ELECTROPHORESIS

Inspector Instructions:

	• Sampling of electrophoresis policies and procedures • Sampling of electrophoresis QC logs
	• Electrophoretic patterns (appropriate separations)

CHM.33500 Daily QC - Electrophoresis **Phase II**

Suitable control samples are run and reviewed with each batch of patient samples for all electrophoresis procedures for which controls are available.

Evidence of Compliance:

✓ Records of electrophoresis QC

CHM.33600 Electrophoresis Separations **Phase II**

Electrophoresis separations are satisfactory.

CHM.33700 Acceptable Limits - Controls **Phase II**

Acceptable limits are set for controls of procedures where the electrophoretic bands are quantified.

Evidence of Compliance:

✓ Records of defined acceptable limits for control range of each lot

HEMOGLOBIN SEPARATION

For purposes of diagnosing hemoglobinopathies, more than one test may be necessary. As an example, hemoglobin solubility testing alone is not sufficient for detecting or confirming the presence of sickling hemoglobins in all situations.

Inspector Instructions:

	• Sampling of abnormal hemoglobin policies and procedures • Sampling of patient reports (confirmatory testing, comments) • Sampling of QC records

	• Hemoglobin electrophoretic patterns (appropriate separations and controls) • Examine a sampling of medium (media) used to identify hemoglobin variants including alkaline/acid electrophoresis, isoelectric focusing, HPLC or other method
	• What is your course of action when the primary screening method appears to show Hb S? • What is your course of action when the primary Hb electrophoresis method shows Hb variants migrating in non-A/non-S positions?

CHM.33708 Hb S Primary Screen **Phase II**

For patient samples that appear to have Hb S in the primary screening by electrophoresis or other separation methods, the laboratory either: 1) performs a second procedure (solubility testing, or other acceptable method) to confirm the presence of Hb S, or 2) includes a comment in the patient report recommending that confirmatory testing be performed.

NOTE: For primary definitive diagnosis screening by electrophoresis or other separation methods, all samples with hemoglobins migrating in the "S" positions or peak must be tested for solubility or by other acceptable confirmatory testing for sickling hemoglobin(s). Known sickling and non-sickling controls both must be included with each run of patient specimens tested.

CHM.33716 Daily QC - Hgb Separation **Phase II**

Controls containing at least three known major hemoglobins, including both a sickling and a nonsickling hemoglobin (eg, A, F and S) are performed with the patient specimen(s) and separations are satisfactory.

NOTE: There are written procedures for instruments with multiple electrophoretic chambers or capillaries to ensure that QC is performed on each individual chamber or capillary.

Evidence of Compliance:

- ✓ QC records reflecting the use of appropriate controls **AND**
- ✓ Electrophoresis media/separation tracings demonstrating appropriate controls and separation

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CHM.33732 Hemoglobin Variants — Phase II

All samples with hemoglobin variants migrating in "nonA, nonS" positions on alkaline electrophoresis, or other low resolution procedure are further defined with other acceptable methods where clinically and technically appropriate.

NOTE: If all clinically significant variants are not clearly separated by the primary method, additional testing must be performed to further characterize these hemoglobin variants. Examples include:

- *Performance by a complementary, separate methodology*
- *Increasing the duration of the assay (for HPLC) where the hemoglobins migrate/elute at different configurations*

Further workup of such variants, including referral to another laboratory, is dependent upon the patient's overall clinical situation, such as findings of erythrocytosis or a hemolytic anemia.

Evidence of Compliance:

✓ Patient reports and records reflecting further work-up, when appropriate

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2) Sabath DE. Molecular diagnosis of thalassemias and hemoglobinopathies: an ACLPS critical review. *Am J Clin Pathol.* 2017;148:6-15.
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CHM.33764 Hb S Predominant Band — Phase II

All samples that appear to have Hb S as the predominant band by the primary screening and confirmed as sickling by appropriate methods are further examined to ascertain whether the "Hb S" band or peak contains solely Hb S or both Hb S and Hb D, Hb G or other variant hemoglobins.

NOTE: When the predominant hemoglobin component appears to be Hb S, it is necessary to determine whether this represents homozygous Hb S or a heterozygote for Hb S and another variant such as Hb D, Hb G, Hb-Lepore, or other hemoglobin variant(s). Given the clinical implications of homozygous Hb S (or Hb S/ß zero thalassemia) it is imperative to exclude other hemoglobin variants, however rare. Referral of these specimens to another laboratory for further workup is acceptable.

Evidence of Compliance:

✓ Patient records or worksheets showing the exclusion of hemoglobin variants **OR** referral for further work-up

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