DXC 800 (PHE) PHENOBARBITOL

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PURPOSE

To provide instructions for the quantitative determination of phenobarbitol on the DXC 800.

PRINCIPLE

PHE reagent, when used in conjunction with UniCel® DxC 600/800 System(s) and SYNCHRON® Systems Drug Calibrator 1, is intended for quantitative determination of Phenobarbital concentration in human serum or plasma.

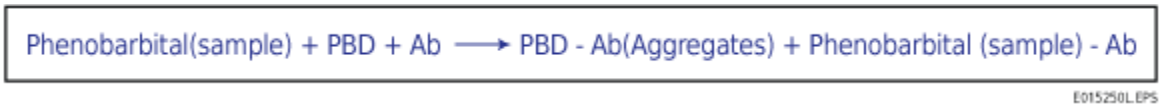
BACKGROUND

Clinical Significance

Phenobarbital is indicated for the treatment of status epilepticus, febrile seizures and seizure disorders (grand mal and psychomotor), except absence (petit mal) seizures. Phenobarbital therapy is monitored for suspected inadequate dose or toxicity.

Methodology

PHE reagent is used to measure the PHE concentration by a particle enhanced turbidimetric inhibition immunoassay method. A particle-bound drug (PBD) binds to PHE specific antibody (Ab) resulting in the formation of insoluble aggregates causing light scatter. Non-particle-bound PHE in the patient sample competes with the PBD for the antibody binding sites, inhibiting formation of insoluble aggregates. The rate and amount of particle aggregation is inversely proportional to the concentration of PHE in the sample.
The SYNCHRON® System(s) automatically proportions the appropriate sample and reagent volumes into a cuvette. The ratio used is one part sample to 98 parts reagent. The system monitors aggregate formation by measuring the change in absorbance at 340 nanometers. This change in absorbance is inversely proportional to the concentration of PHE in the sample and is used by the System to calculate and express the PHE concentration based upon a multi-point calibration curve.



RELATED DOCUMENTS

R-PO-CH-0810	Quality Control Program General Laboratory
R-PO-CH-0809	Quality Control Westgard Rules Statistics
R-PR-AD-0540	Specimen Rejection/Cancellation Protocol
J-F-CH-0820	DXC 800 Controls
J-F-CH-0826	DXC 800 Calibrators
J-F-CH-1940	DXC Analytical Measurement Range

SPECIMEN

Type of Specimen

Biological fluid samples should be collected in the same manner routinely used for any laboratory test. Freshly drawn serum or plasma are the preferred specimens. Acceptable anticoagulants are listed in the PROCEDURAL NOTES section of this chemistry information sheet. Whole blood or urine are not recommended for use as a sample.

Specimen Storage and Stability

1. Tubes of blood are to be kept closed at all times and in a vertical position. It is recommended that the serum or plasma be physically separated from contact with cells within two hours from the time of collection.3

2. Separated serum or plasma should not remain at room temperature longer than 8 hours. If assays are not completed within 8 hours, serum or plasma should be stored at +2°C to +8°C. If assays are not completed within 48 hours, or the separated sample is to be stored beyond 48 hours, samples should be frozen at -15°C to -20°C. Frozen samples should be thawed only once. Analyte deterioration may occur in samples that are repeatedly frozen and thawed.3

Sample Type	Volume	Sample Stability
Plasma/Serum	0.5mL	• Separate serum from cells within 2 hours • Room Temp 8 hours • Refrigerated 48 hours • Frozen 3 months

Criteria for Unacceptable Specimens

See Specimen Rejection/Cancellation Protocol

Sample Volume

A filled 0.5 mL sample cup is the optimum volume. For optimum primary sample tube volumes in primary tube samples and minimum volumes, refer to the Primary Tube Sample Template for your system.

REAGENTS

Contents

Each kit contains the following items:
Two PHE Reagent Cartridges (2 x 100 tests)

Volume per Test	
Sample Volume	3 µL
Total Reagent Volume	295 µL
Cartridge Volumes	A 210 µL B 55 µL C 30 µL

Reactive Ingredients	
Phenobarbital Particle Reagent	4.5 mL
Monoclonal anti-Phenobarbital Antibody (mouse)	8.2 mL
Phenobarbital Reaction Buffer	32.0 mL

Also non-reactive chemicals necessary for optimal system performance.

Reagent Preparation

No preparation is required. Do not mix.

Acceptable Reagent Performance

The acceptability of a reagent is determined by successful calibration and by ensuring that quality control results are within your facility`s acceptance criteria.

Reagent Storage and Stability

PHE reagent when stored unopened at +2°C to +8°C, will remain stable until the expiration date printed on the cartridge label. Once opened, the reagent is stable for 42 days at +2°C to +8°C unless the expiration date is exceeded. DO NOT FREEZE. Do not expose reagent to temperatures above +35°C or to direct sunlight.

CALIBRATION

Calibrator Required

SYNCHRON® Systems Drug Calibrator 1

Calibrator Preparation

No preparation is required.

Calibrator Storage and Stability

SYNCHRON® Systems Drug Calibrator 1 is stable until the expiration date printed on the calibrator bottle if capped and stored in the original container at +2°C to +8°C.

Calibration Information

1. The system must have a valid calibration curve in memory before control or patient samples can be run.

2. Under typical operating conditions the PHE reagent cartridge must be calibrated every 14 days and also with certain parts replacements or maintenance procedures, as defined in the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.

3. This assay has within-lot calibration available. For detailed calibration instructions, refer to the UniCel DxC 600/800 System *Instructions for Use* (IFU) manual.

4. The system will automatically perform checks on the calibration and produce data at the end of calibration. In the event of a failed calibration, the data will be printed with error codes and the system will alert the operator of the failure. For information on error codes, refer to the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.

Traceability

For Traceability information refer to the Calibrator instructions for use.

QUALITY CONTROL

See Related Documents J-F-CH-0820 DXC 800 controls

STEPS

1. If necessary, load the reagent onto the system.

2. After reagent load is completed, calibration may be required.

3. Program samples and controls for analysis.

4. After loading samples and controls onto the system, follow the protocols for system operation. To load samples manually refer to the FHS DXC Series Manual Sample Programming procedure. .For detailed testing procedures, refer to the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual

CALCULATIONS

SYNCHRON® System(s) perform all calculations internally to produce the final reported result. The system will calculate the final result for sample dilutions made by the operator when the dilution factor is entered into the system during sample programming.

ANTICOAGULANT TEST RESULTS

If plasma is the sample of choice, the following anticoagulants were found to be compatible with this method based on a study of 20 healthy volunteers:

Anticoagulant	Level Tested for In Vitro Interference
Lithium Heparin	14 Units/mL
Sodium Heparin	14 Units/mL

PERFORMANCE CHARACTERISTICS

Reference Range

Therapeutic	**15.0 – 40.0 ug/mL**
Critical (Toxic)	**>40.0 ug/mL**

Analytic Range

The SYNCHRON® System(s) method for the determination of this analyte provides the following analytical range:

Sample Type	Conventional Units
Serum or Plasma	5.0 – 80.0 µg/mL

Samples reported out as greater than the analytical range may be confirmed by diluting with saline and reanalyzing. The appropriate dilution factor should be applied to the reported result.

Reporting results outside of analytical range

Lower limit of range: serum / plasma	5.0 ug/mL	Results less than 5.0, report as <5.0 ug/mL
Upper limit of range: serum / plasma	80.0 ug/mL	Results >80.0 should be diluted with 0.9% saline, reanalyzed and dilution factor applied. The maximum allowable dilution is X2. Results >160.0 are reported as >160.0 ug/mL.

Sensitivity

Sensitivity is defined as the lowest measurable concentration which can be distinguished from zero with 95% confidence. Sensitivity for PHE determination is 5.0 µg/mL (21.5 µmol/L).

LIMITATIONS

None identified.

Interferences

1. The following substances were tested for interference with this methodology:

Substance	Source	Level Tested	Observed Effect
Bilirubin (unconjugated)	Bovine	30 mg/dL INDEX of 20	No significant interference (within ± 2.0 ug/mL or 8%)
Hemoglobin	RBC hemolysate	500 mg/dL INDEX of 10	No significant interference (within ± 2.0 ug/mL or 8%)
Lipemia	Intralipid®	500 mg/dL INDEX of 10	No significant interference (within ± 2.0 ug/mL or 8%)

2. Refer to References for other interferences caused by drugs, disease and preanalytical variables.

3. For assays employing mouse antibodies, the possibility exists for interference by human anti-mouse antibodies (HAMA) in the sample. Human anti-mouse antibodies may be present in samples from patients who have received immunotherapy or diagnostic procedures utilizing monoclonal antibodies or in individuals who have been regularly exposed to animals.[12,13] Additionally, other heterophile antibodies, such as human anti-goat antibodies may be present in patient samples. Interpretation of results should be done in the context of the overall clinical presentation of the patient, including symptoms, clinical history, data from additional tests and other appropriate information.

ADDITIONAL INFORMATION

For more detailed information on UniCel DxC Systems, refer to the appropriate system manual.

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