SPECIMEN COLLECTION FORM for Visit 2 and 4 (L21)

CKiD Chronic Kidney Disease in Children Cohort Study

SECTION A: GENERAL INFORMATION

A1. PARTICIPANT ID: AFFIX ID LABEL OR ENTER NUMBER IF ID LABEL IS NOT AVAILABLE

|__| - |__|__| - |__|__|__|

A2. CKiD VISIT #: ___

A3. FORM VERSION: 0 5 / 0 1 / 0 6

A4. SPECIMEN COLLECTION DATE: ___ ___/ ___ ___/ ___ ___ ___ ___
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A5. FORM COMPLETED BY (INITIALS): ___ ___ ___

A6. Is this study visit an accelerated visit? Yes..... 1
No..... 2

The following samples should be collected.

Samples:	**Shipped to:**	**Shipped:**
Serum	**CBL**	**IMMEDIATELY**
Serum	**Cystatin C**	***Batched***

If consent is obtained for biological sample, collect the following:

Samples:	**Shipped to:**	**Shipped:**
Serum (Biological)	***NIDDK Biosample Repository***	***Batched***
Plasma (Biological)	***NIDDK Biosample Repository***	***Batched***
Urine (Biological)	***NIDDK Biosample Repository***	***Batched***

SECTION B: PREGNANCY TEST AND URINE COLLECTION

B1. Is participant a female of child-bearing potential?

Yes..... 1 **(See PROMPT Below)**
No..... 2 **(Skip to B3)**

PROMPT: QUESTION B2 IS FOR FEMALE PARTICIPANTS OF CHILD-BEARING POTENTIAL ONLY. URINE PREGNANCY TEST DATE MUST FALL WITHIN 72 HOURS BEFORE GFR TESTING DATE.

B2. a. Urine pregnancy test date: ___ ___/ ___ ___/ ___ ___ ___ ___
M M D D Y Y Y Y

b. Urine pregnancy results:
Positive..... 1 **(END; COMPLETE DISENROLLMENT FORM)**
Negative..... 2

B3. Is this study visit a Make-Up GFR visit?

Yes..... 1 **(See PROMPT Below)**
No..... 2 **(Skip to B5)**

PROMPT: COLLECT 1.0 mL OF BLOOD IN THE PROVIDED SST LABELED B0 BEFORE THE IOHEXOL INFUSION.

(Remember to waste 1 cc if the blood was not collected from a butterfly.)

B4. Indicate reason(s) for a Make-Up GFR visit: **(Circle "Yes" or "No" for each):**

	Yes	No
IV infiltration.....	1	2
Inability to successfully draw 4 blood samples for iohexol.....	1	2
Other reason.....	1	2 **(Skip to F1)**

Specify:_____**(Skip to F1)**

Obtain urine collected at home. Family was instructed to collect urine at home in a container, such as a jar. IF URINE WAS NOT collected at home, collect FRESH urine into an initial urine collection cup or hat (provided by the site).

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Pour 5 to 14.5 mL of urine into blue top urine collection tube (provided by CBL).

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Check that all information is correct on the urine collection tube and follow packaging instructions and ship to CBL.

Reasons Code List*:	1= Not required	3 = Participant Refused	5 = Inadvertently Destroyed
	2 = Difficult Urine Collection	4 = Collection Contamination	6 = Oversight

Sample Type (Required Volume in Top Color Tube Type):	**(a) Sample Obtained:** Yes / No	**(b) If No, specify reason** *SEE CODE LIST ABOVE	**(c) Additional Requirements:**
B5. 1st Morning Urine (Urine Creatinine, Urine Protein) (5.0 mL–14.5 mL in Blue Top tube)	1 / 2 **(skip to c→)**	___ ___ **(skip to C1)**	i. Was urine collected at home? Yes.....1 No.....2 ii. Time of Collection: __ __ : __ __ 1 = am, 2 = pm

Encourage fluids throughout the visit.

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Place two IV lines (22 gauge or less polyethylene catheters); one in each arm
--OR---
Place one butterfly and one IV line (22 gauge or less polyethylene catheter); one in each arm; use tape to stabilize butterfly for Iohexol infusion

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Complete Pre-iohexol Infusion (B0) blood draw according to MOP instructions/flowchart on page 4.
NOTE: If patient has had a local CBC drawn within the past 30 days, those CBC results may be used instead of drawing another CBC and blood draw amounts on pre-iohexol infusion (B0) blood draw can be decreased by 1 ml.

SECTION C: PRE-IOHEXOL INFUSION (B0) BLOOD DRAW

For Initial Blood Draw with Syringe, Vacutainer OR Butterfly Method:
Select the Type of Consent Obtained (options 1 through 3) That Pertain to the CKiD Participant:

1

If participant consented to BIOLOGICAL samples and local BMP is clinically indicated:
Collect **17.5** mL if participant is **< 30 kg** **OR** **21.5** mL if participant is **≥ 30 kg**.

If **< 30 kg**, immediately transfer **(using 18 gauge needle)** or draw:
- 10.5 mL into (2) Tiger-Top SSTs for CBL, NIDDK BR & CMH
- 3 mL into PST for NIDDK Biosample Repository
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)
- 3 mL in another tube (not provided) for local BMP (**if clinically indicated**)

If **≥ 30 kg**, immediately transfer **(using 18 gauge needle)** or draw:
- 12.5 mL into (2) Tiger-Top SSTs for CBL, NIDDK BR & CMH
- 5 mL into PST for NIDDK Biosample Repository
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)
- 3 mL in another tube (not provided) for local BMP (**if clinically indicated**)

2

If participant consented to BIOLOGICAL samples and local BMP NOT clinically indicated:
Collect **14.5** mL if participant is **< 30 kg** **OR** **18.5** mL if participant is **≥ 30 kg**.

If **< 30 kg**, immediately transfer **(using 18 gauge needle)** or draw:
- 10.5 mL into (2) Tiger-Top SSTs for CBL, NIDDK BR & CMH
- 3 mL into PST for NIDDK Biosample Repository
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)

If **≥ 30 kg**, immediately transfer **(using 18 gauge needle)** or draw:
- 12.5 mL into (2) Tiger-Top SSTs for CBL, NIDDK BR & CMH
- 5 mL into PST for NIDDK Biosample Repository
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)

3

If participant did NOT consent to BIOLOGICAL samples and BMP is clinically indicated:
Collect **11.5** mL from all participants (regardless of weight) and immediately transfer or draw **7.5** mL into Tiger-Top SST for CBL

Immediately transfer **(using 18 gauge needle)** or draw:
- 7.5 mL into (2) Tiger-Top SSTs for CBL & CMH
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)
- 3 mL in another tube (not provided) for local BMP (**if clinically indicated**)

4

If participant did NOT consent to BIOLOGICAL samples and BMP NOT clinically indicated:
Collect **8.5** mL from all participants (regardless of weight) and immediately transfer or draw **7.5** mL into Tiger-Top SST for CBL

Immediately transfer **(using 18 gauge needle)** or draw:
- 7.5 mL into (2) Tiger-Top SSTs for CBL & CMH
- 1 mL in lavender-top tube fore local CBC (tube not provided in CBL kit)

PROCESSING BLOOD FOR CBL, CMH & NIDDK BR

Invert the SST & PST 5-10 times gently to mix.

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Stand SST upright to allow clotting at room temperature for 30 mins.

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Centrifuge SST & PST at MAX SPEED (between 1100-1300 g) for 10 minutes in swinghead units **OR** 15 minutes in fixed angle units (balance tube in centrifuge).*If incomplete separation, centrifuge again for 10-15 minutes.

If sample is GROSSLY HEMOLYZED…

If the sample is **GROSSLY HEMOLYZED (Dark Red),** pipette 2.5 mL of serum into Clear-Top Transport Tube labeled "B0/SST" and send to CBL. If a BMP was NOT collected, then collect 2.5 mL of additional blood at **B1**, **B2**, **B3** or **B4**. Centrifuge and then transfer serum into the extra Clear-Top Tube provided.

If sample is moderately, slightly or NOT HEMOLYZED, proceed with CBL, NIDDK BR and CMH preparation.

CBL

Using the disposable pipette, pipette 4.0 mL of serum into Clear-Top Transport Tube labeled "B0/SST" for CBL chemistries, HPLC Creatinine and lipids. Follow packaging instructions and ship to CBL with accompanying forms, iohexol blood draws and urine. **No FRIDAY shipments.** Refrigerate specimen and ship on next business day.

NIDDK BR

Pipette 1.5 mL (<30kg) or 2.5 mL (≥ 30kg) of serum into clear top cryovial & 1.5 or 2.5 mL of plasma into cryovial into green cap insert.
Store samples in freezer at -70°C or lower, batch up to 40 samples and ship on dry ice every four months to the NIDDK BR. When shipper is needed, complete "NIDDK BR Shipper Request Form" on the CKiD website: http://www.statepi.jhsph.edu/ckid/admin/. Then, follow packaging instructions.

CMH

Using the disposable pipette, pipette 0.5 mL of serum into Blue Screw-Top Cryovial for Cystatin C.
Store sample in freezer at -70°C or lower, batch up to 20 samples and ship on dry ice every four months to Children's Mercy Hospital. When shipper is needed, complete "Cystatin C Shipper Request Form" on the CKiD website: http://www.statepi.jhsph.edu/ckid/admin/Then, follow packaging instructions.

When pickup has been scheduled, complete "On-line Shipping Form" on CKiD website: http://www.statepi.jhsph.edu/ckid/admin/ to notify the appropriate personnel from the CBL, CMH and the NIDDK BR.

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C1. ACTUAL TIME OF PRE-IOHEXOL INFUSION (B0) BLOOD DRAW ____ ____ : ____ ____ 1 = AM 2 = PM

PROMPT: IF SUSBPECTED BLOOD DRAW ADVERSE EVENT (i.e., infection), complete ADVERSE EVENT (ADVR) Form

Reasons Code List*:	1= Not required	3 = Participant Refused	5 = Inadvertently Destroyed
	2 = Difficult Blood Draw	4 = Red Blood Cell Contamination	6 = Oversight

Sample Type (Required Volume in Top Color Tube Type):	**(a) Sample Obtained:** Yes / No	**(b) If No, specify reason** *SEE CODE LIST ABOVE	**(c) Additional Requirements:**
C2. Renal Chemistries and HPLC (5.0 mL in Tiger Top SST)	1 (skip to c→) 2	___ ___ (skip to C3)	i. Indicate the appearance of the serum after centrifuging. Grossly (Dark Red).....1 Moderately (Red/Light Red).....2 Slightly (Pink)..... .....3 Not Hemolyzed (Clear).....4
C3. Cystatin C (1.0 mL in Tiger Top SST)	1 (skip to c→) 2	___ ___ (skip to C4)	Frozen Date: ___ ___ /___ ___ /___ ___ ___ ___ M M D D Y Y Y Y
C4. Local CBC (1.0 mL in Lavender Top tube)	1 (skip to c→) 2	___ ___ (skip to C5)	**N/A**
C5. Serum for Fasting Lipid Panel (1.5 mL in Tiger Top SST)	1 (skip to c→) 2	___ ___ (skip to C6)	**N/A**

C6. Please indicate:
a. Office fax number: _____

b. The recipient's name who receives fax: _____

SITES CAN OBTAIN RESULTS FOR LAB VALUES THAT HAVE BEEN IDENTIFIED AS "KEY VARIABLES". TO OBTAIN RESULTS, GO TO THE CKiD NEPHRON WEBSITE: https://statepiaps.jhsph.edu/nephron/groups/aspproc/, click on "Report Menu" and choose the appropriate lab report (i.e., Selected Renal Panel Lab Variables Report.)

C7. Did the participant consent to have biological samples (i.e., serum, plasma and urine samples) stored at the NIDDK Biosample Repository?

Yes..... 1

No..... 2 **(Skip to E1)**

Reasons Code List*:	1= Not required	3 = Participant Refused	5 = Inadvertently Destroyed
	2 = Difficult Blood Draw	4 = Red Blood Cell Contamination	6 = Oversight

Sample Type (Required Volume in Top Color Tube Type):	**(a) Sample Obtained:** Yes	No	**(b) If No, specify reason** ***SEE CODE LIST ABOVE**	**(c) Additional Requirements:**
C8. Serum for NIDDK Biosample Repository (**3.0 mL or **5.0 mL of blood in Tiger Top SST)	1 **(skip to c→)**	2	___ ___ **(skip to C9)**	Date Frozen: ___ ___ /___ ___ /___ ___ ___ ___ M M D D Y Y Y Y
C9. Plasma for NIDDK Biosample Repository (**3.0 mL of blood in one Green Top or **5.0 mL in two Green Top PSTs)	1 **(skip to c→)**	2	___ ___ **(skip to D1)**	Date Frozen: ___ ___ /___ ___ /___ ___ ___ ___ M M D D Y Y Y Y

** Collect 3.0 mL of whole blood for children < 30 kg and 5.0 mL for children ≥ 30 kg

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SECTION D: URINE COLLECTION AND PROCESSING FOR REPOSITORY

Collect FRESH urine into an initial urine collection cup or hat (provided by the site).

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Pour 60 mL of FRESH urine into blue top urine collection cup with 4 protease inhibitor (PI) tablets. Do not fill the urine past the 60 mL mark on the collection cup. One protease inhibitor tablet should be used for 10-15 mL of urine **(see Table A)**. For example if 30 mL of urine is collected, ONLY 2 PI tablets are needed. (Like all unused supplies, **unused PI tablets should be returned to the CBL**.)

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Invert the urine cup gently 5 – 10 times.

↓

The PI TABLET(s) MUST BE **COMPLETELY** DISSOLVED in the urine.

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Once the PI tablets are completely dissolved, pour urine into six (6) 10 mL urine centrifuge tubes. (**For each tube:** remove clear top cap, pour urine into tube and SNAP cap back onto tube.) Place no more than 10 mL in each tube.
– OR – Sites may also substitute with tubes normally used to centrifuge urine at site.

TABLE A:

Urine Volume	# of PI Tablets
10 – 15 mL	**1**
16 – 30 mL	**2**
31 – 45 mL	**3**
46 – 60 mL	**4**

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Centrifuge urine tube(s) at MAX SPEED (between 1100-1300g) for 10 mins (swinghead units) **OR** 15 mins (fixed angle units).

↓

Decant (pour off) the supernates (liquid reaction) into seven (7) 10 mL urine cryovials. Pour no more than 9 mL of urine into each 10 mL cryovial to allow for expansion.

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Check that all information is correct on the urine transport tube, promptly freeze and store sample(s) at -70ºC or lower. Batch up to 36 samples. When shipper is needed, complete "NIDDK Shipper Request Form" on CKiD website: http://www.statepi.jhsph.edu/ckid/admin/.

↓

When pickup has been scheduled, complete "On-line Shipping Form" on CKiD website to notify Heather Higgins, Rich Frome and Alicia Wentz that sample(s) have been shipped to NIDDK F

Reasons Code List*:			
	1= Not required	3 = Participant Refused	5 = Inadvertently Destroyed
	2 = Difficult Urine Collection	4 = Collection Contamination	6 = Oversight

Sample Type (Required Volume in Top Color Tube Type):	**(a) Sample Obtained:** Yes	No	**(b) If No, specify reason** *SEE CODE LIST ABOVE	**(c) Additional Requirements:**
D1. Urine for NIDDK Biosample Repository (15.0 - 60.0 mL of urine in specimen container and transferred into collection cup with protease inhibitors)	1 **(skip to c→)**	2	__ __ **(skip to E1)**	i. Was supernate decanted into urine transport cryovials? Yes.....1 No.....2 ii. Date Frozen: __ __ /__ __ /__ __ __ __ M M D D Y Y Y Y

SECTION E: OPTIONAL LOCAL LAB TESTS (IF CLINICALLY INDICATED)

Check with the PI at your clinical site to determine whether or not it is **CLINICALLY INDICATED** to obtain additional local labs. These are instances when the PI needs results immediately and/or the participant needs additional local labs performed (i.e., local BMP, local Urine Creatinine and Urine Protein) OR CBL sample is **GROSSLY HEMOLYZED**.

E1. Was a basic metabolic panel (BMP) assay performed at the clinical site's local laboratory?

Yes..... 1 → **Complete Local Basic Metabolic Panel Results Form L03, ONLY if local labs are CLINICALLY INDICATED or CBL Renal Panel Serum is GROSSLY HEMOLYZED**

No..... 2

E2. Was a 1st morning urine protein to creatinine ratio assay performed at the clinical site's local laboratory?

Yes..... 1 → **Complete Local Urine Assay Results Form L06, ONLY if local labs are CLINICALLY INDICATED**

No..... 2

SECTION F: INFUSION SYRINGE WEIGHT

F1. **SCALE MUST FIRST BE ZEROED BEFORE WEIGHING. REMOVE ALUMINUM FOIL PRIOR TO WEIGHING THE SYRINGE. THE SAME SCALE MUST BE USED TO WEIGH THE SYRINGE PRE AND POST IOHEXOL INFUSION.**

a. Syringe Weight **Pre-Iohexol Infusion**: ___ ___ . ___ ___ (g)

b. Syringe Weight **Post-Iohexol Infusion**: ___ ___ . ___ ___ (g) (Post-Infusion Weight should be **at least 6.0g** less than Pre-Infusion Weight. If Post-Infusion Weight is not at least 6g less, please confirm.)

PRE AND POST SYRINGE WEIGHT MUST BE OBTAINED IN ORDER TO CALCULATE CHILD'S GFR.

SECTION G: IOHEXOL – Refer to Instructions for Iohexol Infusion and GFR Blood Draws Flow Chart on Page 10

- **BEFORE** INFUSING IOHEXOL, **SET TIMER = 0**. SIMULTANEOUSLY START TIMER AND BEGIN IOHEXOL INFUSION
- **COMPLETE INFUSION BETWEEN 1 TO 2 MINS**
- LEAVE TIMER RUNNING THROUGHOUT IOHEXOL INFUSION AND SUBSEQUENT BLOOD DRAWS

G1. IOHEXOL INFUSION

a. INFUSION START TIME: ____ ____ : ____ ____ 1 = AM 2 = PM

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- **DO NOT DRAW BLOOD FROM THE IV SITE WHERE IOHEXOL WAS INFUSED. ANOTHER IV SITE MUST BE USED.**
- **WASTE 1 mL OF BLOOD IF DRAWING FROM A SALINE/HEPARIN LOCK.**
- **COLLECT 1 mL OF BLOOD FOR EACH IOHEXOL BLOOD DRAW IN THE PROVIDED SST.**
- **RECORDING THE EXACT NUMBER OF MINUTES ON THE TIMER IS MORE IMPORTANT THAN DRAWING THE BLOOD EXACTLY AT 10, 30, 120 & 300 MINUTES AFTER IOHEXOL INFUSION. FOR EXAMPLE, IF BLOOD IS DRAWN AT 33 MINS INSTEAD OF 30 MINS, DOCUMENT BLOOD DRAWN @ 33 MINS.**
- **TIME SHOULD BE RECORDED IMMEDIATELY AFTER EACH BLOOD SAMPLE IS OBTAINED (i.e., B1, B2, B3, and B4).**

		(i) **ACTUAL MINUTES on TIMER**	(ii) **ONLY** if Timer malfunctions, record Clock Time using the same clock used for G1a	(iii) **Difficult Blood Draw:** Yes	No	(iv) **Blood Volume Collected (1 mL):**	(v) **Centrifuged at Clinical Site:** Yes	No
G2a.	**B1** 10 min:	___ ___ minutes	___ ___ : ___ ___ 1 = AM 2 = PM	1 **(Skip to b)**	2	___ . ___ mL	1 **(Skip to G3a)**	2 **(Skip to G3a)**
b.	**B1** 2nd attempt:	___ ___ minutes	___ ___ : ___ ___ 1 = AM 2 = PM	1	2	___ . ___ mL	1	2

INVERT TUBE 5-10 TIMES AFTER EACH BLOOD DRAW
LET SST TUBE STAND 20-30 MINUTES (BUT NO LONGER THAN 1 HOUR)
CENTRIFUGE FOR AT 1100-1300g for 10 MINUTES IN SWING HEAD OR 15 MINUTES IN FIXED ANGLE

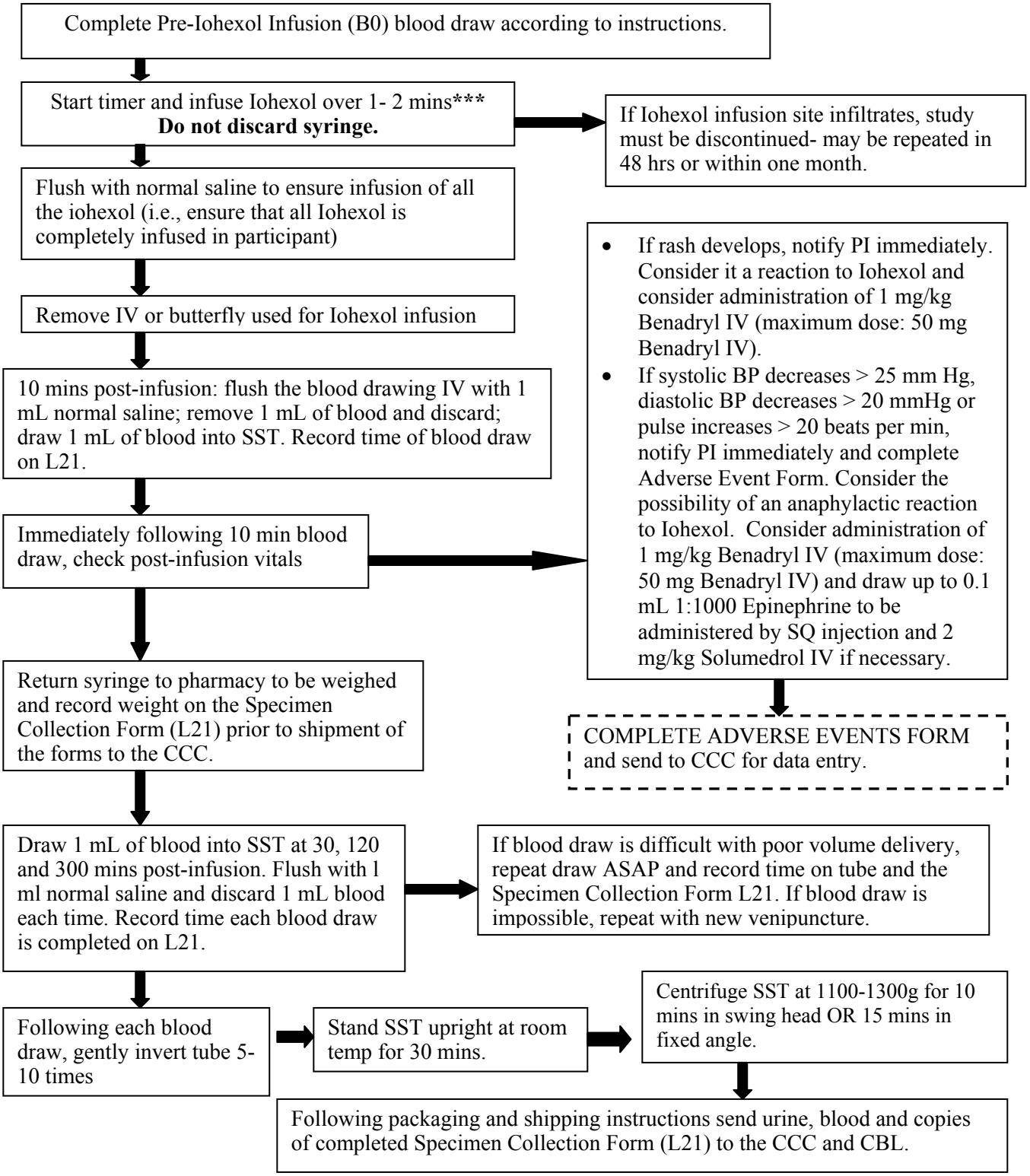
POST VITALS SHOULD BE TAKEN IMMEDIATELY AFTER THE 10 MINUTE BLOOD DRAW USING LOCAL BLOOD PRESSURE MEASUREMENT (i.e. DINAMAP)

- **If rash develops after Iohexol Infusion, consider it a reaction to Iohexol and notify PI immediately. Consider administration of 1 mg/kg Benadryl IV (maximum dose: 50 mg Benadryl IV).**
- **In the rare event that systolic BP decreases more than 25 mm Hg, diastolic BP decreases more than 20 mmHg, or pulse increases more than 20 beats per min, notify PI immediately to evaluate reaction and complete the Adverse Event (ADVR) Form. Consider the possibility of an anaphylactic reaction to Iohexol. Consider administration of 1 mg/kg Benadryl IV (maximum dose: 50 mg Benadryl IV). Draw up to 0.1 mL 1:1000 Epinephrine for SQ injection and 2 mg/kg Solumedrol IV for administration as ordered by physician.**

(i) Post Vitals:		
G3a.	Post- infusion blood pressure:	___ ___ ___ / ___ ___ ___
b.	Post-infusion temperature:	___ ___ . ___ 1 = ºC 2 = ºF
c.	Post-infusion number of heart beats per minute:	___ ___ ___
d.	Post-infusion respirations per minute:	_____

		(i) **ACTUAL MINUTES on TIMER**	(ii) **ONLY** if Timer malfunctions, record Clock Time using the same clock used for G1a	(iii) **Difficult Blood Draw:** Yes	No	(iv) **Blood Volume Collected (1 mL):**	(v) **Centrifuged at Clinical Site:** Yes	No
G4a.	**B2** 30 min:	___ ___ minutes	___ ___ : ___ ___ 1 = AM 2 = PM	1 **(Skip to b)**	2	___ . ___ mL	1 **(Skip to G5a)**	2 **(Skip to G5a)**
b.	**B2** 2nd attempt:	___ ___ minutes	___ ___ : ___ ___ 1 = AM 2 = PM	1	2	___ . ___ mL	1	2
G5a.	**B3** 120 min (2 hrs):	___ hr ___ ___ mins	___ ___ : ___ ___ 1 = AM 2 = PM	1 **(Skip to b)**	2	___ . ___ mL	1 **(Skip to G6a)**	2 **(Skip to G6a)**
b.	**B3** 2nd attempt:	___ hr ___ ___ mins	___ ___ : ___ ___ 1 = AM 2 = PM	1	2	___ . ___ mL	1	2
G6a.	**B4** 300 min (5 hrs):	___ hr ___ ___ mins	___ ___ : ___ ___ 1 = AM 2 = PM	1 **(Skip to b)**	2	___ . ___ mL	1 **(END)**	2 **(END)**
b.	**B4** 2nd attempt:	___ hr ___ ___ mins	___ ___ : ___ ___ 1 = AM 2 = PM	1	2	___ . ___ mL	1	2

Instructions for Iohexol Infusion and GFR Blood Draws



*****Physician should be immediately available (in person or by phone) during Iohexol Infusion**