women of childbearing potential		X (within 48 hours of PET)		X			X (within 48 hours of PET)

Note
[1]: Fasting in morning
[2]: Dopaminergic off state in PD
[3]: Participants will take 12.5g BID for day 1 of intervention, 12.5g TID for days 2 through 7 and increase dose to 25g on day 8 of intervention
[4]: Visit 3 will have optional motor and cognitive testing for individuals who have more than 2 months between their baseline clinical assessment and pre-intervention clinical assessment. For those individuals who do not have to complete motor testing, they will not be required to withhold dopaminergic medication. For individuals taking part in the optional sub-study during the pre-intervention visit, they will still be required to withhold their medication and fast the morning of the visit.
*Visits 1-2 and 4-5. There is no pre-set sequence which assessment (clinical or imaging) comes first. There may be a time interval of up to two months between Visits 1 and Visits 3, depending on scheduling
**Portions of cognitive test battery will be optional for LBD/PDD/MCI participants in anticipation of fatigue
***Keto-mojo usage will be optional if it is deemed too difficult for participant to administer by themselves. It will be required prior to post-intervention imaging and clinical visit (V4/5), but may be performed by research staff as needed
****Clinical Dementia Rating Scale will only be completed by LBD/PDD/MCI subjects

Details of the neuropsychological test battery is shown in Table 3.

Outcome measures: Primary outcome measures will be mean glucose metabolism for the first 7 days of wearing the continuous glucose monitor. Glucose metabolic changes include daily mean over first 7 days of wearing the continuous glucose monitor and/or, as an exploratory measure, fasting glucose based on FDG PET metabolism in the subset undergoing PET imaging. Clinical Dementia Rating scale at baseline and post-intervention will also serve as a primary outcome measure for subjects with LBD, PDD, and MCI.

Table 3: Neuropsychological characterization

Overall mental status	Montreal Cognitive Assessment Scale [3] PD-Cognitive Rating Scale [4]
Intelligence	WAIS-IV: Information[5]***
Attention & Cognitive Control	n-back test [7] (optional), Stroop Color Word Interference Test [9]** WAIS-IV: Digit Span[5]***
Language	Boston Naming Test (30 item) [11]*** Controlled Oral Word Association Test (Phonemic & Semantic)
Learning & Memory	California Verbal Learning Test – II [12]***
Visuospatial/Construction	Clock drawing [13] Benton Judgment of Line Orientation [15]
Executive/Working Memory	D-KEFS: Color Word Test, Verbal Fluency Test, Trail Making Test, Sorting Test [16]*** WAIS-IV Matrix Reasoning***, Digit Backwards [5]***
Information processing speed	D-KEFS: Trail Making Test [16] WAIS-III: Digit Symbol-Coding [5, 18]*** Reaction time

*Neuropsychological test battery will be optional at Visit 3
** Colorblind participants will complete Condition 1 only
***Portions of the neuropsychological test battery will be optional for PDD/LBD patients in anticipation of fatigue

Exploratory outcome measures include: 1) Clinical and lab measures; *Cognition dependent mobility functions:* This test battery will include postural sway using the iSWAY and instrumented Timed Up and Go test (iTUG) Mobility Lab modules (APDM, Inc., Portland, OR) [38-40], dynamic balance using the Mini-BESTest [41] with sensored subtests, and electronic gait mat assessment using the Protokinetics Zeno™ Walkway (Protokinetics LLC, Havertown, PA) for spatiotemporal gait measurements.

KE intervention: The active ingredient of the KE drink used in this study is the ketone monoester or (R)-3-hydroxybutyl (R)-3-hydroxybutyrate or D-β-hydroxybutyrate ester, a substance that was determined to be "Generally Recognized as Safe (GRAS)" by the FDA (GRN No.515). The KE is a drink that, after ingestion, is cleaved by gut esterases into equimolar βHB and (*R*)-1,3-butanediol. Both molecules enter the portal circulation, and the latter is converted in the liver into βHB [42]. Thus, each equivalent of KE yields two equivalents of βHB [43]. Compared to similar ingredients, such as the ketone diester or ketone salts, the ketogenic supplements such as MCTs and ketogenic diets, the KE induces acutely the most robust ketosis. The KE has been administered to one 63-year-old male patient with moderate-severe sporadic Alzheimer during a 20-month period. The patient and caregiver tried a range of doses, including the dose of 25 g p.o. three times daily. This dose level, which was the starting dose for the patient, elicited a robust behavioral improvement when first started and resulted in plasma βHB level of 3 mM. This particular patient tolerated the KE well and exhibited a rapid and sustained behavioral improvement [44]. KE has also been administered to exercising PwPD resulting in improved exercise endurance performance compared to placebo [45]. A common KE dose of 25g t.i.d. p.o., which is being used in this study, can safely elevate plasma βHB up to 4.0 mM or more [36], a concentration that is at least 8-times higher than that achieved with other ketogenic supplements [46]. The acute induction of ketosis achieved with the KE is roughly the equivalent of ketosis achieved after two weeks on a strict (high-fat, low-carb) ketogenic diet [47] or few days of complete fasting (starvation) [48]. A single KE drink used in this study contains 25 g of KE, which in addition to the active ingredient contains water, flavorings, and preservatives. The KE is produced in a GMP-compliant facility in the U.S. (Ketonaid.com).
The pharmacology and safety of KE has been thoroughly studied in animals [49] and, for one month, in healthy human adults [36] and in patients with type 2 diabetes mellitus. Safety and tolerability of sustained exogenous ketosis was assessed using ketone monoester drinks for 28 days in healthy adults. 24 healthy adults, aged 18-70 years, drank 25 ml (26.8 g) of the KE, three times a day for 28

days (a total of 2.1 L). Anthropomorphic measurements, plus fasting blood and urine analyses were made weekly. It was found that elevating blood βHB concentrations from 0.1 to 4.1 (±1.1) mM three times a day for 28 days had no effect on body weights or composition, fasting blood glucose, cholesterol, triglyceride or electrolyte concentrations, nor blood gases or kidney function, which were invariably normal [36]. In addition, a 28-day study in patients with type 2 diabetes also on 26.8 g t.i.d. po found that all participants had >90% adherence, defined as consumption of at least 76 of the maximum 84 KE drinks. Thus, the KE drink was well-tolerated. Adverse symptoms were self-recorded by participants following each of their drinks and were ranked as 'mild', 'moderate' or 'severe'. Overall occurrence of adverse symptoms is expressed relative to the total number, 1,588, of drinks consumed throughout the entire study; mild nausea (5/1,588 = 0.3%), mild headache (5/1,588 = 0.3%) and mild gastric discomfort (9/1,588 = 0.5%) were the only reported symptoms [37]. Mild nausea was reported following 6 of the 2,016 drinks consumed in the 28-day healthy control study [36].

Research staff will administer the first dose of KE drink at Visit 3 in lab to monitor subjects (day 1), and then subjects will be sent home and asked to take a second dose of KE drink with their next meal (day 1). For days 2 through 7, participants will take 12.5g (25mL) TID, and on day 8, the dose will be increased to 25g (50mL) TID as tolerated. KE dose will be titrated down to a tolerated level if necessary. Participants will be asked to take the KE doses with meals to decrease side-effects of hypoglycemia. If participants are experiencing feelings of low glucose levels and/or a blood glucose reading of <65mg/dL (via unblinded continuous glucose monitor and/or KetoMojo reading), participants will be asked to skip the KE dose and wait until their next meal to take a dose. Glucose and ketone body levels will be monitored for the first ~7days using Keto-Mojo (optional) to decrease chances of hypoglycemia, as well as taken weekly for the duration of the KE intervention (optional). Glucose and ketone body levels will also be monitored prior to the post-intervention clinical testing and imaging. Staff will ask participants to limit their alcohol consumption to a maximum of one glass per day and take the final daily dose at least 4 hours prior to sleep.

Magnetic resonance imaging (MRI) will be performed on a 3 Tesla Philips Achieva system (Philips, Best, The Netherlands). A 3D inversion recovery-prepared turbo-field-echo was performed in the sagittal plane using TR/TE/TI=9.8/4.6/1041ms; turbo factor=200; single average; FOV=240x200x160mm; acquired Matrix = 240x200x160 slices and reconstructed to 1mm isotropic resolution. MR-spectroscopy and other sequences will also be performed. Resting-state fMRI scans will serve as a secondary outcome measure and be acquired using a 32-channel head coil and multi-band sequence [50], with nominal parameters: (TR/TE/FA = 720/34/52, 2mm isotropic resolution, 72 slices). Data are collected with eyes fixated on a cross for 10 minutes [51]. Pre-processing follows our standard pipeline that includes physiologic noise reduction and thorough motion correction [52, 53]. Graph theory analysis will be used to identify the most highly connected nodes following published methods. Briefly, ROIs will be represented as nodes and pairwise correlations, based upon the mean time series for each ROI, will serve as weights over corresponding edges, thresholded by FDR correction, $p<0.05$ [52, 53]. Change from baseline will serve as the primary measure of interest. Magnetic resonance spectroscopy (MRS) may optionally complement the MRI sequence. MRS may be performed either at University Hospital or North Campus fMRI Laboratory.

PET imaging will be performed in 3D imaging mode on a Biograph 6 TruePoint PET/CT scanner (Siemens Molecular Imaging, Inc., Knoxville, TN), which acquire 63 transaxial slices (slice thickness: 2.4 mm) over a 15.2 cm axial field-of-view. Images were corrected for scatter and motion. The PET imaging frames will be spatially coregistered within subjects with a rigid-body transformation to reduce the effects of subject motion during the imaging session [54]. Statistical parametric mapping (SPM)

software (SPM12; Wellcome Trust Centre for Neuroimaging, University College, London, England [https://www.fil.ion.ucl.ac.uk/spm/software/spm12/] will be used for PET-MRI registration using the cropped T1-weighted MR volumetric scan. Freesurfer software (http://surfer.nmr.mgh.harvard.edu) will be used to define cortical and subcortical volumes-of-interest (VOI).

MRI-based PET partial volume correction: We will use the Müller-Gärtner partial volume correction technique, which is based on the assumption that white matter uptake is homogeneous and uses gray matter (GM), white matter (WM) and cerebrospinal fluid (CSF) to correct the partial volume effect in GM tissues [55]. Using the segmented images and the assumed PET resolution, the method calculates the spill-out from WM to GM and subsequently subtracts it from the GM. Similarly, it also calculates the spill-out from the GM to the surrounding tissues and compensates for the difference to the GM. The result is a grey matter image with corrected activity values in all voxels.

Statistical analysis
We will use a paired t-test to assess for changes in glucose metabolism before and after the KE supplementation. Glucose metabolic changes include 7-10 daily average and fasting glucose based on continuous glucose monitoring that can be complemented by cerebral FDG PET SUVr in the subset undergoing PET imaging.

As a *post hoc* and only exploratory analyses we will regress differences in with KE -induced changes of the various clinical and laboratory outcome measures.

Sample size estimation: This experiment is a first in human study of this drug for treatment of glucose changes in a neurodegenerative condition. Based on the 28-day t.i.d. KE intervention study performed in patients with type 2 diabetes mellitus [37], and similar effect size in our cohort and using a power of 0.8 and a two-sided alpha of 0.05 for our primary analysis that will compare pre and post KE, we arrive at a total sample size of 30.

Scientific rigor: All tests and neuroimaging procedures used in this proposal have been validated and protocols have been published. Both PET and MR neuroimaging assessments will follow established and published acquisition protocols. Neuroimaging analysis will include well-validated and widely used software packages and processing pipelines such as SPM12, FreeSurfer, FSL, and AFNI. All equipment for mobility testing is commercially available and vendor-provided (validated) outcome parameters will be used. The neuropsychological test battery includes tailored components for use in PwPD [56]. We will rigorously guard against Type I and Type II errors and correct all our statistical inferences with family-wise error and false discovery rate corrections where appropriate. This reproducible and rigorous methodological and analysis approach will ensure results and conclusions that can be replicated by other investigations.

Pitfalls, problems, and alternative strategies

Ineligibility and attrition rate:

Protection of Human Subjects

i) Human Subjects
Description of subject populations:

i) Healthy control persons, M/F, age 45 years and over without a history of neurological or psychiatric disease or significant GI disease or uncontrolled medical comorbidity will be recruited (net n=4).
ii) PD (net n=16) Diagnosis of PD in PD participants (M/F, age 45 years and over) will be based on the United Kingdom Parkinson's Disease Society Brain Bank Diagnostic Research Criteria (patients with clinically significant dementia will be excluded from PD subset and may participate in LBD/PDD subset (i.e., fails informed assent screening criteria and has significant evidence of iADL functions, such as ability to take own medications while administering the pre-consent screening questionnaire).
iii) Persons diagnosed with Lewy Body Dementia (LBD) based on the Fourth consensus report of the DLB Consortium inclusion criteria for probable DLB or Parkinson's Disease Dementia (PDD) based on the criteria set by Emre *et al.* for PDD (M/F, age 45 years and over; net=10).

Exclusion criteria:

1. *Exclusion for all subjects*: (a) Evidence of large vessel stroke or mass lesion on MRI; (b) Regular use of anti-cholinergic drugs or taking high dose (>100mg QD) of quetiapine (c) history of significant GI disease, (d) significant metabolic or uncontrolled medical comorbidity, (e) poorly controlled diabetes, (f) pregnancy or breast feeding, (g) suicidal ideation as determined by Beck Depression Inventory Question 9, (h) current excessive alcohol use as determined by investigator, and (I) subjects with contra-indications to MR imaging, including pacemakers, metal fragments in the body, or severe claustrophobia (for participants undergoing imaging); (i) participants with significant prior radiation exposure may not be eligible for the optional imaging portion of this study.

Recruitment: Subjects will be recruited from the University of Michigan Movement Disorder clinics, UM research studies website, Data Direct, and/or advertisement/flyers.

ii) Sources of Materials:

Information gathered specifically for this research project includes test results from clinical test scales, psychometric cognitive measures, neurobehavioral rating scales, clinical motor data, questionnaires, and data derived from PET and brain MRI.

iii) Potential Risks (as described in informed consent document):

Confidentiality of Research Information: The research data to be collected from subjects will consist of confidential information relating to clinical, motor, cognitive, neurobehavioral, and neuro-imaging functions. These research data are not intended for entry into the subjects' clinical medical records. However, the data remain potentially discoverable. This may lead to violation of privacy and embarrassment of the subject.

Clinical Testing: Risks regarding the clinical assessment are limited to fatigue, frustration and momentary embarrassment that may occur when one experiences difficulty performing a task or learning a new skill. There is an infrequent risk that the blood sample method may cause minor bruising at the injection site. There is a very rare risk for infection. There is a rare risk that you may feel dizzy, lightheaded, or faint after the blood draw. There is an infrequent risk of pain or infection associated with finger prick using Keto-Mojo.

Motor Testing: Many of the tests are comparable to normal standing and walking conditions that you may experience in everyday-life. Nonetheless, there is an infrequent risk of falling or near-falling during these tests which may result in fall-related injuries. There is a very rare risk that the sensors to measure overall movement and balance may become detached and that you may trip. You may also trip on the pressure sensitive mat.

Risks associated with clinical, neuropsychological and behavioral testing: This study involves cognitive tests and questionnaires. The cognitive tests are not harmful, but some people find them frustrating and concerning. Risks regarding the neuropsychological and behavioral assessment are limited to fatigue, frustration and momentary embarrassment that may occur when one experiences difficulty performing a task or learning a new skill. Some people may be uncomfortable/embarrassed disclosing personal information or become nervous about memory testing and may experience discomfort or become tired. Study team members are very well experienced in the assessment of older individuals and persons with dementia, understanding the need for breaks, gentle reassurance, or reinforcement. All assessments will be stopped if requested by the participant.

MRI scan: There is a substantial risk to persons who have metallic objects inside their bodies, as the magnet in the MR scanner can cause these to move. Consequently, participants with pacemakers or metallic objects located in the eye, ear, brain, or blood vessel walls will be excluded from the optional imaging portion. Participants who developed claustrophobic anxiety during scanning found that this fear dissipated within 15 min while remaining in the scanner, or as necessary, after exiting the scanner. There also is the potential that imaging could reveal a previously unrecognized but pre-existing abnormality. Many such abnormalities are not clinically significant, but they may cause anxiety or require further investigation by a personal physician. If one of the investigators identifies such an abnormality, they will contact the personal physician, who will arrange for appropriate care.

PET-CT scans & venipuncture: Insertion of a catheter for intravenous injection of the PET radiopharmaceutical may be commonly associated with slight pain or bruising at the puncture site and rare chance of infection. Participation in this research study will involve low level exposure to radiation associated with the PET, CT, and DEXA scans.

The use of [^{18}F]FDG is considered to be generally safe and effective as approved by the University of Michigan Radioactive Drug Research Committee in accordance with Food and Drug Administration regulations (21 CFR 361.1). Certified staff will be in attendance at all times during the study. A physician will be available, and an emergency cart is located in the PET Facility for treatment of any adverse reactions that may occur.

During the course of this study, the participant will be exposed to radiation from the two [^{18}F]FDG PET-CT and two DEXA scans. The risks associated with the amount of radiation exposure participants receive in this study are considered very rare and comparable to everyday radiation exposure risks.

The biological effect of radiation in humans is measured in terms of Sieverts (Sv) or mSv (1/1000 Sv), which is a unit of uniform whole body exposure. The radiation exposure a participant will receive from the CT, and [^{18}F]FDG PET scan is equivalent to a uniform whole body dose of 12.3 mSv, which is approximately 24.6 % of the annual radiation exposure (50 mSv) permitted to radiation workers by federal regulations. The participant will be instructed to use the bathroom and urinate as soon as possible after the PET scans in order to minimize bladder exposure. The radiation the participant will

be exposed to from each DEXA scan is 0.0310 mSv, which is significantly less than 1% of the permitted annual radiation exposure.

There is no known minimum level of radiation exposure that is recognized as being totally free of the risk of causing genetic defects (cellular abnormalities) or cancer. However, the risk associated with the amount of radiation exposure that participants will receive from this study is considered to be low. The risk of a side effect from this level of radiation exposure is very rare. The risk from radiation exposure of this amount is considered to be similar to other everyday risks, such as driving a car.

No PET studies will be performed on pregnant, nursing, or potentially pregnant women.

A urine pregnancy test will be performed on all women of childbearing potential within 48 hours prior to the PET scanning session.

Risks associated with the ketone ester: The risks to participants are low. Studies in humans have shown evidence of nausea, gastro-intestinal discomfort, abdominal distention, diarrhea, dizziness, lightheadedness, sweating, headache, keto flu-like symptoms, heartburn, shakiness, sleep changes, other symptoms of hypoglycemia, and/or increased glucose levels. Poor or bitter taste is a more common side-effect. Therefore, the KE drink may be diluted. The KE in this study has received GRAS (Generally Regarded As Safe, GRN No.515) certification from the FDA [57], is commercially available in the United States and prior work has shown that the 25g tid po dosing regimen proposed in this study has been found to be safe and generally well-tolerated in healthy controls and patients with type 2 diabetes mellitus [36, 37]. If side effects occur, the study team will decrease the dose and/or ask participants to skip a dose until their next meal.

Data Study Monitoring Plan (DSMP): Drs. Bohnen and Albin will act as the Data Safety Monitoring Plan members for this study. All adverse events and other study related events will be discussed at monthly investigator meetings. Adverse events will be classified according to standard criteria and reported to the IRBMED (relevant University of Michigan IRB) according to standard guidelines. Serious adverse events will be reported within 24 hours.
For risk of suicidality (if participants select answer responses 2 or 3 for question 9 on the Beck Depression Inventory, i.e. 'I would like to kill myself' (answer 2) or 'I would like to kill myself if I had a chance' (answer 3) then the staff will hand out the UM Depression brochure, refer to the contact information and inform Dr. Bohnen or Dr. Albin who will discuss this with the participant. The face-to-face or video conversation with the physicians will occur during that visit and will not be deferred. There will be a safety plan in place before the patient leaves the building.

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Optional Sub-Study Protocol (to be completed at Visit 3 Pre-intervention)

1. Background

Parkinson disease (PD) is a neurodegenerative disease which leads to progressive motor, neuropsychologic, and autonomic dysfunction. At this time, there are no disease modifying agents available and treatment is primarily through symptomatic management including dopamine replacement therapy. Mitochondrial dysfunctions secondary to electron transport chain deficits are considered to be a major pathogenetic mechanism in sporadic PD with some monogenetic forms of PD resulting directly from mitochondrial dysfunctions [1,2,3]. To mitigate this dysfunction, an individual with PD (William Curtis) has employed a strategy of intermittent fasting, exercise, and dietary modification in combination with medication to manage his disease. Dietary modifications include coffee with grass fed clarified butter and grass-fed coffee creamer and a ketone ester supplement each morning. This is also in addition to daily morning exercise. He has history of PD for 23 years and has used this strategy for over 7 years and during this time has had relative stability of his motor symptoms as measured by UPDRS. Rationale behind this regimen includes increase in NAD and NADPH through entrainment of the circadian rhythm and regular exercise which then drives neurotransmitter synthesis when coupled with sustained ketosis (achieved via slow-release fatty acid ketone precursors in buttered coffee combined with fast-release ketone ester). Moreover, butyric acid or other molecules in the fat-fortified coffee may enact other mechanistic effects that have yet to be fully understood. We would first like to evaluate the target engagement and tolerability of this regimen in people with PD.

2. Study Design

Study Overview

This is a single-arm and open-label test for target engagement of the Modified Curtis Protocol in PD. The Modified Curtis Protocol consists of a single dose of a fatty acid and coffee supplement followed by a single dose of ketone ester (KE) which is then followed by cardiovascular exercise.

Aim 1: To evaluate target engagement and the effect of the Modified Curtis Protocol on ketosis in Parkinson disease as assessed by serum beta hydroxybutyrate (BHB).

- **Hypothesis 1:** Participants will experience an increase in serum BHB levels 60 minutes after ingestion of KE.
- **Aim 1 Outcome Measure:** Change from baseline to 1 hour assessment in BHB levels.
- **Aim 1 Analytic Approach:** We will compare mean baseline serum BHB levels to mean BHB levels measured at the 60 minute assessment. We will use the Wilcoxon rank sum test (non-parametric equivalent of standard two sample t test) to evaluate our Aim 1 hypothesis. We will have values available at 2-hours and 4-hours as well to test the duration of the effect. We will also collect serum glucose values at each of the four time points (baseline, 1 hours, 2 hours, and 4 hours).

Aim 2: To evaluate the tolerability of the Modified Curtis Protocol in PD.

- **Hypothesis 2:** Participants will not experience worsening in Movement Disorder Society Unified Parkinson Disease Rating Scale part III ($MDS\text{-}UPDRS_{III}$) scores.
- **Aim 2 Outcome Measure:** Change from baseline to 1 hour in $MDS\text{-}UPDRS_{III}$ scores.
- **Aim 2 Analytic Approach:** Similarly to Aim 1, we will compare baseline $MDS\text{-}UPDRS_{III}$ scores to those obtained at the 60-minute assessment using the Wilcoxon rank sum test. Similar to Aim 1, we will also have values available at 2-hours and 4-hours as well to test the duration of the effect. We will collect additional clinical measures and measures of tolerability including timed up a go test (TUG), finger tapping test (FTT), mini Addenbrooke's Cognitive Examination (mini-ACE), gastrointestinal symptom rating scale (GSRS), and adverse event report.

Sample Size

For the purposes of calculating a sample size, we used parametric testing by means of a paired sample t-test. Please note that to reduce the risk of type 1 error due to outliers in our dataset, we propose to use analogous non-parametric testing in our final test of Aim 1. Based on previously studies looking at change in BHB after consumption of this ketone ester, we expect an increase in 0.5 mmol [4]. This would also be the minimum change needed to achieve ketosis. Using a two-side alpha of 0.05, a power of 0.80, and a proposed standard deviation of 0.5 mmol in BHB serum levels, a sample size of 10 participants will be sufficient to test our Aim 1 hypothesis.

Schedule of Activities

Evaluation	Screen and Informed Consent	In Person Study Visit	Telephone Follow Up Visit
Setting	In-Person/Phone	In-person	Phone
Discuss Study Overview	x		
Assess for Eligibility	x		
Informed Consent	x		
Modified Curtis Protocol		x	

MDS-UPDRS$_{I, II, IV}$ (Baseline)		x	
MDS-UPDRS$_{III}$ (Baseline, 1, 2, and 4 hours)		x	
Mini-ACE (Baseline, 2, and 4 hours)		x	
TUG (Baseline, 1, 2, and 4 hours)		x	
FTT (Baseline, 1, 2, and 4 hours)		x	
Serum ketones and glucose (Baseline, 1, 2, and 4 hours) via Keto Mojo Meter		x	
GSRS (Baseline, 1, 2, and 4 hours)		x	x
Report of side effects (1, 2, and 4 hours)		x	
Follow Up Report of Potential Side Effects			x

Participants screening and informed consent will occur per the original protocol document and be introduced as an optional sub-study. This will all occur in advance of the in-person study visit and prior to any clinical testing. The in-person study visit will occur in the lab of Dr. Bohnen at Domino Farms. This is the primary site for PD human subjects research visits in our UM Movement Disorders Neurology group. Participants will be asked to withhold their usual parkinsonian medications (dopaminergic medications or amantadine) the morning of the in-person study visit and are to bring them to the study visit in order to take them immediately after completion of the day's activities. This is requested in order to prevent a medication induced fluctuation in motor score from impacting the repeated MDS-UPDRS$_{III}$ clinical assessments. They will also be required to arrive in the fasted state per the Modified Curtis Protocol. They will then undergo baseline testing including the MDS-UPDRS$_{I-IV}$, the Mini-ACE test, the TUG, the FTT, the GSRS, and blood testing. The order of these test may vary depending on the participant and visit context. This is collectively estimated to take 30-60 minutes from arrival.

The next hour will consist of the Modified Curtis Protocol at which time participants will first consume the fat and coffee supplement. Thirty minutes later, they will consume the KE. Fifteen minutes later they will begin 5 minutes of stretching in preparation for 10 minutes of low intensity exercise. The exercise component will be on a stationary bicycle and will be interval training type exercise but at low intensity given possible deconditioned state of participants. It will start with 2 minutes of cycling with minimal added resistance while increasing cadence from 30 to 60 rotations per minute (rpm). There will then be two cycles of 30 seconds of modestly increased resistance with a cadence of 80 rpm followed by 2.5 minutes of minimal resistance at a cadence of 70 rpm. This will be followed by a cool down still at minimal resistance decreasing cadence from 60 to 30 rpm.

Sixty minutes after consumption of KE, or 30 minutes after completion of the Modified Curtis Protocol, participants will undergo repeat testing including the MDS-UPDRS$_{III}$, TUG, FTT, GSRS, a side effect assessment, and serum glucose/ketone body lab testing via Keto-Mojo. This same testing plus the Mini-ACE will also recur after 120 minutes (2 hour) and 240 minutes (4 hour) from the time of time the KE drink was finished. After the 4-hour testing is complete,

participants will be allowed to take their held medications, can resume their normal diet, and will be dismissed for the day.

Our study team will follow up with participants by phone in the 48 hours following the in-person Study Visit to ask about potential side effects and to repeat the GSRS as well as ask the closing question which is a Likert scale survey question regarding likeliness that the participant feels they could repeat this protocol on a daily basis. Once the follow-up phone call ends, the sub-study will be complete.

Visit 3 in the original study document consists of psychiatric clinical testing as well as optional motor and cognitive testing. Participants who opt to take part in this sub-study will be exempt from completing the original planned assessments for this day as per the Schedule of Activities (SoA) (besides the Beck Depression Inventory II and Spielberger Trait Anxiety scale which will be completed by all participants at visit 3). The administration of the ketone product and blood lab testing for this visit would be per the sub-study protocol as opposed to the original protocol. Participants would still receive the sleep tracking ring, pregnancy testing (if applicable), and drug accountability before starting the original intervention protocol.

For individuals who cannot tolerate the Curtis protocol intervention, study staff will send home the participants without the KetoneAid supplement and call them within 48 hours to monitor any side effects from testing. Participants may be asked to come back in to the lab to take the KE supplement without any additional testing to assess tolerability of the ketone ester drink alone.

Modified Curtis Protocol Components:

Ketone Ester:
Please see original document for additional information regarding the exogenous ketone supplement by the brand KetoneAid (D-β-Hydroxybutyrate-(R)-1,3 Butanediol). There will be no change to the dose of D-β-Hydroxybutyrate-(R)-1,3 Butanediol which, per the original protocol, is a single 12.5 g dose.

Fat and Coffee Supplement:
The original protocol uses a combination of coffee, grass-fed butter and cream, and coconut oil. Coffee has been suggested to have multiple benefits in PD [6]. Coconut oil is a source of medium chain triglycerides which serve as precursors to ketone bodies. Lastly, grass fed dairy is a source of low carbohydrate and high fat calories which are lower in unsaturated fat. Ingredients for the chosen supplement include coconut oil powder, grass fed butter powder, instant organic coffee powder, medium chain triglyceride oil powder, natural coffee flavor, and real salt. The supplement is called Keto Coffee and made by the brand Fat Fuel. One serving is 234 calories and contains 120 mg of caffeine.

3. Enrollment Criteria

Inclusion criteria:

1. Diagnosis of PD (M/F, age 45 years and over) will be based on the United Kingdom Parkinson's Disease Society Brain Bank Diagnostic Research Criteria (patients with clinically significant

dementia will be excluded, i.e., fails informed assent screening criteria and has significant evidence of difficulty with iADL functions, such as ability to take own medications while administering the pre-consent questionnaire).
2. Persons diagnosed with Lewy Body Dementia (LBD) based on the Fourth consensus report of the Dementia with Lewy Body (DLB) Consortium inclusion criteria for probably DLB or Parkinson's Disease Dementia (PDD) based on the criteria set by Emre *et al.* for PDD (M/F, age 45 years and over)

Exclusion criteria:
1. All exclusion criteria from original protocol applies to sub-study
2. Significant metabolic or uncontrolled medical condition, including significant cardiovascular disease or significant musculoskeletal disorder which would make it unsafe to participate in cardiovascular exercise
3. Lactose intolerance

4. Potential Risks

Please see original document for risks associated with confidentiality of research information, clinical testing, motor testing, neuropsychological testing, and the ketone ester supplement.

Exercise:
Common risks of cardiovascular exercise include fatigue, general discomfort, temporary muscle soreness and pain. Infrequent risks include muscle strain and rare events include musculoskeletal injury or cardiovascular event. Risks to participants have been minimized by limiting the duration of exercise and by limiting participation of those with higher risk of these events through exclusion criteria. Moreover, participants will be guided through gentle stretching prior to initiating exercise to minimize the risk of injury. The HIIT intervals have been intentionally designed with a gentle level of intensity to ensure tolerability for PwP. HIIT is a type of cardiovascular exercise which was shown to have no difference in adverse events when compared to moderate intensity exercise in middle age and older adults with diabetes. [7]

Fat and Coffee Supplement:
Common risks with consumption of this coffee supplement primarily include possibility of poor taste and worsening of tremor. Infrequent side effects of caffeine include anxiety, palpitations, headaches, dizziness, and insomnia. Infrequent side effects of this supplement include stomach discomfort, nausea, bloating, and diarrhea. These risks are limited by limiting the participants to a single serving in the morning which consists of 120 mg of coffee which is equivalent to a standard cup of coffee.

5. Data Study Monitoring Plan:

Drs. Bohnen and Albin will act as the Data Safety Monitoring Plan members for this study. All adverse events and other study related events will be discussed at monthly investigator meetings. Adverse events will be classified according to standard criteria and reported to the IRBMED (relevant University of Michigan IRB) according to standard guidelines. Serious adverse events will be reported within 24 hours.

6. References

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