U.S. Department of Commerce
Juanita M. Kreps
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate

Standard Reference Material 900

Antiepilepsy Drug Level Assay Standard

This Standard Reference Material (SRM) is certified for concentrations of four antiepilepsy drugs--phenytoin, ethosuximide, phenobarbital, and primidone--in a processed human serum base. It is intended for use (1) in the calibration and standardization of procedures employed in clinical laboratories for the determination of these drugs in serum, and (2) for the critical evaluation of working or secondary reference solutions prepared either in-house or supplied commercially. The certified concentrations apply to the four materials supplied, after each serum is reconstituted, following the procedures described in "Instructions for Use."

	Concentration Level, μg/mL			
Drug	Toxic	Therapeutic	Subtherapeutic	Blank
Phenytoin	60.7 ± 0.9	16.7 ± 0.3	4.2 ± 0.1	0
Ethosuximide	174.7 ± 0.6	75.9 ± 0.5	11.8 ± 0.4	0
Phenobarbital	103.6 ± 0.3	21.6 ± 0.2	5.3 ± 0.2	0
Primidone	18.6 ± 0.7	8.1 ± 0.2	3.6 ± 0.1	0

The uncertainties represent one standard error for the above certified values. The imprecisions observed both within and between the liquid and gas chromatographic analyses used for this certificate are included in the standard errors. The statistical analyses were made by R. C. Paule and J. Mandel.

The modified human serum base was processed, vialed and packaged by Microbiological Associates, Walkersville, Maryland. Analyses leading to certification were performed in the NBS Center for Analytical Chemistry by R. Angeles, R. G. Christensen, A. Cohen, B. Coxon, D. Enagonio, D. J. Reeder, and L. T. Sniegoski. Valuable correlating analyses were also performed by: E. Berman, Cook County Hospital, Chicago, Illinois; K. Dudley, The University of North Carolina at Chapel Hill; and H. J. Kupferberg, National Institute of Neurological and Communicative Disorders and Stroke, NIH.

The overall direction and technical measurements leading to the certification were under the chairmenship of D. J. Reeder and R. Schaffer.

The technical and support aspects concerning the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by R. Keith Kirby.

Washington, D.C. 20234
April 5, 1979
(Editorial Revision of Certificate
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J. Paul Cali, Chief
Office of Standard Reference Materials

(over)

The analytical techniques used in the certification of this Standard Reference Material were liquid chromatography, calibrated with external standards, and gas chromatography, calibrated by bracketing and internal standards. The drug concentrations have also been determined with commercially available reagents that are based on immunoenzymatic assay principles. The mean values obtained by this methodology were close to the certified values; however, the precision was not sufficient to be included in the certification data.

The Standard Reference Material is intended for "in vitro" diagnostic use only.

WARNING: HANDLE AS IF CAPABLE OF TRANSMITTING HEPATITIS. Source material from which this serum base was derived was found non-reactive for Hepatitis B antigen when tested with licensed third-generation reagents. No known test method can provide complete assurance that products derived from human blood will not transmit hepatitis.

Instructions for Use

This Standard Reference Material is supplied as a set of four different freeze-dried preparations. They should be stored at refrigerator temperature (~ 4 °C) and should not be exposed to sunlight or ultraviolet radiation. Under such storage, the SRM is expected to be stable for at least 2 years. Samples of this SRM will be monitored. Should statistical evidence indicate a degradation of the certified properties, purchasers will be notified by NBS. It is recommended that the material not be used after 2 years from the date of purchase.

For use, it is necessary to reconstitute the freeze-dried materials with high-purity water.* When a vial is opened, remove the rubber stopper carefully so as not to dislodge any serum particles that may adhere to the stopper. Add 5.0 mL of water to the vial from a calibrated volumetric pipet or other dispenser of known accuracy. Replace the stopper and allow the contents to stand at room temperature for 20-30 min. Finally, mix the contents by gentle swirling. DO NOT shake vigorously because denaturation and frothing may result.

After reconstitution, the contents should be used within one day; otherwise, the certified values cannot be assured. Storage of the reconstituted material beyond 12-24 hours may result in deterioriation of the drugs or in degradation of the serum base.

This Standard Reference Material has been measured and certified at the Laboratories of the National Bureau of Standards, Gaithersburg, Maryland. All inquiries should be addressed to:

Office of Standard Reference Materials
Room B311, Chemistry Building
National Bureau of Standards
Washington, D.C. 20234

The date of issuance and certification of this Standard Reference Material is November 17, 1978.

*Equivalent to Type II reagent grade water as specified by the College of American Pathologists (CAP).