# PROTOCOL TITLE:

*Physiologic determinants of PPI disposition in children*

# PRINCIPAL INVESTIGATOR:

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*V20.0_12.27.22*

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## 1.0 Study Summary

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|---|---|
| **Study Title** | *Physiologic determinants of PPI disposition in children* |
| **Study Design** | Prospective, single center, comparative pharmacokinetic study |
| **Primary Objective** | Assess impact of liver adiposity on PPI pharmacokinetics |
| **Secondary Objective(s)** | Assess impact of genotype, age, inflammation, obesity, and resting energy expenditure on PPI pharmacokinetics (PK) and pharmacodynamics (PD) in children, as well midazolam PK if receiving midazolam for pH probe placement |
| **Research Intervention(s)/ Investigational Agent(s)** | Peroral (PO) or Intravenous (IV) PPI administration, PK sampling, magnetic resonance imaging (MRI), indirect calorimetry via hand-held device and optional gastric pH Probe Monitoring (with or without IV Midazolam administration for pH probe placement) |
| **IND/IDE #** | NA |
| **Study Population** | Children and adults (6-21years) |
| **Sample Size** | n=300 |
| **Study Duration for Individual Participants** | Up to 31 days, depending on whether visits are combined |
| **Study Specific Abbreviations/ Definitions** | PCRU-Pediatric Clinical Research Unit<br>GERD-gastroesophageal reflux disease<br>NAFLD-non-alcoholic fatty liver disease<br>MRI-magnetic resonance imaging<br>PO-per oral<br>PPI-proton pump inhibitor<br>PK-pharmacokinetics<br>PD-pharmacodynamics<br>CYP-cytochrome P450<br>DME-drug metabolizing enzyme<br>PAN-pantoprazole<br>LAN-lansoprazole<br>MIDAZ-midazolam<br>BMI-body mass index<br>FFM-fat free mass<br>$AUC_{0\text{-}inf}$-total area under the plasma concentration-time curve<br>CL/F-apparent oral drug clearance<br>Vd/F-apparent volume of distribution<br>$C_{max}$-peak concentration<br>$T_{max}$-time to peak concentration<br>$t_{1/2}$-half life |

## 2.0 Objectives

**The objective** of this investigation is to identify the biophysiologic mechanisms that contribute to our previously-observed differences in pantoprazole pharmacokinetics in obese children[1], and to determine whether obesity influences the pharmacokinetics (PK) and pharmacodynamics (PD) of proton pump inhibitors.

Utilizing proton pump inhibitors (PPIs) pantoprazole and lansoprazole, as drug substrates to assess hepatic drug metabolizing enzyme (DME) CYP2C19 and CYP3A4 activity, I will test the hypothesis that three obesity-related changes in physiology (e.g., hepatic fat infiltration, low-grade inflammation and disease-associated changes in basal metabolic rate) drive variability in DME activity, and therefore PPI exposure and response, in obese children. I will test this central hypothesis by executing four Specific Aims (SA):

**SA1:** To test the hypothesis that liver adiposity affects systemic exposure to PPIs, I will use magnetic resonance imaging technique (LiverLab, Siemens) to quantify percent liver adiposity and its association with variability in PPI disposition, and DME activity, in children.

**SA2:** To test the hypothesis that chronic low-grade inflammation affects systemic exposure to PPIs, I will measure and compare traditional clinical markers of inflammation (e.g., ESR, CRP), recently developed biomarkers of inflammation (e.g., INF-γ, IL-1β, IL-17α, etc.), and plasma lipidomic profiles in obese and non-obese children, controlling for DME genotype.

**SA3:** To test the hypothesis that basal metabolic rate affects variability in DME activity, I will compare resting energy expenditure (REE) in obese and non-obese children who demonstrate variable systemic exposure to PPIs and DME-mediated PPI metabolites, controlling for DME genotype.

**SA4:** To test the hypothesis that changes in PPI pharmacokinetics affect PPI efficacy (pharmacodynamics), I will measure gastric acid pH using continuous pH probe monitoring after single dose PPI administration.

## 3.0 Background

In the United States, one in six children meets body mass index (BMI) criteria for obesity.[2] Children with obesity are more likely to suffer gastroesophageal reflux disease (GERD)[3], a condition increasingly recognized as chronic, and one that frequently requires long-term acid-suppression therapy with proton pump inhibitors (PPIs).[4] Neither clinical guidelines, nor labeling from the U.S. Food and Drug Administration (FDA), are available for dosing PPIs for children with obesity, as children with obesity are traditionally excluded from clinical trials, leaving prescribers guessing the initial drug dose and, often, increasing doses empirically, to compensate for increased body size.

In the only study of PPI pharmacokinetics in children with obesity, we recently demonstrated decreased apparent oral drug clearance (CL/F) for the PPI pantoprazole, compared to historical, pediatric or adult values, previously reported for individuals without obesity.[1] The observed decrease in pantoprazole CL/F translated to higher systemic drug exposures (area under the concentration-time curve ($AUC_{0\text{-}inf}$)) and peak drug concentration ($C_{max}$)) for children with obesity, for every milligram-per-kilogram of drug received, compared to historical pediatric controls without obesity. Growing concerns emerge regarding the long-term consequences of prolonged, systemic, PPI overexposure in children (e.g., pneumonia, enteric infections, vitamin/mineral deficiencies)[5], with certain adverse events (e.g., osteopenia, fractures) postulated to be specifically related to high drug $AUC_{0\text{-}inf}$ and/or $C_{max}$ over time.[6] Understanding of the biophysiologic mechanisms underlying the observed differences in PK in obese children is the first step toward

developing accurate clinical algorithms for optimizing and individualizing the dose selection of PPIs, and other medications, commonly prescribed to pediatric patients with obesity.

The majority of medications prescribed to children undergo biotransformation by polymorphically-expressed drug metabolizing enzymes (DMEs) that are members of the human hepatic cytochrome P450 (CYP) family.[7] Genetic polymorphisms in DMEs are established sources of inter-individual variability in drug dose – exposure,[7] as observed for PPIs, which serve as substrates for hepatic CYP2C19 and CYP3A4.[8]

PPI's have become the number 12 medication prescribed to children,[9] with >500,000 pediatric prescriptions filled in one year for the PPI lansoprazole.[10] In addition to genetic variation, recently published data from our group suggest that obesity *also* is an important determinant of PPI exposure in children.[1] Obesity is associated with visceral adiposity in metabolically-active tissues (e.g., liver),[11] alterations in organ blood flow,[12] and decreased lean-to-fat body mass ratio,[13] all of which may impact drug disposition, metabolism and exposure.[14] Obesity is also thought to represent a chronic state of low-grade inflammation,[11] which may down-regulate the expression[15,16] and function[16] of DMEs (e.g., CYP3A4), leading to slower drug clearance. The objective of this investigation is to identify the relevant biophysiologic mechanisms that contribute to previously-observed differences in pantoprazole PK in obese children[4], to assess whether these differences affect drug efficacy, and to determine whether these obesity-related alterations in pharmacology influence other PPIs (e.g., lansoprazole).

## 4.0 Study Endpoints

**Primary study endpoints:** PK parameters (e.g., $t_{1/2}$, $T_{max}$, $C_{max}$ , CL/F, $AUC_{0\text{-}inf}$, etc ) for pantoprazole (PAN) and lansoprazole (LAN), and their CYP2C19- and CYP3A4-dependant metabolites, in plasma and urine, and hepatic fat fraction (HFF) and Goodness-of-fit (Rsq) output generated by LiverLab (Seimens) magnetic resonance image sequencing.

**Secondary study endpoints:** Patient demographics (e.g., age, sex, BMI, etc), DME genotype, REE, inflammatory cytokines (e.g., INF-γ, IL-1β, IL-17α, etc.), gastric pH; PK of midazolam, if receiving midazolam for pH probe placement.

## 5.0 Study Intervention/Investigational Agent

**Study drug**: Commercially available, solid formulations of lansoprazole (Prilosec; 15mg and 30mg tablets), pantoprazole (Protonix; 20mg and 40mg tablets), IV pantoprazole (Protonix), and IV midazolam (Versed; optional for pH probe placement) will be utilized; stored and dispensed by IDS pharmacy.

- No IND required

**Study device:** 1) Resting energy expenditure (REE), a measure of the body's metabolic rate, will be measured by indirect calorimetry, using the FDA-approved handheld indirect calorimeter, MedGem® (Microlife, Inc., Golden, CO; Figure 1), validated for use in children ≥ 6 years of age.[17] Our group has extensive experience with using this device in other research investigations in children. 2) pH probe placed intra-nasally and advanced down the esophagus and into the stomach. Our team has extensive gastroenterology experience in placing these devices for routine clinical care in children.

**Figure 1.** Handheld calorimeter MedGem®

- No IDE required

# 6.0 Procedures Involved

6.1

This is a single center, comparative pharmacokinetic and pharmacodynamic study that includes individuals 6-21 years of age, with or without obesity, who may or may not have non-alcoholic fatty liver disease (NAFLD), and who are or are not receiving pantoprazole or lansoprazole as part of their routine medical care.

6.2

Screening visit: On a screening visit (which can be combined with the first PK study visit or can occur within 31 days of study drug administration), prospective volunteers will be provided with a description of the proposed study. After all their questions have been answered, they will be given a copy of the permission/assent form to review and sign. They will then have their medical history and use of medicines reviewed, and will be given a physical examination, including vital signs (blood pressure, heart rate, respirations, temperature, height and weight). Blood will be drawn by needle stick for serum chemistries (which can occur during screening visit or prior to drug dosage), liver function tests and a hematology panel, including complete blood count; a portion will also be retained for *CYP2C19* and CYP3A4 genotyping using the ADME gene panel available through CMH (approximately 6 ml of blood total). If screening visit in the AM, an additional approximately 4 ml of blood will be obtained for lipid panel, insulin level, and metabolomics analysis. If screening not in the AM, the metabolomics sample will be obtained with an AM PK blood draw on day of PK/PK-PD study visit because consistent timing of this sample in the AM is essential for scientific rigor.

A topical anesthetic will be used when appropriate, prior to the blood being drawn. A urine sample will be collected for urinalysis. For female participants a urine pregnancy test will be conducted.

PK visit: Subjects will be admitted to the study unit the morning after an overnight fast (water only permitted after midnight). REE will be determined using a conventional, FDA approved, handheld indirect calorimeter device (MedGem, Microlife, Inc., Golden, CO), which requires the subject to breathe through their mouth for approximately 10 minutes, while reclining flat.

A baseline (pre-dose) blood sample and urine sample will be obtained. A single IV dose of pantoprazole and/or a single oral dose of pantoprazole and/or lansoprazole will be administered as a tablet or combination of tablets, in accordance with the child's routine medical care (if already on pantoprazole or lansoprazole) or at 1.2 mg/kg fat free mass (FFM), calculated via a validated pediatric equation[18] and rounded to the nearest whole tablet.

$$\text{FFM (females)} = \left[1.11 + \left(\frac{(1-1.11)}{\left[1+\left(\frac{\text{Age}}{7.1}\right)^{-1.1}\right]}\right)\right] \times \left[\frac{(9270 \times \text{WT})}{8780 + (244 \times \text{BMI})}\right]$$

$$\text{FFM (males)} = \left[0.88 + \left(\frac{(1-0.88)}{\left[1+\left(\frac{\text{Age}}{13.4}\right)^{-12.7}\right]}\right)\right] \times \left[\frac{(9270 \times \text{WT})}{6680 + (216 \times \text{BMI})}\right]$$

Total dose administered will not exceed 80mg for pantoprazole or 60mg for lansoprazole.

For the pharmacokinetic PPI study, up to 12 blood samples (of up to 2 ml each) will be collected after ingestion or dosing of study drug. Blood samples will be obtained via syringe from an indwelling venous catheter (IV) and placed into blood collection tubes containing sodium heparin, immediately mixed by inversion and centrifuged (1,000 g for 10 minutes at 4ºC). Plasma will be collected by manual aspiration and immediately stored at -70ºC until analysis.

Subjects will be provided with a meal or meal replacement shake (e.g., Premier Protein, Boost Max, Ensure Max) 2 hours after PPI study drug administration and at mealtimes throughout the study visit.

All urine produced over the study period will be collected and a 40-ml aliquot stored at -20ºC until analysis. PAN, LAN, and their primary and secondary metabolites will be quantitated from the urine samples, using the bioanalytical method applied to plasma. Briefly, concentrations of PAN, LAN, and metabolites will be quantified by high performance liquid chromatography with ultraviolet detection (HPLC-UV) as previously described.[20,21] Internal standard (50 g/ml phenacetin) is added to 100 µl of plasma, and the sample deproteinized with 200 µl of acetonitrile. After centrifugation at 3000*g* for 10 min, the supernatant is evaporated to dryness, the residue is reconstituted in 50 µl of 50 mM sodium perchlorate and acetonitrile [80:20 (v/v)] (solvent A), and a 25 µl volume is injected into an Agilent high-performance liquid chromatography system. Separation is achieved using a Chiralcel OJ column (5.0 x 150 mm, 5 µm; Chiral Technologies, Inc., Exton, PA), and a mobile phase delivered by a gradient pump. Column effluent is monitored with the UV detector set at 290 nm. Liquid chromatography with tandem mass spectrometry (LC/MS/MS) methods may also be used.

For those study participants who volunteer for the PK/PK-PD study of PO and/or IV pantoprazole and PO lansoprazole, study drug administration will be within 7-31 days of each other. An update of medical history and physical exam will be performed prior to dosing. A separate screening visit (i.e., vitals, physical exam, safety labs) will be performed for the second and/or third PK/PK-PD study visits, if it falls outside the 31 days window.

PK-PD visit: Subjects will be admitted to the study unit fasting at least 2hrs (water permitted during fast). All other study procedures will be the same as the PK visit except: 1) participants will also have a pH Probe placed prior to PPI study drug administration; and 2) up to 13 PPI PK samples will be drawn, such that a PK sample can be drawn at the time of pH probe removal. To minimize discomfort of pH probe placement, participants will be offered a one-time dose of IV midazolam (0.05mg/kg up to 2mg)) to assist with probe placement. For those subjects who elect to have a one-time dose of IV midazolam (MIDAZ), concentrations of MIDAZ and its relevant metabolites will be measured in blood and urine samples that are already being collected for PPI PK analysis. An additional MIDAZ PK blood sample (up to 2ml) will be drawn every 10-15 minutes after MIDAZ administration and before PPI dosing, during pH probe placement and calibration (approx. 30-45 min; up to 4 PK samples).

Continuous pH probe monitoring: A pH probe, lubricated at the tip, will be introduced into the left or right nostril and advanced into the esophagus and down into the stomach, with placement distance calculated using the formula provided below. Briefly, for placement in the stomach fundus, the probe is advanced 10cm beyond the lower esophageal sphincter (LES).[29] LES placement is calculated using the standard Nowak's formula routinely used in pediatric gastroenterology (3.2 + 0.2 x height in cm).[28] Thus, the gastric placement formula is:

**Gastric pH probe placement = 13.2 + 0.2 x height (in cm)**

Intragastric placement will be confirmed by pH <4 for 5 consecutive minutes, after which participants will receive PPI study drug administration. To avoid undue fasting, while minimizing food interference with the pH probe sensor, participants will be offered a meal replacement shake 4 hours after PPI study drug administration. The pH probe will remain in place for up to 12 hours or overnight, with participants fasting (water permitted). The probe will be removed after collection of the last PK sample. Once the pH probe is removed, the participants will be offered a meal of their choosing and be discharged from the PCRU.

MRI and/or Ultrasound Visit: All participants may undergo non-radioactive magnetic resonance (MR) imaging of the liver, using LiverLab (Siemens), in accordance with standard radiologic safety practices, overseen by the PI and radiology collaborators. LiverLab technology offers a comprehensive magnetic resonance (MR) imaging package for liver fat evaluation, without the need for liver biopsy.[19] It consists of three MR sequences that require brief breath-holding (up to 15 seconds) to generate whole-liver imaging, iron content quantification (R2*) and hepatic fat fraction (HFF), as well as a goodness-of-fit indicative of residual error for HFF and R2*. Goodness-of-fit (Rsq) ≤5% is considered reliable. The imaging sequence takes approx. 45 minutes to perform, does not require sedation or IV contrast, and is non-radioactive. The primary outcome measures generated, HFF and Rsq (Figure 2), will be correlated with PPI PK parameters of interest (e.g., $AUC_{tot}$, CL/F, $V_d$).

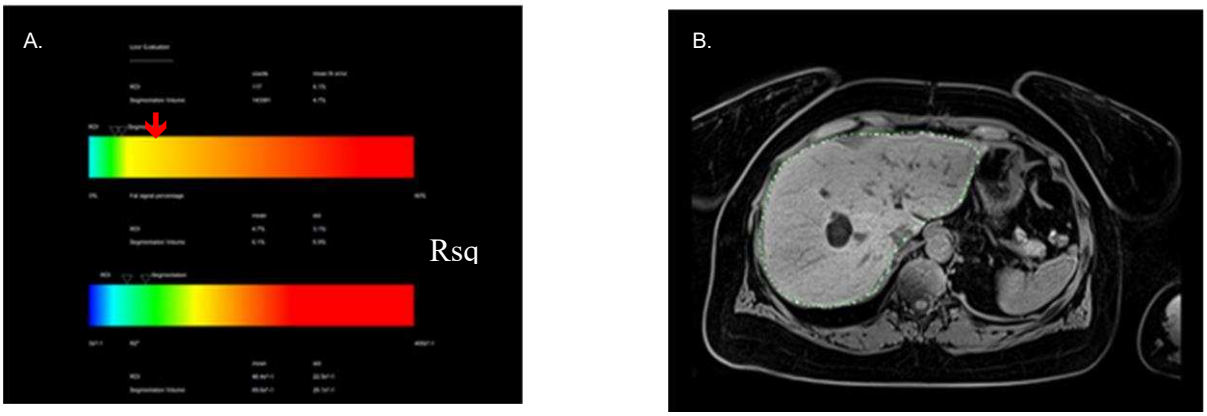



**Figure 2.** LiverLab output provides hepatic fat fraction (arrow) and goodness-of-fit (Rsq), along the fat signal percentage bar (A), and whole organ imaging (B)

All participants may also volunteer to undergo Ultrasound-Guided Attenuation Parameter (UGAP) Liver Steatosis Grading. UGAP imaging is designed to quantify hepatic steatosis using attenuation parameter methods in just 5-10 minutes. UGAP (LOGIQ™E10 Series, General Electric Company) measures fat by quantifying the amount of energy decrease (i.e., attenuation) in the ultrasound beam throughout the selected region of interest (ROI). UGAP output (Figure 3) includes both a continuous measure of attenuation and a categorical gradation of steatosis (grade 1, 2 and 3), along with a measure of consistency in imaging to ensure high quality data (termed IQR/Med). IQR/Med

<30% is considered good. Compared to MRI, ultrasound-based techniques are generally more accessible, more readily-available, and more cost-effective, making UGAP an ideal future imaging biomarker for clinical practice.[33,34]


Shear Elasto / UGAP

| | Stiffness | Velocity | Attenuation Coefficient | B. Attenuation Rate |
|---|---|---|---|---|
| | Site1 | Site1 | Site1 | SEGMENT 5 |
| Med | | | 0.47 dB/cm/MHz | 183.10 dB/m |
| IQR | | | 0.13 dB/cm/MHz | 22.78 dB/m |
| IQR/Med | | | 27.1 % | 12.4 % |
| N | | | 11 | 11 |

| C. | Grade 1 | Grade 2 | Grade 3 |
|---|---|---|---|
| AUROC (95% CI) | 0.900 (0.834 – 0.967) | 0.950 (0.894 – 0.993) | 0.959 (0.920 – 0.999) |
| Attenuation coefficient cutoff value (dB/cm/MHz) | 0.53 | 0.60 | 0.65 |
| Attenuation rate cutoff value (dB/m) | 186 | 210 | 228 |

**Figure 3.** UGAP imaging output provides (A) attenuation in the region of interest (yellow); (B) continuous attenuation measures with imaging consistency assessment (IQR/Med; blue); and (C) conversion of attenuation parameters into a categorical grade of hepatic steatosis. Concomitant ultrasound elastography sequencing ensures there is no liver fibrosis.

Unless imaging results within 31 days of PK visit are already available in the EMR, hepatic fat will be measured with MRI and/or UGAP, within 30 days of PK visit. Participants who already have hepatic fat quantification available via one of the imaging modalities (MRI or UGAP) may be asked to have the other imaging modality performed for the study purposes within 30 days of original hepatic fat imaging.

UGAP ultrasound is being used for what it is approved for in the FDA. We are using the FDA approved labeling but doing a comparison with MRI and drug clearance in kids, which has not been studied.

Subjects may choose to participate in one, two or three of the PK visits. Subjects may be enrolled in the study more than once if there have been significant changes to their BMI or medical history.

The aforementioned data will be obtained and recorded in the case report forms for this study. UGAP and MRI data, including: Patient Name, Medical Record Number and Date of Service will be shared with the UGAP study (STUDY00002487).

## 7. Data and Specimen Banking

Biological specimens (plasma, urine and blood) will be obtained for the purpose of fulfilling the scientific goals of the study. Plasma and urine samples will have the concentration of PAN, LAN, MIDAZ and their major and minor metabolites measured in them (standard HPLC-UV or MS/MS methodologies). One blood sample will be used to isolate DNA. This DNA specimen will be used to determine the genotype of enzymes and/or transporters known, or believed, to be important to understanding the pharmacokinetics (disposition) of PPIs (ADME gene panel). All samples will be labeled with a study identifier, the date of collection, and no PHI. Once all participants have been enrolled in the study and their data analyzed, the aforementioned specimens will be stored for a period of time not to exceed 5 years from the publication of the last manuscript describing the results from the study. The purpose for their retention would be to enable a re-analysis of the specimen for the intended purposes described in the protocol should the experimental data indicate

that this is necessary. The specimens will not be used for scientific purposes other than those related to this investigation, unless subjects consent to having their left over, deidentified biospecimens used for other future research.

If all study procedures and sample collections have been completed, subjects who turn 18 years of age during the lifetime of the study will not be re-contacted to obtain consent (Waiver of Consent at Age 18 requested). Subjects who turn 18 between study visits, will be re-consented using the adult informed consent form.

## 8. Genetic Analysis Information

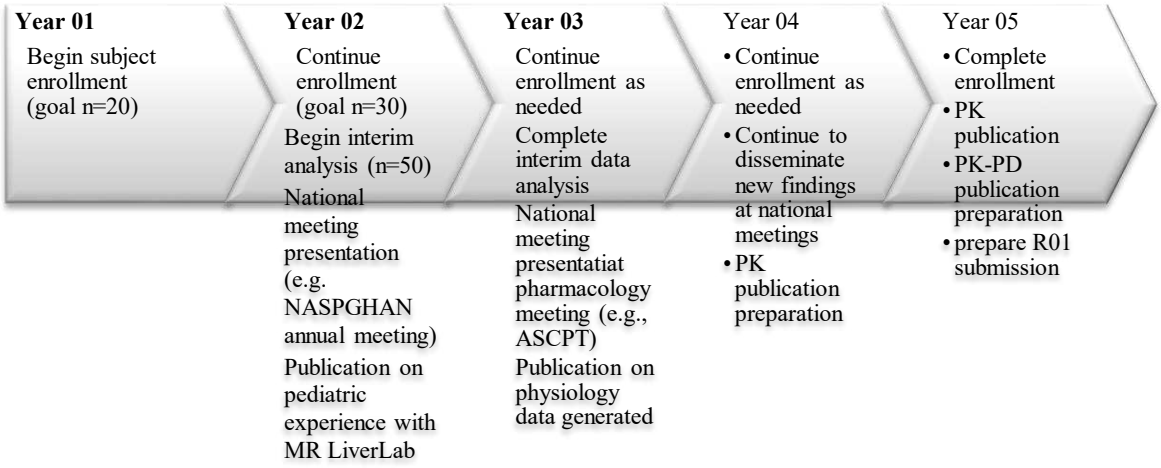
Sequencing of genes important to drug **a**bsorption, **d**isposition, **m**etabolism, and **e**limination will be performed using the ADME gene panel (CLIA-approved lab; CMH Genome Center) and/or may be confirmed by single nucleotide polymorphism analysis (SNPs; non-CLIA approved lab).

Genetic analysis will be restricted to gene analysis of ADME genes; thus, incidental findings pertaining to paternity, disease, pathologic conditions, etc. are not anticipated.

## 9. Sharing of Results with Subjects

Study results other than screening laboratories, REE and MRI findings will not be shared with research subjects because analyses and interpretation of genetic, pharmacologic and other physiologic information generated by this investigation will be batched, to reduce research cost and expedite efficiency.

## 10. Study Timeline




## 11. Inclusion and Exclusion Criteria

Inclusion Criteria

- 6-21 years of age
- Obese and non-obese individuals
  - BMI ≥10$^{th}$ percentile for age (6-20 years of age)
  - BMI ≥18.5 (>20 years of age)
- Otherwise healthy; or otherwise healthy with diagnosis of GERD, NAFLD, chronic abdominal pain or obesity, according to report of medical history and/or review of the medical record

- Receiving or not receiving pantoprazole or lansoprazole for routine medical care
- Clearance for MRI by Hoop Test or by Radiology

Exclusion Criteria

- Unable or unwilling to give written permission/assent/consent
- Treatment with PPIs (e.g. pantoprazole, lansoprazole, omeprazole, esomeprazole, dexlansoprazole) in the last 3 days
- **For PO Study Drug:** Any anatomic abnormality of the GI tract as defined by history, PE, or radiographic findings, including Bariatric surgery, Nissen fundoplication or equivalent surgery.
- **For IV Study Drug:** Any anatomic abnormality of the GI tract as defined by history, PE, or radiographic findings, except Bariatric surgery, Nissen fundoplication or equivalent surgery.
- **For pH Probe Study:** unable to abstain from acid suppressive medications (e.g., PPIs, H2 Blockers, tums, etc) for at least 3 days prior to study
- **For subjects not undergoing weight management**, treatment in the last 3 days with medications known to clinically significantly inhibit (e.g., fluoxetine, fluvoxamine, ketoconazole, ticlopidine, felbamate, trazodone, valproic acid, topiramate) or induce (e.g., phenobarbital, carbamazepine, phenytoin) CYP2C19, and those known at therapeutic doses to significantly inhibit (e.g., erythromycin, clarithromycin, grapefruit juice, verapamil, diltiazem, cimetidine, ketoconazole) or induce (e.g., oxcarbazepine, carbamazepine, phenytoin, phenobarbital, St. John's Wort, rifampin, rifapentine). CYP3A4 activity.
- Unable to have blood drawn for the screening lab tests
- Unable or unwilling to fast prior to the study session
- Metal in the body or any foreign bodies that precludes MRI sequencing
- Claustrophobia
- Exceeds 500lbs or 227 kg in Body Weight
- Demonstrated adverse reaction to previous pantoprazole or PPI exposure
- For optional MIDAZ administration only, demonstrated adverse reaction to previous midazolam exposure
- Impaired hepatic activity as determined by routine liver function testing and defined as values ≥ 5 times the age-specific upper limit of normal (ULN) for AST, ALT, total bilirubin >2.0mg/dl, alkaline phosphaste ≥ 5 times the age-specific ULN
- Impaired renal function defined as creatinine ≥ 3 times the age-specific ULN
- Females of child-bearing age who are pregnant or breast-feeding
- Any known infection with hepatitis B, C, or human immunodeficiency virus (HIV)

Individuals who are not yet adults will be included in the study (i.e., children and adolescents). The following will not be included:

- *Adults unable to consent*
- *Infants*

- *Pregnant women*
- *Prisoners*
- *Wards of the state*

## 12.Vulnerable Populations

This study involves vulnerable subjects (i.e., children and adolescents) and is compliant with HRP-416-Checklist. As this study involves many back-up research personnel (e.g., research coordinators), children of research study members may be eligible to participate, but permission/assent will be obtained by study personnel not related to the child.

## 13.Local Number of Subjects

This is a single-center study; thus, all subjects will be enrolled locally. Up to 300 female and male children, adolescents, and adults (6-21 years) will be enrolled in the study. Subjects may choose to participate in one, two or three of the PK visits. Subjects may be enrolled in the study more than once, if there have been significant changes to their BMI (e.g., weight loss) or medical history (e.g., development of NAFLD, bariatric surgery), at the discretion of the PI.

## 14.Screening and Recruitment Methods

Subjects will be recruited from the Gastroenterology clinic at Children's Mercy Kansas City. Subjects will also be recruited from a pool of previous research participants, in the Divisions of Gastroenterology, Hepatology and Nutrition or Clinical Pharmacology, Toxicology and Therapeutic Innovation (the PI's home divisions), who have opted in to be contacted for future research opportunities. A notice will also be publicly displayed in the waiting areas of outpatient clinics at CMH, Center for Children's Healthy Lifestyles & Nutrition and posted on the Scope. This notice may also be emailed to staff members in the Divisions of Gastroenterology, Hepatology and Nutrition or Clinical Pharmacology, Toxicology and Therapeutic Innovation to remind them about this study opportunity throughout the lifetime of the study. All study related procedures will be conducted in the PCRU, except for MR LiverLab, which will be conducted in the MRI suite at Children's Mercy Kansas City.

PHI for potential subjects will be recorded in a subject pre-screening log, maintained on designated parts of the CMH server that are password protected and restricted to the use by members of the study team. Upon completion of study enrollment, PHI on non-participants will be destroyed.

## 15. Reimbursement, Payment and Tangible Property provided to subjects

Given the time commitment involved in this study, study compensation by Greenphire/ClinCard will be provided to study participants as follows:

- For completion of the screening visit - $25.00
- For completion of PK study IV PAN - $100.00
- For completion of PK study IV MIDAZ - $100.00
- For completion of PO Pantoprazole or Lansoprazole study - $100.00
- For completion of PH Probe study $250.00

- For completion of MRI - $50.00
- For completion of UGAP Ultrasound - $50.00

Social Security Number (SSN) or Individual Tax Identification Number (ITIN) will be collected to make these payments and this is stipulated in the research permission/assent/consent forms. Subjects may choose to complete one or more study visits.

**15.2 Reimbursement:** PK-PD 2-day consecutive visits that require the family to travel a distance of over 15 miles from their home to the hospital will be given the option of a one night CMH approved hotel stay. Hotel reservation will be initiated by the study coordinator and booked by CMH's Participant Engagement & Research Operations Project Manager and paid by the hospital with study funds. No reimbursement funds will need to be made directly to the participant or family if this option is used. This study will not cover any additional fees, charges or expenses made to the hotel room outside the cost of a one-night stay.

## 16. Withdrawal of Subjects

This study is voluntary, and subjects can withdraw from the study at any time. If a subject withdraws from the study within the first three years, information and samples collected during the study before withdrawal will still be used, unless the subject request that they be destroyed. No new study samples or information will be collected. The investigator(s) or the subjects' doctor may remove a subject from the study, at any time, if it is not in the subject's best medical interests to continue the study or if the subject fails to follow study directions.

## 17. Risks to Subjects

Potential risks to subjects are related to, (1) phlebotomy, (2) blood sampling, (3) PPI administration, (4) MRI, (5) loss of confidentiality, (6) pH probe placement and (7) midazolam administration

(1) Risks of *phlebotomy* include of feeling faint, discomfort and/or bleeding in the skin with bruising in the area where the blood sample was taken and a very small risk of infection at the site of skin puncture. These are not considered serious risks. To minimize the risk of infection, correct aseptic (germ free) techniques will be used whenever blood is drawn. To minimize discomfort, blood will be drawn by experienced technicians and topical numbing cream will be offered.

(2) For *blood sampling*, an indwelling venous catheter will be placed in the forearm of each subject. The risks of catheter placement are similar to those of phlebotomy. Blood samples will be obtained via syringe from the indwelling catheter and placed into tubes containing sodium heparin. For the pharmacokinetic study, blood samples (2 ml) will be collected prior to, and at 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 6, and 8 hours after ingestion of study PPI drug, and at the time of pH probe removal for those participating in the PK-PD study. Thus, the total blood volume drawn for the PPI pharmacokinetics is 38 ml (approximately 2.5 tablespoonfuls). Up to 4 additional samples (approx. ½ tablespoonful) may be drawn for those subjects who elect to have a one-time dose of MIDAZ for the PK-PD study.

(3) *Pantoprazole* and *lansoprazole* are PPIs found in remedies for GERD, erosive esophagitis and pathological acid hypersecretory conditions, available by prescription

and over the counter. In pediatric and adult patients, the most commonly reported (> 4%) adverse reactions include: upper respiratory infection, headache, fever, diarrhea, vomiting, rash, and abdominal pain. In this study, a single weight-based dose of 1.2 mg/kg FFM will be administered.

The current recommended pantoprazole dose for children 5 years and older, ≥ 15 kg to < 40 kg body weight, is 20 mg and ≥ 40 kg body weight is 40 mg, once daily for up to 8 weeks for GERD treatment. Pharmacokinetic studies conducted in children have used varying pantoprazole doses ranging from approximately 0.6 mg/kg to 1.2 mg/kg of pantoprazole granules in a study of in neonates and preterm infants,[22] fixed doses of 20 mg and 40 mg corresponding to approximately 0.6 to 1.1 mg/kg in children and adolescents aged 6 through 16 years,[23] and single oral doses of 20 or 40 mg (corresponding to 0.8 mg/kg) or intravenous doses of 0.8 mg/kg and 1.6 mg/kg[24]. Published studies of the pantoprazole in adults have utilized a fixed 100 mg dose of racemic [$^{13}$C]-pantoprazole, corresponding to average doses of 1.4 mg/kg to 1.6 mg/kg of racemic pantoprazole to subjects with average body weights between 60-65 kg.[25,26]

The current recommended lansoprazole dose for children 1 years and older, ≥ 15 kg to < 30 kg body weight, is 30 mg and ≥ 30 kg body weight is 30 mg, once daily for up to 12 weeks for GERD treatment. Pharmacokinetic studies conducted in children have used varying pantoprazole doses ranging from approximately 0.6 mg/kg to 1.3 mg/kg and fixed doses of 15 mg and 60 mg in children aged 1 through 11 years.[27]

It has been noted that almost all metabolic functions of the body, including drug clearance, take place within lean tissues. To avoid giving excessively large doses of drug to subjects who are obese, due to their increased proportion of body fat, administered doses will be based on fat free mass.[18] PPIs have a wide therapeutic index and a single dose of 1.2 mg/kg FFM is not expected to be associated with any form of toxicity.

(4) *Magnetic Resonance Imaging* (MRI) is a safe and commonly employed imaging method in pediatric and adult medicine, as it does not expose patients to radiation, is non-invasive, and does not require sedation or IV contrast for the study-specific use of MR LiverLab. Every precaution will be taken to ensure that all metal from the body is removed prior to MRI procedure and those subjects with implanted metal devices that cannot be easily removed, and are not compatible with MR imaging of the abdomen, will be excluded from the study. When performed properly, by experienced radiology staff, as in the context of this investigation, the risks of MRI are minimal and include discomfort, fear or claustrophobia associated with being confined to the MRI cylinder. All subjects will be screened for history of claustrophobia prior to study participation. MR Liverlab sequencing is brief (approx. 45 min) and all efforts will be made to minimize subject discomfort during imaging, including the use of virtual reality goggles and other distraction methods routinely employed in pediatric radiology.

(5) *UGAP Imaging* is a safe US-based imaging technique that takes approx. 5-10 min to perform. No added risk to patients is anticipated.

(6) *Loss of confidentiality*: Efforts will be made to keep all personal information confidential. All data will be stored with numerical identification and files will be kept in locked offices and/or pass-word protected internal server, to which only select members of the study team will have access. Personal identity will be protected in any publication.

(7) *pH probe placement* is a minimally invasive procedure, similar to nasogastric tube placement. The procedure is very safe when performed by trained and experienced

staff, such as the PI and her team. The PI is a pediatric gastroenterologist, the primary research coordinator is a registered nurse with 7 years of clinical experience in pediatric gastroenterology procedures at CMH, and the study team is well-versed and familiar with pH probe placement and monitoring procedures in children. Procedural complications from pH probe placement and monitoring are rare. Most common complications are self-limited and include nose/throat discomfort and/or nosebleed. To minimize these potential complications, the pH probe tip will be lubricated and participants offered a one-time dose of IV midazolam prior to probe placement, to minimize discomfort.

Participants will be closely monitored during probe placement and calibration to verify placement in the stomach. Distance for advancing the probe into the stomach will be calculated using a standard equation prior to probe placement. To confirm gastric placement, pH will be monitored and confirmed to be <4 (representative of the acidic environment of the stomach) for 5 minutes consecutively after probe placement. If gastric placement cannot be confirmed, the probe will be repositioned and recalibrated. In the highly unlikely event that the probe is advanced into the trachea, instead of the esophagus during placement, participants would develop immediate and obvious respiratory symptoms (e.g., cough) and the probe would be removed and repositioned.

Once the pH probe is placed successfully, the most common complication of continuous pH probe monitoring is accidental dislodging/displacement of the probe. To minimize this risk, the probe will be secured to the participant's cheek with medical tape.

(7) *Midazolam* is a short-acting benzodiazepine frequently used for its anti-anxiety and analgesia properties for short, medical procedures, such as IV starts and pH probe placement.[30] It is the most commonly used benzodiazepine in pediatrics[31] and routinely offered to children to mitigate the discomfort of nasogastric tube or pH probe placement for clinical purposes at our institution. So as not to exceed the maximum recommended dose by the U.S. Food and Drug Administration (FDA), midazolam dosing will be based on total body weight, 0.05 mg/kg up to a maximum one-time dose 2mg (lower side of the maximum recommended range of 2-4mg per dose for children).[30] In pediatric and adult patients, the most commonly reported acute adverse events associated with midazolam administration are airway compromise and hypoventilation. Most often, these occur when midazolam is co-administered with opioids or other anesthetic agents[30] (drugs that are not used in this study). The next most common adverse event is withdrawal syndrome, associated with long-term use of the drug[30] and, therefore, not anticipated from one-time dose administration for the purposes of this study.

In a recent study of one-time IV dose administration in overweight and obese children and adolescents, a patient population analogous to our study population, 2 and 3mg doses of IV midazolam were tolerated well.[32] The maximum one-time dose proposed for our study (2mg) is on the lower end of this dosing range. As an added precaution, all study subjects will be awake during midazolam administration and monitored by an experienced pediatric nurse with procedural experience, with continuous pulse oximetry monitoring available at the bedside, if needed.

**Investigator's Assessment of Risk:** (according to 45 CFR Part 46 Subpart D)

Category 1 [X] Research not involving greater than minimal risk.

Category 2 ☐ Research involving greater than minimal, risk but presenting the prospect of direct benefit to the individual subjects.

Category 3 ☐ Research involving greater than minimal risk and no prospect of direct benefit to the individual subjects, but likely to yield generalizable knowledge about the subject's disorder or condition.

Category 4 ☐ Research not otherwise approvable, which presents an opportunity to understand, prevent, or alleviate a serious problem affecting the health or welfare of children.

**Provide statement explaining your choice**:

MR LiverLab and UGAP imaging are a safe, non-invasive, non-radioactive modality that does not require sedation or IV contrast and, therefore, poses minimal risk. The PI's study team, including established collaborators in radiology, are committed to executing this study procedure safely and with minimal discomfort to the subjects.

GERD is a disease prevalent in normal weight, overweight and obese children and adults. Pantoprazole and lansoprazole are medications commonly used to treat GERD and related conditions in children and adults, approved for that use, and available over the counter. Some study participants will already be on one of these medications for clinical purposes and the same dose they receive for clinical purposes will be administered for study purposes. Furthermore, only a single dose of pantoprazole/lansoprazole will be administered to each subject for the purposes of this study. The risks associated with a one-time dose of PPI are minimal and comparable to placebo; therefore, the overall risk to subjects is minimal.
Continuous pH probe monitoring is routinely used to monitor signs/symptoms of GERD. The risks of pH probe placement are minimal and primarily related to discomfort/irritation in nose or throat, particularly during probe placement. To minimize discomfort/irritation from the pH probe, the probe tip will be lubricated prior to introducing it into the nostril. Participants will also be offered a one-time IV dose of midazolam to minimize discomfort.

Furthermore, subjects and their parents will be provided with contact numbers to enable them to contact an investigator 24 hours daily in the event of an adverse effect. All subjects will be carefully monitored for the development of adverse experiences throughout the course of each study and records of these events will be maintained. The reporting of adverse events will be in compliance with the Institutional Review Board's regulations. All staffs participating in human subjects' research at Children's Mercy Hospital are required to have successfully completed training and passed the certification for performing research in human subjects. The Clinical Research Unit at Children's Mercy Hospital has significant experience with pharmacokinetic studies in pediatric patients and is well equipped to carry out these studies. As well, the PI and her study team have recently completed two PK studies of pantoprazole in children and are, therefore, knowledgeable and familiar with PK studies of PPIs. For these reasons, we feel confident that this study will not place study subjects at undue risk.

## 18. Potential Benefits to Subjects

We do not anticipate that subjects will directly benefit from taking part in the study. The benefits to participation in the study are that participation may help investigators to determine how obesity affects the disposition of certain types of medication in obese/overweight children, and ultimately how the doses of medication should be adjusted in obese/overweight children to adequately treat disease and reduce the risk of side effects.

# 19.Data Management and Confidentiality

19.1 Statistical Analysis

For the pharmacokinetic studies, plasma PAN, LAN and MIDAZ concentration–time data will be analyzed using standard compartmental and noncompartmental methods. Peak drug concentration (Cmax), time to $C_{max}$ ($t_{max}$), area under the concentration–time curve from time zero to the time of the last measurable concentration ($C_T$) ($AUC_T$) and to infinity ($AUC_{tot}$), terminal-phase disposition half-life ($t_{1/2}$), apparent oral clearance (CL/F), and terminal-phase volume of distribution (Vz/F), where F is a bioavailability factor reflecting the fraction of the dose absorbed will be estimated using Kinetica software or comparable software. The disposition rate constant (λz) will be determined as the slope of a log-linear least squares of at least 3 concentration-time points judged, by visual inspection, to be in the apparent terminal elimination phase. $AUC_T$ will be calculated using the linear up/log down method. Half-life will be calculated as $t_{1/2} = \ln 2/\lambda z$, and total AUC ($AUC_{TOT}$) will be estimated using $AUC = AUC_T + C_T/\lambda z$. Apparent oral clearance (CL/F) and apparent steady state volume of distribution (Vz/F) will be calculated as $CL/F = dose/AUC_{TOT}$ and $Vz/F = CL/\lambda z$. Values for CL/F and Vz/F will be normalized by body weight. The lag time (tlag) is defined as the time to the first observable plasma concentration. Similar techniques will be applied for metabolite analyses.

For each PK parameter for the parent and metabolite compound, descriptive statistics will be calculated and compared for each study group, obese vs. overweight vs. non-obese individuals (ANOVA), across DME genotypes (ANOVA), as well as across the entire study population (regression analysis and/or correlation) as a function of hepatic fat fraction (HFF). Relationships between PK parameters and the physiologic determinants generated from SA 1-4 (e.g., REE, HFF, cytokines, gastric pH) will be interrogated by multiple linear regression analyses.

Agreement between MR-PDFF and UGAP measurements will be assessed using concordance coefficients and Bland-Altman analyses. All statistical analyses will be performed in SPSS or comparable statistical software; $\alpha \leq 0.05$.

19.2 Sample size and power calculation

A power calculation, informed by the PI's previous work with pantoprazole in obese and non-obese children, informed the power calculation, which suggests that a sample size of 50 subjects provides 90% power to detect a linear effect of liver adiposity on pantoprazole $AUC_{tot}$, assuming a correlation of 0.5. Since three medications will be used in this study (pantoprazole,lansoprazole, and midazolam), we will aim to enroll additional number of subjects (n=300) to account for any added variability introduced by multiple study drugs and in case the relationship is not linear. Therefore, we will aim to enroll 300 subjects.

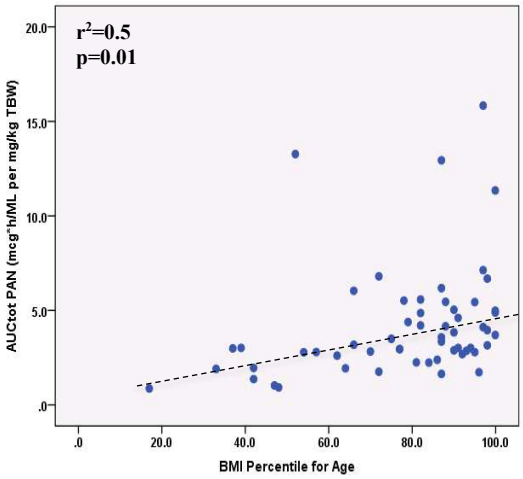



**Figure 3.** Spearman's correlation between dose-for-weight adjusted systemic pantoprazole exposure ($AUC_{tot}$) and BMI percentile, after single oral dose pantoprazole administration (n=57; $r^2$=0.5, p=0.01).

19.3

All data will be deidentified and labeled with a study ID number only (No PHI) for the purposes of data analysis, to minimize breaches in confidentiality. A master linking list, linking PHI to the study ID number, will maintained on a separate, secure sector of the CMH server that is password protected and accessible to specific study staff.

19.4

A Certificate of Confidentiality has not been issued for this study, as this is not an NIH-funded study.

19.5

All biologic study specimens retained by the study team with be labeled with the study ID and the date collected (No PHI). They will be kept in the Clinical Pharmacology Lab. All unused samples will be destroyed within 10 years of study completion.

19.6 Identifiable Data:

*PHI chart:

| | | |
|---|---|---|
| 1. Name/Initials | ☐ Accessed only | ☒Recorded |
| 2. *All* elements of date (except year) directly related to an individual (e.g., date of birth, admission date, discharge date, date of death) | ☐ Accessed only | ☒ Recorded |
| 3. Medical record number | ☐ Accessed only | ☒ Recorded |
| 4. Account number | ☒ Accessed only | ☐ Recorded |
| 5. Health plan identification number | ☐ Accessed only | ☐ Recorded |
| 6. Social Security Number | ☒ Accessed only | ☐ Recorded |
| 7. Device identifiers and serial number | ☐ Accessed only | ☐ Recorded |
| 8. Certificate/License number | ☐ Accessed only | ☐ Recorded |
| 9. Telephone number | ☐ Accessed only | ☒ Recorded |
| 10. Fax number | ☒ Accessed only | ☐ Recorded |
| 11. Email addresses | ☐ Accessed only | ☒ Recorded |
| 12. Web addresses (URLs); Internet IP addresses | ☐ Accessed only | ☐ Recorded |
| 13. Street address, city, county, precinct, zip code or equivalent geographical codes | ☒ Accessed only | ☐ Recorded |
| 14. Full face photographic images and any comparable images | ☐ Accessed only | ☐ Recorded |
| 15. Biometric identifiers, including finger and voice print | ☐ Accessed only | ☐ Recorded |
| 16. Vehicle identifiers and serial numbers, including license plate number | ☐ Accessed only | ☐ Recorded |
| 17. Any other unique identifying number, characteristic or code that may help | ☐ Accessed only | ☐ Recorded |

| | | |
|---|---|---|
| identify individual participants including their initials (e.g., student or employee ID number) | | |
| 18. Elements of date, including year, for persons 90 years or older | ☐ Accessed only | ☐ Recorded |

## 20. Provisions to Monitor the Data to Ensure the Safety of Subjects

This study is similar to previous PK studies conducted by the PI and her research team (Risk Category 1). It involves the same class of study drugs, PPIs. As with previous studies, to ensure safety, subjects and their parents will be provided with contact numbers to enable them to contact an investigator 24 hours daily in the event of an adverse effect. All subjects will be carefully monitored for the development of adverse experiences throughout the course of each study visit and records of these events will be maintained. The reporting of adverse events will be in compliance with the Institutional Review Board's regulations. All staffs participating in human subjects' research at Children's Mercy Hospital are required to have successfully completed training and passed the certification for performing research in human subjects. The Clinical Research Unit at Children's Mercy Hospital has significant experience with pharmacokinetic studies in pediatric patients and is well equipped to carry out these studies. For these reasons, we feel confident that this study will not place study subjects at undue risk.

## 21. Provisions to Protect the Privacy Interests of Subjects

1. **What subject identifiers are used in:**
   a. **study documents to funding agency-** No subject identifiers will be included in any datasets that may be required to be deposited in public repositories as a condition of federal (NIH) funding. Participants will only be identified by their unique study identification number.

   b. **study documents retained at CMH –** Patient identifiers such as patient initials and date of birth, name, address and phone number, medical record number, study number, hospital account number, social security number and dates of service, per p/a and data collection sheets will be retained.

2. **Where will data be stored and how will confidentiality be maintained?** Division of Clinical Pharmacology, Toxicology and Therapeutic Innovation and/or the Division of Gastroenterology. All paper records are in a locked cabinet in a locked office, when not attended by the study team, with restricted access to study staff. For electronic data, all computers are located in a secure (ID swipe access) area and are password-protected.

3. **Who will have access to data?** PI and dedicated study staff.

4. **Will tissue/blood samples be stored beyond the end of the study?** [ ] Y [X] N

**a. If yes, explain, and specify what subject identifiers will be retained on the samples**: Specimens for DNA testing will be assigned a subject identification code that does not contain any patient identifying information.

**b. Is this clearly stated in the consent form?** Yes

**5. What will happen to data and samples if subject withdraws prior to completing the study?** Samples and data will be destroyed, per subject request.

## 22. Compensation for Research-Related Injury

*NA*

## 23. Economic Burden to Subjects

Subjects are not responsible for any costs associated with this research

## 24. Permission/Assent/Consent Process

CM research policies on informed permission/assent/consent will be followed and written documentation of assent/consent will be obtained prior to any study procedures. A Waiver of Consent at Age 18 is requested for subjects who turn 18 years of age, during the lifetime of the study, but for whom all study procedures and sample collections have been completed prior to age 18. Subjects who turn 18 between study visits, will be re-consented using the adult informed consent form at their next study visit.

## 25. Process to Document Permission/Assent/Consent

CM Research Policy 10.04 Obtaining Permission/Assent/Consent will be followed.

## 26. Setting

This is a single-center study that will take place at Children's Mercy Kansas City. All study visits will be conducted in the PCRU at CMH except the MRI visit, which will occur in one of the MRI suites at Children' Mercy Kansas City. For the purposes of this investigation, there is an established collaboration between the PI and the Radiology Department.

See section 14.0 for recruitment strategies and identification of subjects.

## 27. Resources Available

This study is externally funded by the NASPGHAN Foundation (PI), NIDDK (5K23DK115827; PI) and the Frontiers Clinical and Translational Research Pilot Study Grant (PI); thus, the study team is motivated to complete enrollment for this investigation within the timeframe of these grants. The PI and her research team have a strong track record in successfully conducting PK studies in children at CMH (n=106, combined, for two recent PK investigations of pantoprazole in children). MR LiverLab sequencing is readily available for study purposes and key, invested, individuals from the CMH Radiology Department are co-investigators on this research collaboration, to ensure timely completion of all research procedures.

The preliminary data generated during the course of the NASPGHAN Foundation award will serve as a springboard for additional external funding for the PI, including her NIH K23 career development award, as well as other research funding opportunities. In the meantime, any additional study costs will be offset by discretionary funds available to the PI.

## 28. Multi-Site Research

*NA*

## 29. International Research

*NA*

## 30. Provisions to Monitor the Data to Ensure the Safety of Subjects

*30.1* In addition to the Principal Investigator, which individual or group will be responsible for monitoring the data and safety for this study?

☐ Sponsor or Sponsor Designee (including the Sponsor CRO)
☒ Data and Safety Monitoring Board (DSMB) or Data Safety Monitoring Committee (DSMC)
☐ Independent Monitor (s)
☐ Internal Committee at CM
☐ Other: _____

***Data Safety Monitoring Plan:*** *See monitoring plan uploaded under "Other Attachments" on the "Local Documents" page in the myIRB submission*

## References


1. Shakhnovich V, Smith PB, Guptill JT, et al. Obese children require lower doses of pantoprazole than nonobese peers to achieve equal systemic drug exposures. *J Pediatr* 2018;193:102-8
2. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of childhood and adult obesity in the United States. *JAMA Pediatr* 2013;311:06-14
3. Koebnick C, Getahun D, Smith N, Porter AH, Der-Sarkissian JK, Jacobsen SJ. Extreme childhood obesity is associated with increased risk for gastroesophageal reflux disease in a large population-based study. *Int J Pediatr Obes* 2011;6:e267-63
4. Gold B. Gastroesophageal reflux disease: could intervention in childhood reduce the risk of later complications? *Am J Med* 2004;5(suppl 1):23-29
5. Stark CM, Cade MN. Side effects and complications of proton pump inhibitors: a pediatric perspective. *J Pediatr* 2016;168:16-22
6. Dubcenco E, Beers-Block PM, Kim L, et al. A proton pump inhibitor in the reformulation setting: bioequivalence and potential implications for long-term safety. *Clin Transl Sci* 2017;10:387-94
7. Zanger UM, Schwab M. Cytochrome P450 enzymes in drug metabolism: Regulation of gene expression, enzyme activities, and impact of genetic variation. *Pharmacol Ther* 2013;138:103–141
8. Kearns GL, Winter HS. Proton pump inhibitors in pediatrics: relevant pharmacokinetics and pharmacodynamics. *J Pediatr Gastroenterol Nutr* 2003;37:S52-59

9. Chai G, Governale L, McMahon AW, et al. Trends of outpatient prescription drug utilization in US children, 2002-2010. *Pediatr* 2012;130:23-31
10. National ambularotry medical care survey: 2012 state and national summary tables. http://www.cdc.gov/nchs/fastats/drug-use-therapeutic.htm. Published March 29, 2014. Updated October 7, 2016. Accessed October 17, 2016
11. Esser N, Legrand-Poels S, Piette J, et al. Inflammation as a link between obesity, metabolic syndrome and type 2 diabetes. *Diabetes Res Clin Pract* 2014;105:141-150
12. Ijaz S, Yang W, Winslet MC, Seifalian AM. Impairment of hepatic microcirculation in fatty liver. *Microcirculation* 2003;10:447-456
13. Hanley MJ, Abernethy DR, Greenblatt DJ. Effect of obesity on the pharmacokinetics of drugs in humans. *Clin Pharmacokinet* 2010;49:71-87
14. Harskamp-van Ginkel MW, Hill KD, Becker K, et al. Drug dosing in obese children: a systematic review of current pharmacokinetic data. *JAMA Pediatr* 2015;169(7):678-85
15. Shakhnovich V, Vyhlidal C, Friesen C, et al. Decreased pregnane X receptor (PXR) expression in children with active Crohn's disease." *Drug Metab Dispos* 2016;44(7):1066-69
16. Aitken AE, Richardson TA, Morgan ET. Regulation of drug-metabolizing enzymes and transporters in inflammation. *Annu Rev Pharmacol Toxicol* 2006;46:123-49
17. Fields DA, Kearney JT, Copeland KC. MedGem hand-held indirect calorimeter is valid for resting energy expenditure measurement in healthy children. *Obesity* 2006;14(10):1755-61
18. Al-Salami HS, Goulding A, Grant A, Taylor R, Holfod N, Duffull SB. Prediction of Fat-Free Mass in Children. *Clin Pharmacokinet* 2015;54:1169-78
19. Sellers R. MR LiverLab. *MAGNETOM Flash* 2016;66(3):39-43
20. Tanaka M, Yamazaki H. Direct determination of pantoprazole enantiomers in human serum by reversed-phase high-performance liquid chromatography using a cellulose-based chiral stationary phase and column-switching system as a sample cleanup procedure. *Anal Chem* 1996;**68**:1513-1516.
21. Aoki I. High performance liquid chromatographic determination of lansoprazole and its metabolites in human serum and urine. *J Chromotograph* 19911;**571**:283-290
22. Ward RM, Tammara B, Sullivan SE, Stewart DL, Rath N, Meng X, et al. Single-dose, multiple-dose, and population pharmacokinetics of pantoprazole in neonates and preterm infants with a clinical diagnosis of gastroesophageal reflux disease (GERD). *Eur J Clin Pharmacol* 2010;**66**:555-561
23. Ward RM, Kearns GL, Tammara B, Bishop P, O'Gorman MA, James LP, et al. A multicenter, randomized, open-label, pharmacokinetics and safety study of pantoprazole tablets in children and adolescents aged 6 through 16 years with gastroesophageal reflux disease. *J Clin Pharmacol* 2011;51:876-87
24. Kearns GL, Blumer J, Schexnayder S, James LP, Adcock KG, Reed MD, et al. Single-dose pharmacokinetics of oral and intravenous pantoprazole in children and adolescents. *J Clin Pharmacol* 2008;**48**:1356-1365.
25. Desta Z, Modak A, Nguyen PD, Lemler SM, Kurogi Y, Li L, et al. Rapid identification of the hepatic cytochrome P450 2C19 activity using a novel and noninvasive [13C]pantoprazole breath test. *J Pharmacol Exp Ther* 2009;**329**:297-305.
26. Furuta T, Kodaira C, Nishino M, Yamade M, Sugimoto M, Ikuma M, et al. [13C]-pantoprazole breath test to predict CYP2C19 phenotype and efficacy of a proton pump inhibitor, lansoprazole. *Aliment Pharmacol Ther* 2009;**30**:294–300.
27. Tolia V, Fitzgerald J, Hassal E, et al. Safety of lansoprazole in the treatment of gastroesophageal reflux disease in children. *J Gastroenterol Hepatol Nutr* 2002;35(Suppl4):S300-S307

28. J V Arcos-Machancoses, D García Tirado, V Vila Miravet, G Pujol Muncunill, S Pinillos Pisón, J Martín de Carpi. What is the best method for calculating the optimal position of an esophageal pH probe in children? *Diseases of the Esophagus* 2019;32(5):doz014
29. Fackler WK, Vaezi MF, Richter JE. Ambulatory gastric pH monitoring: proper probe placement and normal values. *Aliment Pharmacol Ther* 2001;15(8):1155-62
30. Midazolam Product Monograph. Pfizer. October 25, 2017. Last accessed 27 February 2020. https://www.pfizer.ca/sites/default/files/201712/2017.10.25_Midazolam_PM_PF_E_210318.pdf
31. Reed MD, Rodarte A, Blumer JL, et al. The single-dose pharmacokinetics of midazolam and its primary metabolite in pediatric patients after oral and intravenous administration. *J Clin Pharmacol* 2001;41:1359-1369
32. Van Rogen A, Vaughns JD, Moorthy GS, Barrett JS, Knibbe CAJ, et al. Population pharmacokinetics of midazolam and its metabolites in overweight and obese adolescents. *Br J Clin Pharmacol* 2015;80(5):1185-96
33. Friesen CS, Chan SS, Wagner JB, Hosey-Cojocari C, Csanaky IL, Shakhnovich V. Critical need for pharmacologic treatment options in NAFLD: A pediatric perspective. *Clin Transl Sci* 2021;00:1-3
34. Tada T, Kumada T, Toyoda H, Kobayashi N, Sone Y, et al. Utility of Attenuation Coefficient Measurement Using an Ultrasound- Guided Attenuation Parameter for Evaluation of Hepatic Steatosis: Comparison With MRI-Determined Proton Density Fat Fraction. *AJR* 2019;212:332-341