Report of Analysis

Reference Material 8419

Inorganic Constituents in Bovine Serum

Prepared by the United States Department of Agriculture

Distributed by the National Bureau of Standards

This Reference Material (RM) is intended primarily for use in calibrating instrumentation and evaluating the reliability of analytical methods for the determination of major, minor, and trace inorganic constituents in blood serum, plasma, and similar biological fluids.

Preparation: RM 8419 was prepared from blood that was collected directly from the severed carotid artery of an unconscious dairy cow in a plastic pail, then immediately poured into plastic centrifuge bottles. After clotting, the blood was centrifuged, serum collected, and centrifuged again, to remove any remaining red blood cells.

Homogeneity: All of the serum was pooled in a large plastic container, stirred for one hour, aliquoted by plastic siphon tube into sterile polypropylene tubes (4.2-4.3 mL/tube), capped, labeled, bagged with ice cubes to maintain 100% humidity, and stored at -20 °C. All operations except the initial collection were carried out in a Class-100 area of a clean room facility.

For the homogeneity assessment, numerous tubes, selected at random, were analyzed for the major elements, Na, K, Ca and Mg. Percent relative standard deviations (% RSD) between 1.0 and 1.8 were observed. Since these values include both homogeneity and overall precision of the methodology, the homogeneity can be no worse than these values. For the minor element, Cu, a % RSD of 1.6 was observed at the 0.75 mg/L level, while for the trace element, Cr, a % RSD of about 7 was observed at the 0.26 μ g/L level, all of which indicate good material homogeneity.

Comparison with human serum: The reported major (matrix) constituents of human and bovine serum are compared in Table 1. In addition, the measured specific gravity of each is identical (1.026 g/mL) as is the moisture content (90.7%). An advantage of a bovine serum over a human serum RM is the reduced risk of transmission of dangerous diseases such as hepatitis and AIDS (acquired immune deficiency syndrome).

Table 1. Comparison of major (matrix) components in human and bovine sera

Components	Human ^a	Bovine ^b
Total Protein, g/L	60-80	54-70
Albumin, g/L	35-50	34-50
Fat, g/L	1.8-4.1	1.9-5.1
Glucose, g/L	0.70-1.10	0.9-5.1
Na, mmol/L	135-148	130-142
Cl, mmol/L	98-106	93-112
P(i), mg/L ^c	30-45	31-49

^a Fundamentals of Clinical Chemistry, Tietz, N.W., ed., W.B. Saunders, Philadelphia, PA 1976.

^bBlood and Other Body Fluids, Dittmer, D.S., ed., Fed. Am. Soc. Exp. Biol., Washington, DC, 1961.

^cInorganic phosphorus.

Notice and Warnings to Users

IN VITRO TESTING ONLY!

Storage: It is recommended that the material be stored in the original containers at -20 °C. It is recommended that the freezer not be of the "frost-free" variety. For long term storage, it is recommended that the material be sealed in a plastic bag containing ice cubes to maintain 100% humidity.

Sample size: Each of the tubes contains approximately 4.2-4.3 mL, which should permit retrieval of 4.0 mL of serum per tube.

Aliquots: Tubes should be opened only in a clean area with precautions taken against contamination during sampling. Tubes should be thawed at room temperature and mixed well prior to sampling. It is recommended that aliquots be measured by weight, rather than volume, due to the viscous nature of serum. Volume can be computed from the specific gravity (1.026 g/mL) at 23 °C.

Cooperating Analysts: This material was developed under the technical guidance of C. Veillon, USDA, Beltsville, MD. The collaboration of the following is gratefully acknowledged:

- S.A. Lewis, K.Y. Patterson, D.C. Reamer and N.A. Bryden, USDA, Beltsville, MD.
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Special thanks to R. Alvarez (NBS), M. Ihnat (Agriculture Canada), and W.R. Wolf (USDA), for helpful discussions.

Recommended Values: The recommended concentrations and estimates of uncertainty are given in Table II. These concentrations are those with which the authors feel most comfortable, considering the values reported by the collaborators, and what is known about the laboratories, analysts, and techniques used. Naturally, these are subject to scrutiny by the scientific community, but form a reasonable starting point. Criteria used in assigning the estimates of uncertainty are based primarily on ranges encompassing values for which the authors are reasonably confident, centered near the means of such values. Reasonable experimental reproducibilities, and standard deviations of the means were also considered. While this is not a statistically meaningful way to select recommendations, insufficient data were available for a statistically valid assessment. Detailed results reported by the collaborators will be presented in a forthcoming publication (Veillon, C., et al., Anal. Chem., submitted).

Table II. Recommended values and estimated uncertainties for Inorganic Constituents in Bovine Serum, Reference Material 8419.*

Recommended Value		
Element	± Estimate of Uncertainty	Methods
Na	141 ± 2 mmol/L	3,9,11
K	$5.1 \pm 0.2 \text{ mmol/L}$	3,9,11
Ca	$2.5 \pm 0.1 \text{ mmol/L}$	1,2,11
Mg	$0.85 \pm 0.1 \text{mmol/L}$	1,2,11
Fe	$2.0 \pm 0.4 \text{ mg/L}$	1,2,4,6,7,10,11,12
Cu	$0.75 \pm 0.1 \text{mg/L}$	2,4,5,6,7,10,11,12,13
Zn	$1.1 \pm 0.1 \text{mg/L}$	2,5,6,7,10,11,12,13
Al	$13 \pm 5 \mu g/L$	4,5,6,12
Co	$1.2 \pm 0.3 \mu g/L$	4,10,12,13
Cr	$0.30 \pm 0.05 \; \mu\mathrm{g/L}$	4,5,8,10,12
Mn	$2.6 \pm 0.5 \ \mu g/L$	4,5,6,7,10,12
Mo	$16 \pm 4 \mu g/L$	5,6,10,12
Ni	$1.8 \pm 0.6 \mu g/L$	4,6,12,13
Se	$16 \pm 2 \mu g/L$	5,8,10
V	<2 μg/L	6,12

- 1. Colorimetry.
- 2. Flame Atomic Absorption Spectrometry.
- 3. Flame Atomic Emission Spectrometry.
- 4. Electrothermal Atomic Absorption Spectrometry (Conventional).
- 5. Electrothermal Atomic Absorption Spectrometry (Zeeman).
- 6. Inductively Coupled Plasma Atomic Emission Spectrometry.
- 7. Inductively Coupled Plasma Atomic Fluorescence Spectrometry.
- 8. Isotope Dilution Mass Spectrometry.
- 9. Ion Selective Electrode.
- 10. Neutron Activation Analysis.
- 11. Simultaneous Multi-Element Atomic Absorption Spectrometry (flame).
- 12. Simultaneous Multi-Element Atomic Absorption Spectrometry (furnace).
- 13. Voltammetry.
- *U.S.D.A. designation: 7292

As more data become available, the recommended values may be updated and additional elements listed. Data, comments, and inquiries from users will be welcomed, and should be directed to:

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