ANTIMICROBIAL AGENT AND CHEMOTHERAPY, Oct. 1977, p. 550

Vol. 12, No. 4
Printed in U.S.A.

Discrepant Results of Amphotericin B Assays on Fresh Versus Frozen Serum Samples

R. M. BANNATYNE* AND R. CHEUNG

Department of Bacteriology, The Hospital for Sick Children, Toronto, Ontario, M5G 1X8 Canada

Received for publication 6 May 1977

Amphotericin B assays on frozen serum samples (−20°C) generally underestimate the serum levels obtained on promptly processed samples. The degree of the discrepancy is variable and independent of the length of time stored.

Serum assays for amphotericin B may not be within the capabilities of many hospital laboratories. In such circumstances, specimens for assay are often frozen until they can be transported to a laboratory that provides this service. Because of the notorious instability of amphotericin B under various physical conditions, including those of solution and temperature (2–4), we compared the activity of the drug in divided serum samples processed both immediately and after prolonged storage at −20°C.

Eighty-five clinical serum samples for amphotericin B assay received in the laboratory over a 10-month period were split into two portions. One portion was assayed for its amphotericin B concentration immediately on receipt, and the other portion was assayed after storage of the sample at −20°C for periods from 3 to 10 months. Assays on the frozen samples were all performed within a few weeks of each other. All assays were performed by the accurate micromethod previously described (1) using the same batch and lot numbers of nystatin assay agar and amphotericin B (Fungizone, E.R. Squibb and Sons Ltd.), respectively. The results of the assays on the frozen samples were then compared with those on the fresh samples.

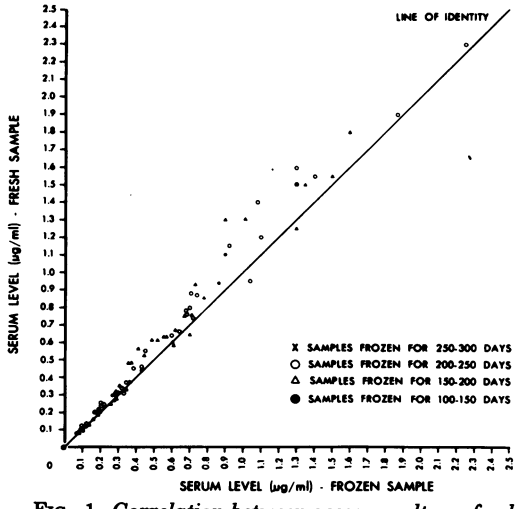

FIG. 1. *Correlation between assay results on fresh versus frozen samples. Each point represents the results of duplicate assays performed on split serum samples, one value plotted on the ordinate and the other on the abcissa. The line of identity is included for reference.*

The correlation between the results obtained on fresh versus frozen samples was poor (Fig. 1). Complete agreement was obtained in only 6 of 85 samples. The assay values obtained in the majority of the frozen samples were below those observed on the same samples processed fresh. Of these, 29 of 85, 26 of 85, 8 of 85, and 1 of 85 were within 10, 20, 30, and 87.5%, respectively, of the expected value. Assays of 14 of 85 frozen samples gave slightly higher levels than expected, but all were within 10% of the expected value. No relationship was noted between the length of time frozen (3 to 10 months) and the extent of the drop in activity (Fig. 1).

LITERATURE CITED

1. **Bannatyne, R. M., R. Cheung, and H. R. Devlin.** 1977. Microassay for amphotericin B. Antimicrob. Agents Chemother. 11:44–46.
2. **Block, E. R., and J. E. Bennett.** 1973. Stability of amphotericin B in infusion bottles. Antimicrob. Agents Chemother. 4:648–649.
3. **Cheung, S. C., G. Medoff, D. Schlessinger, and G. S. Kobayashi.** 1975. Stability of amphotericin B in fungal culture media. Antimicrob. Agents Chemother. 8:426–428.
4. **Hamilton-Miller, J. M. T.** 1973. The effect of pH and of temperature on the stability and bioactivity of nystatin and amphotericin B. J. Pharm. Pharmacol. 25:401–407.