TRAINING UPDATE

Lab Location:	SGMC	**Date Distributed:**	4/19/2018
Department:	Core Lab	**Due Date:**	5/8/2018
		Implementation:	**5/8/2018**

DESCRIPTION OF PROCEDURE REVISION

Name of procedure:

Fetal Hemoglobin (APT) SGAH.C22 v3

Fetal Hemoglobin (APT) Log AG.F148.1

Description of change(s):

***Note**: significant change to how results will be reported – why?*

- *CAP requires a reference range be reported with results*
- *The text (interpretation of NFH) is too long to display in Cerner*
- *Changing the name of test to more accurately reflect what is being tested, means we can now use 'NEG' and 'POS' to result*
- *The reference range will display in Cerner as 'negative'*

Other changes are format updates

Section	Reason
Page 1	Change title
4,6	Remove individual section labeling instructions and add general one
10.1	Change SQ reporting codes
10.5	Review data moved from section 6
11.1	Change reference range to "negative"
15	Update to new standard wording

LOG: edited to match SOP reporting changes

This revised SOP and log will be implemented on May 8, 2018

Document your compliance with this training update by taking the quiz in the MTS system.

Technical SOP

Title	**Fetal Hemoglobin (APT)**	
Prepared by	Marilyn Van Degrift	Date: 2/23/2010
Owner	Robert SanLuis	Date: 10/6/2015

Laboratory Approval	**Local Effective Date:**	
Print Name	**Signature**	**Date**
Refer to the electronic signature page for approval and approval dates.		

Review		
Print Name	**Signature**	**Date**

TABLE OF CONTENTS

1. TEST INFORMATION

Assay	Method/Instrument	Local Code
Fetal Hemoglobin (APT) Test	Colorimetric	APT

Synonyms/Abbreviations
APT - Downey

Department
Chemistry

2. ANALYTICAL PRINCIPLE

Adult hemoglobin denatures to a brownish hematin in an alkaline environment. Fetal hemoglobin is alkali resistant with a persistent pink color.

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Random stool or gastric contents
Special Collection Procedures	N/A
Other	N/A

3.2 Specimen Type & Handling

Criteria		
Type	**-Preferred**	Grossly bloody (red) non-tarry stool or bloody vomitus or bloody mucous
	-Other Acceptable	Blood stained diaper. Note: For diapers, sheets, etc., cut out the soiled area and elute in 2-5 ml of distilled water to obtain a sufficient sample for testing. Specimen for testing should have a visible "pink" hue. See procedure notes in section 13.
Collection Container		Plastic container, tube or blood stained diaper
Volume	**- Optimum**	1 mL
	- Minimum	0.5 mL
Transport Container and Temperature		Collection container at room temperature
Stability & Storage Requirements	Room Temperature:	(15-30°C) 30 min
	Refrigerated:	(2-8°C) 1 week
	Frozen:	Unacceptable
Unacceptable Specimens & Actions to Take		Specimens that are unlabeled, improperly labeled, or those that do not meet the stated criteria are unacceptable. Request a recollection and credit the test with the appropriate LIS English text code for "test not performed" message. Examples: Quantity not sufficient-QNS; Unacceptable specimen-UNAC. Document the request for recollection in the LIS.

Criteria	
Compromising Physical Characteristics	N/A
Other Considerations	None

NOTE: Labeling requirements for all reagents, calibrators and controls include: (1) Open date, (2) Substance name, (3) Lot number, (4) Date of preparation, (5) Expiration date, (6) Initials of tech, and (7) Any special storage instructions. Check all for visible signs of degradation.

4. REAGENTS

4.1 Reagent Summary

Reagents	Supplier & Catalog Number
10% Sodium Hydroxide (NaOH)	Ricca Chemical Co. Cat # 7260-32
NERL Ultra Purified Water	Thermo Scientific Cat #0015

4.2 Reagent Preparation and Storage

Reagent	**10% NaOH**
Container	Sterile plastic container as supplied by the manufacturer
Storage	Room temperature
Stability	1 year
Preparation	N/A

Reagent	**NERL Ultra Purified Water (distilled)**
Container	Sterile plastic container as supplied by the manufacturer
Storage	Room temperature
Stability	Unopened: until expiration date on bottle Opened: 30 days at room temperature
Preparation	N/A

5. CALIBRATORS/STANDARDS

N/A

6. QUALITY CONTROL

6.1 Controls Used

Controls	Supplier and Catalog Number
Adult Blood	N/A
Cord Blood	N/A

6.2 Control Preparation and Storage

Control	Adult Blood
Preparation	Ready to use from Lavender top tube obtained in Hematology or Blood Bank. **Note**: Care should be taken to select a healthy adult blood sample.
Storage/Stability	Use fresh sample

Control	Cord Blood
Preparation	Ready to use from Cord Blood specimen obtained from Blood Bank.
Storage/Stability	Use fresh sample

6.3 Frequency

Run Adult and Cord Blood controls with each patient test.

6.4 Tolerance Limits and Criteria for Acceptable QC

With the addition of NaOH, Hemoglobin in the Adult Control should change from pink to brown immediately while the Cord Blood Control (Fetal Hemoglobin F) will retain a pinkish hue. Note that all hemoglobin, including alkali resistant Hgb F, will denature with time in an alkali environment. Results must be read within the designated time frame.

Fetal Hemoglobin may be elevated in adult oncology patients and individuals with homozygous Hgb S or Thalassemia. If the APT adult control result is not as expected, another (healthy) adult control should be selected and the test repeated.

Cord bloods may range from 60% to 90% Fetal Hemoglobin. If the cord blood control result is not as expected, select another cord blood and repeat test.

6.5 Documentation

- Record Quality Control and patient results on appropriate worksheet.
- Refer to complete policies and procedures for QC documentation and for record retention requirements in the Laboratory QC Program.

6.6 Quality Assurance Program

- The laboratory participates in CAP proficiency testing. All proficiency testing materials must be treated in the same manner as patient samples.

7. EQUIPMENT and SUPPLIES

7.1 Assay Platform

N/A

7.2 Equipment

N/A

7.3 Supplies

- Test tubes
- Pipettes

8. PROCEDURE

NOTE: For all procedures involving specimens, buttoned lab coats, gloves, and face protection are required minimum personal protective equipment. Report all accidents to your supervisor.

1. For each specimen (Adult and Cord Blood controls and the Patient) label two tubes: Test and Reference.

	Adult Control	Cord Control	Patient Specimen
Test			
Reference			

2. Add 2 mL of distilled water to each tube.

3. Add 1 drop of control or bloody patient specimen to their respective tubes.

4. Starting with the Adult Control, add 0.1 mL of 10% NaOH to the "Test" tube, mix and compare to the adult "Reference" tube. Look for an immediate color change from pink to brown/beige. Read result within 15 seconds after adding the NaOH and mixing.

5. Record result and interpretation on the APT Log.

6. Repeat steps 4-5 for the Cord Blood Control and then for each Patient Specimen. A pink hue will persist when fetal hemoglobin is present. Do not let the reaction sit before reading as even alkali resistant Fetal Hemoglobin will denature in an alkali environment over time.

NOTE: In the event that the test system becomes inoperable, notify supervision or designee for further direction. Patient specimens must be stored in a manner that maintains the integrity of the specimen.

9. CALCULATIONS

N/A

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

10.1.1 If solution turns from pink to brown or beige, without a persistent pink hue, adult hemoglobin is present.

~~REPORT AS: NFH "Negative for fetal hemoglobin"~~

REPORT AS: **NEG** "Negative"

10.1.2 If solution does not immediately turn brown, i.e. it stays pink or a pink hue persists, fetal hemoglobin is present.

~~REPORT AS: PFH "Positive for fetal hemoglobin"~~

REPORT AS: **POS** "Positive"

10.1.3 If solution was not visibly bloody (See section 13 Procedure Notes).

CANCEL AS: **UNAC** "specimen unacceptable"

10.2 Rounding

N/A

10.3 Units of Measure

N/A

10.4 Clinically Reportable Range (CRR)

N/A

10.5 Review Patient Data

N/A

10.6 Repeat Criteria and Resulting

Repeat test using a larger volume (if available), whenever color development is uncertain. Repeat controls using a different sample when color development is not as expected or uncertain.

10.7 Resulting

Record results and interpretation on the APT Log and enter in LIS using function **MEM**.

Enter Shift (**1, 2, or 3**)
Worksheet: **SCH1**
Test: <Enter>

Enter "**A**" (Accept)
Enter Accession number
Press <Enter> until Result screen displayed

11. EXPECTED VALUES

11.1 Reference Ranges

Negative ~~for fetal hemoglobin~~

11.2 Critical Values

None established

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

The APT test is useful in distinguishing between Melena Neonatorum (hemorrhage from an infant's gastrointestinal tract) and the passing by the infant of maternal blood swallowed during labor.

13. PROCEDURE NOTES

- **FDA Status:** Exempt
- **Validated Test Modifications:** None

The specimen requirement for this test as stated in section 3.2 is a grossly bloody stool, vomitus or mucous. **In the case of blood stained diapers, sheets, etc.**:

- If after adding distilled water a pink hue is not seen, the specimen is unacceptable for testing.
- Call a caregiver and explain the following: 'The specimen we received did not appear to be bloody, nor were we able to elute any pink or red color from it; therefore we are unable to perform this test.'
- Document the call and cancel the test. Indicate the inability to perform this test on the APT QC log along with the documentation of the notification.

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

N/A

14.2 Precision

Reactions should be read immediately after adding NaOH, within 15 seconds. All hemoglobins will ultimately denature (turn brownish) in a strong alkali environment.

14.3 Interfering Substances

Some oncology patients and individuals with hemoglobinopathies (Thalassemia, Hgb S) may have elevated levels of fetal hemoglobin and should not be used as an adult control.

14.4 Clinical Sensitivity/Specificity/Predictive Values

N/A

15. SAFETY

Refer to your local and corporate safety manuals and Safety Data Sheet (SDS) for detailed information on safety practices and procedures and a complete description of hazards.

16. RELATED DOCUMENTS

1. Laboratory QC Program
2. APT procedure SC.505 v006, Special Chemistry, Chantilly
3. Fetal Hemoglobin (APT) Log (AG.F148)

17. REFERENCES

- M. Crook, Haemoglobin in stools from neonates: measurement by a modified APT-test, Med Lab Sci. 1991 Oct;48(4):346-7
- Apt. L and Downey, W.S., Melena Neonatorum – J. Pediatrics 47, 6, 1955.
- Bray's Clinical Laboratory Methods, 7th Edition.
- H. Franklin Bunn, Induction of Fetal Hemoglobin in Sickle Cell Disease, Blood, Vol 93 No.6, 1999
- McRury JM & Barry RC: A modified APT test: a new look at an old test. Pediatr Emerg Care 1994; 10:189-191
- Apt L, et al, Melena Neonatum, The Swallowed Blood Syndrome, J Pediat, 45:5, 6-12

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
			Supersedes SOP SGAHC002.002		
000	11/28/11	3.2	Change Room Temp range to 15-30C Refer to section 13 for procedural	C Reidenauer	C Reidenauer

			notes on acceptability.		
000	11/28/11	4.1, 4.2,7.3	Add NERL Water	C Reidenauer	C Reidenauer
000	11/28/11	10.1.3	Change instruction 'Report' to 'Cancel'	C Reidenauer	C Reidenauer
000	11/28/11	11.2	Update title to local terminology	L Barrett	C Reidenauer
000	11/28/11	13	Add procedure notes on Unacceptable Specimen handling.	C Reidenauer	C Reidenauer
000	11/28/11	15	Update to standard content	L Barrett	C Reidenauer
000	11/28/11	16	Add QC Program and SC.505 SOP	L Barrett	C Reidenauer
001	10/6/15		Update owner	L Barrett	R SanLuis
001	10/6/15	4.2	Change open NERL stability from 7 to 30 days	L Barrett	R SanLuis
001	10/6/15	10.6	Add LIS reporting steps	L Barrett	R SanLuis
001	10/6/15	11.1	Change from none to negative	L Barrett	R SanLuis
001	10/6/15	16	Move log from section 19	L Barrett	R SanLuis
001	10/6/15	17	Add reference for reference range	L Barrett	R SanLuis
001	10/6/15	Footer	Version # leading zero's dropped due to new EDCS in use as of 10/7/13	L Barrett	R SanLuis
2	4/13/18	Page 1	Change title	L Barrett	R SanLuis
2	4/13/18	4, 6	Remove individual section labeling instructions and add general one	L Barrett	R SanLuis
2	4/13/18	10.1	Change reporting codes	L Barrett	R SanLuis
2	4/13/18	10.5	Review data moved from section 6	L Barrett	R SanLuis
2	4/13/18	11.1	Change to negative	L Barrett	R SanLuis
2	4/13/18	15	Update to new standard wording	L Barrett	R SanLuis

19. ADDENDA

None

Quest Diagnostics

Fetal Hemoglobin (APT) Log

Shady Grove Medical Center

Month _____ Year_____

Adult Blood (negative for Fetal Hgb) and Cord Blood (positive for Fetal Hgb) Controls are tested with each patient.
Read results ~~immediately~~ within 15 seconds after adding NaOH. Record as negative (turns brown), or positive for Hgb F (remains pinkish).

Note: † A complete color change to brown/beige indicates adult hemoglobin is present, specimen is Negative for Fetal Hgb **(NEG ~~NFH~~)**
* Persistence of a pink hue is indicative of alkali resistant fetal hemoglobin, specimen is Positive for Fetal Hgb **(POS ~~PFH~~)**
Interpretation is difficult with scant or non-bloody specimen. Cancel as an unacceptable specimen, "UNAC".

Date	Adult Blood Control Color Results: (Acceptable: Color changes to Brown / Beige)	Cord Blood Control Color Results: (Acceptable: Color remains Pink or Pink Hue Persists)	Patient Name / MR#	Patient Color Results: † "Brown" * "Pink"	Select LIS code: NEG: Negative (Complete Color Change) POS: Positive (Pink / Pink Hue Persists)	Tech

Weekly review:	Weekly review:	Weekly review:
Weekly review:	Weekly review:	Monthly review:

Fetal Hemoglobin (APT) Log