PROTOCOL TITLE:
An Open-Label, Single- and Multi-Dose Study to Evaluate the Relationship between the Pharmacokinetics, Pharmacodynamics, and Clinical Outcomes of Atomoxetine in CYP2D6 Extensive, Intermediate, and Poor Metabolizers in Children with Attention Deficit/Hyperactivity Disorder

PRINCIPAL INVESTIGATORS:

James Steven Leeder (Primary contact)
Clinical Pharmacology, Toxicology, and Therapeutic Innovation
816-302-3001
sleeder@cmh.edu

Sarah Soden
Developmental and Behavioral Sciences
816-302-8589
ssoden@cmh.edu

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Version 19.0/April 1, 2023

REVISION HISTORY

Revision #	Date of IRB Approval	Consent Change? Describe plan/timeline	Summary of Changes
1.0	11/29/2016	N/A	Initial submission
2.0	06/29/2017	Y	1) Expand enrollment to include ADHD Summer Treatment Program (STP) 2) Med history, physical exam, Tanner staging added 3) Typographical error fixed (Echo to EKG) 4) Modified Vanderbilt form 5) Drug manufacturer, NDC, lot # to be collected 6) Urine to be collected 7) Additional blood draws during PK visits 8) Sweat patches to be collected 9) Study booklet cosmetic changes 10) Vitals to be obtained with PK visit blood draws 11) Pupillometry/eye-tracking frequency changes
3.0	08/16/2017	N	1) Changing CDISC to KSADS instrument
4.0	12/18/2017	Y	1) Changes in recruitment pool and recruitment numbers

			2) Add EKG to last PK visit 3) Change to consent form regarding continuing care for ADHD after study completion
5.0	01/10/2018	Y	1) Changes to CRFs 2) Tanner staging can be maxed out or does not need to be done if down in last 4 weeks 3) Clarifications to P/A/C about physical exam, Tanner staging, pupillometry
6.0	07/05/2018	Y – notification of DSQ	1) Change inclusion/exclusion criteria to allow children who have taken other ADHD medications 2) Addition of DSQ 3) Distribute blood sample into 2 types of tubes for processing plasma and serum 4) REDCap changes 5) Allow physicians to titrate ATX slower 6) Added SCARED questionnaire
7.0	11/12/2018	Y – reconsent at next available opportunity	1) Allowance of a small snack to alleviate GI symptoms as needed during research visits 2-4 2) Added side effects known to include GI upset, somnolence 3) Add edited study booklet to accommodate participants recruited from the ADHD Clinic and the PCC (change in other docs) 4) Addition of Sluggish Cognitive Tempo (SCT) scales 5) A section for disposal of drug and situations when drug does not need to be returned to IDS Pharmacy 6) Allowing omission of safety labs if prior safety labs were collected within the previous week
8.0	12/21/2018	Y	1) Adding Recruitment Flyer 2) New washout period procedures 3) Information on when a snack may be given
9.0	07/25/2019	Y	1) Modified inclusion/exclusion criteria 2) Detailed pre-screening and recruitment methods 3) Updated all CRFs and created checklists for each study visit 4) Created CRFs for visits with study physicians 5) Reformatted entire protocol to use new protocol template 6) Creating of DSMP, DSMB, and CQMP

			7) Clarified KSADS use 8) Added 2-week follow-up after last study visit 9) Added formal adherence and AE checks to study physician visits 10) Added optional wean of study drug to avoid abrupt discontinuation 11) Formalized templates for sharing research findings 12) Changed subject reimbursement schedule 13) Created CHADIS handouts, STP presentation, recruitment flyers, and washout handout Updated potential participant log and enrollment log
10.0	02/05/2020	Y – reconsent at next available opportunity	1) Removed Jennifer Lowry from protocol. 2) Added Wendy Wright to protocol. 3) Added PCC ADHD Clinic on Broadway as a participant recruitment site. 4) Modified Prescreening and Recruitment Methods to add PCC ADHD Clinic located on Broadway as a recruitment site. This is the 6th recruitment track. 5) Clarified duration between Research Visit 2 and Research Visit 3. 6) Updated Study Timeline for clarity and addition of a new 6th recruitment track. 7) Updated Study Timeline to clarify timing of Study Physician Visits 8) Added option for not repeating entire Intake Visit for participants referred to the research study by a Study Physician and if the participant had a clinical appointment with Study Physician within 6 weeks of enrollment. 9) Clarified ADHD Care Following Conclusion of the Study and moved to section 11.5. 10) Changed J-tip to “topical lidocaine,” to allow participants the option of requesting other topical lidocaine formulations. 11) Expanded time frame to call families for adherence checks prior to Research Visit 3 and Research Visit 4. 12) Updated Schedule of Events to be consistent with Study timeline. 13) Described how to return genotyping results to families. Created letter to return research

			genotyping results to participants and their families and explain the limitations of these research results. This is to be compliant with the following policy: *Research Documentation in the Electronic Health Record*, which was published on 9/26/2019 14) Provided further details regarding genetic analysis, including future genetic analysis, and iGO-PK model refinement in protocol and permission/assent and consent forms. Additional formatting changes were included so that the permission/assent and consent forms are consistent with one another. 15) Updated Study Setting to include ADHD Clinic on Broadway 16) Updated Message Center Template to include provider at PCC ADHD Clinic as a Study Physician. 17) Updated Study Coordinator/Research Assistant Recruitment Script for better comprehension and to better address participants recruited from different tracks. 18) Updated permission/assent and consent forms for better clarity and comprehension. 19) Updated permission/assent and consent forms to revise benefits of the study with regards to returning of genotyping information. 20) Updated extra cost section in permission/assent and consent forms. 21) Updated contact information on permission/assent and consent forms 22) Revised formatting and wording of questions for contact about future research for better clarity on permission/assent and consent forms. 23) Updated contact information and added ADHD Clinic on Broadway information in U54 Study Booklet. 24) Modified Checklist for Research Visit 1 to include adding Certificate of Confidentiality for research. 25) Checklists for Research Visit 2, Research Visit 3, and Research Visit 4 for both Study Design 1 and Study Design 2 updated to include creation

			of calendar invites, notes to describe if/when a study procedure does not need to be repeated, and ensuring accuracy of information on labels of biospecimens. 26) Updated Research Visit 3 Study Design 1 and Study Design 2 checklists and Research Visit 4 – Study Design 1 and Study Design 2 checklists to reflect date range change for adherence checks 27) Intake Visit CRF – updated to include "none" option for comorbidities 28) Updated Research Visit 1 – Part 2 CRF to include Child Medical History. Correction made to respondents of Vanderbilt questionnaire. 29) Updated page numbers on Serious Adverse Events CRF 30) Updated Research Visit 4 – Study Design 1 and Study Design 2 CRFs to add EKG to procedures 31) Research Visit 4 Adherence Check and Research Visit 3 Adherence Check CRFs updated to reflect changes in date range for adherence checks 32) Updated Research Visit 1 – Part 1 CRF to add "N/A" option for creating CHADIS account and "N/A" for participant willingness to washout for those unable to provide assent.
11.0	3/19/2020	N	1) Removed Benjamin Black and Bridgette Jones from the protocol. 2) Protocol modified to allow participants to take medication with a small amount of soft food to aid with capsule swallowing. 3) Updated Date When Version 10.0 was approved in Revision History Table. 4) Updated "topical lidocaine," to specify that this refers to both J-tip and cream formulations. 5) Updated protocol to allow topical lidocaine to be administered at either Research Visit 1 Part 1 or Research Visit 1 Part 2 for venipuncture procedures. 6) Updated permission/assent form with regards to Future Research section. This section now includes a yes/no box for data to be submitted to national database.

			7) Uploaded most applicable version of NICHQ Vanderbilt Assessment questionnaires being used in the study (Version 2). 8) Changed "Vanderbilt ADHD Rating Scales," to official name, "NICHQ Vanderbilt Assessment Scales." 9) Uploaded NICHQ Vanderbilt Assessment Scales, both initial and follow-up, for parent and teacher informant. 10) Uploaded School Intervention Questionnaire on myIRB. 11) Updated CYP2D6 genotype-to-phenotype interpretation based on new Clinical Pharmacogenetic Implementation Consortium Guidelines (CPIC). 12) Updated CYP2D6 Study Design Assignment based on new CPIC guidelines and data generated from participants who have completed ATX pharmacokinetic studies in Ped Develop Pharm U54 Study (IRB# 16010069) and this study.
12.0	3/26/2020	N	1) COVID-19 Addendum to Approved Study Protocol. This addendum was enacted as a part of the Children's Mercy Hospital response to the global COVID-19 pandemic. 2) Briefly, the addendum made the following changes to follow social distancing guidelines: a. Contacting all active participants and their families to alert them to upcoming changes and to ascertain interest in continuing the study. b. Prescreening, recruitment, and new enrollment in this study was paused. c. Participants who have not started on pharmacological intervention were advised to continue with their current ADHD therapy (if applicable) until it has been determined to be safe enough to progress with the remainder of the study. d. For participants who have started on pharmacological intervention (i.e. participants who have completed Research Visit 2 at a minimum), study

			procedures were modified to allow social distancing. i. Full PK research visits are to be delayed or cancelled, based on the timing of the visit and the status of the pandemic. Research visits in full are only to be conducted if Children's Mercy Hospital and Children's Mercy Resesarch Institute have determined the COVID-19 situtation has improved such that these visits may occur. Modified research visits are to occur in lieu in certain scenarios, and the procedures for these modified visits are limited to those which may be conducted remotely. ii. Study physician visits are to proceed according to schedule to provide continuity of ADHD care for study participants. However, the majority of these visits were conducted via telehealth. Study physicians are able to request in-person visits on a case-by-case basis. For telehealth visits, a physical exam and collection of vitals were not required to be collected. iii. Compensation was adjusted for participants completing modified research procedures.
13.0	7/6/2020	N	1) Modification to COVID-19 Addendum to Approved Study Protocol. This modification is part of Children's Mercy Hospital plan to restart research during the global COVID-19 pandemic. 2) In-person Study Physician visits are allowed at the discretion of the Study Physician and do not need to be deemed, "clinically necessary" for these to be scheduled. 3) Research Visit 3 and Research Visit 4 may now occur at the PCRU. Participants will be asked

			prior to scheduling Research Visit 3 and Research Visit 4 if the participant and/or parent/LAR would like to conduct these in person or cancel (pertains to Research Visit 3) or have a modified visit (pertains to Research Visit 4). 4) Addition of COVID-19 symptom screening for in-person Research Visit 3. Updated "Adherence Check" CRF to "Research Visit 3 (RV3) Screen" CRF which included adherence and COVID-19 Symptom Screen. 5) Addition of COVID-19 symptom screening for in-person Research Visit 4. Updated "Adherence Check" CRF to "Research Visit 4 (RV4) Screen" CRF which included adherence and COVID-19 Symptom Screen. 6) An updated letter titled, "Temporary Study Changes in Response to COVID-19 Pandemic – Updated," has been written to describe the changes to participants and families. 7) Updated Research Visit 3 and Research Visit 4 checklists to include COVID-19 symptom screening. 8) Added "direct hand-off" as a method of transferring medication and study materials between the parent/LAR and study personnel. 9) These changes will only be applicable to active participants who are currently taking atomoxetine (U54-035, U54-037, and U54-038).
14.0	9/8/2020	Y	1) Personnel changes: a. Added Jamie Neal-Lewis, RN, APRN, CPNP, CPN, to the protocol in the role of Study Physician. b. Removed Wendy Wright, MSN, RN and Madeline DeShazer, BS from the protocol. 2) Permission/assent and consent forms changes: a. Added certificate of confidentiality language b. Modified injury language per IRB template c. Change in window for Research Visit 3 d. Removed PCC ADHD Clinic as a site for Study Physician Visits.

			e. Included option for telehealth Study Physician Visits. 3) Protocol changes: a. Removed Track 6 for recruitment and the PCC ADHD Clinic on Broadway as a study site. b. Updated the Study Coordinator/Research Assistant Recruitment and Scheduling Scripts to remove Track 6. c. Inclusion/Exclusion criteria: i. Updated inclusion criteria to include "inferred" ADHD diagnosis. This is to account for the fact that obtaining typical diagnostic data for ADHD may not be feasible during the COVID-19 pandemic. ii. Updated exclusion criteria to include evidence of intellectual disability from review of EMR. d. Expanded window for Research Visit 3 to make scheduling flexible. The timeframe for Research Visit 3 can now be scheduled anytime after the participant has been taking medication regularly for 2 weeks and up until the 18-Week Visit with Study Physician. e. Amended Study Physician visits to occur in person or through telehealth. The type of visit will be at the Study Physician's discretion. f. Updated Schedule of Events Table to reflect changes to RV3 and Study Physician visits. g. Drug handling and study drug timeline changed in to be compliant with IDS delivery policies developed to address specific scenarios, such as with the COVID-19 response. h. Methods of drug handling and the study drug timeline have been amended to include practices to reduce foot-traffic at CMH.

			i. Pharmacy Policies cited updated to include latest revision date – Section 5.2, pg. 34. j. CYP2D6 Phenotype, Activity Score, and example diplotype table headings updated. k. Updated Procedures to Reduce Risk section to include COVID-19 precautions and Certificate of Confidentiality changes in the permission/assent and consent forms. l. Removed "Appendix P," "18-week Visit with Study Physician – Research 4 Delay Approval," and the COVID-19 addendum because these documents were all associated with the COVID-19 response. These documents no longer needed with Version 14 of the protocol because this protocol has been updated to include COVID-19 precautions. Therefore, these document were removed from Smartform. These documents were never included in full version of the protocol because they were part of the COVID-19 addendum. 4) Study Booklet Changes: a. New study timeline figure b. Removed PCC ADHD Clinic as Study Physician site. 5) Updated checklists to include COVID-19 instructions prior to research visits and changed "Madeline" to "RA/clinical psychologist," where applicable: a. RV1 Part 1 and 2 Checklist No Washout Required (3.0) i. Changed "Madeline" to "RA/clinical psychologist" ii. Changed "Remind family to bring participant's SSN" to "Call family 1-2 days before and remind them of appointment time, to bring participant's SSN, and go over COVID-19 instructions."

			b. RV1 Part 1 Checklist Washout Required (3.0) i. Changed “Remind family to bring participant’s SSN” to “Call family 1-2 days before and remind them of appointment time, to bring participant’s SSN, and go over COVID-19 instructions.” c. RV1 Part 2 Checklist Washout Required (3.0) i. Changed “Madeline” to “RA/clinical psychologist” ii. Added “Call family 1-2 days before and remind them of appointment time and go over COVID-19 instructions.” d. RV2 – Study Design 1 Checklist (3.0) i. Changed “Call family 1-2 days before and remind to arrive at 0700, fast after midnight, water is okay, bring study booklet” to “Call family 1-2 days before and remind to remind them of appointment time, go over COVID-19 instructions, fast after midnight (water is okay), and bring the study booklet.” e. RV2 – Study Design 2 Checklist (3.0) i. Changed “Call family 1-2 days before and remind to arrive at 0700, fast after midnight, water is okay, bring study booklet” to “Call family 1-2 days before and remind to remind them of appointment time, go over COVID-19 instructions, fast after midnight (water is okay), and bring the study booklet.” f. RV3 – Study Design 1 Checklist (4.0) i. Changed, “Call patient 1-2 days prior to visit to check adherence, COVID-19 symptom screen, remind to bring study med and calendar, and fast after

			midnight" to "Call patient 1-2 days prior to visit to check adherence, go over COVID-19 instructions, remind to bring study med and calendar, and fast after midnight" ii. Removed the "Log CMH COVID-19 symptom screen for participant and family member(s) activity. g. RV3 – Study Design 2 Checklist (4.0) i. Changed, "Call patient 1-2 days prior to visit to check adherence, COVID-19 symptom screen, remind to bring study med and calendar, and fast after midnight" to "Call patient 1-2 days prior to visit to check adherence, go over COVID-19 instructions, remind to bring study med and calendar, and fast after midnight" ii. Removed the "Log CMH COVID-19 symptom screen for participant and family member(s) activity. h. RV4 – Study Design 1 Checklist (4.0) i. Changed, "Call patient 1-2 days prior to visit to check adherence, COVID-19 symptom screen, remind to bring study med and calendar, and fast after midnight" to "Call patient 1-2 days prior to visit to check adherence, go over COVID-19 instructions, remind to bring study med and calendar, and fast after midnight" ii. Removed the "Log CMH COVID-19 symptom screen for participant and family member(s) activity. i. RV4 – Study Design 2 Checklist (4.0)

			i. Changed, “Call patient 1-2 days prior to visit to check adherence, COVID-19 symptom screen, remind to bring study med and calendar, and fast after midnight” to “Call patient 1-2 days prior to visit to check adherence, go over COVID-19 instructions, remind to bring study med and calendar, and fast after midnight” ii. Removed the “Log CMH COVID-19 symptom screen for participant and family member(s) activity. 6) Updated Research Visit 3 and 4 “Screens” to “Adherence Check”: a. RV3 – Adherence Check (6.0) i. Removed COVID-19 Symptom Phone Screen with parent/LAR/guardian section. **ii.** Removed Children’s Mercy Hospital COVID-19 Symptom Screen with participant, parent/LAR/guardian, and accompanying family members section. iii. Changed name of CRF from “Screen” to Adherence Check b. RV4 – Adherence Check (6.0) i. Removed COVID-19 Symptom Phone Screen with parent/LAR/guardian section. **ii.** Removed Children’s Mercy Hospital COVID-19 Symptom Screen with participant, parent/LAR/guardian, and accompanying family members section. iii. Changed name of CRF from “Screen” to Adherence Check 7) Updated Study Physician checklists: a. Intake Visit – Checklist (2.0)

			i. Changed “Ensure baseline CHADIS questionnaires are all completed” to “Ensure baseline CHADIS questionnaires have been sent out” ii. Changed “Collect physical exam data” to “Collect physical exam data if visit conducted in person. These measures are omitted for telehealth visits.” b. 6-week Study Physician Visit Checklist (2.0) i. Changed “Ensure baseline CHADIS questionnaires are all completed” to “Ensure baseline CHADIS questionnaires have been sent out” ii. Changed “Collect physical exam data” to “Collect physical exam data if visit conducted in person. These measures are omitted for telehealth visits.” iii. Added, “If needed, ask participant’s parents/LAR to send pictures of adherence calendar and remaining pills. Conduct pill count/adherence check.” c. 12-week Study Physician Visit Checklist (2.0) i. Changed “Ensure baseline CHADIS questionnaires are all completed” to “Ensure baseline CHADIS questionnaires have been sent out” ii. Changed “Collect physical exam data” to “Collect physical exam data if visit conducted in person. These measures are omitted for telehealth visits.” iii. Added, “If needed, ask participant’s parents/LAR to send pictures of adherence calendar and remaining pills.

			Conduct pill count/adherence check." d. 18-week Study Physician Visit Checklist (2.0) i. Changed "Ensure baseline CHADIS questionnaires are all completed" to "Ensure baseline CHADIS questionnaires have been sent out" ii. Changed "Collect physical exam data" to "Collect physical exam data if visit conducted in person. These measures are omitted for telehealth visits." iii. Added, "If needed, ask participant's parents/LAR to send pictures of adherence calendar and remaining pills. Conduct pill count/adherence check." 8) Updated Study Physician Visits CRFs to document telehealth visits and omit physical exam findings when these visits are telehealth. a. Intake Visit – CRF (3.0) i. Updated "Physical Exam Finding" section to include the following instructions: "Check box below if this is a telehealth visit. Physical exam findings are omitted for telehealth visits." ii. Included a checkbox to indicate whether the visit was a telehealth visit. b. 6-week Study Physician Visit CRF (2.0) i. Updated "Physical Exam Finding" section to include the following instructions: "Check box below if this is a telehealth visit. Physical exam findings are omitted for telehealth visits." ii. Included a checkbox to indicate whether the visit was a telehealth visit. c. 12-week Study Physician Visit CRF (2.0)

			i. Updated "Physical Exam Finding" section to include the following instructions: "Check box below if this is a telehealth visit. Physical exam findings are omitted for telehealth visits." ii. Included a checkbox to indicate whether the visit was a telehealth visit. d. 18-week Study Physician Visit CRF (2.0) i. Updated "Physical Exam Finding" section to include the following instructions: "Check box below if this is a telehealth visit. Physical exam findings are omitted for telehealth visits." ii. Included a checkbox to indicate whether the visit was a telehealth visit.
15.0	03/16/2021	Y – reconsent at next available opportunity	1) Personnel Changes (MOD00006449, approved 12/28/2020): a. Added Sarah Beals-Erickson, Simone Moody, and Lynne Ray b. Removed Carla Allan 2) Follow-up personnel changes: a. Removal of Sarah Beals-Erickson 3) Changes to permission/assent and consent form: a. Update Krista Ladue-Wright's phone number b. Update IRB's phone number c. Update language on shipping medication to participants 4) Changes to recruitment: a. Allow for advertising of study on CMH internal website and on CMRI public facing website. 5) Update to shipping medications: a. Changed shipping protocol to reflect IDS Pharmacy's primary role in shipping medications.

			6) Updated KSADS questionnaire format to include use of KSADS-COMP, a computerized version of the KSADS questionnaire.
16.0	3/17/21	Yes – Reconsent not needed; verbally notify currently enrolled participants at next study visit	1. Personnel change – Jennifer Wagner, MD replacing Jean Dinh, PharmD, PhD as PI (primary contact); add James Steven Leeder to myIRB. 2. Changes to P/A/C forms and recruitment flyer to reflect change in PI. 3. Changes to CRFs to provide instruction clarity for study team members on completion of study procedures a. Adverse Events CRF, RV3 CRF (Study Designs 1 and 2), 6-Week, 12-Week, and 18-Week Visit with Study Physician CRFs, and RV4 CRF (Study Designs 1 and 2)
17.0	10/7/2021	Y – This has been completed	1) Intervention assignment to include Study Design 3 2) Visit length by study design table (Figure 5) updated 3) Research Visit 3 and 4 to describe possible switch to Study Design 3 4) RV3 and RV4 confirmation of study design 5) RV3 and RV4 addition of Study Design 3 dosing and collection time points 6) RV3 and RV4 addition of Study design 3 urine collection instructions 7) RV3 and RV4 addition of Study Design 3 sweat patch collection instructions 8) RV3 and RV4 clarification on fasting for Study Design 3 9) Procedures to reduce risk updated to include blood collection estimates and mitigation strategies for Study Design 3 10) Payments addition of clarification for hours in the table 11) Payments addition to maximum compensation for participants in Study Design 3 12) Clarified number of doses needed from IDS pharmacy since option of Study Design 3 13) Changes to P/A/C forms to reflect addition of Study Design 3

			14) Changes to CRFs including checklists and data collection sheets for Study Design 3 15) Changes to CRFs to include documentation of time of prior day before research visits 16) Changes to Research visit Checklist forms to include securing participant binders and samples following the study 17) Personnel Changes: Added Rachael Robertson to study
18.0	TBD	No – Will verbally tell particpants at next Research Visit, but protocol change potentially confers less risk to the particpant	1) Tanner staging changed to optional procedure per participant preference 2) P/A/C forms updated to reflect that Tanner staging is now an optional procedure 3) Personnel Changes: Remove Tamorah Lewis and Susan Abdel-Rahman from study 4) P/A/C and protocol updated to remove verbiage listing Susan Abdel-Rahman as a PI.
19	TBD	Yes	1. Remove Jennifer Wagner from PIs.

Table of Contents

1.0 Study Summary

Study Title	An Open-Label, Single- and Multi-Dose Study to Evaluate the Relationship between the Pharmacokinetics, Pharmacodynamics, and Clinical Outcomes of Atomoxetine in CYP2D6 Extensive, Intermediate, and Poor Metabolizers in Children with Attention Deficit/Hyperactivity Disorder
Study Design	This is a non-randomized, un-blinded, exposure escalation efficacy trial of the effects of atomoxetine in children with attention deficit/hyperactivity disorder (ADHD).
Primary Objective	1. To investigate the effect of genetic variability on the pharmacodynamics of ATX by evaluating biomarkers of inhibition of the drug target, the norepinephrine re-uptake transporter, following ATX exposure control. 2. To explore how changes in pharmacodynamic markers correspond to clinical response to medication.
Secondary Objective(s)	1. To establish the relationship between ATX systemic exposure, inhibition of the norepinephrine uptake transporter, and therapeutic response 2. To determine global baseline patient characteristics differences between ATX responders and non-responders. 3. To explore how baseline oral microbiome make-up influences clinical outcomes and to describe changes in the oral microbiome with ATX therapy. 4. To refine the iGO-PK ATX dosing algorithm. 5. To evaluate comprehension of the study procedures by study participants and their families when an illustrated study booklet is supplied with a consent form. 6. To validate a urinary CYP2D6 phenotype biomarker.
Research Intervention(s)/ Investigational Agent(s)	Atomoxetine – a norepinephrine reuptake transporter inhibitor which is FDA approved for the treatment of ADHD in children. iGO-PK dosing algorithm – a dosing algorithm to normalize atomoxetine exposure regardless of *CYP2D6* genotype.

IND/IDE #	NA
Study Population	Children, both males and females between the ages of 6-18 with ADHD or a high likelihood of having ADHD, electing to receive care at Children's Mercy Hospital and Clinics and children enrolling in the CMH&C ADHD Summer Treatment Program (STP).
Sample Size	Target enrollment of 160 research participants, in anticipation of 120 participants completing the research study.
Study Duration for Individual Participants	6-8 months
Study Specific Abbreviations/ Definitions	1. ADHD = attention deficit/hyperactivity disorder 2. ATX = atomoxetine 3. DHPG = dihydroxylphenol glycol 4. iGO-PK = individualized genetic and ontogeny pharmacokinetic prediction 5. PK = pharmacokinetic 6. PD = pharmacodynamic 7. Research Visit 1 = RV1 8. Research Visit 2 = RV2, or may referred to as pharmacokinetic study 1 (PK1), or first dose PK 9. Research Visit 3 = RV3, or may referred to as pharmacokinetic study 2 (PK2), or steady state PK 10. Research Visit 4 = RV4, or may be referred to as pharmacokinetic study 3 (PK3), or end of study PK

2.0 Objectives

2.1 Primary Objectives(s):

- To investigate the effect of genetic variability on the pharmacodynamics of ATX by evaluating biomarkers of inhibition of the drug target, the norepinephrine re-uptake transporter, following ATX exposure control.
- To explore how changes in pharmacodynamic markers correspond to clinical response to medication.

Secondary Objective(s):

- To establish the relationship between ATX systemic exposure, inhibition of the norepinephrine uptake transporter, and therapeutic response
- To determine global baseline patient characteristics differences between ATX responders and non-responders.
- To explore how baseline oral microbiome make-up influences clinical outcomes and to describe changes in the oral microbiome with ATX therapy.
- To refine the iGO-PK ATX dosing algorithm.
- To evaluate comprehension of the study procedures by study participants and their families when an illustrated study booklet is supplied with a consent form (please see Supplementary Documents, “U54 Study Booklet” and “U54 Study Booklet Information Handout”).
- To validate a urinary CYP2D6 phenotype biomarker.

2.2 Hypotheses to be Tested:

- Hypothesis 1: Genetic variability in the *SLC6A2* gene is associated with variability in 1) concentration-time curves of DHPG and 2) pupil size changes, when evaluating tonic and phasic data.
- Hypothesis 2: Variability in pharmacodynamics markers, i.e. plasma DHPG and pupil size changes, are associated with clinical response to atomoxetine.
- Hypothesis 3: The atomoxetine response cohorts (i.e. response versus non-response and tolerant versus non-tolerant) will have differing global baseline characteristics, such as age, gender, ethnicity, ADHD subtype, genetic polymorphisms, etc.

3.0 Background

3.1 Atomoxetine Background and Gaps in Knowledge

Atomoxetine (ATX), Strattera®, is a norepinephrine re-uptake transporter inhibitor that is approved by the Food and Drug Administration (FDA) for the treatment of attention deficit/hyperactivity disorder (ADHD) (Seneca et al., 2006; Michelson et al., 2007). The drug is often considered a second- or third-line agent, due to the perception that the drug does not work very well. In fact, in a review of studies submitted to the FDA, Newcorn *et* al reported that there appeared to be discrete classes of response to atomoxetine. After 6-9 weeks of treatment, 47% of the patients were considered "responders" based on changes in the rating scales used to measure ADHD symptoms whereas 40% of patients were considered non-responders (Newcorn et al., 2009). Statistically significant ($p<0.001$) differences in scores between responders and non-responders were apparent after the first week of treatment (Fig. 1).

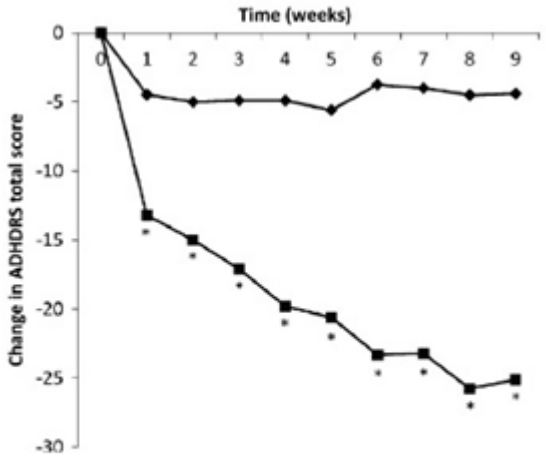


Fig 1. Discrete classes of response to ATX based on patterns of temporal change in ADHD-RS in an analysis of pooled data [2].

At the relatively low starting doses of the titration scheme, this suggests that there may be a subgroup of patents who are particularly responsive to ATX. We hypothesize that there could be two reasons for this: 1) variability in drug pharmacokinetics (i.e., inadequate drug concentrations in the blood over time could lead to poor response) and 2) variability in drug pharmacodynamics (i.e. differences at the level of the target of drug action that limit the response to a drug, regardless of concentration of drug present in the blood). The interplay of pharmacokinetics and pharmacodynamics is presented in Figure 2 below:

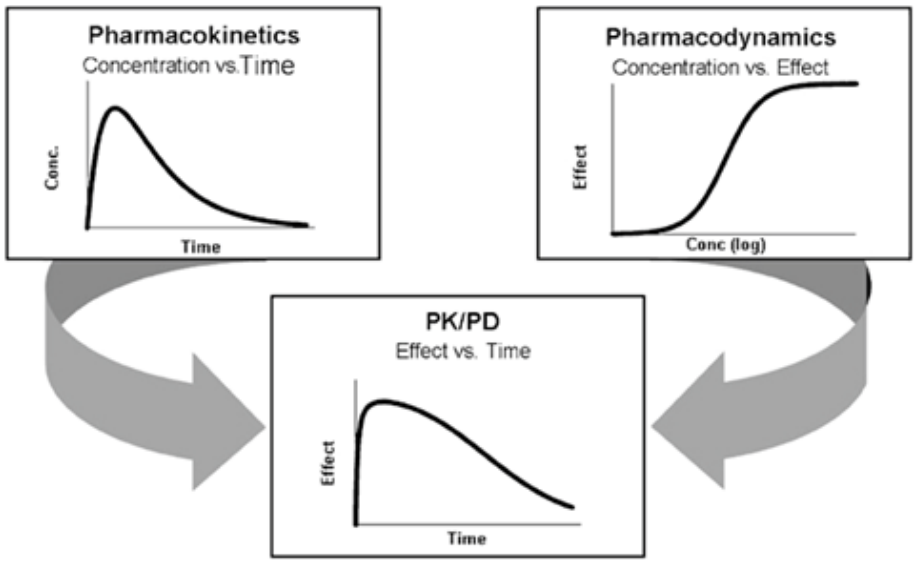


Fig 2. Interplay of Pharmacokinetic and Pharmacodynamics

The *CYP2D6* gene, which encodes for the drug metabolizing enzyme CYP2D6 responsible for the clearance of ATX from the body, is highly polymorphic (Gaedigk et al., 2007; Gaedigk et al., 2010; Gaedigk et al., 2010; Gaedigk, 2013). ATX metabolism by CYP2D6 protein is one of the major routes of clearance (i.e., removal) of this drug. Genetic variability in the *CYP2D6* gene leads to wide inter-individual variability in the activity of the enzyme, ultimately resulting in differing amount of drug in the body (also referred to as "exposure," and is a component of drug pharmacokinetics). Secondly, the *SLC6A2* gene which encodes for the norepinephrine reuptake transporter, the drug target for ATX, is also subject to genetic variation. Reported genetic variants of *SLC6A2* have been associated with decreased abundance of the transporter (Kim et al., 2006). The consequences of *SLC6A2* genetic variation with regards to ATX clinical response are currently unknown. In the context of distinct "responder" and "non-responder" groups with a population of atomoxetine-treated patients, non-response could be due to definable differences at the level of the drug target (patients unlikely to respond regardless of the ATX concentrations achieved), or simply a consequence of inadequate exposure in a substantial proportion of population. The goal of this study is to address this issue.

3.2 Relevant Preliminary Data

In a previous single-dose ATX pharmacokinetic study conducted at the Children's Mercy Hospital, we investigated the impact of CYP2D6 genetic variability on drug pharmacokinetics as an initial step in better understanding the poor clinical response to the drug (Brown et al., 2016). The results from the single-dose ATX study found that ATX exposures varied greatly between children (30-50 fold) when administered the same weight-adjusted dose (0.5 mg/kg) with the highest concentrations/exposure in participants deficient in CYP2D6 activity (so-called

"poor metabolizers") and the lowest exposure in participants who had two or more functional copies of the gene, and the highest CYP2D6 activity. Furthermore, simulations conducted to investigate the concentrations (exposure) achieved with chronic dosing indicated that a majority of patients with at least one functional copy of the CYP2D6 gene were unlikely to achieve the level of exposure associated with clinical response.

A more important implication of these data is that it is impossible to investigate the effect of differences at the level of the drug target on variability in drug response unless we are able to minimize the variability in drug response that is a consequence of the normal variability in exposure when the same dose (0.5 mg/kg) is given to patients who differ in their ability to eliminate the drug. In other words, we need to develop models and systems by which we can individualize the dose of ATX so that every person has the same exposure. The data collected from this single-dose pharmacokinetic study were subsequently used to build a dosing algorithm to determine a dose that would lead to a desired exposure, defined initially as the peak concentration achieved (C_{max}) with *a priori* knowledge of a child's CYP2D6 genetic information, which can be used to predict activity, in addition to other patient characteristics such as weight and obesity status. The process for validation of this model, termed the **i**ndividualized **G**enetic and **O**ntogenic **P**harmaco**k**inetics algorithm (iGO-PK), is described in the ATX PBPK Model Validation Study (Appendix A). The major strength of this dosing strategy is that it allows us to ensure that drug exposures do not greatly differ in children, and therefore allows us to describe quantitatively how genetic variability at the drug target alone affects ATX pharmacodynamics and clinical outcomes.

3.3 Study Significance

Prior studies evaluating variability in drug response are confounded by the highly variable ATX exposures between children, even when given the same weight-based dose, and hence are difficult to interpret (e.g., is a patient truly non-responsive to the medication, or is "non-response" simply due to having inadequate concentrations of the medication in the body). In this investigation we propose to individualize ATX dosing in a cohort of children with ADHD to a standardized exposure goal (based on the maximum concentration achieved, or C_{max}), initially using a iGO-PK model derived from our original genotype-stratified PK study, and assign responder or non-responder status according to response criteria established *a priori* by the clinical members of the study team. Subsequent analyses will investigate predictors of response (metabolomics analysis of pretreatment plasma samples; assessment of biomarkers of ATX inhibition of SLC6A2, such as dihydroxyphenylglycol (DHPG) concentrations and pupil diameter and eye-tracking studies; *SLC6A2* genotype; pharmacokinetic-pharmacodynamic (PK/PD) analysis of ATX and DHPG data, etc), as well as differences in exposure→response relationships for different DHPG strata or *SLC6A2* genotypes. In addition, the dosing algorithms (iGO-

PK) will be updated and revised as the body of pharmacokinetic data increases through the duration of the study.

4.0 Study Endpoints and Rationale

4.1 Primary Study Endpoints

The primary aims of this study focus on characterizing the relationship between changes in pharmacodynamic markers and clinical outcome measures, when inter-subject variability in ATX pharmacokinetics has been minimized (Fig. 3). ATX pharmacodynamics will be measured using plasma DHPG and metabolomic data, pupillometry during attention tasks, and changes in blood pressure and heart rate . Clinical outcomes will be assessed by standardized ADHD measures used in a clinical setting, of which include: NICHQ Vanderbilt Assessment Scale (ADHD symptom rating scale); Strengths and Difficulties Questionnaire; Child Behavior Checklist; the School Intervention Questionnaire; Medication Side Effects; and Clinical Global Impression. Some of these questionnaires will also capture comorbidity information, such as Oppositional Defiant Disorder (ODD). We will also use the Kiddie Schedule for Affective Disorders and Schizophrenia for School-Age Children – Present and Lifetime Version (K-SADS-PL) or K-SADS-COMP (the computerized version of the K-SADS-PL) to diagnose ADHD and commonly associated comorbidities. The K-SADS-PL is a semi-structured interview that is extensively used in clinical psychology research to diagnose an extensive array of psychopathology based on the DSM-5. Given the length of this interview, common research practices are to select only the modules that are expected to be pertinent to the study participant (Black et al., 2012; Kariuki et al., 2018; Caudle et al., 2020). The K-SADS-PL modules selected for this research study are as follows: agoraphobia, separation anxiety disorder, social anxiety disorder/selective mutism, specific phobias, generalized anxiety, obsessive compulsive disorder, and post-traumatic stress disorder. The other research comorbidities or impairments that we will assess for and associated psychometric tests we will use are as follows: impairment in executive function using the Behavior Rating Inventory of Executive Function (BRIEF); anxiety using the Screen for Child Anxiety and Related Disorders (SCARED); intelligence quotient using the Kaufman Brief IQ Test (KBIT); and sluggish cognitive tempo using the Barkley Sluggish Cognitive Tempo Scale (SCT). Pharmacodynamic data will be obtained: baseline (Research Visit 1); when ATX is first initiated in a participant with the first dose (Research Visit 2); when the participant has taken the medication chronically and thus, expected to be at steady-state (Research Visit 3); and at the end of study when the dose has been optimized for the participant (Research Visit 4). Concurrent pharmacokinetic analysis will take place to ensure that ATX plasma and urine exposure variability has been minimized using methods described in a previous ATX pharmacokinetic study conducted at Children's Mercy Hospital (Brown et al., 2016). Additionally, we will measure ATX and metabolites in sweat as surrogate for fraction of drug unbound (Marchei et al., 2012; Marchei et al., 2013). The clinical ADHD questionnaires will be administered at the Intake Visit and at follow-up visits following initiation of atomoxetine treatment (6-weeks, 12-weeks, and

18 weeks). These questionnaires ask the participant's parents and teachers to rate behaviors characteristic of ADHD. Changes in other comorbidities associated with ADHD, such as anxiety or sluggish cognitive tempo, will be evaluated at Research Visit 1 and at Research Visit 4. We will also simultaneously monitor side effects of ATX, another measure of clinical outcomes, and categorize study participants on their ability to tolerate ATX. Adverse events will be assessed at all encounters between the study personnel and study participant. At Research Visit 4, we will also administer the Dietary Survey Questionnaire, given that long-term dietary habits may affect both ATX pharmacokinetics and pharmacodynamics.

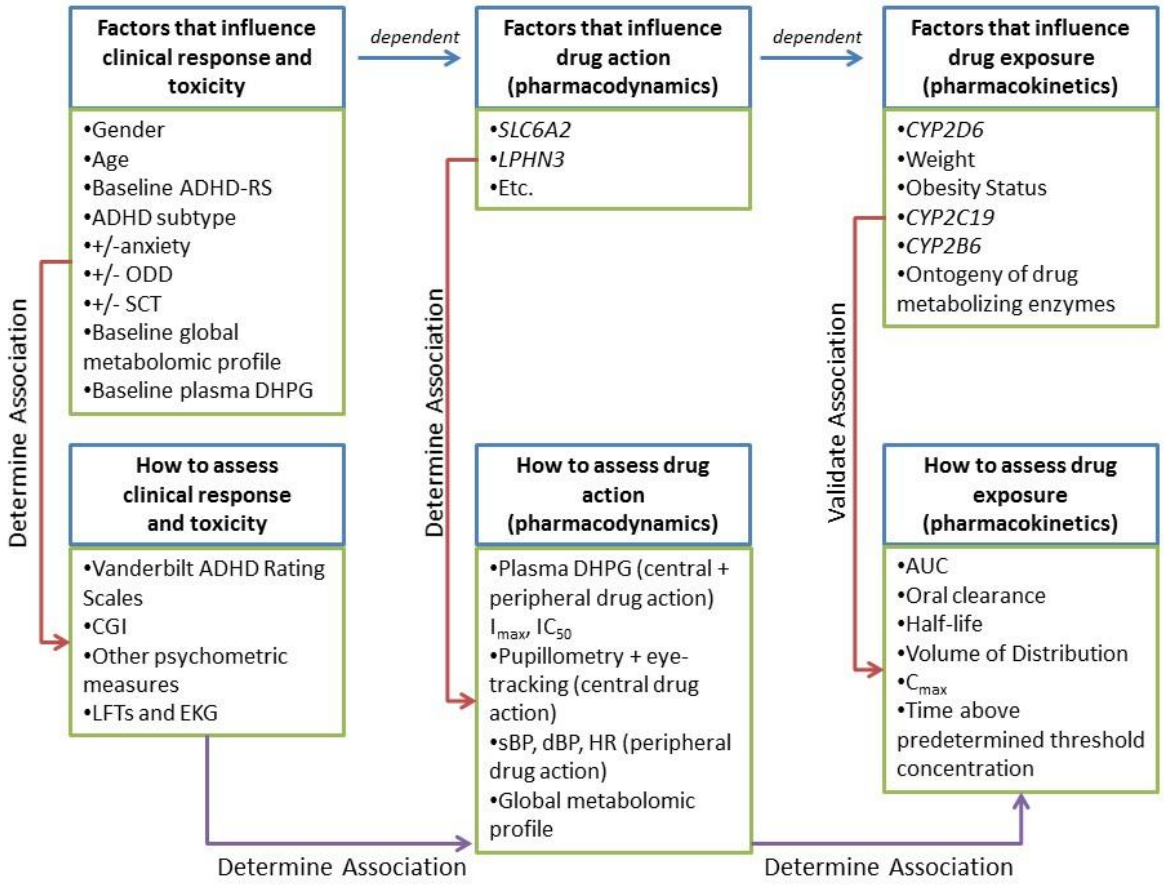


Fig 3. - Analysis of Study Endpoints

ATX is thought to exert its therapeutic effect in ADHD through inhibition of the presynaptic norepinephrine (NE) transporter (NET) encoded by the *SLC6A2* gene. NE is involved in several neuropsychological functions, including visual attention, general alertness, sustained attention, learning and memory and executive functions; dysregulation of NE homeostasis has been implicated in the development of ADHD. Consequently, there has been considerable interest in *SLC6A2* as a candidate gene for ADHD, but the results of studies investigation associations between *SLC6A2* allelic variants and ADHD and related phenotypes have been inconsistent and discordant. Of

interest is rs28386840, also known as *SLC6A2 -3081 A>T*, in the regulatory region of the *SLC6A2* gene. The minor -3081 T allele has been associated with a reduction in promoter activity, which would result in lower expression of the NET transporter.

In vitro, as well as *in vivo* animal and human studies have provided evidence that plasma concentrations of 3,4-dihydroxyphenylglycol (DHPG) are derived predominantly from NE inside nerve cells. As a consequence, inhibition of NE reuptake by ATX is accompanied by decreases in NE inside presynaptic nerve cells, and subsequently, decreases in plasma DHPG concentrations. Published studies in adults indicate that DHPG concentrations in plasma progressively decrease for the first four hours following ATX administration and gradually recover in 24 hours; comparable data in children with ADHD receiving ATX have not been reported. In contrast, single doses of a structurally related norepinephrine reuptake inhibitor, edivoxetine to adolescents produced an average reduction in baseline DHPG of 28% across all tested doses. With chronic dosing, the reduction in DHPG concentration persisted for at least 12 hours. Of particular interest is the observation that no reduction from baseline was observed in two subjects, both of whom had relatively low baseline concentrations of 431 pg/ml and 657 pg/ml and for whom the lack of response could not be attributed to low drug exposure; no information regarding clinical response was provided. These considerations provide the basis for assessing DHPG as a potential biomarker of clinical response to ATX, and specifically for investigating the relationship between the magnitude of reduction in plasma DHPG concentration as a function of ATX concentration, and how it translates into clinical response in children with ADHD. As ATX selectively binds to NET in the prefrontal cortex and modulates the reuptake of not only norepinephrine, but also dopamine, we propose to conduct a comprehensive analysis of neurotransmitters, including norepinephrine, dopamine, DHPG and several of the breakdown products (see Secondary Aims). (Kielbasa et al., 2009; Kielbasa and Lobo, 2015; Kielbasa et al., 2015). A similar approach has been applied to serotonin selective reuptake inhibitors for adults with major depressive disorder. Responders and non-responders citalopram/escitalopram displayed significantly different baseline metabolomics signatures (Kaddurah-Daouk et al., 2011).

In addition to DHPG as a pharmacodynamics marker, we also propose to investigate a biomarker of SLC6A2/NET inhibition in the brain. The SLC6A2/NET transporter is expressed both in the central nervous system (i.e., the brain) and peripheral nervous systems (i.e, the rest of the body), the measurement of change in plasma DHPG likely represents a combination of drug binding at both sites. To differentiate between SLC6A2/NET peripherally and in the brain, we will collect additional data. To assess peripheral inhibition we will also monitor heart rate and blood pressure as measures of ATX drug action in the peripheral nervous system. Current information suggests that controlled release of norepinephrine by a region of the brain called the locus coeruleus is responsible for optimal attention and alertness during cognitive tasks. Functional MRI data collected in 20 healthy male adults administered with a single dose of 40 mg

atomoxetine found activation in the right inferior frontal gyrus, a region that is associated with inhibitory control (Chamberlain et al., 2009). Furthermore, there is a U-shaped relationship between norepinephrine release and task performance insofar that too little or too much norepinephrine leads to decreased attention and problematic task performance, while low-to-moderate levels of norepinephrine are considered optimal for maintaining alertness and attention during cognitive tasks. Moreover, pupil diameter changes mirror norepinephrine release in the locus coeruleus in non-human primates (Gilzenrat et al., 2010). In a carefully controlled experiment, where light levels were held constant during a well-established test of attention, one research group demonstrated that more difficult cognitive tasks were associated with decreases in pupil diameter. In contrast, larger pupil diameters were associated with exploratory behavior, poorer task performance and disengagement. Thus, the magnitude of increase in pupil size in response to standardized tasks during ATX treatment offers potential as a biomarker of central inhibition of NET activity. See the section below for further details regarding this test of attention.

Attentional Orienting and Pupil Size Measurement

Eye movement and dilation in response to attentional-shift requiring tasks will be used to measure ATX drug action in the CNS. The tasks are based on a modified Posner Cuing Task, which measures both endogenous and exogenous orientation of attention, which correspond to tonic and phasic locus coeruleus activation. This is an established non-invasive method to measure norepinephrine signaling in the brain. Assessment will take place at both the ADHD Specialty clinic and at the PCRU. Participants will be asked to sit 57 cm from a computer monitor, which is housed in a light dome to maintain a consistent level of illumination. Volunteers will be asked to place their chin in a mounted chin rest to stabilize the facial area. Each study participant will be asked to complete target localization and target discrimination tasks. Both tasks begin with the participants looking at two white squares on black background. In between these two boxes will be a white fixation cross. Participants will be asked to begin each trial by focusing on the fixation cross. Next, a cue is given. In this case, one of two boxes brightening will serve as the cue. Following the cue, a target will appear in one of the two boxes by presentation of an “O” or a “Q” in one of the two boxes. Participants will be asked to push colored buttons on the game controller that correspond with the correct answer when presented with the target. If the target appears in the box that was brightened, this is defined as a valid trial. When this does not occur, this is considered an invalid trial. Half of the trials will be designed to be invalid. The valid and invalid cues serve to measure directing of attention by study participants. Volunteers will be measured for their ability to localize the target (*e.g.* did the target appear on the left or right box?) and discriminate the target (*e.g.* is the target a “Q” or an “O”?) following the cue. In localization tasks, if the target appears on the left side, volunteers will be asked to push the blue button. When the target is on the right side, a successful trial would mean the volunteer pushed the red button. In discrimination tasks, if the target presented is a “Q” the volunteer will be asked to push the red button, while an “O” target means the volunteer should press the blue button. Volunteers will be given

the opportunity to practice the tasks (3 trials). Measurements will be recorded for 162 trials which for both localization and discrimination tasks. Eye-moment, dilation, and reaction time will be measured during all tasks. Please see **Figures 1 and 2** in the Appendix H of this document. **Figure 1** illustrates an example of a valid and invalid trial for successful completion of the localization task, while **Figure 2** illustrates an example of a valid and invalid trial for successful completion of the discrimination task.(Aston-Jones and Cohen, 2005; Alnaes et al., 2014)

Secondary Study Endpoints:

The secondary aims focus on: exploring baseline patient characteristics that may predispose an individual to have beneficial clinical response from ATX therapy; refinement of the iGO-PK dosing algorithm; exploration of the role of the microbiome in impacting ATX pharmacokinetics and pharmacodynamics; validation of a urinary CYP2D6 biomarker; and exploring ways improve study participant understanding of research procedures. A previous study with an anti-depressant, citalopram, suggests that the ability to respond positively to a medication may be predicted given an understanding of the individuals' composite of signaling molecules, i.e. the global neurotransmitter profile. We are interested in comparing the baseline global neurotransmitter profile between children who do and do not respond. We are also interested in understanding how the neurotransmitter profile changes with chronic ATX use and correlating these changes with clinical benefit of toxicity. To do this, we will evaluate a panel of approximately 30 neurotransmitters (Gupta et al., 2016)(refer to Appendix I for example list) prior to treatment and during treatment with ATX.

Because there is evidence that the gut microbiome can to influence host neurotransmitter concentrations (Petra et al., 2015), we would like to evaluate this as a possible confounder to the global neurotransmitter profile. Given that the oral and gut microbiome landscape correlate well with one another (Zhang et al., 2015) we will also sample saliva to account for factors that could contribute to an individual's neurotransmitter make-up.

The pharmacokinetic data that we collect on each individual participant allows us to provide more information for modeling, and for further refinement. We recognized in our previous pharmacokinetic study that CYP2D6 activity, as measured using a phenotypic marker (urinary dextromethorphan/dextrorphan ratio), was the best predictor of ATX exposure. This urinary ratio was associated with ~75% of variability observed in ATX exposure in the study. We would like to explore a urinary biomarker that is reflective of CYP2D6 activity that has previously been studied (Tay-Sontheimer et al., 2014) for possible inclusion in the iGO-PK model. Validation of this biomarker with ATX exposure is necessary prior to consideration as a variable in the iGO-PK algorithm.

Lastly, we recognize that our patient population will include young children and that lengthy consent forms may not be optimal for conveying information. We plan to

distribute a supplemental illustrated booklet (please refer to Supplementary Documents, “U54 Study Booklet”) explaining study procedures along with a standard consent form to all participants approached for the study. A short questionnaire will accompany the booklet, asking study participants and their families to evaluate the study booklet (please refer to Appendix G) in helping with their clarity and comprehension of the study.

4.2 Primary Safety Endpoints

Participant baseline health, including behaviors, will be recorded at Research Visit 1 with a physical exam, baseline electrocardiogram, medical history, and concomitant medication history. Additionally, baseline health and behavior data will be captured at the Intake Visit with the Study Physician. Adverse events will also be closely monitored. These are defined by Children’s Mercy Hospital Reportable Events Research Policy and Procedures as “untoward or unfavorable occurrences in a human subject, including an abnormal sign [e.g., abnormal physical exam or laboratory finding], symptom, or disease, temporally associated with the subject’s participation in the research, whether or not considered related to the subject’s participation in the research.” Additionally, an adverse event is also defined as behaviors or symptoms that occur with increased frequency or severity as consequence of the subject’s participation in the research study.

Both types of adverse events will be documented in this study. Adverse events are reported in many ways in this study. These include but are not limited to: emails to study coordinators/clinical trial manager, phone calls on study cell phone, phone calls to ADHD Clinic or Developmental and Behavioral Clinic, reported to study physician at clinic visit, reported on Medication Side Effects questionnaires, reported at research visits when asked, or via a Cerner notification of admission to the hospital to the study physician. Additionally, adverse events will be assessed and documented at Research Visit 2, Research Visit 3, Research Visit 4, 6-week Visit, 12-week Visit, and 18-week Visit.

Adverse events will be categorized and reported to the IRB as outlined in CM Policy regarding adverse events. These events will be documented consistently, regardless of how they were reported. This includes the delegation of an individual member of the study personnel to handle all adverse event reporting. All adverse events will be recorded at all research visits and in between visits as needed on the Adverse Event CRF kept in the subject’s study binder.

In addition, subjects will be classified based on their tolerance of ATX. A participant is considered tolerant of ATX if the side effects did not lead to discontinuation of ATX therapy. A participant is considered intolerant of ATX if the side effects lead to the discontinuation of medication and withdrawal of a subject from the study.

5.0 Study Intervention/Investigational Agent

5.1 Description

ATX is a norepinephrine reuptake transporter inhibitor which is approved by the FDA for use in children with ADHD. The medication displays high inter-individual pharmacokinetic variability, which has been associated with *CYP2D6* genotype. An individual's predicted CYP2D6 phenotype can be estimated from their genotype. This estimation is quantified as a CYP2D6 activity score (AS). The CYP2D6 predicted phenotype and the participant's height, weight, habitus, and gender are used to inform the ATX dosing tool, the individualized genetic and ontogeny pharmacokinetic model (iGO-PK). Both the CYP2D6 genotype-predicted phenotype activity score and iGO-PK are described below. Doses that are expected to result in $C_{max,ss}$ values of 400 ng/mL, 600 ng/mL, and 800 ng/mL will be calculated for each participant. Study physicians will start with a dosing regimen that is predicted to achieve a $C_{max,ss}$ of 400 ng/mL and will titrate the dose to reach $C_{max,ss}$ values of 600 ng/mL or 800 ng/mL according to participant response. Study physicians may choose to titrate slowly to $C_{max,ss}$ of 400 ng/mL when initiating ATX therapy to avoid side effects.

CYP2D6 Genotype-Predicted Phenotype Activity Score
Participants will be assigned a CYP2D6 phenotype based on the CYP2D6 activity score, which is calculated using *CYP2D6* genotype information. These data are used to determine the design and length of stay of the PK studies. Calculation of an activity score is done first by determining the functional score for each copy of CYP2D6 present in a patient.

When this study was first submitted, a *CYP2D6* allelic variant is given a score of 1 if it is considered fully functional when compared to the wild-type reference sequence. If an allelic variant is reported to result in decreased activity, the functional score assigned is 0.5. It is important to note that a functional score of 0.5 does mean the allelic variant exhibted 50% reduction in activity; rather this designation is meant to convey that a variant had less activity compared to wild-type, while at the same time not being devoid of catalytic capability. An allelic variant that results in complete loss of CYP2D6 activity was given a functional score of 0. The activity score is a summation of the functional score for all *CYP2D6* gene copies. An activity score of 0 or 0.5 would have originally placed an individual in the poor metabolizer (PM) or intermediate metabolizer (IM) study design (i.e. Study Design 1), and a score ≥ 1 would assign an individual to an extensive metabolizer (EM) study design (i.e. Study Design 2). It should be noted that individuals in the latter study design were comprised of EM1s, EM1.5s, EM2s, and ultra-rapid metabolizers (UM). (Gaedigk et al., 2008; Hicks et al., 2013; Twist et al., 2013).

New standardized guidelines for *CYP2D6* genotype-to-phenotype interpretation were recently published in 2020 by the Clinical Pharmacogenetic Implementation Consortium

(CPIC) (Caudle et al., 2020), which recommended a different functional score for the *CYP2D6*10* allelic variant (reduced score to 0.25) and also different phenotype assignments based on activity score. The different phenotype designations are listed in the table below.

Inferred CYP2D6 Phenotype	Consensus contiguous CYP2D6 Activity Score Definition	Examples of CYP2D6 diplotypes for consensus translation method[1]
Ultra-rapid Metabolizer (UM)	> 2.25	**1/*1xN*, **1/*2xN*
Normal Metabolizer (NM)	1.25 ≤ x ≤ 2.25	**1/*10*, **1/*1*, **1/*2*
Intermediate Metabolizer (IM)	0 < x < 1.25	**4/*10*, **41/*41*, **1/*4*
Poor Metabolizer (PM)	0	**4/*6*, **3/*5*

Table is edited from (Caudle et al., 2020).
[1]Functional scores for the reference: **1* = 1, **2* = 1, **3* = 0, **4* = 0, **5* = 0, **6* =0, **10* = 0.25, **41* = 0.5.

Additionally, data generated from studies conducted at Children's Mercy Hospital has suggested that changes in study design assignment are warranted, particularly for children with an activity score of 1, which could be a result of having two reduced function CYP2D6 alleles (0.5/0.5) or one functional copy and one non-functional copy of CYP2D6 (1/0). The studies mentioned above are: Ped Develop Pharm U54 (IRB# 16010069) and this study, ATX-PBPK-PD-Clinical Outcomes (U54) (IRB# 16100728). Initial analysis suggests that 0.5/0.5 children have higher ATX exposure compared to 1/0 children, suggesting slower ATX apparent oral clearance. Additionally, 1/0 children more closely resembled children with two fully functional copies of CYP2D6 in their pharmacokinetics.

This preliminary data generated from previous ATX studies and the revised CPIC guidelines have prompted the study team to revise study design assignment. Therefore, to ensure that each participant's ATX pharmacokinetics are accurately characterized, all children with an activity < 1 and those who have a two reduced function alleles will be assigned to Study Design 1. All other participants will be initially assigned to Study Design 2.

Additional genotype testing may be conducted if the participant's pharmacokinetic profile and genotype are inconsistent with one another. The additional genotype testing, if needed, will be documented on the provided CRF.

iGO-PK Model
This method was developed using data generated from a previous ATX single-dose PK study conducted at Children's Mercy Hospital (Brown et al., 2016). This model stratifies participants into two distinct populations: individuals who are CYP2D6 PM and those who non-PMs, i.e. IM, EM1, EM2, and UM (read above for more details on *CYP2D6* genotype and phenotype prediction). With the new changes to *CYP2D6* genotype-to-

phenotype interpretation and study design, the only major change is that children with two reduced function alleles will be modeled as an IM. Analysis of the previous PK study found that in non-PM participants, the dose-exposure relationship was best described using a two-compartment model with first order absorption and first order clearance. In PM participants, dose-exposure was best described using a one compartment model with first order absorption, first order clearance, and a secondary peaking component. Additionally, lean body weight (LBW) and obesity status (positive or negative) variables were found to significantly impact variability in ATX exposure in both models. Retrograde extrapolation of an initial dose of ATX for PK Study 1 will be determined by setting the desired C_{max} goal at steady-state ($C_{max,ss}$) to 400 ng/mL, and incorporating patient specific parameters CYP2D6 AS, LBW, and obesity status. The actual dose dispensed will be as close as possible to the model generated dose, given the available atomoxetine capsule formulations. Additionally, we will also consider the most convenient combination of capsules for a child to take and how a prescriber would dose this medication in a naturalistic setting. During the course of the study, we will also be monitoring clinical response and toxicity. The exposure ($C_{max,ss}$) may be titrated down to below 400 ng/mL to reduce toxicity or up to 800 ng/mL to increase response as tolerated.

5.2 Drug Handling

The study drug, ATX, will be stored, prepared, labeled, and dispensed by the CMH Investigational Drug Service (IDS) pharmacy. Study file documentation, accountability, and records for dispensing of atomoxetine will be maintained by IDS pharmacy personnel.

The study physician will submit a signed and dated prescription for ATX to IDS Pharmacy. Alternately, designated study personnel will deliver a prescription that has been signed and dated prescription by the study physician to IDS Pharmacy. A copy of all prescriptions will be retained in the participant's study binder. IDS Pharmacy will deliver the ATX prescription to the PCRU for Research Visit 2. Subsequent prescriptions for ATX will be filled as needed and will typically occur in conjunction with 6-week, 12-week, and 18-week visits. Medication refills may also occur outside of the 6-week, 12-week, and 18-week visits. Medication refills may be handed off or delivered to the participant to reduce foot traffic at Children's Mercy Hospital, as needed. Dose adjustments may also occur outside of the 6-week, 12-week, and 18-week visits.

All investigational drug will be returned to the study team at the end of the study for disposal by the IDS pharmacy, unless the child and their family would like to continue using ATX. In addition, if the participant would like to discontinue the study drug, but he or she will require an ATX wean to reduce risk of side effects from abrupt discontinuation as directed by his or her study physician, the participant may keep the remaining drug to complete this wean.

Refer to the Investigational Drug Service Policy and Procedure (Last Revision 04/2020) for the standard operating procedure (SOP) outlining details regarding the control of the study drug used in this protocol. Transportation of study drug will be handled by both IDS pharmacy and study personnel. Refer to the Investigational Drug Product Transport Policy (Last Revision 12/2019.) The locations of drug transportation and how this will be conducted are summarized below. Further details for drug handling may be found in Appendix J, "Study Drug Timeline."

- IDS to participant at CMH Adele Hall – IDS personnel or designated study team members, as specified on the Delegation of Authority, may transfer investigation drug from IDS pharmacy to clinics located in CMH Adele Hall. Direct hand-off of medication to the participant or their LAR may also occur outside of CMH Adele Hall, such as at the front entrance of the hospital, to reduce foot traffic. Lastly, IDS personnel may transfer the investigational drug to the CMH Adele Hall outpatient pharmacy for the study participant/family to pick up.

- IDS to PCRU – IDS personnel or designated study team members may transfer investigation drug from IDS pharmacy to the PCRU.

- IDS to Children's Mercy Hospital Kansas (CMK) – IDS personnel may use a courier service to deliver a filled prescription to the outpatient pharmacy at CMK for study participants to pick up at their convenience.

- IDS to participant's residence – IDS Pharmacy may ship medication to the participant's residence. The party responsible for shipping medication will be determined by applicable IDS policies at the time of shipment. During the consent process, the participant and/or guardian providing consent will provide written authorization for IDS Pharmacy to ship study medication to the participant's home, if needed.

6.0 Inclusion and Exclusion Criteria

6.1 Inclusion Criteria

- Males and females 6-18 years of age at the time of enrollment
- Diagnosis of ADHD, as confirmed or inferred by a Study Physician at Intake visit.
- Intention of the Study Physician to begin therapy with ATX at intake visit
- Willing to provide written permission/assent to participate
- ADHD Medication Status is one of the following:
    - ADHD medication naïve or not currently taking ADHD medication including stimulants, α_2-agonists, and ATX, or
    - Currently taking a stimulant for ADHD and is willing to wash out of stimulants prior to starting ATX. This washout is also approved by a Study

Physician, or other qualified study personnel (see Section 11.0 for Procedures Involved).

Exclusion Criteria

- An IQ < 70 or evidence of intellectual disability from review of EMR
- A diagnosis of Autism Spectrum Disorder
- Inability or unwillingness to have blood drawn as described in the protocol schedule of events and consent
- Underlying risk for cardiotoxicity, such as presentation of structural cardiac abnormalities, cardiomyopathy, or arrhythmias
- Clinically significant abnormal safety laboratory values as determined by treating physician
- Diagnosis that may cause abnormal absorption or gastric emptying, such as reflux, inflammatory bowel disease, or Crohn's disease
- For females, a positive urine pregnancy test
- Previous history of adverse drug reaction to ATX
- Use of drugs known to inhibit CYP2D6:
  - Concurrent therapy with sertraline, venlafaxine, imipramine, nortriptyline, quinidine, propafenone, cimetidine, tamoxifen, bupropion, over-the-counter medications containing diphenhydramine, codeine, tramadol, hydrocodone, or oxycodone
  - Concurrent or previous therapy with fluoxetine or paroxetine in the last 2 months
  - Concurrent or previous therapy with terbinafine in the last 6 months
- Unwillingness or inability to washout of stimulant ADHD medications
- Concurrent or recent use of other psychiatric/behavioral health drugs including SSRIs, SNRIs, antipsychotics, anxiolytics, anti-epileptics, and α_2-agonists that would impact the participant's pharmacokinetic and/or pharmacodynamic baseline
- Subject is considered by PI to be unsuitable for participation in the study for any reason

6.2 Special Populations

We will include individuals who are not yet adults (children and teenagers who up to 18 years of age). We will exclude adults unable to consent, pregnant women, prisoners, and wards of the state.

7.0 Vulnerable Populations

7.1 Additional Safeguards

This research study aims to enroll children and teenagers with ADHD between the ages of 6-18 years of age. Because children are vulnerable to coercion or undue influence, appropriate provisions will made to obtain and document:

- Permission to participate in the study from the participant's legally authorized representative.
- Assent to participate in the study from the participant. An illustrated study booklet, see supplemental document, "U54 Study Booklet," that complements the permission/assent form will be provided for children to better understand study procedures.
- For children age 7 and older, a signature is required to document assent to study procedures. Signatures may be waived if the child is capable of assenting but unable to sign. Documentation of the reason is required.
- For children under age 7, only the permission of the legally authorized representative is required for study participation. A study participant under age 7 who wishes to revoke assent and withdraw from the study may do so at any time.
- The PI may also remove a child under the age of 7 without the assent of the legally authorized representative for reasons that include, but are not limited to, safety, participant not responding well to medication, or issues with participant non-compliance, etc.

8.0 Local Number of Subjects

8.1 Multiple Cohorts

A total of 160 subjects will be accrued locally. The study does not contain multiple cohorts.

8.2 Target Participants

The research proposal targets 160 participants who are expected to be enrolled and screened. The target number of subjects needed to complete research procedures is 120. Participants who complete at least Research Visits 1-3, the Intake Visit, and the 6-Week Visit with the Study Physician will be considered to have "partially completed" the study. Completion of these visits will include obtaining baseline measurements, confirmation of eligibility, single and steady-state PK and PD measurements, and at least one post-treatment clinical response assessment. Since data from these participants will meet the study aims, those who partially complete the study will be counted toward the 120.

9.0 Prescreening and Recruitment Methods

9.1 Recruitment Overview

Participants will be recruited over four years from patients and families seeking care for ADHD at Children's Mercy Hospital and Clinics. These participants may be recruited from six different tracks:

Track 1: Patients referred to ADHD Clinic on College Boulevard
Track 2: Patients at Pediatric Care Clinic (PCC) at the Broadway and West locations
Track 3: All other patients seeking ADHD care from Children's Mercy Hospital and Clinics providers
Track 4: External provider referral to Children's Mercy Hospital and Clinics and patient self-referral
Track 5: Children's Mercy Summer Treatment Program (STP) participants

Active Recruitment: Participants will be actively recruited from **Track 1** and **Track 2.** Active recruitment entails study personnel performing regular chart review of patients seen through **Track 1** and **Track 2** (see section 9.2). These two clinics were selected for active recruitment because patients and families generally seek care for ADHD at these Children's Mercy Hospital and Clinics locations. As such, these clinics represent the largest population of potentially eligible participant population.

Passive Recruitment: Participants will be passively recruited from **Track 3**, **Track 4** and **Track 5**. With passive recruitment, potential participants or their healthcare providers self-identify or refer, respectively, potential research participants to the study. Potential participants may learn about this study from information posted on the internal Children's Mercy Hospital website and on the Children's Mercy Research Institute (CMRI) public facing website. Social media platforms, such as Twitter and Facebook, may be used to disseminate study information. The social media platforms will reference or link to study information available on the CMRI website. Use of social media to share study information will be compliant with any existing CMH policies. The use of social media to share study information will not begin until these policies have been developed. Presentation of study information and content posted on the CMRI website will be at the discretion of the Digital Team, Research Administration, and the research team. Modification of images may be applied by the Digital Team and Research Administration to in be consistent with the aesthetic of the CMRI webpage. All study information will be approved by the IRB (Supplementary document, "CMRI Website Information")

Track 3 allows a mechanism for potential participants within the Children's Mercy Hospital and Clinic systems to participate in this research study, who are not actively being recruited. **Track 4** allows a mechanism for potential participants outside of the

Children's Mercy Hospital and Clinic systems to participate in this research study. The STP is a summer program for children with ADHD where both behavioral and medical therapies are provided. **Track 5** provides a mechanism for STP participants to enroll in this study.

9.2 Chart Review

The following prescreening information will be identified from chart review when possible for Tracks 1-5:

- 6-18 years of age
- Diagnosis or indication of intellectual disability (IQ <70)
- Diagnosis or indication of Autism Spectrum Disorder (ASD)
- Diagnosis or indication of underlying risk for cardiotoxicity, such as presentation of structural cardiac abnormalities, cardiomyopathy, or arrhythmias
- Diagnosis or indication of disease that may cause abnormal absorption or gastric emptying
- Previous history or indication of prior adverse drug reaction to ATX
- Current or previous therapy with drugs used to treat ADHD
- Concurrent or previous therapy with drugs known to inhibit CYP2D6
- Concurrent therapy with other psychiatric/behavioral drugs
- Presence of any of the following questionnaires:
  - Child Medical History
  - NICHQ Vanderbilt Assessment Scale Initial Assessment
  - Child Behavior Checklist
  - Strengths and Difficulties Questionnaire
  - School Intervention Questionnaire
  - Please note: any version of the above questionnaires are permissible for chart review, screening, and recruitment given that these only inform the research team of the *likeliness* of participant suitability for the study.

9.3 Participant Notification of Study Opportunity

Participants may be informed of the study opportunity in multiple ways, depending upon the recruitment track.

Active Recruitment: **Track 1** and **Track 2**

1. The potential participant's assigned provider will be contacted regarding eligibility for potential participants meeting eligibility criteria upon initial chart review. For message center template, please see Appendix B. If the provider approves, the family will be contacted by phone and the study

opportunity will be presented (for the phone script, please refer to Appendix C or D).

2. If the family is interested in learning more, study personnel will either email or mail a copy of the study consent or permission/assent form (whichever is applicable) and a recruitment flyer with FAQs (see Supplementary Documents, "Recruitment Flyer") to the family based on their preferred communication method.
    a. Study personnel will specify that the family/potential participant are not to sign the form to provide consent, but rather, the form is used for information purposes at this juncture and study personnel will review this formally with the family/participant if he or she choose to enroll. (See Appendix E for the email/letter sent with the consent or permission/assent form and recruitment flyer.)
    b. This conversation will be documented in an electronic Potential Participant Log located on the Hospital Share Drive in the Clin Pharm folder, U54 sub-folder.
    c. Study personnel will include contact information for the family/participant for any questions, concerns, etc with the email or mailing.
    d. Study personnel will ask permission to make a follow-up call one week after the email/mailing is sent out.
3. A follow-up call will be made one week later.
    a. If families/potential participants are interested, study personnel will proceed to enrollment (section 11.1).
    b. If families/potential participants are not interested, no further contact attempts will be made.
    c. Up to three follow-up calls will be made until the family/participant is reached. All potential participant contact will be documented in the "Potential Participant Log" (see subsection 9.4 below; see Supplementary Documents "U54 Potential Participant Log").

<u>Passive Recruitment</u>: **Track 3**

1. CMH provider notifies study personnel of the potential participant;
2. OR an individual self-identifies themselves or their child, as a potential participant to study personnel and the potential participant is patient of provider at CMH.
3. Study personnel perform a chart review. If upon chart review,
    a. The potential participant meets prescreening criteria, study personnel will contact the family by phone and the study opportunity will be presented (for the phone script, please refer to Appendix D). Subsequent steps are the same as steps 2 and 3 in "Active Recruitment" above.

b. The potential participant does not meet prescreening criteria, study personnel will notify the referring provider or the individual and/or family. If the family would like the results from the prescreen for follow-up care, the study team will write a letter to be given to the participant's primary care provider. (See Appendix M).

Passive Recruitment: **Track 4**

1. An external provider refers their patient OR an individual self-identifies themselves or their child as a potential participant to study personnel and the potential participant is not currently being seen by a provider at CMH. Potential participants and families will be made aware of the study through listing on ClinicalTrials.gov, posting of the study on the CMRI externally facing website, the CMRI website, or social media referencing the CMRI website.
2. Study personnel will contact the participant by phone to introduce the study.
    a. Study personnel will either email or mail a copy of the study consent or permission/assent form (whichever is applicable) and a recruitment flyer with FAQs (see Supplementary Documents, "Recruitment Flyer") to the family based on their preferred communication method along with the Recruitment Letter or Email (Appendix E).
        i. This conversation will be documented in an electronic Potential Participant Log located on the Hospital Share Drive in the Clin Pharm folder, U54 sub-folder.
        ii. Study personnel will include contact information for the family/participant for any questions, concerns, etc with the email or mailing.
        iii. Study personnel will ask permission to make a follow-up call one week after the email/mailing is sent out.
    b. Study personnel will specify that the family/potential participant are not to sign the form to provide consent, but rather, the form is used for information purposes at this juncture and study personnel will review this formally with the family/participant if he or she choose to enroll.
    c. A follow-up call will be made one week later.
        i. If families/potential participants are interested, study personnel will conduct a phone consent or via telehealth appointment.
            1. Families may fax signed and document permission/assent or consent forms. Study personnel must instruct families to bring original signed and dated forms at Research Visit 1.

2. Families may also mail signed and document permission/assent or consent forms to designated study personnel.
3. Once a faxed copy or a mailed original version of the consent has been received, the study team will proceed with research procedures.

ii. If families/potential participants are not interested, no further attempts to contact will be made.

3. After enrollment and receipt of faxed or mailed signed and dated permission/assent form or consent form, study personnel will send out questionnaires to gather preliminary information.
    a. The following questionnaires will be sent via CHADIS. These questionnaires are administered as part of the standard-of-care intake process at ADHD Clinic on College Boulevard. Information collected on these questionnaires provides necessary information for prescreening to guage the likelihood these children meet the eligibility criteria that would be identified from chart review in other tracks. The questionnaires are:
        i. Child Medical History (parent informant)
        ii. NICHQ Vanderbilt Assessment Scale – Initial Assessment (parent and teacher informant)
        iii. Child Behavior Checklist (parent and teacher informant)
        iv. Strengths and Difficulties Questionnaire (parent and teach informant)
        v. School Intervention Questionnaire (teacher informant)
        vi. Please note that all questionnaires (and other versions of these questionnaires) are validated clinical measures that aid in the diagnosis. However, these data are used in conjunction with Study Physician clinical impressions and do not directly determine a diagnosis. Because this, any version of the questionnaires listed are permissible in aiding diagnosis determination.
    b. After these questionnaires are submitted and returned, study personnel will review and determine if prescreening criteria are met.
        i. If initial prescreening criteria are met, study personnel will proceed to schedule Research Visit 1 (section 11.1).
        ii. If initial prescreening criteria are not met, study personnel will notify the family. If the family would like the results from the prescreen for follow-up care, the study team will write a letter to be given to the participant's primary care provider. (See Appendix M).

<u>Passive Recruitment:</u> **Track 5**

1. All participants and their families at STP will be provided the study information handout (please see Supplementary Documents, “Recruitment Flyer”).
2. A study team member will present study information briefly at the end of a weekly behavioral parent training meeting and answer any parent/participant questions. See Supplementary Documents, “STP presentation” for presentation content.
3. Interested participants may call the study cell phone listed on the handout.
    a. Subsequent steps are the same as steps 2 and 3 in “Active Recruitment” above.
    b. Baseline results from psychometric questionnaires administered prior to beginning STP (as a requirement of participation in the STP program) will be obtained from the participant’s electronic medical record and/or STP research records

9.4 PHI Documentation

PHI will be recorded for potential participants prior to obtaining informed permission/assent/consent during pre-screening procedures outlined above. This information will be stored securely on the Hospital Share Drive under the file name “Potential Participant Log.” (see Supplementary Documents, under “U54 Potential Participant Log.”)The PHI collected will consist of:

- Patient first and last name
- Appointment date and appointment location (PCC or ADHD Clinic)
- Parent Name
- Parent Phone Number

Other non-PHI collected on this spreadsheet will include attempts to call the family including date, time, notes from the conversation, interest, if consent form was sent, follow-up call attempts after consent form was sent, and enrollment status. This spreadsheet of potential participants will only be kept until completion of enrollment to avoid duplicate recruitment.

A partial waiver of HIPAA Authorization (waiver for recruitment purposes only) was approved at the time of the initial protocol approval (see Notification of Initial Approval, 11/29/2016).

Data for participants who are determined to be ineligible for the study after enrollment/upon screening will be stored in the “U54 Master Enrollment Log” and will be documented as “screen fails.” (see Supplementary Documents, under “U54

Master Enrollment Log"). The reason for screen fail will also be documented in the participant's study binder.

10.0 Study Timelines

10.1 Study Duration

- The duration of an individual subject's participation in the study will be approximately 6-8 months.
- The duration anticipated to enroll all subjects and complete participant study procedures is approximately 5 years (June 2022)
- The estimated date for the investigators to complete this study (complete primary analysis) is approximately 5 years from when enrollment began (December 2022).

11.0 Procedures Involved

11.1 Study Design Overview

This is a longitudinal exposure escalation study of atomoxetine in children with ADHD. This study simultaneously measures participants' pharmacokinetic and pharmacodynamic profiles after administration of a single-dose of ATX, approximately 2-8 weeks, and approximately 18 weeks of chronic use as well as the therapeutic efficacy/clinical response after chronic use. The study design is best illustrated by categorizing the chronological visits by phase as illustrated in Figure 4. The phases occur in the following order:

1) Prescreen
2) Enrollment
3) Screen
4) Intervention

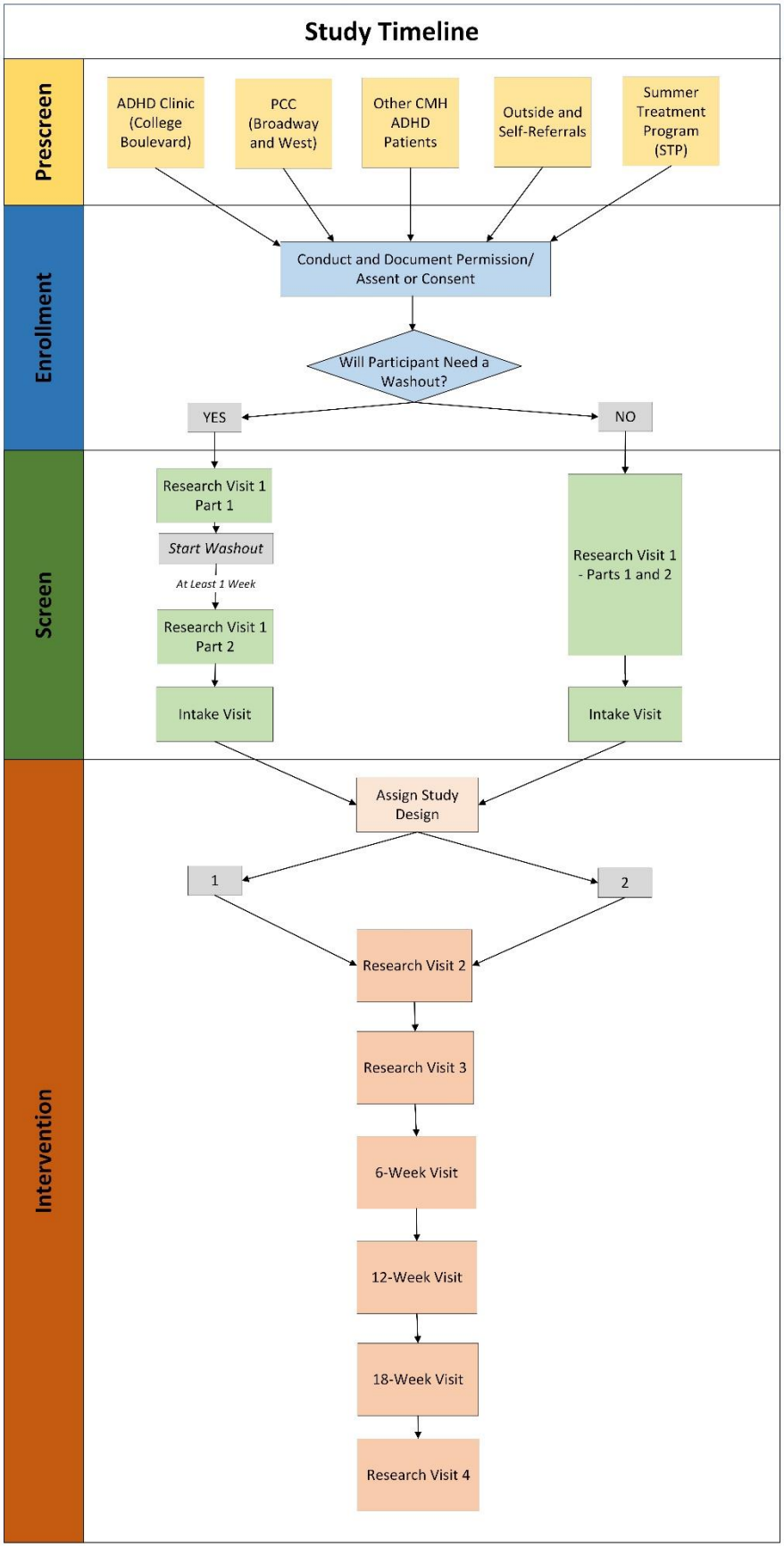


Fig. 4 – Study Design Overview

1. *Prescreen*

   This phase consists of identifying and contacting eligible study participants and presenting the study opportunity. See section 9.0 for details.

   After a family expresses interest in enrolling, study personnel will contact the participant/family to review key parts of the study in preparation for the first research visit. See Appendix F "Study Coordinator/Research Assistant Scheduling Script" for details. This discussion with review:

   a. Frequency and duration of study visits
   b. If a medication washout is required, explanation about washout safety and instruction to not begin the washout until written permission/assent or consent has been obtained
   c. The study drug is only available in capsule form, so the child will need to be able to swallow pills
   d. The study will require multiple blood draws and an IV

2. *Enrollment*

   Enrollment typically takes place during Research Visit 1 Part 1. Enrollment can occur prior to Research Visit 1 Part 1 when a diagnosis of ADHD has not been established or the diagnosis cannot be verified by review of EMR. Additionally, enrollment may occur prior to Research Visit 1 Part 1 by phone or telehealth to minimize in-person procedures to prevent SARS-CoV-2 viral transmission. Research Visit 1 Part 1 visit takes place in the PCRU and includes obtaining informed consent or permission/assent, gathering information about a participant's comfort with needlesticks and pill swallowing, obtaining a blood sample to determine the participant's genotype, collecting medical history, demographics, medication history, and initiating an ADHD medication washout if needed.

3. *Screen*

   After enrollment at Research Visit 1 Part 1, the remaining screening procedures will occur during Research Visit 1 Part 2 and the Intake Visit with Study Physician.

   If study personnel determine no washout will be required, Research Visit 1 Parts 1 and 2 may occur in tandem. However, if a washout will be required, the 2 parts of Research Visit 1 will occur separately. After the participant has completed the washout, he or she will return for Part 2 to initiate screening procedures.

   Research Visit 1 Part 2 takes place in the PCRU and includes obtaining baseline research psychometric measures and baseline safety and research measures to assess for inclusion/exclusion criteria. Psychometric measures will be administered via paper questionnaires, electronically (as with K-SADS-COMP), or obtained using the Comprehensive Health and Decision Information System (CHADIS). K-SADS-COMP was

developed using Microsoft ASP.NET 4.0 framework and Microsoft SQL Server 2016 database. These platforms are stable, reliable and secure. CHADIS is a secure web-based patient questionnaire tool to assist healthcare professionals with their patient clinical care decisions. Parents and caregivers of the study participant are able to complete certain questionnaires online. These results are then imported into the participant's electronic medical record.

The Intake Visit with Study Physician takes place in the Developmental and Behavioral Health Sciences Clinic at the Adele Hall campus, the ADHD Clinic on College Boulevard, or via telehealth. During this visit, the participant and family undergo a routine standard of care visit with a study physician to confirm/infer a diagnosis of ADHD, determine an ADHD subtype, assess for co-morbidities, perform a physical exam, and determine if ATX is an appropriate therapy for the participant. If the Intake Visit occurs via telehealth, all in-person procedures may be omitted. Further details are described in section 11.2. The results from questionnaires will be reviewed. For participants without a formal diagnosis (i.e. documented on participant's electronic medical record), questionnaires must be received from both parent and teacher.

If the subject was seen by a study physician within 6 weeks prior to enrollment and at baseline (meaning not taking any ADHD medication), the visit does not need to be repeated in its entirety. The study physician may amend his or her visit note in the EMR to document the following for research purposes:

a. Confirm/infer ADHD diagnosis
b. Assess and confirm/infer diagnosis of other comorbidities
c. Assess and confirm appropriateness of atomoxetine therapy for participant
d. Complete Clinical Global Impression (CGI).

After this, the Principle Investigator(s) (PI) will review information from these visits to determine if the participant meets all eligibility criteria.

4. *Intervention assignment*

Once all inclusion and exclusion criteria have been assessed during screening procedures, the PI will document that the subject meets all eligibility criteria to qualify for study participation. The PI will then assign the participant to one of three study designs based on his or her CYP2D6 activity score (see section 5.1). The length and data collection procedures of Research Visit 2, Research Visit 3, and Research Visit 4 will vary based on the participant's CYP2D6 activity score, genotype, and frequency of dosing. During the course of the study following Research Visit 2, some study particpants may be changed from the usual once daily dosing to twice a day (bid) dosing per their study physician. These participants initially assigned to Study Design 2 may require switching to Study Design 3 as determined by the research team to capture pharmacokinetic data

of non-Q12-hour bid dosing. Additionally, Study Design 3 participants taking twice daily dosing may be switched back to daily dosing over the course of the study, thus necesaitating assignment back to Study Design 2. Given the decreased drug clearance of poor metabolizers, particpants in Study Design 1 are not anticipated to be prescribed bid dosing and thus would not require switching to Study Design 3. These length variations are illustrated in Figure 5 below. Data collection procedure variations are illustrated in the specific visit descriptions in section 11.2.

Visit Length by Study Design			
	Study Design 1 Activity Score < 1 or participants have 2 reduced function alleles	**Study Design 2** Activity Score ≥ 1 (i.e. all other scores)	**Study Design 3** Partcipant previously in Study Design 2, on non-Q12-hour bid dosing
Research Visit 2	72 Hours	12 Hours	Non applicable
Research Visit 3	24 Hours	12 Hours	Up to 24 Hours
Research Visit 4	24 Hours	12 Hours	Up to 24 Hours

Figure 5 – Please note, visit length is expressed in hours post-drug administration.

After study design assignment, the participant will begin the study intervention procedures, starting with Research Visit 2. See below for a brief description of each visit.

Research Visit 2 – This visit is also referred to as pharmacokinetic study 1 (PK1), or first dose PK. This visit takes place in the PCRU and also includes procedures occurring in preparation of this visit, in addition to procedures occurring after administration of the first dose of ATX.

Research Visit 3 – This visit is also referred to as pharmacokinetic study 2 (PK2) or steady state PK. This visit takes place in the PCRU and also includes procedures occurring in preparation of this visit, in addition to procedures after administration of medication when the participant is at steady-state. Steady-state is assumed to have been achieved once the participant has been taking atomoxetine routinely for two weeks. This visit may occur any time after the participant has been taking study drug routinely for 2 weeks and prior to the 18-Week Visit with Study Physician. If the participant is prescribed non-Q12-hour twice daily dosing at this time, the study team will notify paritipant and/or family prior to visit, and the participant will be switched to Study Design 3 for this research visit. Compliance will be documented by checking the capsule count against the medication calendar and documented in corresponding CRFs. Additionally, study personnel will reach out participants and/or their families ~1 week

prior to remind them that all medications need to be taken in the week prior to Research Visit 3. Study personnel will call prior to Research Visit 3 to ensure that participants have been compliant for a week.

6-Week Visit with Study Physician – The participant and family meet with a study physician approximately 6 weeks after ATX therapy was initiated. A ± 2-week window is allowed for ease of scheduling and to ensure Research Visit 3 occurs prior to the visit. Prior to this visit, questionnaires will be sent out via CHADIS to parents and teachers to assess participant's behavior. During this visit, safety, efficacy, and side effects will al be assessed and dose adjustments will be made accordingly. Dose adjustments may be outside research and visits with study physicians as needed.

12-Week Visit with Study Physician – The participant and family meet with a study physician approximately 6 weeks after the 6-Week Visit with Study Physician. A ± 2-week window is allowed for ease of scheduling. Prior to this visit, questionnaires will be sent out via CHADIS to parents and teachers to assess participant's behavior. During this visit, safety, efficacy, and side effects will be assessed and dose adjustments will be made accordingly. Dose adjustments may be outside research and visits with study physicians as needed.

18-Week Visit with Study Physician – The participant and family meet with a study physician approximately 6 weeks after the 12-Week Visit with Study Physician. A ± 2-week window is allowed for ease of scheduling. Prior to this visit, questionnaires will be sent out via CHADIS to parents and teachers to assess participant's behavior. During this visit, safety, efficacy, and side effects will be assessed and dose adjustments will be made accordingly. In addition, baseline questionnaires will be re-administered to gauge symptom and impairment changes post-treatment initiation. Dose adjustments may be outside research and visits with study physicians as needed.

Research Visit 4 – This visit is also referred to as pharmacokinetic study 3 (PK3) or end of study PK. This vist occurs after the 18-Week Visit with Study Physician. This visit takes place in the PCRU and consists of the same procedures as Research Visit 3. Determination of Study Design will be confirmed prior to visit, as some participants may require switching to a different study design if frequency of dosing has changed. During this visit, baseline psychometric measures will be re-administered to measure change post-treatment initiation. An end of study EKG will be obtained, and a Dietary Survey Questionnaire will be administered.

- Description of Research Procedures

1. Research Visit 1 – Part 1

a. When scheduling this visit, the research team will ensure that the participant and their family are aware of the safety measures at

Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage and any PCRU guidance in response to the COVID-19 pandemic. The research team will discuss with the participant and their family that safety procedures to reduce SARS-CoV-2 transmission will be in place for all in-person visits for the foreseeable future.

b. Conduct permission/assent with participant and legally authorized representative (LAR), respectively. Obtain a signed and dated, written informed permission/assent form.
    i. If permission/assent conducted via phone or virtually prior to Research Visit 1 – Part 1, ensure that original copy with participant and LAR is received prior to beginning research procedures.
    ii. Make a copy of all permission/assent forms for the participant and LAR.
c. Obtain demographic data: age, gender, race.
d. Obtain medical history, including prior surgeries.
e. Obtain history of prior and concomitant medication. Add to Concomitant and Prior Medications Log.
f. Provide participants and their families the illustrated U54 Study Booklet that will be used to:
    i. Supplement the information about the study procedures given in the permission/assent form;
    ii. Record medication administration, and;
    iii. Detail answers to any questions they have about the study.
    iv. A questionnaire will be included with the booklet asking families to evaluate how helpful the U54 Study Booklet was in their understanding of the research study. The participant and their parents may also provide feedback for the U54 Study Booklet as well;
    v. Because this questionnaire and survey are a nested study within the longitidunal study, participants and their families will also be given a handout, (please see supplemental document, "U54 Study Booklet Information Handout,") to explain the purpose of the U54 Study Booklet, the purpose of the questionnaire, and it describes that the responses to the booklet will be anonymous.
    vi. The U54 Study Booklet Information Handout serves as a permission/assent or consent document and the voluntary return of the questionnaire and survey serves as assumed permission/assent or consent.
g. Provide handout explaining CHADIS.

h. Collect information regarding the participant's comfort with needlesticks and ability to swallow pills. Questions include:
    i. Parent impression about child's anticipated comfort with needlestick
    ii. History of needlesticks
    iii. General fears about healthcare settings
    iv. Fears about unfamiliar situations
    v. Ability to swallow pills

i. Collect blood sample for genotyping (~1 mL).
    i. The study team may order topical lidocaine (which for this study refers to either a J-tip or cream) if the study participant requests this for procedures requiring a needlestick.

j. For <u>enrolled participants</u> where a washout is needed prior to beginning atomoxetine:
    i. A washout will only be considered for participants currently taking stimulant medication for ADHD. Examples of this include methylphenidate or amphetamine salts.
    ii. A study physician or qualified study personnel will determine if the washout may be conducted safely. If so, he or she will document approval of the washout on associated CRF. If this conversation occurs via telehealth, the study physician or qualified study personnel may document the conversation electronically and send an approval via email. The CRF and email will be printed and stored in the participant's binder. Persons who are able to approve a washout will be identified in the Delegation of Authority/Roles.
    iii. Once the washout has been approved, study personnel will review the washout plan with the participant. Study personnel will:
        1. Document any questions or concerns that the participant and family have regarding washout.
        2. Document participant and family's understanding and willingness to complete washout.
        3. Educate families on who to call if issues arise during washout. See "Washout Handout" in Supplementary Documents.
        4. Educate families to restart on medications (particularly if participant was on stimulants) if behaviors while off medication could cause risk to the participant.
    iv. A 1-week washout period of at least one week is sufficient for children taking stimulants.
    v. If the child is unable to washout of medication or the study physician does not approve the washout's safety, they will be removed from the study.

vi. Schedule Research Visit 1 – Part 2 after washout period is anticipated to be completed.

k. If it is determined that the participant was previously taking and subsequently discontinued any of the following medications (see below) prior to notification of study opportunity, an appropriate time to administer baseline questionnaires and to begin ATX intervention must be determined.
   i. ADHD medication other than a stimulant, e.g. ATX or α_2-agonists
   ii. Selective Serotonin Reuptake Transporter Inhibitor (SSRI)
   iii. Selective Serotonin/Norepinhephrine Reuptake Transporter Inhibitor (SNRI)
   iv. Any medication that could reasonably be expected to affect neurotransmitter concentrations, such as anxiolytics, antiepileptics, antipsychotics.
   v. A CYP2D6 inhibitor
   vi. The following table will be used as guidance for when to administer baseline questionnaires (i.e. return to baseline neurotransmitter levels and/or behavior) and when to begin ATX therapy (i.e. drug and/or active metabolites excreted from participant). However, this only addresses the most common medications that children with ADHD or behaviorial concerns are likely to be trialed on.
      1. The time indicated in “Medication Washout Interval,” is the minimum amount of time needed for medication to be removed and thus, the amount of time needed before beginning ATX intervention.
      2. Administering baseline questionnaires relates to estimating when the participant is likely to return to baseline behavior, which could be longer than the time needed for drug to be removed from systemic circulation. When it is noted that the, “length of pharmacodynamic effects can be variable,” the research team must rely on parent, teacher, and or participant report of symptoms and function. Evaluation of the participant’s medication chart may be also assessed for this information.
   vii. A detailed literature search must be conducted for medications not listed here. The process by which this occurs will be described in more detail in Appendix O.

Medication Class/Group	Examples *also CYP2D6 inhibitor (indication)	When to Administer Baseline Questionnaires	Medication Washout Interval (time expected for drug to be removed from systemic circulation)
ADHD Medications (non-stimulants)	atomoxetine	2-4 weeks	CYP2D6 genotype dependent (2- 4 weeks)
	clonidine (sleep)	1-2 weeks	CYP2D6 genotype dependent (1-2 weeks)
	clonidine (ADHD)	2-4 weeks	CYP2D6 genotype dependent (2-4 weeks)
	guanfacine (sleep)	1-2 weeks	May be CYP2D6 genotype dependent (1-2 weeks)
	guanfacine (ADHD)	2-4 weeks	May be CYP2D6 genotype dependent (2-4 weeks)
SSRIs	fluoxetine*	Length of PD effects can be variable	2 months, (inhibits 2D6, 3A4, 2C19)
	sertraline*	Length of PD effects can be variable	1 week, weak inhibitor of CYP2D6
SNRI	bupropion*	Length of PD effects can be variable	2 – 4 weeks (metabolites primary inhibitors)
	venlafaxine*	Length of PD effects can be variable	1 week, weak inhibitor of CYP2D6
Antipyschotics	aripiprazole	Length of PD effects can be variable	CYP2D6 genotype dependent (1-2 months)
	risperidone	Length of PD effects can be variable	CYP2D6 genotype dependent (1 -2 weeks)
Antiepileptics	levetiracetam	Length of PD effects can be variable	1 week
	valproic acid	Length of PD effects can be variable	1 week (2-4 years, t1/2 3.5 – 20 hours)
Anxiolytics	buspirone	Length of PD effects can be variable	1 week
CYP2D6 Inhibitors	imipramine	Length of PD effects can be variable	2 weeks
	nortriptyline	Length of PD effects can be variable	2 weeks (t1/2 14 -51 hours)
	quinidine	immediately after washout	1 week
	propafenone	immediately after washout	2 weeks
	cimetidine	immediately after washout	1 week (weak inhibitor of CYP2D6)
	tamoxifen	immediately after washout	2 months (may inhibit through competition at active site)
	terbinafine	immediately after washout	6 months
	cetirizine	most likely immediately after washout, however histamine can affect neurotransmitter profile	1 week
	loratidine	most likely immediately after washout, however histamine can affect neurotransmitter profile	1 week

2. Research Visit 1 – Part 2

a. If Research Visit 1 – Part 1 and Research Visit 1 – Part 2 are scheduled on different dates, the research team will ensure that the participant and their family are aware of the safety measures at Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage and any PCRU guidance in response to COVID-19.

b. For participants where washout was needed: ensure that medication washout (with or without taper) has been completed. Ask about any changes to medical history, including if surgeries occurred between Research Visit 1 – Part 1 and this visit. Add this data to the CRF for Research Visit 1. Ask about any changes to history of prior and concomitant medication. Add to Concomitant and Prior Medications Log.

c. Measure vital signs: height, weight, blood pressure, heart rate, respiration rate, and temperature.

d. Perform physical exam.

e. Perform Tanner Staging. This procedure is optional according to participant preference.

f. Obtain electrocardiogram reading. Send to cardiologist for interpretation. Sign off that participant has no underlying cardiac disease.

g. Collect blood samples for safety laboratory tests (processed immediately at Children's Mercy Hospital): CBC with differential, basic metabolic panel, and liver function test.

i. The study team may order topical lidocaine (either J-tip or cream) if the study participant requests this for procedures requiring a needlestick.

h. Collect blood samples for global neurotransmitter metabolomics analysis (pharmacodynamic sample) (2 mL).

i. Perform urinary pregnancy test for female subjects age 10 and above.

j. Collect urinary sample for safety laboratory tests (processed immediately at Children's Mercy Hospital): urinalysis with microbiology.

k. Collect urinary sample for CYP2D6 biomarker measurement (~10 mL).

l. Collect baseline saliva for microbiome analysis (~2 mL).

m. Perform assessment of attention tasks (Pupillometry during Posner's Cuing Task). Further details can be found in Appendix H.

n. Perform psychometric measures to confirm ADHD diagnosis and possible comorbidities:

i. Kiddie Schedule for Affective Disorders and Schizophrenia (KSADS) (either paper or K-SADS-COMP). The following sections from the screen interview will be used: agoraphobia, separation anxiety disorder, social anxiety disorder/selective mutism, specific

phobias, generalized anxiety, obsessive compulsive disorder, and post-traumatic stress disorder. Supplemental sections will be filled for the above selected psychopathology as needed (parent and participant informant).

ii. Kaufman Brief Intelligence Test (KBIT) (participant informant).

iii. Behavior Rating Inventory of Executive Function (BRIEF) (parent informant. Teacher informant is optional).

iv. Screen for Child Anxiety Related Disorders (SCARED) (parent and if participant ≥ 8 years of age)

v. Barkley's Sluggish Cognitive Tempo (SCT) (parent informant)

o. Administer the following questionnaires:

i. Medication Side Effects (parent and teacher informant)

ii. NICHQ Vanderbilt Assessment Scale – Initial Assessment (parent and teacher informant)

iii. Strengths and Difficulties Questionnaire (parent and teacher informant)

iv. Child Behavior Checklist (parent and teacher informant)

v. School Intervention Questionnaire (teacher informant)

vi. Child Medical History$^{\$}$ (parent informant)

vii. Of note, if these questionnaires were obtained within one year of this visit date AND were completed while the participant was not taking ADHD medication, these do not need to be readministered. Additionally, these are clinically validated questionnaires for ADHD. Different versions of these questiononaires are permissible to use to aid the Stuey Physician in a determination a diagnosis.

p. Record adverse events. Document in Adverse Events log.

q. Create Greenphire account.

r. Add compensation to participant's Greenphire account.

s. Update Prior Medical History from data collected on questionnaire marked with $^{\$}$ above.

3. Intake Visit with Study Physician

a. This visit may occur in-person or via telehealth, at the Study Physician's discretion.

b. If this visit occurs in-person:

i. Conduct physical exam. Documentation will be in EMR.

ii. Assess and confirm/infer ADHD diagnosis.

1. For participants without a formal diagnosis of ADHD (e.g. documentation of ADHD in participant's electronic medical record), responses to questionnaires from parent and teacher will be sought prior to diagnosis. During the COVID-19 pandemic, teacher response are will not always available. In the event that teacher responses are not

available, the Study Physician may infer a diagnosis with the information that is available to the Study Physician, (e.g. school reports, previous teacher's feedback) at their clinical discretion. Follow-up for a formal diagnosis is at the discretion of the Study Physician and is dependent on the pandemic situation and the available tools for diagnosis available while the participant is active in the study.

2. For participants with a pre-existing diagnosis of ADHD, data from participant EHRs may be used to support confirmation of a diagnosis. EHRs from clinics outside of Children's Mercy Hospital may be requested in the course of clinical practice.

iii. Assess and confirm diagnosis of other comorbidities.

iv. Assess and confirm appropriateness of atomoxetine therapy for participant.

v. Assess for adverse events. Document in Adverse Events log.

vi. Complete Clinical Global Impression (CGI).

c. If this visit occurs via telehealth, the physical exam will be omitted.

d. This visit may occur before enrollment if participant is referred from a Study Physician. This visit can occur within 6 weeks enrollment.

e. If Children's Mercy Hospital develops procedures for drive-up collection of vital signs (e.g. heart rate, blood pressure), the Study Physician may ask the participant to use this service to supplement their evaluation.

4. Research Visit 2

Prior to Visit

a. Obtain *CYP2D6* genotyping results for participant to calculate *CYP2D6* activity score and determine a CYP2D6 predicted phenotype.

b. Determine whether the First Dose Pharmacokinetic Study of atomoxetine for participant will be Study Design 1 or Study Design 2.

c. Determine an atomoxetine dose using participant CYP2D6 predicted phenotype, height, weight, obesity status, and gender obtained from Research Visit 1. This dose will be simulated to target a plasma $C_{max,ss}$ of 400 ng/mL. This will be determined using the iGO-PK model (see section 5.1 above) .

d. Obtain a signed and dated prescription from a Study Physician for a dose of atomoxetine that is expected to result in a plasma $C_{max,ss}$ of 400 ng/mL.

e. Optional: Obtain a signed and dated prescription for topical lidocaine (either J-tip or cream) to alleviate pain with needlestick for participant.

f. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.
g. Confirm placement in Study Design 1 or Study Design 2.
h. Contact participant and participant's family prior to the visit to:
    a. Remind them of their scheduled Research Visit 2.
    b. Inform as to which Study Design the participant will be under for Research Visit 2 and all remaining research visits.
    c. Instruct participant to begin fasting at midnight prior to coming in for Research Visit 2. Water is permissible while fasting.
    d. The research team will ensure that the participant and their family are aware of the safety measures at Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage and any PCRU guidance in response to COVID-19.

On Day of Visit

a. Confirm participant's willingness to continue in the research study.
b. Obtain medication history and concomitant medications. Add to Concomitant and Prior Medications Log.
c. Measure vital signs: height, weight, blood pressure, heart rate, respiration rate, and temperature.
d. Perform physical exam.
e. Place indwelling venous catheter to obtain blood samples for safety labs and for atomoxetine pharmacokinetic and pharmacodynamic analysis.
f. Collect blood samples for the following safety laboratory tests (to be processed immediately at Children's Mercy Hospital): CBC with differential, basic metabolic panel, and liver function test.
    i. These tests may be omitted if these data were collected in the week prior, at the discretion of the study coordinator, the physician performing the physical exam, or any person designated in the delegation of authority log.
g. Collect urine for urinalysis with microbiology.
h. Perform urinary pregnancy test on female subjects.
i. Collect baseline urine sample (~10 mL) for predose urine sample and CYP2D6 biomarker analysis.
j. Confirm prior to dosing that participant is:
    i. Healthy enough to continue Research Visit 2 (sign off on safety labs and physical exam sheet by physician).
    ii. Not pregnant, females only (urine pregnancy test).
k. Place two sweat patches on the participants back. The sweat patch is 15.4 cm^2 in size and consists of a cellulose blotter paper collection pad

(PharmChek®, PharmaChem Laboratories, Menlo Park, CA). Prior to patch placement, clean the area of skin to which the patch will be applied with rubbing alcohol.

l. Collect pre-dose blood, blood pressure, and heart rate prior to dosing medication.

m. Administer dose of atomoxetine according to prescription.

n. Collect 3 mL of blood, blood pressure, and heart rate at the following time points after dose administration:

- *i.* Timeframe:
  - *i.* Study Design 1: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, 16, 20, 24, 48, and 72 hours.
  - *ii.* Study Design 2: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 hours.
  - *iii.* A window of ± 5 minutes is acceptable for blood draws at time points 0.5 – 2 hours.
  - *iv.* A window of ± 15 minutes is acceptable for blood draws at all other time points.
- *ii.* Collection Instructions: 2 mL of blood will be collected in a EDTA tube and 1 mL of blood will be collected in a serum gel separator tube.
- *iii.* Processing: Within an hour of collecting sample, plasma and serum will be separated by centrifugation at 4°C (1000 – 2000 g x 10 min), aspirated manually, and transferred to polypropylene cryovials for storage at -70°C until analysis.

o. Collect and pool urine starting after dose administration.

- *i.* Study Design 1: collect all urine for the first 24 hours in the PCRU. Discharge participant with urine jug at 24 hour and 48 hour time point and instruct participants to return urine jug 24 hours later to PCRU. Record volume of urine excreted for the intervals of 0-24 hrs, 24-48 hrs, and 48-72 hours post drug administration. Collect a ~10 mL sample from each urine interval. NB: The study team will instruct the participant that every attempt should be made to collect all urine, however the team recognizes that collecting urine while at school or at work may not be feasible.
- *ii.* Study Design 2: collect all urine 12 hours post drug administration. Record the volume of urine excreted. Collect a ~10 mL sample.

p. Collect ~ 2 mL of saliva. Instruct participant not to eat or drink for 30 minutes prior to beginning to spit.

q. Remove sweat patches

- *i.* For Study Design 1, remove one sweat patch 12 and 24 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

ii. For Study Design 2, remove one sweat patch 8 and 12 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

r. The research team will ask the participant to fast for 2 hours after drug administration. After this 2-hour mark, the participant is free to eat and drink as desired. Document all meal times and content. Collect food receipts when available. The following are scenarios where the research team may deviate from fasting for 2 hours post-administration:

i. A participant may request to take atomoxetine with a small amount of soft food (e.g., pudding, applesauce, etc.) to aid with capsule swallowing. Document the food and approximate volume the participant used to aid with capsule swallowing (e.g., participant took capsule with ~1 teaspoon of chocolate pudding).

ii. If participant reports GI upset, a small snack may be given to alleviate symptoms, even in the event that the fasting time has not been completed. Document GI upset as an adverse event in the adverse events log. If snack is given, document the snack time and content.

s. Perform assessment of attention tasks (Pupillometry during Posner's Cuing Task). Further details can be found in Appendix H.

t. Perform Tanner Staging exam. This procedure is optional according to participant preference. The participant does not need to have Tanner Staging if this was performed previously within 4 weeks prior of Research Visit 2 or if the participant has already reached maximum stage in a prior Tanner staging exam.

u. Dispense medication to participant and discuss instructions on atomoxetine prescription and for medication calendar.

v. Assess for adverse events. Document in Adverse Events log.

w. Add compensation to participant's Greenphire account.

5. Research Visit 3

Prior to Visit

a. Optional: Obtain a signed and dated prescription from a Study Physician for topical lidocaine (either J-tip or cream) to alleviate pain with needlestick for participant.

b. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.

c. Confirm participation in Study Design 1, Study Design 2, or need to switch to Study Design 3.

d. If participant will be changing to Study Design 3, discuss with participant and/or family changes from Study Design 2.

e. Contact participant and participant's family between 8-10 days prior to Research Visit 3 to remind participant to take all prescribed doses the week prior to Research Visit 3.

f. Contact participant and participant's family 1 -2 days prior to the visit to:

i. Remind them of their scheduled visit.

ii. Ensure that participant has been compliant on medication for 1 week prior to scheduled Research Visit 3.

i. If the participant has not been compliant, the visit will need to rescheduled.

iii. Instruct participant to begin fasting at midnight prior to the visit. Water is permissible while fasting.

iv. Remind participant to bring their home atomoxetine medication to the PCRU, but to not take the medication in the morning.

v. Remind participant to bring U54 Study Booklet, which contains the participant's medication calendar.

vi. If participant has been switched to evening dosing of ATX, schedule Research Visit 3 to mirror how the participant typically takes their medication. (i.e. if the participant takes their medication in the evening, the beginning of their pharmacokinetic study should also be in the evening).

vii. The research team will ensure that the participant and their family are aware of the safety measures at Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage and any PCRU guidance in response to COVID-19.

On Day of Visit

a. Confirm participant's willingness to continue in the research study.

b. Obtain medication history and concomitant medications. Add to Concomitant and Prior Medications Log.

c. Verify medication calendar with remaining capsules count.

d. Make copies of pages in medication calendar.

e. Measure vital signs: height, weight, blood pressure, heart rate, respiration rate, and temperature.

f. Perform physical exam.

g. Place indwelling venous catheter to obtain blood samples for safety labs and for atomoxetine pharmacokinetic and pharmacodynamic analysis.

h. Collect blood samples for the following safety laboratory tests (to be processed immediately at Children's Mercy Hospital): CBC with

differential, basic metabolic panel, and liver function test. These tests may be omitted if these data were collected in the week prior, at the discretion of the study coordinator, the physician performing the physical exam or any person designated in the delegation of authority log.

i. Collect urine for urinalysis with microbiology.

j. Perform urinary pregnancy test on female subjects 10 years of age or older.

k. Collect baseline urine sample (~10 mL) for predose urine sample and CYP2D6 biomarker analysis.

l. Confirm prior to dosing that participant is:

- *i.* Healthy enough to continue Research Visit 3 (sign off on safety labs and physical exam sheet by physician).
- *ii.* Not pregnant, females only (urine pregnancy test).
- *iii.* Has been compliant with medication (medication calendar check). If the participant has not been compliant, the visit will need to rescheduled.

m. Place two sweat patches on the participant's back. The sweat patch is 15.4 cm^2 in size and consists of a cellulose blotter paper collection pad (PharmChek®, PharmaChem Laboratories, Menlo Park, CA). Prior to patch placement, clean the area of skin to which the patch will be applied with rubbing alcohol.

n. Collect pre-dose blood, blood pressure, and heart rate prior to dosing medication.

o. Administer dose of atomoxetine according to prescription.

p. Collect 3 mL of blood, blood pressure, and heart rate at the following time points after dose administration:

- *i.* Timeframe:
  - *i.* Study Design 1: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12 16, 20, and 24 hours.
  - *ii.* Study Design 2: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12 hours.
  - *iii.* Study Design 3:
    1. Following first dose: Collect at 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 hours as applicable following first dose and will continue only until the second dose is given.
    2. Second dose will be given at the same time increment between when participant routinely takes their first and second dose ± 5 minutes (eg. If 6 hours are between routine dosing for participant, the second study dose will be given 6 hours ± 5 minutes after the first dose).

3. Collect predose blood, blood pressure, heart rate prior to dosing 2nd dose
4. Once the second dose is given, continued blood draws, blood pressure, and heart rate collection based on the timing of the *first* dose will stop.
5. Timing of blood, blood pressure, and heart rate will now be timed according to *second* dose only.
6. Timing of collections will restart, with timepoints 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 12, and 16 hours after the second dose.

iv. A window of ± 5 minutes is acceptable for blood draws at time points 0.5 – 2 hours.

v. A window of ± 15 minutes is acceptable for blood draws at all other time points.

ii. Collection Instructions: 2 mL of blood will be collected in an EDTA tube and 1 mL of blood will be collected in a serum gel separator tube.

iii. Processing: Within an hour of collecting sample, plasma and serum will be separated by centrifugation at 4°C (1000 – 2000 g x 10 min), aspirated manually, and transferred to polypropylene cryovials for storage at -70°C until analysis.

q. Collect and pool urine starting after dose administration.

i. Study Design 1: collect all urine for the first 24 hours in the PCRU. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

ii. Study Design 2: collect all urine 12 hours post drug administration. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

iii. Study Design 3: collect all urine starting after the first dose and continuing through 16 hours post second dose administration. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

r. Collect ~ 2 mL of saliva. Instruct participant not to eat or drink for 30 minutes prior to beginning to spit.

s. Remove sweat patches

i. For Study Design 1, remove one sweat patch 12 and 24 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

ii. For Study Design 2, remove one sweat patch 8 and 12 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

iii. For Study Design 3, remove one sweat patch 8 and 16 hours post second drug dose administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

t. The research team will ask the participant to fast for 2 hours after drug administration. After this 2 hour mark, the participant is free to eat and drink as desired. Document all meal times and content. Collect food receipts when available. The following are scenarios where the research team may deviate from fasting for 2 hours post-administration

i. A participant may request to take atomoxetine with a small amount of soft food (e.g., pudding, applesauce, etc.) to aid with capsule swallowing. Document the food and approximate volume the participant used to aid with capsule swallowing (e.g., participant took capsule with ~1 teaspoon of chocolate pudding).

ii. If participant reports GI upset, a small snack may be given to alleviate symptoms, even in the event that the fasting time has not been completed. Document GI upset as an adverse event in the adverse events log. If snack is given, document the snack time and content

iii. Fasting will not be required for participants taking medication in the evening who are participating in an overnight pharmacokinetic study. The research team does not believe it is feasible to expect a child to fast for 8-12 hours while awake.

iv. If participant is assigned Study Design 3, fasting prior to and/or after the second dose of medication will not be required. The research team does not believe it is feasible to expect a child to fast prior to both the first dose and second dose.

u. Perform assessment of attention tasks (Pupillometry during Posner's Cuing Task). Further details can be found in Appendix H.

v. Perform Tanner Staging. This procedure is optional according to participant preference. The participant does not need to have Tanner Staging if this was performed previously within 4 weeks prior of Research Visit 3 or if the participant has already reached maximum stage in a prior Tanner staging exam.

w. Assess for adverse events. Document in Adverse Events log.

x. Add compensation to participant's Greenphire account.

6. 6-Week Visit with Study Physician

Prior to Visit

a. Dose adjustment may be required before this visit, particularly if there are reports of severe adverse events (e.g. unremitting GI issues) or lack of efficacy. When this occurs, the study physician may adjust the ATX regimen as needed. Documentation of changes will be recorded in the participants electronic medical record. If a new prescription is required, the study physician will follow the procedures outlined in "Other visit related activities," (see below).

b. Send out the following psychometric questionnaires 1-2 weeks prior to 6-Week Visit with Study Physician:

i. Medication Side Effects (parent and teacher informant)

ii. NICHQ Vanderbilt Assessment Scales follow-up assessment (parent and teacher informant)

iii. Please note: Responses will always be sought, but the study team recognizes the potential difficulties with returning results, particularly during the summer break or the pandemic for teachers. The study team will remind the parent to complete his or her questionnaires a maximum of 3 times if applicable to the participant's school situation. Additionally, the study team will encourage parents to reach out to their child's teacher(s) to return questionnaires as well. A reminder for parents to reach out to teachers will occur a maximum of 3 times.

iv. Questionnaires should be returned ± 2 weeks of study visit with physician.

v. If questionnaires are late or are not received, this will be noted on CRFs and are considered "missing data."

vi. Both the Medication Side Effects and NICHQ Vanderbilt Assessment scales are validated questionnaires that are used to aid Study Physician in determining treatment response. Use of different versions of these questionnaires are permissible to aid the Study Physician, in additional to clinical impression, with medication management and determination of response.

c. Contact participant and participant's family 1 -2 days prior to 6-Week Visit with Study Physician to:

i. Remind them of their scheduled 6-Week Visit with Study Physician.

ii. Remind participant to bring their home atomoxetine medication to 6-Week Visit with Study Physician (in-person visit only).

iii. Remind participant to bring U54 Study Booklet, which contains the participant's medication calendar (in-person visit only).

Day of Visit

a. This visit may occur in-person or via telehealth, at the Study Physician's discretion.

b. If this visit occurs in-person:

i. The research team will ensure that the participant and their family are aware of the safety measures at Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage.

ii. Conduct physical exam. Documentation will be in EMR.

iii. Assess ADHD symptoms/review questionnaires.

iv. Assess other comorbidities and concurrent medications. Add to Concomitant and Prior Medications Log.

v. Assess for atomoxetine side effects.

vi. Make atomoxetine dose changes as necessary, with guidance from study team if necessary and document reason for changes.

vii. Document prescription change (if made) in CRF.

viii. Assess for adverse events. Document in Adverse Events log.

ix. Verify medication calendar with remaining capsules count.

x. Make copies of pages in medication calendar.

xi. Complete Clinical Global Impression (CGI)

c. If this visit occurs via telehealth proceed with all procedures stated above with the following exceptions:

i. The physical exam will be omitted.

ii. Ask the participant to email pictures of the pertinent adherence calendar pages and a picture of the remaining medication.

Other Visit Related Activities

a. Obtain a signed and dated prescription for atomoxetine from Study Physician. This may occur prior, during, or after this visit.

b. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.

c. Deliver medication to patient, or leave medication at Children's Mercy Outpatient Pharmacy for family to pick up medication at their convenience. Medication may be mailed (see section 5.2) at the request of the participant's family.

d. Add compensation to participant's Greenphire account if calendar and medication bottle brought to visit.

e. If Children's Mercy Hospital develops procedures for drive-up collection of vital signs (e.g. heart rate, blood pressure), the Study Physician may ask the participant to use this service to supplement their evaluation.

7. 12-Week Visit with Study Physician

Prior to Visit

a. Dose adjustment may be required before this visit, particularly if there are reports of severe adverse events (e.g. unremitting GI issues) or lack of efficacy. When this occurs, the study physician may adjust the ATX regimen as needed. Documentation of changes will be recorded in the participants electronic medical record. If a new prescription is required, the study physician will follow the procedures outlined in "Other visit related activities," (see below).

b. Send out the following psychometric questionnaires 1-2 weeks prior to 12-Week Visit with Study Physician:

i. Medication Side Effects (parent and teacher informant)

ii. NICHQ Vanderbilt Assessment Scales follow-up assessment (parent and teacher informant)

iii. Please note: Responses will always be sought, but the study team recognizes the potential difficulties with returning results, particularly during the summer break or the pandemic for teachers. The study team will remind the parent to complete his or her questionnaires a maximum of 3 times if applicable to the participant's school situation. Additionally, the study team will encourage parents to reach out to their child's teacher(s) to return questionnaires as well. A reminder for parents to reach out to teachers will occur a maximum of 3 times.

iv. Questionnaires should be returned ± 2 weeks of study visit with physician.

v. If questionnaires are late or are not received, this will be noted on CRFs and are considered "missing data."

vi. Both the Medication Side Effects and NICHQ Vanderbilt Assessment scales are validated questionnaires that are used to aid Study Physician in determining treatment response. Use of different versions of these questionnaires are permissible to aid the Study Physician, in additional to clinical impression, with medication management and determination of response.

c. Contact participant and participant's family 1 -2 days prior to 12-Week Visit with Study Physician to:
  i. Remind them of their scheduled 12-Week Visit with Study Physician.
  ii. Remind participant to bring their home atomoxetine medication to 12-Week Visit with Study Physician (in-person visit only).
  iii. Remind participant to bring U54 Study Booklet, which contains the participant's medication calendar (in-person visit only).

Day of Visit

a. This visit may occur in-person or via telehealth, at the Study Physician's discretion.
b. If this visit occurs in-person:
  i. Conduct physical exam. Documentation will be in EMR.
  ii. Assess ADHD symptoms/review questionnaires.
  iii. Assess other comorbidities and concurrent medications. Add to Concomitant and Prior Medications Log.
  iv. Assess for atomoxetine side effects.
  v. Make atomoxetine dose changes as necessary, with guidance from study team if necessary and document reason for changes.
  vi. Document prescription change (if made) in CRF.
  vii. Assess for adverse events. Document in Adverse Events log.
  viii. Verify medication calendar with remaining capsules count.
  ix. Make copies of pages in medication calendar.
  x. Complete Clinical Global Impression (CGI)
c. If this visit occurs via telehealth, proceed with all procedures stated above with the following exceptions:
  i. The physical exam will be omitted.
  ii. Ask the participant to email pictures of the pertinent adherence calendar pages and a picture of the remaining medication.

Other Visit Related Activities

a. Obtain a signed and dated prescription for atomoxetine from Study Physician. This may occur prior, during, or after this visit.
b. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.
c. Deliver medication to patient, or leave medication at Children's Mercy Outpatient Pharmacy for family to pick up medication at

their convenience. Medication may be mailed (see section 5.2) at the request of the participant's family.
Add compensation to participant's Greenphire account if calendar and medication bottle brought to visit.

d. If Children's Mercy Hospital develops procedures for drive-up collection of vital signs (e.g. heart rate, blood pressure), the Study Physician may ask the participant to use this service to supplement their evaluation.

8. 18-Week Visit with Study Physician

Prior to Visit

a. Dose adjustment may be required before this visit, particularly if there are reports of severe adverse events (e.g. unremitting GI issues) or lack of efficacy. When this occurs, the study physician may adjust the ATX regimen as needed. Documentation of changes will be recorded in the participants electronic medical record. If a new prescription is required, the study physician will follow the procedures outlined in "Other visit related activities," (see below).

b. Send out the following psychometric questionnaires 1-2 weeks prior to 18-Week Visit with Study Physician:

i. Medication Side Effects (parent and teacher informant)

ii. NICHQ Vanderbilt Assessment Scales follow-up assessment (parent and teacher)

iii. Strengths and Difficulties Questionnaire (parent and teacher informant)

iv. Child Behavior Checklist (parent and teacher informant)

v. School Intervention Questionnaire (teacher informant)

vi. Please note: Responses will always be sought, but the study team recognizes the potential difficulties with returning results, particularly during the summer break or the pandemic for teachers. The study team will remind the parent to complete his or her questionnaires a maximum of 3 times if applicable to the participant's school situation. Additionally, the study team will encourage parents to reach out to their child's teacher(s) to return questionnaires as well. A reminder for parents to reach out to teachers will occur a maximum of 3 times.

vii. Questionnaires should be returned ± 2 weeks of study visit with physician.

viii. If questionnaires are late or are not received, this will be noted on CRFs and are considered "missing data."

vii. All clinical questonnaires are validated tools that are used to aid the Study Physician in determining treatment response. Use of different versions of these questionnaires are permissible to aid the Study Physician, in additional to clinical impression, with medication management and determination of response.

b. Contact participant and participant's family 1 -2 days prior to 18-Week Visit with Study Physician to:

i. Remind them of their scheduled 18-Week Visit with Study Physician.

ii. Remind participant to bring their home atomoxetine medication to 18-Week Visit with Study Physician (in-person visit only).

iii. Remind participant to bring U54 Study Booklet, which contains the participant's medication calendar (in-person visit only).

Day of Visit

a. This visit may occur in-person or via telehealth, at the Study Physician's discretion.

b. If this visit occurs in-person:

i. Conduct physical exam. Documentation will be in EMR.

ii. Assess ADHD symptoms/review questionnaires.

iii. Assess other comorbidities and concurrent medications. Add to Concomitant and Prior Medications Log.

iv. Assess for atomoxetine side effects.

v. Make atomoxetine dose changes as necessary, with guidance from study team if necessary and document reason for changes.

vi. Document prescription change (if made) in CRF.

vii. Assess for adverse events. Document in Adverse Events log.

viii. Verify medication calendar with remaining capsules count.

ix. Make copies of pages in medication calendar.

x. Complete Clinical Global Impression (CGI)

c. If this visit occurs via telehealth, proceed with all procedures stated above with the following exceptions:

i. The physical exam will be omitted.

ii. Ask the participant to email pictures of the pertinent adherence calendar pages and a picture of the remaining medication.

Other Visit Related Activities

a. Obtain a signed and dated prescription for atomoxetine from Study Physician. This may occur prior, during, or after this visit.
b. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.
c. Deliver medication to patient, or leave medication at Children's Mercy Outpatient Pharmacy for family to pick up medication at their convenience. Medication may be mailed (see section 5.2) at the request of the participant's family.
d. Add compensation to participant's Greenphire account if calendar and medication bottle brought to visit.
e. If Children's Mercy Hospital develops procedures for drive-up collection of vital signs (e.g. heart rate, blood pressure), the Study Physician may ask the participant to use this service to supplement their evaluation.

9. Research Visit 4

Prior to Visit

a. Optional: Obtain a signed and dated prescription by a Study Physician for topical lidocaine (either J-tip or cream) to alleviate pain with needlestick for participant.
b. Submit signed and dated prescription to Children's Mercy Hospital Investigation Drug Services (IDS) pharmacy.
c. Confirm participation in Study Design 1, Study Design 2, or need to switch to Study Design 3.
d. If participant will be changing to Study Design 3 or back to Study Design 2, discuss with participant and/or family changes from prior study design.
e. Contact participant and participant's family 8-10 days prior to Research Visit 4 to remind participant to take all prescribed doses of medication in the week prior to Research Visit 4.
f. If participant has been switched to evening dosing of ATX, schedule Research Visit 4 to mirror how the participant typically takes their medication (i.e. if the participant takes their medication in the evening, the beginning of their pharmacokinetic study should also be in the evening).
g. Contact participant and participant's family 1 -2 days prior to Research Visit 4 to:
i. Remind them of their scheduled visit.

ii. Ensure that participant has been compliant on medication for 1 week prior to scheduled Research Visit 3.
   i. If the participant has not been compliant, the visit will need to rescheduled.

iii. Instruct participant to begin fasting at midnight prior to visit. Water is permissible while fasting.

iv. Remind participant to bring their home atomoxetine medication to the PCRU, but to not take the medication in the morning.

v. Remind participant to bring U54 Study Booklet, which contains the participant's medication calendar.

vi. The research team will ensure that the participant and their family are aware of the safety measures at the Children's Mercy Hospital to prevent transmission of the SARS-CoV-2 virus. These resources and information to be shared will be detailed on the Children's Mercy Hospital homepage and any PCRU guidance in response to COVID-19.

On Day of Visit

a. Confirm participant's willingness to continue in the research study.

b. Obtain medication history and concomitant medications. Add to Concomitant and Prior Medications Log.

c. Verify medication calendar with number of remaining capsules.

d. Take back medication calendar.

e. Measure vital signs: height, weight, blood pressure, heart rate, respiration rate, and temperature.

f. Perform physical exam.

g. Place indwelling venous catheter to obtain blood samples for safety labs and for atomoxetine pharmacokinetic and pharmacodynamic analysis.

h. Collect blood samples for the following safety laboratory tests (to be processed immediately at Children's Mercy Hospital): CBC with differential, basic metabolic panel, and liver function test. These tests may be omitted if these data were collected in the week prior, at the discretion of the study coordinator, the physician performing the physical exam or any person designated in the delegation of authority log.

i. Collect urine for urinalysis with microbiology.

j. Perform urinary pregnancy test on female subjects 10 years of age or older.

k. Collect baseline urine sample (~10 mL) for predose urine sample and CYP2D6 biomarker analysis.

l. Confirm prior to dosing that participant is:

i. Healthy enough to continue Research Visit 4 (sign off on safety labs and physical exam sheet by physician).

ii. Not pregnant, females only (urine pregnancy test).

iii. Has been compliant with medication (medication calendar check). If the participant has not been compliant, the visit will need to rescheduled.

m. Place two sweat patches on the participant's back. The sweat patch is 15.4 cm^2 in size and consists of a cellulose blotter paper collection pad (PharmChek®, PharmaChem Laboratories, Menlo Park, CA). Prior to patch placement, clean the area of skin to which the patch will be applied with rubbing alcohol.

n. Collect pre-dose blood, blood pressure, and heart rate prior to dosing medication.

o. Administer dose of atomoxetine according to prescription.

p. Collect 3 mL of blood, blood pressure, and heart rate at the following time points after dose administration:

i. Timeframe:

i. Study Design 1: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, 16, 20, and 24 hours.

ii. Study Design 2: 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 hours.

iii. Study Design 3:

1. Following first dose: Collect at 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, and 12 hours as applicable following first dose and will continue only until the second dose is given.
2. Second dose will be given at the same time increment between when participant routinely takes their first and second dose (eg. If 6 hours are between routine dosing for participant, the second study dose will be given 6 hours after the first dose).
3. Collect predose blood, blood pressure, heart rate prior to dosing 2nd dose
4. Once the second dose is given, continued blood draws, blood

pressure, and heart rate collection based on the timing of the first dose will stop.

5. Timing of blood, blood pressure, and heart rate will now be timed according to second dose only.
6. Timing of collections will restart, with timepoints 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 12, and 16 hours after the second dose.

iv. A window of ± 5 minutes is acceptable for blood draws at time points 0.5 – 2 hours.

v. A window of ± 15 minutes is acceptable for blood draws at all other time points.

ii. Collection Instructions: 2 mL of blood will be collected in an EDTA tube and 1 mL of blood will be collected in a serum gel separator tube.

iii. Processing: Within an hour of collecting sample, plasma and serum will be separated by centrifugation at 4°C (1000 – 2000 g x 10 min), aspirated manually, and transferred to polypropylene cryovials for storage at -70°C until analysis.

q. Collect and pool urine starting after dose administration.

i. Study Design 1: collect all urine for the first 24 hours in the PCRU. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

ii. Study Design 2: collect all urine 12 hours post drug administration. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

iii. Study Design 3: collect all urine starting after the first dose and continuing through 16 hours post second dose administration. Record the volume of urine excreted. Collect a ~10 mL sample and store in 15 mL polypropylene falcon tubes for storage at -70°C until analysis.

r. Collect ~ 2 mL of saliva. Instruct participant not to eat or drink for 30 minutes prior to beginning to spit.

s. Remove sweat patches

i. For Study Design 1, remove one sweat patch 12 and 24 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

ii. For Study Design 2, remove one sweat patch 8 and 12 hours post drug administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

iii. For Study Design 3, remove one sweat patch 8 and 16 hours post second drug dose administration. Place sweat patch in labeled ziplock bag for storage at -70°C until analysis.

t. Obtain end of study electrocardiogram reading. Send to cardiologist for interpretation.

u. The research team will ask the participant to fast for 2 hours after drug administration. After this 2-hour mark, the participant is free to eat and drink as desired. Document all meal times and content. Collect food receipts when available. The following are scenarios where the research team may deviate from fasting for 2 hours post-administration:

i. A participant may request to take atomoxetine with a small amount of soft food (e.g. pudding, applesauce, etc.) to aid with capsule swallowing. Document the food and approximate volume the participant used to aid with capsule swallowing (e.g. participant took capsule with ~1 teaspoon of chocolate pudding).

ii. If participant reports GI upset, a small snack may be given to alleviate symptoms, even in the event that the fasting time has not been completed. Document GI upset as an adverse event in the adverse events log. If snack is given, document the snack time and content.

iii. Fasting will not be required for participants taking medication in the evening who are participating in an overnight pharmacokinetic study. The research team does not believe it is feasible ot expect a child to fast for 8-12 hours while awake.

iv. If participant is assigned Study Design 3, fasting prior to and/or after the second dose of medication will not be required. The research team does not believe it is feasible to expect a child to fast prior to both the first dose and second dose.

v. Perform assessment of attention tasks (Pupillometry during Posner's Cuing Task). Further details can be found in Appendix H.

w. Perform Tanner Staging. The participant does not need to have Tanner Staging if this was performed previously within 4 weeks prior of Research Visit 3 or if the participant has already reached maximum stage in a prior Tanner staging exam.

x. Administer end of study psychometric questionnaires:

i. Screen for Child Anxiety Related Disorders (SCARED parent and participants ≥ 8 years old informant).

ii. Barkley's Sluggish Cognitive Tempo (SCT) (parent informant).

iii. Behavior Rating Inventory of Executive Function (BRIEF) (parent informant. Teacher informant is optional).

y. Administer Dietary Survey Questionnaire (participant informant).

z. Please note that the SCARED and the BRIEF are validated tools that are used to aid the Study Physician in determining treatment response. Use of different versions of these questionnaires are permissible to aid the Study Physician, in additional to clinical impression, with medication management and determination of response.

aa. Assess for adverse events. Document in Adverse Events log.

bb. Determine and document participant's adherence to ATX medication regimen.

i. High adherence is defined as the participant taking ≥ 80% of their scheduled doses

ii. Medium adherence is defined as the participant taking between 40 – 79% of their scheduled doses.

iii. Low adherence is defined as a participant taking < 40% of their scheduled doses.

11.1.1.1 Add compensation to participant's Greenphire account.

10. ADHD Care Following Conclusion of the Study

Research personnel will call research participants and/or their parents two weeks (± 1 week) following study conclusion or withdrawal from the study to check if any adverse events occurred in the interim. The research team will also take the opportunity to re-educate the family if a taper of medication was needed. This information will be documented on the 2-week Post-Study Completion Check CRF.

See the Schedule of Events (SOE) table below outlining all of the above procedures by visit.

Schedule of Events	**Prescreen**	**Research Visit 1*ɵ**	**Intake with Study Physicianᴪ**	**Research Visit 2***	**Research Visit 3***	**6-Week Visit with Study Physicianᴪ**	**12-Week Visit with Study Physicianᴪ**	**18-Week Visit with Study Physicianᴪ**	**Research Visit 4***	**2-week Post-Study Check**
Time point	N/A	N/A	N/A	Start Med/Day 0	Day 0 + at least 2 weeks€	Day 0 + 6 weeks and after RV3	6-Week Visit with Study Physican + 6 weeks	12-Week Visit with Study Physican + 6 weeks	After 18-Week Visit	After Research Visit 4
Visit Window	N/A	N/A	N/A	N/A	See note€	± 2 weeks	± 2 weeks	± 2 weeks	N/A	± 1 weeks
Type of Visit/Event	Phone	Phone/In Person	In person/ telehealth	In Person	In Person	In person/ telehealth	In person/ telehealth	In person/ telehealth	In Person	Phone
Potential Participant Identified	X									
Introduce the Study	X									
Schedule Research Visit 1	X									
Obtain Consent or Permission/Assent		X[1]								
Obtain participant continued assent (verbal)				X	X				X	
Document continued assent in research note				X	X				X	
Provide U54 Study Booklet		X								

Schedule of Events	Prescreen	Research Visit 1*ɵ	Intake with Study PhysicianΨ	Research Visit 2*	Research Visit 3*	6-Week Visit with Study PhysicanΨ	12-Week Visit with Study PhysicanΨ	18-Week Visit with Study PhysicanΨ	Research Visit 4*	2-week Post-Study Check
Needlestick/Pill Swallowing Questions		X								
Explain and Begin Washout (if needed)		X								
Demographic Data		X								
Prior/Concomitant Meds		X		X	X	X	X	X	X	
Prior Medical History		X								
Vital Signs		X		X[2]	X[2]				X[2]	
Physical Exam		X	X	X	X	X	X	X	X	
Electrocardiogram		X							X	
Tanner Staging (optional)		X		X	X				X	
Pregnancy Test		X		X	X				X	
Safety Laboratory Tests (CBC w/diff; Base Met; LFTs; UA w/micro)		X		X	X				X	
Confirm ADHD Diagnosis			X							
Assess for Comorbidities			X			X	X	X		

Confirm ATX appropriateness for participant			X							
Schedule of Events	**Prescreen**	**Research Visit 1*ɵ**	**Intake with Study Physicianψ**	**Research Visit 2***	**Research Visit 3***	**6-Week Visit with Study Physicianψ**	**12-Week Visit with Study Physicianψ**	**18-Week Visit with Study Physicianψ**	**Research Visit 4***	**2-week Post-Study Check**
Interpret genotyping results				X[3]						
Assign study design				X[3]						
Determine medication dose				X[3]						
Pre-Visit Reminder Call				X[3]	X[3,4]	X[3]	X[3]	X[3]	X[3,4]	
Administer medication				X	X				X	
Dispense medication				X		X	X	X		
Dispense topical lidocaine (optional)				X	X				X	
Place IV				X	X				X	
Genotyping sample (blood)		X								
Pharmacokinetic sample (blood)				X	X				X	
Pharmacodynamic sample (blood)		X		X	X				X	
CYP2D6 Biomarker (urine)		X		X	X				X	

Pharmacokinetic sample (urine)				X	X				X	
Microbiome sample (saliva)		X		X	X				X	
Schedule of Events	**Prescreen**	**Research Visit 1*ɵ**	**Intake with Study Physicianψ**	**Research Visit 2***	**Research Visit 3***	**6-Week Visit with Study Physicianψ**	**12-Week Visit with Study Physicianψ**	**18-Week Visit with Study Physicianψ**	**Research Visit 4***	**2-week Post-Study Check**
Sweat patches (sweat)				X	X				X	
Pupillometry during Posner's Cuing Task		X		X	X				X	
Dietary Survey Questionnaire									X	
Kiddie Schedule for Affective Disorders and Schizophrenia (KSADS)		X								
Kaufman Brief IQ Test (KBIT)		X								
Behavior Rating Inventory of Executive Function (BRIEF)		X							X	
Barkley Sluggish Cognitive Tempo Scale (SCT)		X							X	
Screen for Child Anxiety and Related Disorders (SCARED) – *parent*		X							X	

Screen for Child Anxiety and Related Disorders (SCARED) – *child* (≥ 8)		X							X	
Schedule of Events	**Prescreen**	**Research Visit 1*ɵ**	**Intake with Study Physicianψ**	**Research Visit 2***	**Research Visit 3***	**6-Week Visit with Study Physicianψ**	**12-Week Visit with Study Physicianψ**	**18-Week Visitwith Study Physicianψ**	**Research Visit 4***	**2-week Post-Study Check**
NICHQ Vanderbilt Assessment Scales – *parent*			X[5,6]			X	X	X		
NICHQ Vanderbilt Assessment Scales – *teacher*			X[5,6]			X	X	X		
Child Behavior Checklist – *parent*			X[5,6]					X		
Child Behavior Checklist – *teacher*			X[5,6]					X		
Strengths and Difficulties Questionnaire – *parent*			X[5,6]					X		
Strengths and Difficulties Questionnaire – *teacher*			X[5,6]					X		
Medication Side Effects – *parent*			X[5,6]			X	X	X		
Medication Side Effects – *teacher*			X[5,6]			X	X	X		

School Intervention Questionnaire			X[5,6]					X		
Child Medical History Questionnaire			X[5,6]							
Schedule of Events	**Prescreen**	**Research Visit 1*Θ**	**Intake with Study PhysicianΨ**	**Research Visit 2***	**Research Visit 3***	**6-Week Visit with Study PhysicianΨ**	**12-Week Visit with Study PhysicianΨ**	**18-Week Visit with Study PhysicianΨ**	**Research Visit 4***	**2-week Post-Study Check**
Clinical Global Impression			X			X	X	X		
Adherence Check					X	X	X	X	X	X[7]
Adverse Events		X	X	X	X	X	X	X	X	X
Take Back U54 Study Booklet/Survey									X	

[1] Execution of permission/assent may occur via phone to allow participants to fill out psychometric questionnaires.
[2] Heart and blood pressure to take with each blood draw.
[3] These events occur prior to the visit for preparation.
[4] These phone calls occur in 2 parts.
[5] These questionnaires must be completed by the Intake Visit; however, they may be assigned at Research Visit 1 to allow participants ample time to complete.
[6] For participants recruited from Track 4, these questionnaires are assigned immediately after consent has been obtained.
[7] For participants who require a taper off medication.
*These visits will occur in the Pediatric Clinical Research Unit (PCRU).
ΨThese visits will occur at either the ADHD Clinic on College Boulevard or in the Developmental and Behavioral Sciences Clinic. If these visits are telehealth visits, all in-person procedures, such as obtaining vitals or a physical exam, may be omitted.
ΘThis visit can occur in two parts to minimize time of washout.
€The window for this visit is flexible and has been expanded from the previous versions of this protocol. This visit can occur anytime after the participant has been taking medication regularly for 2 weeks and up until prior to the 18-Week Visit with Study Physician.

- Procedures to Reduce Risks

1. Medical Chart Review during Prescreen will be used to determine if a potential participant has any of the following:
    a. Diagnosis or indication of underlying risk for cardiotoxicity, such as presentation of structural cardiac abnormalities, cardiomyopathy, or arrhythmias.
    b. Diagnosis or indication of disease that may cause abnormal absorption or gastric emptying.
    c. Previous history or indication of prior adverse drug reaction to atomoxetine
    d. Concurrent therapy with drugs known to inhibit CYP2D6 or previous treatment with strong CYP2D6 inhibitors.
    e. Concurrent therapy with other psychiatric/behavioral drugs
2. The two parts of Research Visit 1 may occur at separate times for participants who require a washout prior to beginning atomoxetine therapy. Research Visit 1 Part 1 includes a blood stick for genotyping purposes. Genotyping analysis, which takes approximately one week to complete and interpret, will be conducted while the participant is washing out. Were we to collect genotyping sample when the participant is at baseline, the participant could potentially be off of medication for ~ 2 weeks, while waiting for genotyping results such that ATX can be dosed in the participant using the iGO-PK model.
3. Needlestick comfort and pill-swallowing ability questions will be asked to reduce discomfort and anxiety associated with needlesticks and taking the study drug.
4. Baseline electrocardiogram during Research Visit 1 to assess presence of any underlying risks for cardiotoxicity prior to starting participant on ATX.
5. End of study electrocardiogram during Research Visit 4 to assess for any cardiac injury that may have occurred while the participant was taking ATX chronically.
6. Safety Labs – CBC with differential, hepatic function test, basic metabolic panel, and urinary analysis with microbiology will be conducted at Research Visit 1, Research Visit 2, Research Visit 3, and Research Visit 4.
7. Pregnancy Test will be conducted at Research Visit 1, Research Visit 2, Research Visit 3, and Research Visit 4 to ensure the subject is not pregnant.
8. Assessment of adverse events
9. For blood collection:

a. Blood will be obtained either through needle stick (Research Visit 1) or through an indwelling venous catheter (Research Visit 2, Research Visit 3, and Research Visit 4).

b. With our study design, the maximal amount of blood drawn from any participant for clinical and research purposes for a research visit assigned Study Design 1 or 2 is 54 mL in a 72 hour period. This would mean that the lower limit for weight to complete study procedures is 11 – 15 kg. Our study population typically exceeds this weight range. However, given that there are multiple research visits where blood is drawn, for younger child and for those who are approximately 11- 15 kg, we will schedule research visits such that they are greater than 30 days a part.

c. For subjects who are less than 16.5 kg, the total amount of blood drawn from the subjects may exceed the 3 ml/kg/day limit, particularly if the individual is a CYP2D6 intermediate or poor metabolizer. The maximum amount of blood drawn within a 24 hour period is 48 mL.

d. With our study design, the maximal amount of blood drawn for a participant for clinical and research purposes for a research visit assigned Study Design 3 is 75ml in a 24 hour period. The majority of patients will not be enrolled under Study Design 3. The lower limit of weight to complete all possible blood draws for Study Design 3 is 15kg – 25kg (3-5ml/kg = 75ml). Our study population typically exceeds this weight range. Additionally, participants in Study Design 3 may not be required to have all blood draws adding up to 75ml, as the number of draws depends on the timing of their dosing.

e. For Study Design 3 subjects who are less than 25 kg, research visits will be scheduled to not exceed predicted blood draw totals of 9.5ml/kg or more over an 8 week period.

10. Incidental findings which are discovered during the course of this study may be released to families in a letter at their request for the purposes of seeking further care (please refer to Appendix M).

11. During the COVID-19 pandemic, the research team will make every attempt to reduce the number of in-person procedures while providing ADHD care to the participant and achieving the aims of this research study.

12. The research team will adhere to all Children's Mercy Hospital and Children's Mercy Research Institute guidelines for reducing transmission of SARS-CoV-2 virus.

13. The research team will ensure that participants and their LARs have resources for reducing SARS-CoV-2 virus tranmission from Children's Mercy Hospital and Children's Mercy Research Institute.

14. A Certificate of Confidentiality has been issued for this study by the National Institutes of Health which protects the privacy of research participants by prohiting disclosure of identifiable and sensitive information. However, disclosure is permitted for the following scenarios:

a. Required by Federal, State, or local laws (e.g., as required by the Federal Food, Drug, and Cosmetic Act, or state laws requiring the reporting of communicable diseases to State and local health departments), excluding instances of disclosure in any Federal, State, or local civil, criminal, administrative, legislative, or other proceeding;
b. Necessary for the medical treatment of the individual to whom the information, document, or biospecimen pertains and made with the consent of such individual;
c. Made with the consent of the individual to whom the information, document, or biospecimen pertains; or
d. Made for the purposes of other scientific research that is in compliance with applicable Federal regulations governing the protection of human subjects in research.

11.4 Source Records Used

a. Permission/Assent or Consent Forms (Research Visit 1 or Phone Consent)
b. Demographics (Research Visit 1 CRF)
c. Prior/Concomitant Medications (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
d. Prior Medical History (Research Visit 1 CRF, Child Medical History Questionnaire)
e. Needlestick and Pill Swallowing Questions (Research Visit 1 CRF)
f. Vital Signs (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
g. Data from physical exam (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
h. Pregnancy status (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
i. Baseline and end of study electrocardiogram (Research Visit 1 and Research Visit 4 CRFs)
j. Tanner Staging (Optional) (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
k. Genotyping data (Research Visit 2 and Research Visit 3 CRFs)
l. Atomoxetine dose (IDS pharmacy prescriptions)

m. Pharmacokinetic Samples – plasma, urine, and sweat (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
n. Pharmacodynamic Samples – plasma and serum (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
o. Microbiome Samples – saliva (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
p. CYP2D6 biomarker analysis – urine (Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
q. Attention Tasks – Pupillometry during Posner's Cuing Task data (data stored on computer hard drive, documentation in Research Visit 1, Research Visit 2, Research Visit 3 and Research Visit 4 CRFs)
r. Long-term dietary patterns – Dietary Survey Questionnaire (paper questionnaire)
s. Intelligence Quotient – Kaufman Brief IQ Test (KBIT) (paper questionnaire)
t. ADHD and comorbidities –
    i. Kiddie Schedule for Affective Disorders and Schizophrenia Present and Lifetime (K-SADS-PL) (paper questionnaire or KSADS-COMP);
        1. Subject Information (pages 1-3)
        2. Attention Deficit/Hyperactivity Disorder (pages 4-12)
        3. Generalized Anxiety Disorder (pages 13-17)
        4. Social Anxiety/Selective Mutism Disorder (pages 18-21)
        5. If KSADS-COMP is used, the above data are available electronically. These data will be downloaded and added to the participant binder.
    ii. Behavior Rating Inventory of Executive Function (BRIEF) (paper questionnaire);
    iii. Bradley Sluggish Cognitive Tempo Scale (paper questionnaire);
    iv. Screen for Child Anxiety and Related Disorders (CHADIS questionnaire);
    v. NICHQ Vanderbilt Assessment Scales for parent and teacher (CHADIS questionnaire);
    vi. Child Behavior Checklist (CHADIS questionnaire);
    vii. School Intervention Questionnaire (CHADIS questionnaire);
    viii. Clinical Global Improvement (Study Physician Note)

u. Medication side effects –
  i. Medication Side Effects (CHADIS questionnaire);
  ii. In-person assessment (study CRFs and participant EMR);
  iii. Parental reports (study CRFs and participant EMR if parent calls in and reports to a RN or physician on-call)

v. Adverse events –
  i. in person assessment (study CRFs and participant EMR);
  ii. parental reports (study CRFs and participant EMR if parent calls in and reports to a RN or physician on-call)

w. Study medication adherence – in person assessment (Research Visit 3, 6-Week Visit with Study Physician, 12-Week Visit with Study Physician, 18-Week Visit with Study Physician, and Research Visit 4 CRFs, Drug Adherence Calculator Excel Workbook)

11.5 Continuing Care

Additional clinical ADHD care will be at the discretion of the study physician, participant, and participant's family. This may include, but is not limited to, the following: 1) immediate transition of atomoxetine prescribing to the primary care physician or other specialist identified by the family, 2) continuation of atomoxetine prescribing and follow-up care by the study physician, 3) discontinuation or weaning of atomoxetine when deemed clinically appropriate. If the participant's ADHD care will be transitioned to their primary care provider or another community provider. A letter describing the course of the participant's care in the research trial will be provided to the family and the care provider. (See "End of Study ADHD Care Transition Letter" in the Appendix N)

11.6 IBC

The following IBC protocols will be used in this study:

- IBC Protocol #18-27 "Processing of Blood, Urine, Saliva, and Cerebral Spinal Fluid Samples from Clinical/Research Studies in the Clinical Pharmacology Research Laboratories"
- IBC Protocol #18-10 "Processing of Blood, Saliva, and Urine Samples from Clinical/Research Studies in the Pediatric Clinical Research Unit"

12.0 Data and Sample Banking

- Data Banking Procedures: All data collected on study related CRFs and participant data on the EMR assessing participant outcomes (in

particular, participant safety, efficacy, and toxicity) will serve as source documents for this study. For ease of analysis and logistics of sharing data with the study team members, a subset of the complete dataset that are pertinent to assessing the research aims will be entered in a REDCap database. This is currently being developed and is tentatively titled, "U54 Longitudinal Atomoxetine – Clinical Outcomes 2.0." All study personnel, research associates, and U54 collaborators may have access to the data, however on a need to know basis.

- Sample Storing Procedures: Samples will be stored in –80 degree C freezers housed within the department of Clinical Pharmacology, Toxicology, and Therapeutic Innovation. These samples will be kept and stored here until all data analysis is complete and all findings from this research study have either been finalized or published. All study personnel, research associates, and U54 collaborators may have access to the samples. Following completion of this study and processing of all samples, these samples will be moved to the Pharmacokinetics/Pharmacodynamics (PK/PD) Repository (see the corresponding bullet point below)

- Procedures to release data or samples: Researchers within Children's Mercy Hospital, or their collaborators, who may find these data and/or specimens useful to answering future research questions may submit a written request to the PI of this study (in the event that data analysis and collection are still occurring) or the PI of the PK/PD Repository (following study completion). Permission will be granted by written letter by the respective principal investigator(s) (AppendixK). The distribution of data and/or samples will be facilitated by the PI and study personnel in the event that data and samples are shared prior to the conclusion of this study. Following the conclusion of this study and transfer to the PK/PD repository, release of data and specimens are the responsibility of the PK/PD Repository PI and Manger and will follow the protocol Pharmacokinetics/Pharmacodynamics Repository (IRB# TBD).

- Transfer to the Pharmacokinetics/Pharmacodynamics (PK/PD) Repository: Following conclusion of this study, samples will be transferred to the Pharmacokinetics/Pharmacodynamics Repository which will be maintained by the Children's Research Institute Biorepository (CRIB). The Pharmacokinetics/Pharmacodynamics Repository will be an identified repository which is comprised of clinical data, research data, and biological samples (e.g. blood, urine, tissue

samples, etc.) that may be used prospectively to answer research questions relating to pediatric clinical pharmacology. The data distributed for prospective future research will be de-identifed by CRIB personnel as is described in the PK/PD Repository protocol (IRB # TBD).

- Samples from minors, plan for re-contacting and obtaining consent: The study population will include minors whose samples will be retained for long term research. The permission/assent and consent forms for this study will provide participants with the option of allowing retention of their samples and data in the PK/PD Repository for prospective future studies. If the participant turns 18 at the time of data collection, the participant will be reconsented. If the participant turns 18 after data collection has taken place, no attempt shall be made to recontact the participant (see section 25.0).

13.0 Genetic Analysis Information

- Genome-wide analysis and single nucleotide polymorphism (SNP) analysis will be conducted on participant samples.

- We do not expect to discover paternity findings, as we are not enrolling trios in this study. However, given that we are assaying for genetic variants in biochemical pathways that affect neurotransmitter levels and activity, it is possible that findings may be made regarding other diseases. In particular, findings may be made where there is significant overlap between ADHD and the disease in question, such as autism or anxiety.

- Genetic testing (microarray SNP analysis, PCR sequencing, etc.) to determine ATX dose and study design will not be conducted in a CLIA-certified lab. Genome-wide testing will be conducted at the Children's Mercy Hospital Genome Center, which is CLIA-certified. Additional testing may be conducted should more information regarding genes associated with ATX pharmacokinetic, pharmacodynamic, and clinical outcomes become available. The assays may be conducted in either the Clinical Pharmacology laboraties or at the Genome Center, depending on assay availability.

14.0 Sharing of Results with Subjects

- Sharing Results with Participants: Results from genetic testing of pharmacogenes, such as, *CYP2D6, CYP2C19, CYP2B6*, etc. will be shared with participants and their families. In particular, *CYP2D6* genetic information will be pertinent to research participants as this result determines the duration of Research Visit 2, Research Visit 3, and Research Visit 4. *CYP2C19* data will also be shared given the number of available medication guidelines written for this pharmacogene. Should additional actionable pharmacogene information become available during the course of this study, further information may be shared with the family. Genetic information will be explained in a letter (Appendix L) to the participant/family. The letter will state that the genotyping information obtained from this study are research results that are used to achieve the research goals of this study. These results were obtained from our research lab which does not adhere to CAP/CLIA guidelines. Because of this, these results cannot be included in the participant's medical record and should not be used to make medical decisions. The research team will inform participants/families that the Genotype- and Ontogeny Linked Dose Individualization and Clinical Optimization for Kids (GOLDILOKs) Clinic can facilitate obtaining genetic testing that is CAP/CLIA certified at the expense of the participant. Additonally, the GOLDILOKs Clinic can interpret the genetic results obtained from CAP/CLIA labs for the participant and their families. The results obtained by the GOLDILOKs clinic can be documented in the participant's medical record for future medical decision-making. This practice complies with the CRI Policy and Procedure, "Research Documentation in the Electronic Health Record" approved on 09/26/2019.

- Sharing Incidental Findings with Participants: Incidental findings that a discovered during the course of this study may be released to participants and their families for the purposes of seeking further care (Appendix M).

- Sharing Results with Providers: Participant results from genetic testing of pharmacogenes will be documented in the EMR if the family requests a referral to the GOLDILOKs Clinic for further interpretation of genetic results. However, this note will include a statement that these data were not obtained from a CLIA-certified lab. At the end of the study, or when the study participant withdraws or is lost to follow-up, a final research note will be included to describe the atomoxetine regimen for the participant while in the study. This note will be forwarded to

the Study Physician to aid in the transition of care of the participant from the study to a healthcare provider.

15.0 Reimbursement, Payment and Tangible Property provided to subjects

15.1 Payments

Below is the payment schedule for activities related to participation in this study. Payments to subjects will be made via Greenphire. Families will be asked to provide a social security number for their child so that their child may be reimbursed.

Activity	Compensation
Completion of Research Visit 1	$50.00
Completion of Research Visit 2, 0 –12 hours	$200.00
Completion of Research Visit 2, 12 – 24 hours	$60.00
Completion of Research Visit 2, return 48 hours	$30.00
Completion of Research Visit 2, return 72 hours	$30.00
Completion of Research Visit 3, 0 –12 hours	$200.00
Completion of Research Visit 3, 12 – 24 hours	$60.00
Completion of Research Visit 4, 0 –12 hours	$200.00
Completion of Research Visit 4, 12 – 24 hours	$60.00
Completion, return of drug taking calendar, and drug bottle at Research Visit 3	$15.00
Completion, return of drug taking calendar, and drug bottle at Research Visit 4	$15.00
Return of drug taking calendar and drug bottle at 6-Week Visit with Study Physician	$10.00
Return of drug taking calendar and drug bottle at 12-Week Visit with Study Physician	$10.00
Return of drug taking calendar and drug bottle at 18-Week Visit with Study Physician	$10.00

Please note, visit length is expressed in hours post-drug administration.

The maximum compensation for participants in Study Design 1 is: $950.00
The maximum compensation for participants in Study Design 2 is: $710.00
The maximum compensation for particpants in Study Design 3 is: $830.00

16.0 Withdrawal of Subjects

16.1 Anticipated circumstances in which subjects will be withdrawn from the research study

- Participant experiences a serious adverse event and there is a probable association between the serious adverse event and study drug and/or research procedures.
- Participant experiences an adverse event, and there is a high probability of an association between the adverse event and study drug and/or research procedures and the adverse event interferes significantly with the participant's well-being (e.g. daily stomach aches).
- Participant requires care beyond what the research study team can provide while still meeting eligibility criteria (e.g. participant requires medication for tics that interferes will study drug).
- Participant is lost to follow-up.
- Participant is not compliant with study procedures.
- Participant revokes consent/assent.
- Participant no longer meets inclusion/exclusion criteria (e.g. abnormal safety labs).
- Participant does not have adequate response from ATX at 12-week visit and 18 week visit (<10% reduction in symptoms).

The PI may remove the participant from the study without the LAR's permission or participant's consent/assent if the above circumstances occur.

16.2 Procedures for orderly termination/subject withdrawal

1. PI, Study Physician, or individual designated in the Delegation of Authority will contact the study participant and their family.
    a. If the participant initiates the withdrawal, reasons will be documented and if applicable, intervention or plans for follow-up will made.
    b. If the study team terminates the subject's participation in the study, study personnel will clearly explain the reasons for removal from the study and if applicable,

discuss next steps for necessary interventions or plan for follow-up care (e.g titration schedule for discontinuing study drug, etc).

2. If applicable, adverse events related to withdrawal will be promptly reported to the IRB per CM Policy 5.11 "Reportable Events". Adverse events will continue to be followed until resolution.
3. Study personnel will complete end-of-study and withdrawal documentation in the subject binder.
4. All previously collected samples and data, regardless of whether participant completed the research study in its entirety, will be saved for data analysis.
5. Transfer of care procedures from the study physician to the participant's primary care provider will take place in accordance with those outlined in section 11.6 for participants completing the study in its entirety.

17.0 Risks to Subjects

- Confidentiality Risk: There is a risk of loss of confidentiality. Confidentiality will be protected to the greatest extent possible. There is also a risk of loss of confidentiality when using the internet. This is especially pertinent when filling out questionnaires using a web interface. The study team will make every effort to protect the participant's confidentiality such that this risk is minimal.

- Blood Draw: Risks of drawing blood include discomfort and/or bruising. A needlestick can be greater than minimal risk. However, numbing spray and topical lidocaine can be provided to minimize pain associated with needle sticks. In addition, the study team will inquire about needlestick fears and comfort techniques prior to the first needlestick.

- Allergic reaction: It is possible to have an allergic reaction to atomoxetine. Currently, there is no way to predict whether an individual will have an allergic reaction to atomoxetine *a priori*. However, the percentage of individuals experiencing allergic reaction is low. The first dose of atomoxetine will be administered

to the participant in the PCRU. Medications and procedures are in place should a participant have an allergic anaphylactic reaction to atomoxetine.

- Genetic Risks: The unauthorized disclosure of genetic information may have an impact on a participant's employability, insurability, immigration status, paternity suits, or social reputation. These risks include the chance of discrimination. The study team will make every effort to protect the participant's genetic such that this risk is minimal. Additionally, the genes investigated in the study have thus far only been associated with drug pharmacokinetics and pharmacodynamics. The risk of discrimination with this type of genetic information is lower compared to genes linked to predisposition of disease.

- Side Effects Risks: Participants may experience side effects of atomoxetine related to the way the medication is dosed. Typical side effects include increased heart rate, headache, stomach upset, nausea, sleepiness, and emotional lability. In addition, there are reports of suicidal ideation with atomoxetine use. The level of risk is variable given that the severity of side effects can be different among participants.

- Psychological Risks: There may be psychological risks associated with answering questions about the participant's mood and behaviors at home and at school. The level of risk in answering psychological questionnaires would be equal to an individual seeing care for ADHD and comorbidities associated with ADHD, as many of these questionnaires are used to assess symptoms and improvement. We therefore expect the psychological risks to pose no increase in psychological risks over baseline.

18.0 Potential Benefits to Subjects

- Personalized Medication Dosing: Participants will receive more personalized dosing of atomoxetine therapy through participation in study.

- Genetic Information: Participants will have genetic information returned to them (as it is related to duration of study visits) and will be given information about the GOLDILOKs Clinic at Children Mercy's Hospital where they can learn about the impact that their unique genetic information has on the way their body processes ATX and numerous other medications. As mentioned previously in section 14.0, it will be explained to families that genetic testing results obtained as part of the study cannot be used by GOLDILOKs Clinic for interpretation, but rather, that the testing will need to be repeated in a CLIA-certified lab. If the participant and/or family would like to be seen in GOLDILOKs Clinic and repeat genetic testing, these services will not be paid for by the study and will be the responsibility of the participant/family.

19.0 Investigator Assessment of Risk/Benefits Ratio

Select as applicable:	**Pediatric Risk Category:**	
	Category 1	Research not involving greater than minimal risk (45 CFR §46.404 and 21 CFR §50.51)
x	Category 2	Research involving greater than minimal risk but presenting the prospect of direct benefit to the individual subjects. (45 CFR §46.405 and 21 CFR §50.52)
	Category 3	Research involving greater than minimal risk and no prospect of direct benefit to individual subjects, but likely to yield generalizable knowledge about the subject's disorder or condition. (45 CFR §46.406 and 21 CFR §50.53)
	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. (45 CFR §46.407 and 21 CFR §50.54)
Select if applicable:	**Adult Risk Category:**	
	Not Greater than Minimal Risk	
x	Greater than Minimal Risk	

20.0 Data Management

20.1 Data Analysis Plan

Population to be Analyzed:

- The pharmacokinetic population will consist of all participants with sufficient plasma concentrations for calculation of all pharmacokinetic parameters.
- The pharmacodynamic population will comprise of all patients with sufficient DHPG plasma concentrations, adequate data from pupil dilation and eye-tracking measurements, and cardiovascular measures (heart rate and blood pressure). Additional metabolites analyzed via metabolomics platform will also be analyzed (as discussed below).
- All participants who meet criteria for determination of both pharmacokinetic and pharmacodynamic parameters and have at least partially completed the study will be evaluated for pharmacodynamic linked clinical outcomes analysis. A partial completion is any participant where at least Research Visit 1, Research Vist 2, Research Visit 3, Intake Visit with Physician, and 6-week Visit with Study Physician have been completed. Clinical outcomes will be analyzed in four ways:
    - Response: responder vs. non-responder
    - Toxicity: tolerant vs. non-tolerant
    - Adherence: low, medium, and high
    - Adverse event reporting
- In addition, all participants for which demographic and background characteristics and concomitant medication history were collected will be analyzed.
- All completed and returned U54 Study Booklet questionnaires and surveys will be used to assess the effectiveness of the U54 Study Booklet in study comprehension. Feedback from the survey will be compiled for analysis.

Pharmacokinetics:

Estimation of the pharmacokinetic parameters and generation of the statistical analyses on the PK parameters will be performed by The Division of Clinical Pharmacology Toxicology & Therapeutic Innovation at the Children's Mercy Hospital, Kansas City, MO. Pharmacokinetic analyses will be conducted using Kinetica™ version 5.0 (ThermeElectron, Philadelphia, PA). Plasma ATX concentration versus time data will be curve fit using a peeling algorithm to generate initial polyexponential parameter estimates. Final parameter estimates will be determined from an iterative, nonlinear weighted least squares regression algorithm with reciprocal ($1/y^2calc$) weighting. Model-dependent pharmacokinetic parameters will be calculated from final polyexponential parameter estimates. Final model selection will be performed by evaluating the

goodness-of-fit criteria including; the objective function, Akaike & Schwartz criteria, standard deviations and coefficients of variation for the polyexponential parameters estimates and the correlation matrix detailing the degree of interdependence between the parameter estimates for each model. In the event that an insufficient number of post-peak ATX plasma concentration time points are available, a model-independent approach will be used and parameters of interest calculated using standard techniques. Specifically, C_{max} and T_{max} will be obtained by direct examination of the plasma concentration versus time profile. The area under the plasma concentration versus time curve during the sampling period (AUC_{0-n}) will be calculated using the mixed log-linear rule and extrapolation of the AUC to infinity ($AUC_{0-\infty}$) calculated by summation of AUC_{0-n} + Cp_n/λ_z, where Cp_n represents the final plasma concentration predicted from the fitted apparent terminal elimination phase and λ_z is the apparent terminal elimination rate constant. Model-independent pharmacokinetic parameters will then be calculated using standard (i.e., statistical moment theory) techniques. ATX metabolites *N*-desmethylatomoxetine (NDM-ATX), 4-hydroxyatomoxetine (4-OH-ATX) and hydroxyl-2-carboxylatomoxetine (OH-2-COOH-ATX) pharmacokinetics will be evaluated in a fashion analogous to that described for the parent compound.

The following PK parameters will be derived by non-compartmental analysis from plasma concentration data following a single dose study drug administration:

C_{max}	Peak plasma concentration determined from the observed data
T_{max}	Time to observed maximum drug concentration
$AUC_{0-\tau}$	Area under the plasma concentration versus time curve, determined by log-linear trapezoidal estimation, to the 12 or 24 hour time point (depending on CYP2D6 AS designated study design).
$AUC_{0-\infty}$	Area under the concentration versus time curve to infinity, determined by log-linear trapezoidal estimation $AUC_{0-\tau} + C_T/k$ Where k is the elimination rate constant defined as the negative slope following maximum concentration determined by least squares regression from terminal phase data.
$T_{½}$	Terminal half-life determined by quotient 0.693/k
CL/F	Oral clearance determined by quotient dose/ $AUC_{0-\infty}$
Vd/F	Terminal phase volume of distribution determined by Dose/k· $AUC_{0-\infty}$

fm	Fraction of drug metabolized through different pathways of biotransformation

The following PK parameters will be derived by non-compartmental analysis from plasma concentration data following chronic dosing to steady-state of study drug:

$C_{max,ss}$	Peak plasma concentration determined from the observed data at steady-state
$T_{max, ss}$	Time to observed maximum drug concentration at steady-state
$AUC_{0-\tau,ss}$	Area under the plasma concentration versus time curve, determined by log-linear trapezoidal estimation, to the 12 or 24 hour time point (depending on CYP2D6 AS designated study design).
$T_{½}$	Terminal half-life determined by quotient 0.693/k
CL/F_{ss}	Oral clearance determined by quotient dose/ $AUC_{0-¥}$
Vd/F_{ss}	Terminal phase volume of distribution determined by Dose/k· $AUC_{0-¥}$

<u>iGO-PK Model Update</u>: The current iGO-PK Model (Version 1.0) uses ATX pharmacokinetic data from 24 pediatric participants (Brown et al., 2016). Since the building of Version 1.0, more ATX pharmacokinetic data has been collected from studies conducted at Children's Mercy Hospital (IRB# 16010069) and this current study. The data will be used to update and refine the iGO-PK model when it has been determined that sufficient new data has been collected. This determinination will be made by the Principal Investigators and other Co-Investigators.

<u>Pharmacodynamics:</u>
Estimation of the pharmacodynamics parameters and generation of statistical analyses will be performed by The Division of Clinical Pharmacology, Toxicology & Therapeutic Innovation at the Children's Mercy Hospital, Kansas City, MO. An indirect model of pharmacodynamic changes, using change in plasma DHPG as a function of systemic plasma ATX concentration will be evaluated using Phoenix NLME (Certara, Princeton, NJ, USA), NONMEM (Icon, Dublin, Ireland), MATLAB (Natick, MA, USA), SAS version 9.4 (SAS, Cary, NC, USA), and R (The R Foundation). Concentration changes in DHPG as a function of change in systemic ATX levels will be fit to an indirect model (Equation A). Pharmacodynamic parameters k_{in}, k_{out}, I_{max}, and IC_{50} will be estimated for each metabolite studied using a nonlinear mixed effects model to take into account variability of pharmacodynamic marker change within the study population. Statistical regression models will be constructed to determine which patient covariates are

predictive of pharmacodynamic response. A maximal effect (E_{max}) model describing the relationship between ATX concentration and pharmacodynamic effect will also be assessed in this study (Equations B and C). Models will be compared using statistical tests (Sum of Squares, Loglikelihood, AIC, standard error of estimates, and distribution of residuals). Changes in ATX C_{max} and $AUC_{0-¥}$ will also be plotted against pupillometry and eye-tracking data gathered when testing participants with localization and discrimination tasks. To describe the relationship between attentional orientating as function of ATX exposure, the data will be regressed both linearly and non-linearly and best of fit statistics (Akaike Information Criteria and Bayesian Information Criteria) will be employed to determine the most appropriate model.

Global Neurotransmitter Analysis

$k_{in,\ DHPG}$	Rate at which DHPG is created endogenously
$k_{out,\ DHPG}$	Rate at which DHPG is catabolized endogenously
$I_{max,\ DHPG}$	Maximal fractional inhibition produced by ATX
$IC_{50,\ DHPG}$	Concentration of ATX that results in 50% of maximal inhibition
$E_{max,\ DHPG}$	Maximum response through interaction of drug and drug target
$EC_{50,\ DHPG}$	Concentration of ATX that results in 50% of maximal response

Rates of formation and elimination (k_{in} and k_{out}), maximal inhibition (I_{max}), concentration at which 50% maximal inhibition is achieved (IC_{50}), maximal response (E_{max}), and concentration at which 50% maximal response is achieved (EC_{50}) will be determined for all neurotransmitters assessed in the in the global neurotransmitter panel (see Appendix I for metabolite list). The Dietary Survey Questionnaire and microbiome information from saliva samples will be analyzed as possible covariates of the neurotransmitter profiles.

Genetic Analysis: Genome-wide analysis and single nucleotide polymorphism (SNP) analysis will be conducted on participant samples. Genetic testing (microarray SNP analysis, PCR sequencing, etc.) will be conducted to characterize variants in genes involved in disposition and response of ATX. Some examples of genes that may affect drug disposition and response include, but are not limited to, *CYP2D6*, *CYP2C19*, *ABCB1*, and *SLC6A2*. Genes associated with ADHD phenotype may also be analyzed, especially if ATX response (or lack thereof), is associated with a particular sub-population of individuals with ADHD. Additional testing may be conducted should more information become available regarding genes that may affect the prescribed drug's pharmacokinetic profile, pharmacodynamic characteristics, or clinical outcomes (i.e. safety, efficacy, or toxicity). Information on variants will be analyzed to determine impact on ATX safety, efficacy, or toxicity.

Pupillometry Analysis:

Changes in performance during Posner Cuing Task will be measured as a function ATX exposure and C_{max} concentrations. Pupil size will be measured during two types of activities: localization and discrimination tasks. Pupil size differences during localization tasks report on the individuals' baseline (tonic) release of norepinephrine. This corresponds to endogenous, or volitional attention. Pupil size differences during discrimination tasks is a composite of the individuals' baseline (tonic) and situational (phasic) release of norepinephrine. Currently it is unclear whether tonic, phasic, or both are impaired in individuals with ADHD.

Cardiovascular Marker Analysis:
Changes in heart rate will also be measured as a function ATX exposure and C_{max} concentrations. One side effect of ATX that is directly related to the pharmacodynamics of the drug is an increased heart rate due to increased concentration of norepinephrine in blood. We would like to understand the extent of this side effect as a function of increased systemic ATX exposure and C_{max}. By studying these two components, we will be able to distinguish ATX drug action in central and peripheral nervous systems.

Pharmacodynamic Changes Correlated with change in NICHQ Vanderbilt Assessment Scales:
All changes markers of pharmacodynamic action will be correlated with whether a child is classified as a responder or non-responder; and tolerant or intolerant (see Clinical Outcomes below for details). Extent of adherence will be used as a confounding factor in both of these analyses.

Clinical Outcomes:

Determine and document participant response to ATX therapy:

- Responders will be defined as:
    - Participants, who after approximately 18 weeks of continuous treatment with ATX, demonstrate ≥ 40% reduction of baseline ADHD symptom score;
    - OR the Clinical Global Improvement rating is 1 or 2 (Very Much Improved or Much Improved).
- Non-responders will be defined as:
    - Participants, who after approximately 18 weeks of continuous treatment with ATX, demonstrate < 40% reduction of baseline ADHD symptom score, AND the Clinical Global Improvement score ≥ 3.
    - Participants who discontinue ATX prior to the planned end of study (18 weeks) due to perceived lack of efficacy despite dose escalation.

Determine and document participant tolerance of ATX therapy:

- ATX intolerant participants are:
    - Participants who present with side-effects that significantly interfere with daily functioning or outweigh therapeutic benefit (CGI Efficacy Scale categorization)
    - Participants who must discontinue ATX for any of the following reasons: AST or ALT three times the upper limit of normal (ULN) for age, alkaline phosphatase five times the ULN, total bilirubin two times ULN, QT prolongation or dysrhythmia, hypertension (systolic or diastolic BP above 95% for age), tachycardia (HR above 95% for age)

Parent and teacher responses for the NICHQ Vanderbilt Assessment Scales will be obtained at baseline and ± 2 weeks of 6-week Visit with Study Physician, 12-week Visit with Study Physician, and 18-week Visit with Study Physician. If questionnaires are returned outside of this window, or are unreturned, these will be considered, "missing data." Determination of response, i.e. responder vs. non-responder, will be done using parent responses to questionnaires. Teacher response to questionnaires will also be analyzed to evaluate symptom reduction in a secondary setting. Partial return of teacher questionnaires are still useful in that these data may be used to correct for bias in parental response.

Questoinnaires conducted at baseline and at either 18-week visit with Study Physician or a Research Visit will be analyzed for change pre- and post-intervention. These include: BRIEF – parent, SCT, SCARED, CBCL, and School Intervention Questionnaire.

Statistical analyses of clinical response in the population studied will be performed by The Division of Health Services and Outcomes Research at the Children's Mercy Hospital, Kansas City, MO. SAS version 9.4 (SAS, Cary, NC, USA) and R (The R Foundation) will be used to determine if ATX responders and non-responders can be differentiated by a subset of patient specific covariates. Variables that will be included in this analysis are data collected at the patient's initial clinic visit: DHPG, *SLC6A2* genotype, ADHD subtype, ADHD severity, co-morbid anxiety or oppositional defiant disorder, age, and gender. Predictive power of classification models will be assessed by computing the area under the receiver operating characteristic curve and/or by k-fold cross-validation. Two-sample t-tests or non-parametric alternatives will be utilized to test for group differences in percent decrease in DHPG and a differences in proportions test will be used to assess the association between response and *SLC6A2* genotype. In additional, global metabolomic analyses will be conducted to determine whether non-responders and responders are intrinsically different in their biochemical make-up. This will further understanding of how differences in

response phenotype could be a result of different biological pathway involvement in ADHD. Digital maps will be constructed for partial least square-discriminant analysis (PLS-DA) models with prior knowledge of which volunteers were categorized as responders and non-responders. The variable influence on the projection (VIP) parameter will be used to identify which metabolites (i.e. pathways) are the most predictive of differentiating between the responder and non-responder populations. Analyses will also be conducted to determine mechanistic reasons for why certain individuals are non-responders or responders. Changes in pharmacodynamic endpoints will be compared between responders and non-responders. Specifically pharmacodynamic parameters such as I_{max}, IC_{50}, differences in pupil diameter changes in localizations and discrimination tasks, and overall change in DHPG will be compared between the two populations using MANOVA and/or appropriate two-sample test.

ATX pharmacodynamic data for each *SLC6A2* activity group will be examined using standard descriptive statistics. Analysis of variance will be used to compare parameters between pre-defined genotype populations followed by application of the appropriate parametric or non-parametric post-hoc test to determine where inter-group differences occur. Linear and non-linear least squares regression analysis will be used to examine the correlation between DHPG fitted parameters and changes in eye-tracking and pupillometry differences in *SLC6A2* genotype groups. Univariate analysis of variance, nonlinear regression and analysis of covariance will be used to evaluate the relationship between demographic variables (alone or in combination) with corresponding pharmacodynamic parameter estimates. All analyses will be performed in SPSS version 11.5 (SPSS, Chicago, IL) with the significance limit set at α=0.05. A sample size of 120 was deemed reasonable with the volume of patients being referred to the ADHD sub-specialty over a four year period. Powering of statistical tests was done assuming 20% attrition and data simulated for 96 patients. Assuming that 30% of the participants completing the study are categorized as responders, an α = 0.05, and power = 80%, a mean difference in covariates assessed of 0.6 standard deviations is detectable with a one-sided t-test. With zero attrition, the mean detectable difference in covariates of interest drops to 0.5 standard deviations.

Study Comprehension:
Responses to the U54 Study Booklet questionnaire will be summarized by frequency counts and percentages to each question asked. Improvement to the study booklet will be categorized by type of improvement suggested and summarized by frequency counts and percentages.

Background and Demographic Characteristics: Background and demographic characteristics will be listed by participant. Continuous variables will be summarized by sample size (n), mean, median, standard deviation (SD),

minimum, and maximum. Discrete variables will be summarized by frequencies and percentages.

Concomitant Therapy: Descriptive statistics such as frequency counts and percentages of participants who use concomitant medications will be provided by therapeutic classification and specific therapy.

20.2 Sample Size Determination

We hypothesize that there are at least two discrete ATX response phenotypes groups and additionally that these groups may be distinguished by differences in pharmacodynamic markers, and/or baseline patient characteristics. We will use two-sample *t*-tests (if transformed data are approximately Gaussian) or appropriate non-parametric alternatives to test for group differences in baseline and maximal change in DHPG, and a difference in proportions test to assess association between response group and SLC6A2 genotype. To estimate power for these tests we assumed 20% attrition and simulated data for 96 patients. For a 30% response rate among study completers (a conservative estimate), α = 0.05, and power = 80%, a mean difference of 0.6 SDs between responders and non-responders (eg, on DHPG) was detectable using a one-sided *t*-test. With 120 patients (zero attrition) a 0.5 SD difference was detectable. Depending on the rate and nature of attrition and missing data we will consider non-response weighting or data imputation to reduce estimation bias and improve power. We assessed power for the difference in proportions test based on a 45% probability of a given patient having at least one SLC6A2 lower expression "T" allele (*i.e.,* frequency of *SLC6A2* -3081 AT and TT genotypes ~45% based on a minor T allele frequency of 0.061 (http://useast.ensembl.org/Homo_sapiens/Variation/Population?db=core;g=ENSG00000103546;r=16:55655604-55706192;v=rs28386840;vdb=variation;vf=109832868)).

Assuming attrition of 20% and a response rate of 30%, power was 81% to detect a 0.23 difference in allele frequency between responders and non-responders using a one-sided test. With 120 patients a 0.20 difference was detectable with 80% power.

While these analyses are central to our hypothesis, we recognize that other plasma metabolites or genetic differences may contribute to varying drug response. A target metabolite panel will be assayed in each participant and statistical analyses of these will be analogous to DHPG. A panel of genetic variants will also be analyzed for contribution to variable drug pharmacokinetics, pharmacodynamics, and clinical response in the same manner as the *SLC6A2* variant described above.

20.3 Confidentiality

Research records which include, but are not limited to: participant study binders, financial binders, regulatory binders, study logs, etc. These records will be stored in a locked cabinet in the study coordinators' office or will be stored on Children's Mercy Hospital secure server. Any electronic files containing PHI will be password protected. Access to research records and data will be restricted to study personnel and project collaborators. Information shared with project collaborators will be on a need to know basis and will be limited to the information needed to perform thorough data analysis. Study personnel who will be in contact with PHI will be trained on PHI practices in accordance to Children's Mercy Hospital standards and will regularly be certified per institution policy.

20.4 Certificate of Confidentiality

A Certificate of Confidentiality has not been issued for this investigation. The original notice of award for this grant (Funding ID: 1-U54-HD090258-01) was issued on 09/23/2016, which is prior to the date when Certificates of Confidentiality were automatically supplied by the National Institutes of Health.

20.5 Quality Control and Assurance

Quality Control (QC) and Quality Assurance (QA) of data collected as a part of this investigation will be described in a Clinical Quality Management Plan (CQMP) (Supplementary Documents, "Clinical Quality Mangement Plan."). Under this plan, QA and QC procedures will take place at each subject study visit and on a quarterly and annual schedule, and as needed in response to staff or process changes. Key components reviewed in these processes will include: consent and permission/assent processes and documentation, review of participant-related CRFs, review of study drug accountability logs, review and update of the delegation of authority, review and update of the training log, and review and update of the regulatory binder. Documentation of all QC and QA processes outlined in the CQMP except for visit checklists will be maintained in the Quality Management Binder alongside the study regulatory binders. Visit checklists will be maintained in the study subject binder.

20.6 Data and Sample Handling

All samples will be stored within the Clinical Pharmacology laboratories under appropriate storage conditions. Access to the laboratory is secured by card access only. Samples will be associated with a study identification number: U54-XYZ (where XYZ denotes a number assigned by study personnel). Samples will be stored and kept until such time that all samples have been processed, data are

finalized, and manuscripts/publications detailing study findings have been completed. All research personnel, research associates, and laboratory staff will have access to samples and research data. Sample information may be shared with associates of study personnel (e.g. grant collaboraters) for the purposes of data analysis that are relevant to achieving study objectives and aims detailed in the grant funding this project (1-U54-HD090258-01).

Receipt and transmission of data and/or samples will be conducted by study personnel designated in the Delegation of Authority Log. Samples that need to be transported or transferred to a facility outside of Children's Mercy Hospital will be shipped overnight on dry ice, following the Children's Mercy Hospital's Policy, 1200.RLSM Transport of Research Materials Ver1.2.

21.0 Provisions to Monitor the Data to Ensure the Safety of Subjects

21.1 Data Safety Monitoring Board

The PI and study personnel will be responsible for monitoring the safety of participants. Additionally, a Data and Safety Monitoring Board (DSMB) will be organized to review the progress of the study and ensure the safety of study participants per Children's Mercy Hospital Policy 11.01 Data Safety Monitoring. The board will consist of faculty from both the Children's Mercy Hospital, national experts on clinical pharmacology, and physicians who have previously or are actively involved in the care of children with ADHD. The board will consist of:

- James McCracken, MD, PhD
- Erika Nurmi, MD, PhD
- Bridget Clark, DO
- Jennifer Goldman, MD, MS

Dr. McCracken and Dr. Nurmi are child psychiatrists at UCLA Mattel Children's Hospital and Resnick Neuropsychiatric Hospital at UCLA. Bridget Clark is a child psychiatrist at Children's Mercy Hospital. Jennifer Goldman is an infectious disease physician with expertise in adverse drug reactions.

The board will meet on a bi-annual basis and after all participants have been enrolled and completed study procedures. Further ad-hoc meetings will be determined by the PI and DSMB, particularly in the event of new or significant findings. Documentation of the meetings will be recorded and stored in the Division of Clinical Pharmacology, Toxicology & Therapeutic Innovation at Children's Mercy Hospital and Clinics and will be submitted to myIRB. The DSMB will be responsible for monitoring adverse events, serious adverse events,

enrollment numbers, protocol violations/deviations, participant withdrawal(s)/termination(s), and efficacy data.

Collection of Safety Information:
Safety information will include source documents that record measures to reduce risk for participants (section 11.3), QC/QA findings reports (section 20.5), and adverse events (section 4.2).

Please Supplementary Documents, "DSMB Charter," and "DSMB Protocol" for further details.

22.0 Provisions to Protect the Privacy Interests of Subjects

22.1 Privacy Interest

The study will follow standards established by the following Children's Mercy Hospital policy: Request Restrictions to Protected Health Information.

22.2 At Ease

The study team will allow the participant and their families time to decide whether he/she wants to both participate and continue in the study. Families are given at least a week to review the permission/assent forms before deciding whether to participate in this study. During enrollment, families are encouraged to ask questions and additionally, a supplemental illustrated booklet detailing study procedures is provided to the family. The study team will review the type of information that is being collected with the participant and their family. The participant and their family will also be asked at each research visit whether they would like to continue to participate in the study.

22.3 Permission to Access Data

The study will follow standards established by the following Children's Mercy Hospital policies: Confidentiality; HIPAA & Privacy Manual Standard.

22.4 PHI to be Accessed

The electronic medical record will be accessed for the purposes of recruitment and prescreening (see section 9.0). Data related to ADHD and comorbidity symptoms will be collected from the participant EMR for the purposes of data analysis.

22.5 HIPAA Authorization

The study has a partial HIPAA waiver for the purposes of recruitment and prescreening (see section 9.0).

23.0 Compensation for Research-Related Injury

There is no compensation for research-related injury. Should illness or injury occur during participation in this study, treatment is available and will be provided at the usual charge. Payment for this treatment will be the responsibility of the participant.

24.0 Economic Burden to Subjects

Participation in Research Visit 1, Research Visit 2, Research Visit 3, and Research Visit 4 will not result in any extra cost to the participant or their parents aside from personal time and travel costs to come to these visits.

Intake Visit with Study Physician, 6-Week Visit with Study Physician Visit, 12-Week Visit with Study Physician, and 18-Week with Study Physician may incur extra costs to the participant or their families depending on their insurance plan. This is because the Study Physicians involved in the care of the participants are considered consultants and these visits with the Study Physician may be considered as referrals to secondary care.

25.0 Permission/Assent/Consent Process

The research team will be obtaining permission and assent if the potential participant is younger than 18 years of age. Consent will be obtained if the potential participant is over 18 years of age and if the participant turns 18 during the study.

- Permission/assent or consent may be conducted in person with the study team at the PCRU or this may take place over the phone.
- The research team will wait at least for at least a period of one week between informing the prospective subject and obtaining either permission/assent or consent.
- Permission and assent will be reaffirmed prior to any research procedures take place at Research Visit 1, Research Visit 2, Research Visit 3, and Research Visit 4.
- This research study will be following the CM research policies regarding informed permission/assent/consent.
- This research team will be obtaining permission/assent/consent via telephone, and we will be following CM research policy 10.05 Telephone Process regarding telephone consent.

- The permission/assent and consent forms will contain a section for participants to allow their data and samples to be stored in the PK/PD Repository for use in prospective research.

Subjects Who Are Not Yet Adults

Prospective subjects under the age of 18 are considered minors and have not attained the legal age for consent to treatments or procedures involved in the research under the applicable law of the jurisdiction (the state of Missouri) in which the research will be conducted.

- Parental permission need only be obtained from one parent even if the other parent is alive, known, competent, reasonably available, and shares legal responsibility for the care and custody of the child. Permission to participate in the research study is done so by the potential participant's legally authorized representative (LAR). LARs who are not the potential participant's biological parents must provide documentation (e.g. adoption paperwork) prior to or at the time of the permission/assent process. The study team will maintain a copy of these records as a part of the participant's study binder. Assent will be obtained for all children greater than or equal to 7 years of age. For children under the age of 7, assent is not required, but may be obtained at the discretion of the study team.

- Documentation of both permission and assent will be recorded on study specific permission/assent forms and will be signed and dated by the parent or LAR and potential participant (if applicable). If potential participant is unable to sign, the study team will document the reason for this. If assent is not obtained for a child under 7, this will also be documented on the permission and assent form.

Subjects Who Become Adults at 18 Years of Age At the Time of Data Collection

Participants who turn 18 years old while actively participating in this study procedures will be consented at age 18 using a full Adult Consent form.

Subjects Who Become Adults at 18 Years of Age After Data Collection is Completed

The study is seeking a waiver of consent for participants who turn 18 after data collection has been completed due to: the amount of time needed to complete enrollment for this study, study procedures, data processing, and data analysis (~ 5 years) and the potential of not completing study procedures by the time many of the participants turn 18; the fact that participants may move; or may not follow up; and the high number of participants. This study could not be reasonably completed without a Waiver of Consent for children reaching the age of majority after study procedures have been completed.

26.0 Documentation of Permission/Assent/Consent

This research study will be following the CM Research Policy 10.04 Obtaining Permission/Assent/Consent.

27.0 Setting

The following are the main sites where the research team will identify and recruit potential participants:

- ADHD Clinic, Children's Mercy College Boulevard Clinics, 5520 College Boulevard, Ste 365, Overland Park, KS, 66211
- Children's Mercy, Broadway Clinics, 3101 Broadway Boulevard, Kansas City, MO 64111
- Children's Mercy West Clinic, 4313 State Avenue, Kansas City, KS 66102

Research data collection will be performed at the following sites:

- Pediatric Clinical Research Unit, Adele Hall Campus, Children's Mercy Hospital, 2401 Gillham Road, MO 64108
- ADHD Clinic, Children's Mercy College Boulevard Clinics, 5520 College Boulevard, Ste 365, Overland Park, KS, 66211
- Developmental and Behavioral Sciences, Adele Hall Campus, Children's Mercy Hospital, 2401 Gillham Road, MO 64108

28.0 Resources Available

<u>Facilities</u>

- PCRU: The Clinical Research Unit is a six-bed inpatient facility designed for the conduct of pediatric clinical research. The unit is located on the ground floor of the Hall Tower (0460.900). The unit has three private patient rooms and one room containing three patient beds with basic furniture. All rooms are equipped with emergency equipment and supplies, bathroom facilities and couches for parents to stay with

children. The unit has an exam room, two sample processing laboratories, medication room, kitchen, clean utility, dirty utility, a nurses' station with four workstations, photocopying and fax machine, and a Translogic tube system. The unit also has secured storage for medications and supplies.

- Drug Biotransformation/Analytical Laboratory: The analytical facilities of the Division of Clinical Pharmacology, Toxicology, and Therapeutic Innovation occupy approximately 1280 square feet in the CMH Pediatric Research Center and contains several analytical instruments, including three triple quadrupole mass spectrometers (a Waters Xevo TQD, a Waters Xevo TQS, and a Waters Xevo TQ-XS), a Waters Xevo G2-XS QtoF mass spectrometer, each with Acquity UPLC front end systems. Additionally, the lab has access to a Waters Ion key source coupled to an M-class Acquity that can be used for nanoflow type of experiments on all Waters systems. The laboratory also has three Agilent HPLC systems, two of which are equipped with fluorescence and UV/vis detectors and the third with a diode array detector, and two POC-One infrared spectrometers for analyzing [13C]O_2 breath test samples. Cold rooms, centrifuges, electrophoresis and blotting systems, and -20°C and -80°C storage are available to conduct experiments and store samples.

- Pharmacogenetics/Pharmacogenomics Laboratory: The Pharmacogenetic and Pharmacogenomics Core Laboratories has access to a variety of platforms to conduct genotype analysis: TaqMan analysis; High-Resolution Melt Curve analysis (HRM); traditional PCR-RFLP for SNP detection and long-range PCR to detect major gene arrangements. Genotyping panels have been established for many CYP genes, including: *CYP2D6* (the panel established by this group represents one of the most comprehensive genotyping panels for this highly polymorphic gene, which is notoriously difficult to genotype); *CYP2C9*; *CYP2C19*; *CYP2B6*; and the *CYP3A* gene locus. Quantitative PCR procedures have been established to determine copy number variation for *CYP2D6*, *SULT1A1*, and *UGT2B17*. Additional genotyping platforms, such as the Affymetrix DMET chip, are available through the Cytogenetics and Molecular Genetics Laboratory. The Pharmacogenetics/Pharmacogenomics Laboratory is equipped with standard and real-time PCR instruments, a 3730x DNA sequencer (Applied Biosystems), and a Kodak DS Image Station 440 CF (PC-controlled) for photodcumentation, data analysis, and storage. Centrifuges, electrophoresis and blotting systems, and -20°C and -80°C storage are available to conduct and store samples. The unit has nine 96-well PCR instruments, three quantitative real-time PCR cyclers (ABI 7900HT real time thermocycler; an Eppendorf realpex *ep*gradient S

Mastercycler and an MJ Research Opticon 2 Q-PCR instrument) as well as two EcoRealTime PCR instruments (Illumina) to facilitate HRM analysis. The laboratory also has a dedicated MiSeq platform that is housed in the Center for Pediatric Genomic Medicine.

29.0 Multi-Site Research

This section is not applicable to this study.

30.0 International Research

This section is not applicable to this study.

31.0 Appendices

Title	Appendix Title	Section Referenced Within Protocol
ATX PBPK Model Validation Study Summary	Appendix A	3.2
Message Center Template	Appendix B	9.3
Phone Message from ADHD Clinic Nurses	Appendix C	9.3
Study Coordinator/RA Recruitment Script	Appendix D	9.3
Recruitment Email or Letter	Appendix E	9.3
Study Coordinator/RA Scheduling Script	Appendix F	11.1
U54 Study Booklet Questionnaire	Appendix G	4.1, 11.2
Pupillometry Example	Appendix H	11.2
Neurotransmitters in metabolomic panel	Appendix I	20.1
Study Drug Timeline	Appendix J	5.2
Sample Letter to Release Data and/or Specimens by PI	Appendix K	12.0
Sample Letter to Share Genetic Results	Appendix L	14.0
Sample Letters to Release ADHD Results or Incidental Findings to Participants of this Study	Appendix M	9.3, 11.3, 14.0
Sample Letters for End of Study ADHD Care Transition	Appendix N	11.2
Literature Search and Documentation for Washout and Return to Baseline	Appendix O	11.2

Appendix A – ATX PBPK Model Validation Summary

The primary aim of this study is to validate a method to individualize ATX dosing to achieve a desired level of ATX exposure, referred to as the **i**ndividualized **G**enetic and **O**ntogenic **P**harmaco**k**inetic (iGO-PK) algorithm. This dosing algorithm relies on two separate models, one that is used to dose CYP2D6 PMs and a model for the rest of the population (i.e., the non-PM model). This dosing algorithm has been designed to calculate a dose of ATX for individual patients that is expected to result in ATX C_{max} concentrations at steady-state associated with a reduction in ADHD symptoms (Michelson et al, 2007) The model takes into account CYP2D6 genotype, which is converted into an activity score, weight, and obesity status. Validation and refinement of this dose prediction model is required to ensure that a C_{max} goal can be achieved reliably with the patient information that is provided. Additionally, we are interested in obtaining more data for *CYP2D6* Ims and PMs because of the small sample size in these phenotype groups in the initial pharmacokinetic study conducted by Brown et al (2016). The data collected from this study will also be used to refine the model, given that the initial model only utilized data from 23 children who represent a wide range of CYP2D6 activity.

Appendix B – Message Center Template

I am recruiting patients ages 6-18 with suspected or diagnosed ADHD for a study with the medication Atomoxetine (Strattera) through Clinical Pharmacology/GOLDILOKs. Atomoxetine is an FDA-approved medication for ADHD, but its use has been 2nd or even 3rd line for ADHD due to lack of efficacy. In our study, we are interested in using personalized medicine to tailor a dose to each patient to improve how this medication works for each child. In this longitudinal study, patients interested in non-stimulant medication will receive personalized Atomoxetine dosing based on their CYP2D6 genotype (the enzyme that breaks down the medication). Patients referred for this study will receive their ADHD care from a study physician for the next 6-8 months and have intermittent research visits in the Pediatric Clinical Research Unit (PCRU) at the Adele Hall campus to monitor how they are responding to and breaking down the medication. From this study, we hope to learn how to better use this medication for children with ADHD.

After reviewing this patient's chart, he/she is eligible for the study, and I see he/she has an appointment with you [timeframe]. If you, your patient, and his/her family are interested in starting this medication, please feel free to mention there is an ADHD medication study available. If he/she and his/her parents are interested, please feel free to either give them my contact information below to call or email me, provide the study handout available in each provider workroom, or simply reply back and I will reach out to the family.

If your patient is currently on an alpha agonist or atomoxetine for ADHD, unfortunately, he/she cannot enroll in our study. However, if your patient is currently taking a stimulant for ADHD, he/she will need to undergo a 1 week washout period prior to starting the study medicaiton. Please note, the study team must first obtain permission/assent or consent prior to any study procedures, including initiating a medication washout.

There is a slide regarding this study on the workroom monitors for you to refer to, but if you have any further questions, please feel free to reach out.

Thank you,

(signature + contact information)

Appendix C – ADHD Clinic RN

Hi, my name is _____ and I am a nurse/care provider at the Children's Mercy Hospital ADHD Clinic.

We reviewed the results from the questionnaires that you and your child's teachers filled out. Based on the answers we received, we believe that there is a high chance that your child may have attention deficit/hyperactivity disorder (or ADHD for short). *We would like to schedule an appointment for you to meet with our physician and clinical psychologist for further evaluation.* (NB: this italicized section about scheduling may be omitted if the caller has no intention to schedule an appointment.)

Because your child has a high likelihood of having ADHD, he/she **may be able to participate in an ADHD study that is taking place at Children's Mercy Hospital. If you/your child might be are interested in participating in this study, or would like to learn more about this study, we would be more than happy to pass your contact information to the study coordinator of this study for more detailed information.**

Would you be interested in this study, or would you like to learn more about this study?

1. Yes, please have the study coordinator contact me with more information
2. No, I am not interested in participating in the study at this time.

Appendix D – Study Coordinator/Research Assistant Recruitment Script

Hi, my name is _____, and I work in research at Children's Mercy Hospital. My team is working on an Attention Deficit and Hyperactivity Disorder, or ADHD, medication study for children ages 6-18. Do you have 5-10 minutes to talk about a voluntary study opportunity?

(If yes, continue. If no, ask to contact at a later date/time.)

Track 1 – I am contacting parents of new patients with upcoming appointments at the Children's Mercy ADHD Clinic on College Blvd to let you know about this study opportunity. I see that your child has an upcoming appt on MM/DD/YYYY.

Track 2 – Your child's healthcare provider, _____, at PCC on Broadway, contacted me about your interest in our ADHD study.

Track 3 – Your child's healthcare provider, _____, at _____, contacted me about your interest in our ADHD study.

(Track 4 and Track 5 contact us; this would be a return call or incoming call.)

First, I'd like to tell you about what we are studying and why. Then, if you are interested in learning more, I'll share what would happen during the study. First, please note that all research is voluntary, and your decision to participate or not will not affect the care your child receives here at Children's Mercy in any way. Feel free to stop me at any time if you have questions while I explain the study.

We are studying an ADHD medication, atomoxetine, (the brand is name is Strattera), in children ages 6-18 years old. This study would involve your child taking this medication. This medication is approved by the FDA for children with ADHD and has been on the market for about 15-20 years. It isn't used very often though. This is because it doesn't seem to work very well for some children. For some children, they behave much better in school and pay attention in class while taking this medicine. However, for other children, it doesn't seem to have much effect. There are lots of reasons why a medicine might not work for someone, and one of those reasons is how fast or slow they break down the medicine. We think that perhaps one reason this medicine doesn't work as well for some children is because the dose is too high or too low for how fast or slow they break it down. What we would like to do is draw a small amount of blood and run genetic testing to figure out how fast or slow your child breaks down the medicine. Together with other information such as age, weight, height, and ethnicity, we will come up with a dose that we think is just right for your child. We would ask your child to take this medicine for several months and see if it helps treat his or her ADHD symptoms. There is a chance that this medicine will not work for your child regardless of the dose,

and this is because the dose is just one piece of the puzzle. We want to understand why this medicine works for some children and doesn't for others. Ultimately by studying this drug in your child and other children, we hope to learn how to better use this drug for other children with ADHD in the future.

Do you have any questions for me at this point? *(yes/no)*

Next, I'd like to tell you what we would ask from you and your child during the study if you were to enroll.

This study takes place over about 6-8 months. There are 8 total visits—4 research visits and 4 clinic visits. At research visits, we study how your child is breaking down the medicine. At clinic visits, you and your child visit with a study doctor to see how the medicine is working at home and at school and change the dose if needed. First, we would bring you in for a research visit to gather information about your child, including his or her ADHD symptoms, health history, and draw blood for genetic testing and to make sure your child is health enough for the study. Then, we would have you and your child meet with a study doctor at a clinic visit to see if ATX would be a good fit. If so, we would then calculate a dose for your child and start your child on the medicine.
We will ask you and your child to come to the research unit (PCRU) at the Adele Hall campus to give the first dose of medicine. After we give the medicine, we will draw small blood samples multiple times over the course of several hours to measure the amount of drug in your child's blood. This visit will generally last about 12-14 hours. Depending on the information we learn about your child's genes, we may ask your child to stay overnight to measure more samples. This would be a 24 hour study. We will know how long your child's visits will last before you ever start the medicine so you can decide if this will work for your family. This visit is to help us understand how your child breaks down the medicine after just one dose.

Do you have any questions for me at this point? *(yes/no)*

After this, we will send you and your child home with a prescription of atomoxetine to take every day. Your child will see your study physician every month or 2 to see how the medicine is working at home and at school. If needed, the doctor may adjust the dose. In addition to these doctor's visits, we will ask you and your child to come back to repeat the blood samples again two times. One time is anytime between these doctor's visits, and the other time is after all of the doctor's visits are completed. These help us understand how your child breaks down the medicine after your child's body is used to taking the medicine every day.

So altogether, the study will last 6-8 months, include 4 clinic visits, and 4 research visits, and your child will take this medicine every day. This study is voluntary, so if at any point you or your child want to stop, you may and it will not affect the care your child receives at Children's Mercy in any way.

Do you have any questions for me at this point? *(yes/no)*

Please note, there are some medications your child cannot take to participate in this study. In particular, if your child is currently taking a stimulant medication for ADHD, he or she cannot continue this medication during the study. If you and your child choose to enroll in the study, we will ask your child to "washout" of his or her ADHD medication. This means your child will need to stop the ADHD medication and wait at least 1 week to "washout" the effects of the ADHD medication so that your child is back to his or her ADHD symptom baseline. Please note, if you do choose to enroll, we ask that you do not stop your child's ADHD medication until after you and your child have signed the study consent form with a study team member in person at your first research visit.

Do you have any questions about this part? *(yes/no)*

If you are interested in learning more, I would like to email or mail you some further information about the study for you to review with your child. I realize this is a lot of information, so please take the necessary time you need before deciding whether you and your child want to participate in this study.

Would it be okay to check back in with you? *(yes/no)*

When would you like me to contact you? *(record time or date)*

END

Appendix E – Recruitment Email or Letter

Thank you for your interest in our ADHD study. I have attached the study consent form, and a Frequently Asked Questions (FAQ) sheet for you to review. These include details about the study including the purpose of the study, what would happen during the study, risks, benefits, alternatives, etc. The consent form includes sections at the end for your signature, but please do not sign this. This form is simply for your information at this time. If you and your child choose to enroll in our study, a study team member will review this with you and your child in person and answer any questions you may have before we ask you to sign the form.

If your child is currently taking stimulant medication for ADHD, he or she cannot continue this medication during the study. Your child will need to stop, or "wash out" of his or her medication to start atomoxetine. We ask participants to stop ADHD medication for at least a week before starting atomoxetine so that the study team can better understand your child's ADHD symptoms off medicine and how this new medicine will work in your child's body. We ask that you do NOT stop your child's ADHD medication until after you and your child come in to sign the consent form and meet with a study doctor to talk about the washout.

When your child stops his or her medication, his or her ADHD symptoms will likely return. Your child may have trouble focusing and have more impulsive behavior. Some children may handle this brief time off ADHD medicine just fine while other children may have a lot of trouble at school and home. We know that as the parent, you know your child better than anyone else. Therefore, we want you to think about if washing out is okay for your child. These behaviors may impact school performance, after-school sports and activities, and how your child interacts with family, teachers, and other children. Some children may do better washing out of ADHD medicine over a school break when ADHD behaviors are less disruptive. While you look over the consent form and FAQ sheet, please consider if the washout is appropriate for your child.

If you have any questions or concerns, please feel free to reach out to us via phone or email.

Thank you,

ADHD Study Team

(Study Team Email)

(Study Cell Phone)

Appendix F – Study Coordinator/Research Assistant Scheduling Script

Hi, my name is _____ and I am a study coordinator/research assistant at Children's Mercy Hospital. Thank you for your interest in our study. I am calling to schedule your child's first research visit. But first, I'd like to talk to you about a few key points in the study.

First, I want to review how long visits are and how often visits occur.

1) Research Visit 1 lasts about 4-5 hours; Part 1 is about 1 hour and Part 2 is 3-4 hours.
2) Research Visits 2-4 last at least 14 hours. Depending on what we learn about your child, some children have 24 – 72 hour visits, and we will ask that your child stay overnight at the research center.
3) The first 2 study physician visits and first 2 research visits happen close together over the first 2-3 months of the study. After that, visits happen every month or 2.

Second, if your child will need to wash out of his or her ADHD medication, please do not stop the medication until after you sign the consent form.

Do you have any questions for me at this time?

Third, the study medicine, atomoxetine, is only available in capsules (pills), so children who enroll in the study will need to be able to swallow pills to start the medicine. If your child has trouble swallowing pills or has never tried swallowing pills, we are happy to work with your child to help teach him/her how to do this.

Lastly, I want to talk a little bit about the blood draws that happen in this study.

In this study, we ask participants to have multiple blood draws, and we know these needlesticks can be stressful to some children. We want to help make these needlesticks as stress-free as possible for any child who wants to be in the study. It can be hard to predict how a child will react to a needlestick. However, we have learned some things from other children who have enrolled in our study. Children who have never had a needlestick, such as a blood draw at a doctor's office, vaccinations, or even had an IV, tend to be nervous about the blood draw. Also, children who have general fears about hospitals, clinics, and healthcare situations and children who "just shut down" in unfamiliar situations have more trouble than others. I want to encourage you to think about how your child might do with the needlesticks and let us know what we can do to make this a pleasant experience for your child. When you come in for your first research visit, a study team member will talk to you to learn about your child's comfort level with needlesticks before we do any blood draws.

Do you have any questions for me at this time?

Great! If you are ready, I'd like to schedule your research visit.

(Schedule Date and Time)

This visit takes place at the Adele Hall campus, which is the main Children's Mercy campus located downtown. When you arrive, park in the visitor parking garage, check in at security, and go to the Pediatric Clinical Research Unit, or PCRU for short. This is located on the ground level next to the gift shop.

Please remember to bring your child's social security number to the visit. We are required to keep a record of this information for payment to any child participating in this research study.

Do you have any final questions for me?

(END)

Appendix G – U54 Study Booklet Questionnaire (please see PDF attachment for Study Booklet)

1. How much do you agree with each statement?

This booklet:	 Strongly Disagree	 Disagree	 Neutral	 Agree	 Strongly Agree
Helped me understand why this drug works differently in different people.					
Helped understand what a research visit is and what clinic visit is.					
Helped me understand what I would be doing at each visit.					
Helped me understand that I can choose to be, or not be in this study.					
Was useful to me as I was going through this study.					

2. What would be helpful in future study booklets?

Appendix H – Example of pupillometry and eye-tracking tasks

Figure 1 – Valid and invalid examples of the localization task, both with successful responses.

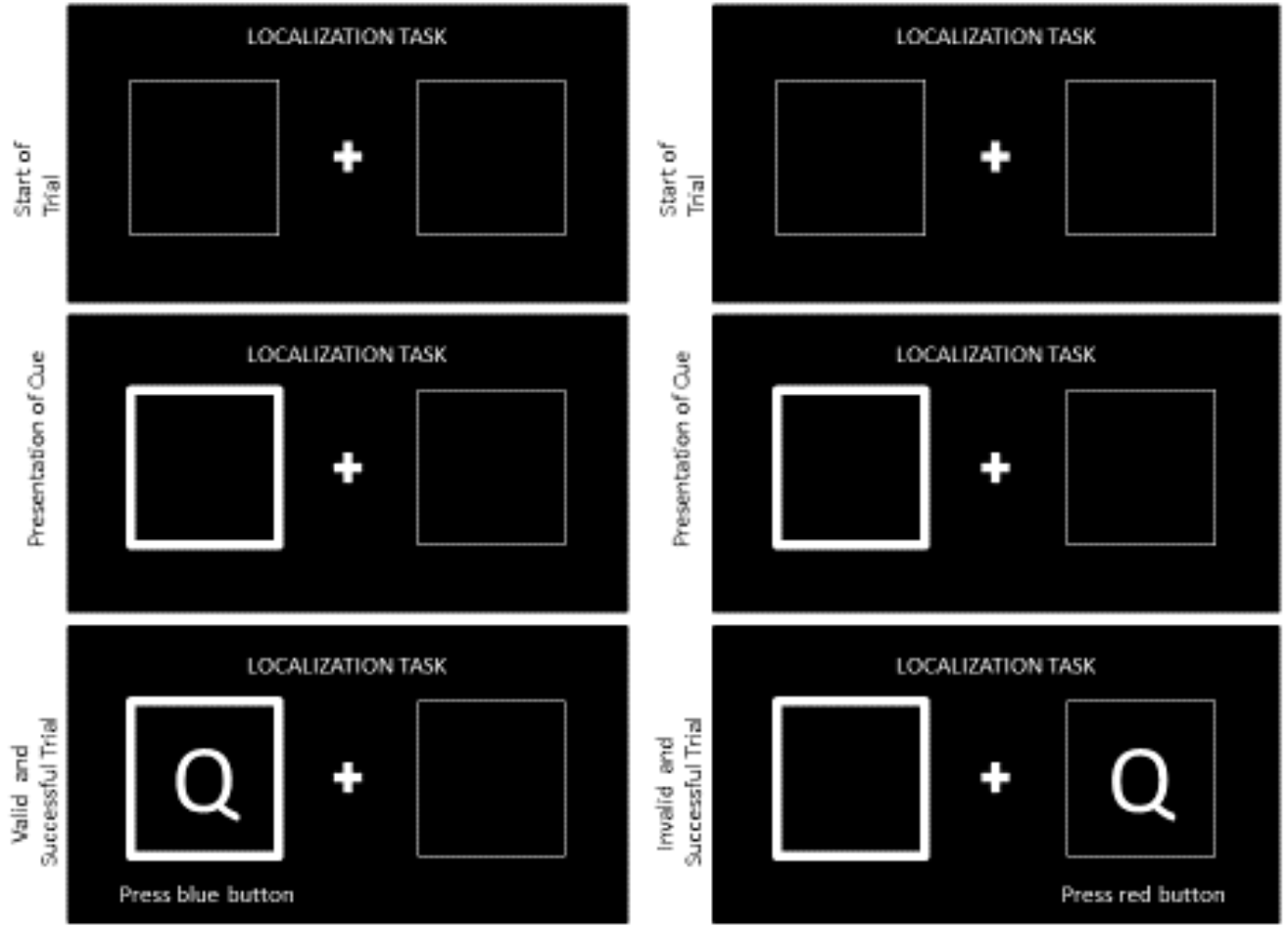
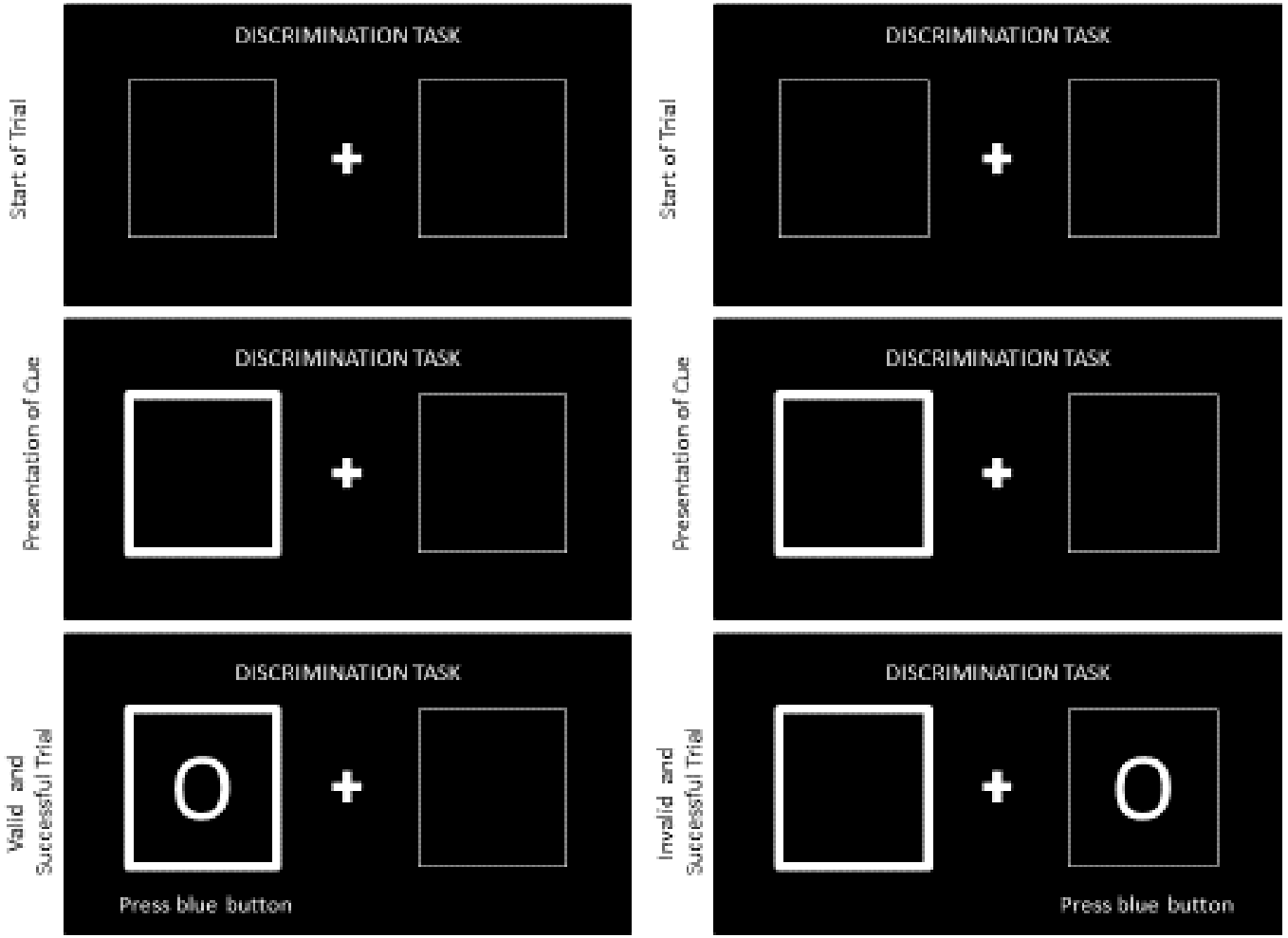


Figure 2 – Valid and invalid examples of the discrimination task, both with successful response.



Appendix I – Example of neurotransmitters in metabolomics panel

1. 3,4-dihydroxyphenylglycol (DHPG)
2. α-methyltryptophan
3. α-tocopherol
4. cysteine
5. δ-tocopherol
6. γ-tocopheral
7. guanine
8. guanosine
9. homogentisic acid
10. homovanillic acid
11. hypoxanthine
12. indole-3-acetic acid
13. indole-3-propionic acid
14. kynurenine
15. methionine
16. salicylic acid
17. serotonin
18. tryptophan
19. tyrosine
20. uric acid
21. vanillylmandelic acid
22. xanthine
23. xanthosine
24. 1,3-dimethyl xanthine
25. 1,7-dimethyl xanthine
26. 3-methoxy-4-hydroxyphenyl glycol
27. 4-hydroxybenzoic acid
28. 4-hydroxyphenylacetic acid
29. 4-hydroxyphenyllactic acid
30. 5-hydroxytrptophan
31. 5-hydroxyindoleacetic acid

Appendix J – Study Drug Timeline, a description of drug handling during the ATX PBPK-PD Clinical Outcomes Study

START of Intervention

↓

Designated Study Personnel/PI calculates the initial dose with the iGO-PK model

↓

Study personnel/PI shares the dose calculation with RA/Study coordinator/Designated Study Personnel and Study Physician following the participant

↓

RA/Study coordinator/Designated Study Personnel prepares IDS prescription for 6-8 weeks for Study Physician to sign.

↓

Study Physician signs prescription, scans, and emails back to RA/Study coordinator/Designated Study Personnel

↓

RA/Study coordinator/Designated Study Personnel emails the prescription to IDS to fill and files the original in the subject binder

↓

IDS brings the Rx to the PCRU for Research Visit 2/PK1

↓

At Research Visit 2/PK1

- 1 dose from the prescription is administered to the participant, medication label from IDS is scanned into MAR at dose administration. RN/Designated personnel doses. Topical lidocaine (optional) may also be dispensed.
- PI/Designated Study Personnel teaches family/participant about medication (review prescription details, study booklet calendar, when to contact, anticipated possible side effects, etc)
- PI/Designated Study Personnel give prescription to the family/participant to take home

- PI/Designated Study Personnel write a note in PowerChart documenting this education, the prescription dispensed, and any questions/concerns. Study physician co-signs this note.

↓

At Research Visit 3/PK2

- A prescription for a single dose (or two doses if Study Design 3) of atomoxetine (optional topical lidocaine) will be dispensed at Research Visit 3/PK2. A medication label is generated by IDS, so documentation of the dose is recorded in the nurse note. RN/Designated personnel doses.
- PI/Designated Study Personnel count the participant's remaining prescription and compare against medication adherence calendar to calculate adherence.

↓

Prior to 6-Week Visit, RA/Study coordinator/Designated Study Personnel prepares IDS prescription for 6-8 weeks for Study Physician to sign

↓

Study Physician signs prescription, scans, and emails back to RA/Study coordinator/Designated Study Personnel

↓

RA/Study coordinator/Designated Study Personnel emails the prescription to IDS to fill and files the original in the subject binder

↓

At 6-Week Visit,

- PI/Designated Study Personnel counts the participant's remaining prescription and compares against medication calendar to calculate adherence.
- Dispensing Rx to family occurs by any of the following ways:
    - RA/Study coordinator/Designated Study Personnel obtains prescription from IDS and to provide to family/participant. Hand-off of medication may occur in clinic or just outside CMH at a pre-determined location (e.g. the front entrance).
    - IDS brings the prescription to the outpatient pharmacy for the family/participant to pick up
- IDS/ RA/Study coordinator/Designated Study Personnel may use a courier service to deliver medication to CMK outpatient pharmacy for the

family/participant to pick up. This use of courier service will be determined by IDS policies and practices.
    - IDS Pharmacy ships the medication to the family in Home Care Pharmacy medication coolers via FedEx using OptiFreight. Shipment of medication will be follow IDS policies and practices.
- If the dose changes, RA/Study coordinator/Designated Study Personnel takes the old prescription and returns to IDS for disposal

↓

Prior to 12-Week Visit, PI/Designated Study Personnel prepares IDS prescription for 6-8 weeks for Study Physician to sign

↓

Study Physician signs prescription, scans, and emails to RA/Study coordinator/Designated Study Personnel

↓

RA/Study coordinator/Designated Study Personnel emails the prescription to IDS to fill and files the original in the subject binder

↓

At 12-Week Visit,

- PI/Designated Study Personnel counts the participant's remaining prescription and compares against medication calendar to calculate adherence.
- Dispensing Rx to family occurs by any of the following ways:
    - IDS brings the prescription to the outpatient pharmacy for the family/participant to pick up
    - IDS/RA/Study coordinator/Designated Study Personnel may use a courier service to deliver medication to CMK outpatient pharmacy for the family/participant to pick up. This use of courier service will be determined by IDS policies and practices.
    - IDS Pharmacy ships the medication to the family in Home Care Pharmacy medication coolers via FedEx using OptiFreight. Shipment of medication will follow IDS policies and practices.
- If the dose changes, RA/Study coordinator/Designated Study Personnel takes the old prescription and returns to IDS for disposal

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Prior to 18-Week Visit, PI/Designated Study Personnel prepares IDS prescription for 6-8 weeks for Study Physician to sign

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Study Physician signs prescription, scans, and emails to RA/Study coordinator/Designated Study Personnel

↓

RA/Study coordinator/Designated Study Personnel emails the prescription to IDS to fill and files the original in the subject binder

↓

At 18-Week Visit,

- PI/Designated Study Personnel counts the participant's remaining prescription and compares against medication calendar to calculate adherence.
- Dispensing Rx to family occurs by any of the following ways:
    - RA/Study coordinator/Designated Study Personnel obtains prescription from IDS and to provide to family/participant. Hand-off of medication may occur in clinic or just outside CMH at a pre-determined location (e.g. the front entrance).
    - IDS brings the prescription to the outpatient pharmacy for the family/participant to pick up
    - IDS/RA/Study coordinator/Designated Study Personnel uses a courier service to deliver medication to CMK outpatient pharmacy for the family/participant to pick up. This use of courier service will be determined by IDS policies and practices.
    - IDS Pharmacy ships the medication to the family in Home Care Pharmacy medication coolers via FedEx using OptiFreight. Shipment of medication will follow IDS policies and practices.
- If the dose changes, RA/Study coordinator/Designated Study Personnel takes the old prescription and returns to IDS for disposal

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At Research Visit 4/PK3

- A prescription for a single dose (or two doses if Study Design 3) of atomoxetine (optional topical lidocaine) will be dispensed at Research Visit 3/PK2. A medication label is generated by IDS, so documentation of the dose(s) is recorded in the nurse note. RN/Designated personnel doses.
- RN/Designated study personnel doses.
- PI/Designated Study Personnel count the participant's remaining prescription and compare against medication adherence calendar to calculate adherence.
- If the participant intends to continue the study medication after study completion, the participant/family keep the remaining prescription (in the protocol)
- If the participant does not intend to continue the study medication, RA/Study coordinator/Designated Study Personnel take the prescription and return to IDS for disposal (Of note, since the new protocol will suggest a taper for those discontinuing the drug, this will change).

At any point during this study

- Medication may be dispensed to the participant and their family. Dispensing does not need to coincide with Study Physician visits, especially if these occur via telehealth in response to the COVID-19 pandemic. Dispensing Rx to family occurs by any of the following ways:
    - RA/Study coordinator/Designated Study Personnel obtains prescription from IDS and to provide to family/participant. Hand-off of medication may occur in clinic or just outside CMH at a pre-determined location (e.g. the front entrance).
    - IDS brings the prescription to the outpatient pharmacy for the family/participant to pick up
    - IDS/RA/Study coordinator/Designated Study Personnel uses a courier service to deliver medication to CMK outpatient pharmacy for the family/participant to pick up
    - RA/Study coordinator/Designated Study Personnel or IDS Pharmacy ships the medication to the family in Home Care Pharmacy medication coolers via FedEx using OptiFreight
        - NB: For certain scenarios, such as during the COVID-19 pandemic, the shipment of medication will be determined by IDS policies.
    - When any member of the study team picks up medication for the participant, they are acting as an, "agent of the patient" for the purposes of facilitating transfer of drug between IDS Pharmacy and study participant.

↓

END

Appendix K – Sample Letter to Release Data and/or Specimens by PI

I, (PI Name), authorize the release of the following data to (Collaborator Name) (list data to be shared) for the purposes of achieving the aims of the following research study (Study Name, IRB# if applicable):

- *Example* 1 (all pharmacokinetic data collected as a part of this)
- *Example* 2 (all genetic information for participants that completed this study)
- *Example* 3 (demographic information for all children enrolled)
- *Etc.*

PI Signature: _____

Date:_____

Collaborator Signature: _____

Date:_____

Appendix L – Sample letters to Share Genetic Information

To the family of (Participant's Name) (DOB MM/DD/YYYY):

Your child had genetic testing as a part of this ADHD study. This was done to help us learn how fast or slow your child "metabolizes"—or breaks down and removes atomoxetine (Strattera®) from his body. We want to return the test results to you and your child.

What are the results?

In general, atomoxetine is metabolized, or broken down, by CYP2D6 (pronounced "sip-2-d-6"). Based on the genetic testing, your child is a(n) _____ **metabolizer** for CYP2D6. This means when your child takes medicines like atomoxetine, your child's body breaks down the medicine _____ than most people in the population.

There are **4** different types of CYP2D6 metabolizers. They are:

Types of CYP2D6 Metabolizers		
Ultra-Rapid Metabolizer		Break down the medicine **much faster** than most people in the population
Normal Metabolizer		Break down the medicine **about the same** as most people in the population
Intermediate Metabolizer		Break down the medicine **a little slower** than most people in the population
Poor Metabolizer		Break down the medicine **much slower** than most people in the population

For most people, CYP2D6 breaks down **almost all** the atomoxetine a person takes. For some people, CYP2D6 doesn't work as well. For these people, another enzyme, CYP2C19 (pronounced "sip-2-c-19"), will help break atomoxetine down, too.

Your child's genetic testing also told us your child is a(n) _____ **metabolizer** for CYP2C19. This means when your child takes medicines normally broken down by this enzyme, your child's body breaks down the medicine _____ than most people in the population.

There are **5** different types of CYP2C19 metabolizers. They are:

Types of CYP2C19 Metabolizers		
Ultra-Rapid Metabolizer		Break down the medicine **much faster** than most people in the population
Rapid Metabolizer		Break down the medicine **a little faster** than most people in the population
Normal Metabolizer		Break down the medicine **about the same** as most people in the population
Intermediate Metabolizer		Break down the medicine **a little slower** than most people in the population
Poor Metabolizer		Break down the medicine **much slower** than most people in the population

What else should I know about the results?

These enzymes, CYP2D6 and CYP2C19, break down several other medicines. Your child may take these other medications in the future. However, your child and his/her regular doctors **cannot** use the research study results to help figure out if and how to use these other medicines if he/she may need them in the future. These test results can only be used for **this ADHD medicine** in **this ADHD study**. This means we can't put the results in your child's medical chart or send the results to his/her regular doctor. This also means your child's regular doctor cannot use the results to help treat your child for any illnesses or diseases outside of this study.

Why can't these results be used outside the study?

By law, we are not able to use these results outside of the study. This is because our tests are run in a "research-grade" lab instead of a "clinical-grade" lab. Clinical grade labs have different rules than research grade labs, and the tests they run are often different. When your child goes to his/her regular doctor and has blood drawn for lab work, the tests are likely run in a clinical grade lab.

For this study, we wanted to look at your child's genetics that control how CYP2D6 and CYP2C19 works. If your child's genetic testing were done in a clinical-grade lab, the test would probably look for only a few well-known differences in your child's genetics that might have a big impact on how CYP2D6 and CYP2C19 work. Our research grade tests look at these same differences, but we also look for other new and rare differences that are not well studied yet. Our research-grade test works better for our study because we want to look for **all** of these differences, not just the well-known ones.

Please know, even though our study testing is not run in a clinical-grade lab, this does not mean they are wrong or unsafe. We feel very confident that these results are accurate enough for the study to help us determine a dose of atomoxetine that works and is safe for your child. However, we are not sure yet what these results mean for other medicines that are broken down by these enzymes. More studies need to be done to fully understand what this means for other medicines.

I want to learn more about my child's test results. What should I do next?

If you would like to have your child's genetic information about CYP2D6 and CYP2C19 in his medical chart for your child and his/her other doctors to make medical decisions outside of the study, you can repeat the test in a clinical grade lab. Before you decide to repeat the test, you and your child should consider what the testing can tell you and how it might help your child's doctors in the future.

What can the testing tell me and my child's doctors?

Learning more information about how fast or slow your child's CYP2D6 enzyme and CYP2C19 enzyme works may help you and your child's doctors figure out which medicines he/she should or should not use and show if he/she may be at risk of side effects or a medicine not working at a normal dose. Your child will likely benefit more from learning more about these medicines if he/she is a:

- CYP2D6 poor metabolizer
- CYP2D6 intermediate metabolizer
- CYP2D6 ultra-rapid metabolizer
- CYP2C19 poor metabolizer
- CYP2C19 intermediate metabolizer
- CYP2C19 ultra-rapid metabolizer

Learning more may help because regular doses of these medicines may be too high or too low for your child or they may not work at all.

Which medicines should I know about?

There are several medicines that are affected by CYP2D6 and CYP2C19. Currently, there are guidelines for only a few of these medicines that say the dose should be higher or lower than normally prescribed if you break down the medicine faster or slower than most people in the population. Below are some examples of medicines where there are guidelines for CYP2D6 and/or CYP2C19 genotype:

Medicine	Used for this Disease or Illness
Atomoxetine (Strattera®)	Attention Deficit Hyperactivity Disorder (ADHD)
Codeine	Pain Management
Ondansetron (Zofran®)	Nausea/Vomiting
Selective Serotonin Reuptake Inhibitors (SSRIs) • Paroxetine (Paxil®) • Fluvoxamine (Luvox®) • Citalopram (Celexa®) • Escitalopram (Lexapro®) • Sertraline (Zoloft®)	Depression and Anxiety
Tricyclic Antidepressants • Amitriptyline (Elavil®) • Nortriptyline (Pamelor®)	Depression and Pain Management
Clopidogrel (Plavix®)	Stroke or Heart Attack (Blood Thinner)

If your child is prescribed any of these medicines in the future, repeating the genetic testing in a clinical-grade laboratory may help your child's doctors decide if the medicine is okay to use and at what dose. Right now, there are only guidelines for these medicines. There may be guidelines for other medicines in the future as more research studies are done. Please talk to your child's regular doctor to decide if genetic testing for metabolizing medicines may be helpful in the future.

Where can the testing be done and how can I learn more?

If you would like to learn more about how your child breaks down medicines with CYP2D6 and CYP2C19, your child can be seen at a clinic here at Children's Mercy Hospital called GOLDILOKs (Genomic-And-Ontogeny-Linked Dose Individualization and Clinical Optimization for Kids) Clinic. At GOLDILOKs clinic, a physician, or doctor, will talk with you and your child and answer your questions. After this discussion, you and your doctor may choose to repeat the genetic test using a clinical-grade lab and discuss what the results mean for other medicines. The study team will not pay for the costs of the GOLDILOKs Clinic visit or the testing GOLDILOKs Clinic does. The costs for these services will be billed to your child's insurer. You will be responsible for any such costs your child's insurer does not cover. If your child is uninsured, you will be responsible for these costs.

If you would like to learn more about GOLDILOKs clinic or make an appointment, please visit their website at https://www.childrensmercy.org/departments-and-clinics/pharmacology-and-toxicology/goldiloks-clinic/ or call the clinic at (816) 234-3059.

What else should I know about?

Please note, there is a chance that the results from the clinical grade testing and the study genetic testing may be different. This is because the tests are run differently. It does not necessarily mean the tests from the study were wrong. As mentioned before, clinical-grade tests are set up to look for the differences in each person's genetics that might have a big impact on how CYP2D6 and CYP2C19 work. Our research-grade tests look at these same differences, but we also look for other new and rare differences that are not well studied yet. Because the tests are not looking for the same differences, the results may come out different.

Who can I talk to if I have questions or concerns?

We understand that you may have questions about this information. Please feel free to reach out to a member of the study team at either the study cell phone (816) 359-2963 or the study email ADHDstudyteam@cmh.edu.

Sincerely,

Sincerely,
(Principal Investigator Signature)

Principal Investigator Name
Principal Investigator Contact Information

Appendix M – Sample letters to Release ADHD Results or Incidental Findings to Participants of this Study

For return of ADHD pertinent results:

To whom it may concern:

Attached are parent and teacher completed NICHQ Vanderbilt Assessment Scales (ADHD rating scales). These scales were completed during the prescreening process for the following research study: An Open-Label, Single- and Multi-Dose Study to Evaluate the Relationship between the Pharmacokinetics, Pharmacodynamics, and Clinical Outcomes of Atomoxetine in CYP2D6 Extensive, Intermediate, and Poor Metabolizers in Children with Attention Deficit/Hyperactivity Disorder. The results have been released back to the family at the family's request. NICHQ Vanderbilt Assessment Scales are interpreted in the context of a clinical evaluation for diagnosis and monitoring of ADHD.

Sincerely,
(Study Physician Signature)

Study Physician Name
Study Physician Contact

For the return of incidental findings:

To whom it may concern:

The following (list study procedures) were completed during the prescreening process for the following research study: An Open-Label, Single- and Multi-Dose Study to Evaluate the Relationship between the Pharmacokinetics, Pharmacodynamics, and Clinical Outcomes of Atomoxetine in CYP2D6 Extensive, Intermediate, and Poor Metabolizers in Children with Attention Deficit/Hyperactivity Disorder.

The (list study procedures) may indicate the following (list incidental findings). Further evaluation is recommended.

The results are being released back to the family to facilitate care for the participant's primary care provider or specialist of the family's choosing.

Sincerely,
(Physician Signature)

Physician Name
Physician Contact

Appendix N – Sample letters for End of Study ADHD Care Transition

To the family of [subject name] (DOB MM/DD/YYYY):

Thank you for allowing the Division of Developmental and Behavioral Sciences to provide care for [subject name]'s Attention Deficit Hyperactivity Disorder (ADHD). I am writing this letter to assist you with transferring care to your regular healthcare provider. [Subject] was enrolled in the following research study: An Open-Label, Single- and Multi-Dose Study to Evaluate the Relationship between the Pharmacokinetics, Pharmacodynamics, and Clinical Outcomes of Atomoxetine in CYP2D6 Extensive, Intermediate, and Poor Metabolizers in Children with Attention Deficit/Hyperactivity Disorder. [Subject]'s current dose of atomoxetine is __ mg [frequency]. For future comparisons, you may wish to share with his regular provider that his/her most recent NICHQ Vanderbilt Assessment Scales (ADHD rating scales) indicated that # of 9 symptoms of inattention and # of 9 symptoms of hyperactivity/impulsivity were in the significant range. This demonstrates _____ improvement from the time of enrollment when rating scales indicated that # of 9 symptoms of inattention and # of 9 symptoms of hyperactivity/impulsivity were in the significant range. It is recommended not to abruptly stop this medication and to make any future changes under the guidance of his physician. I am happy to provide his physician with additional information or guidance if needed. I can be reached through my clinic nursing staff at (816) 855-1927.

Sincerely,
(Study Physician Signature)

Study Physician Name
Study Physician Contact

Appendix O – Literature Search and Documentation for Washout and Return to Baseline

When a literature search must be conducted to determine the length of time needed to wash out of medications and when to administer baseline questionnaires, the follow data should be sought:

- **Pharmacokinetics parameters of the drug in question**. Ideally, these parameters will have been obtained in children, although for many drugs the data do not yet exist. When this data is not available in pediatrics, adult data may be used to estimate half-life and the expected amount of time needed to for medication to be removed from systemic circulation. In some instances, ranges are given for half-life. Use the most conservative value for estimating length of time needed to wash out of medciation. Additionally, consider estimating if ontogeny plays a role in in key processes for drug removal (i.e. CYP2D6, CYP3A4, CYP2C9, CYP2C19 genotype/activity or renal function).
- **Pharmacodynamics parameters of the drug in question**. This information should be sought to determine the amount of time needed for the effects of medication to wear off. The time needed for this to occur may be longer than the washout period for drugs. For many older drugs, this information is not available. Efforts should be made to determine if these parameters are reported in children and/or adults. Please note that for many neuropsychiatric drugs, there may information about the direct action of the drug, however little to no data may exist for the downstream modifications caused as of result of taking medication.
- **Behavior of the participant**. A proxy of return to baseline behaviors is also through evaluation of the participant's behaviors. This can be done when discussing the study with parent and through chart review.
- **Prior trials evaluating the drug in question**. Another source of information is evaluating how previous clinical trials have washed out of the drug in question. Crossover trials with a washout and baseline measure during washout can provide some insight to the length of time needed for drug concentrations and drug effects to return to baseline.
- **Expert opinion.** In addition to the data above, washout duration will be discussed at clinical project meetings by study team members. Study team members include professionals from various backgrounds including developmental and behavioral pediatricians, a pediatric psychologist, and clinical pharmacologists.
- **When little to no data exist for a drug**. If the study team is unable to make an estimation as to how long to wash out of medication, it is advisable that the participant not continue further in the study. The participant will have met exclusion criteria, "Subject is considered by PI to be unsuitable for participation in the study for any reason."

A note to file will written to describe the data available and the research team's conclusions about the appropriateness of washout, length of washout, and when to administer baseline questionnaires.

32.0 List of Supplementary Documents

Title	Section Referenced Within Protocol
Clinical Quality Management Plan	20.5
Data Safety Monitoring Board Charter	21.1
Data Safety Monitoring Board Protocol	21.1
Recruitment Flyer	9.3
Summer Treatment Program Presentation	9.3
U54 Potential Participant Log	9.3, 9.4
U54 Master Enrollment Log	9.4
U54 Study Booklet	2.1
U54 Study Booklet Information Handout	2.1
CHADIS Handout	11.2
Washout Handout	11.2
CMRI Website Information	9.1, 9.3

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