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Correlation of Urine pH with the Detection of Cytomegalovirus by the Shell Vial Technique

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The influence of the pH of urine on the detection of cytomegalovirus (CMV) by the shell vial assay was evaluated. The pH values of 295 urine specimens ranged from 4.7 to 8.5 (mean 6.3) and were not significantly different in culture-positive versus culture-negative samples. None of the urine specimens appeared to be toxic for the cells used in the shell vial assay. We recommend inoculation of urine specimens into shell vials without adjustment of pH.

Cytomegalovirus (CMV) is an important etiologic agent of hepatitis and heterophile-negative mononucleosis-like disease in normal hosts, is the most common infectious cause of a variety of long-term defects in neonates, and produces life-threatening, disseminated disease in immunocompromised hosts (Navaqi, 1984). Because of the potentially fatal outcome of CMV infections and the recent availability of specific antiviral chemotherapy, prompt and accurate detection of this virus in the laboratory is clinically important (Popow-Kraupp and Kunz, 1988; Sorbello et al., 1988).

The detection of CMV by the shell vial assay has become an accepted rapid method for establishing the laboratory diagnosis of this infection. However, to achieve maximal sensitivity of this procedure, several variables must be carefully controlled: the type of cells, preparation of cell cultures, speed of centrifugation, incubation time, monoclonal antibody and other reagents, and staining procedure. Urine is a very productive specimen source for detecting CMV, but the pH of this body fluid can vary almost four units (Bradley and Schumann, 1984). In addition, some viruses, especially those with envelopes, are susceptible to inactivation at extremes in pH (Croughan and Behbehani, 1988; Schieble, 1985; Smith, 1985). Interestingly, urine specimens submitted to some laboratories for the diagnosis of CMV infection are adjusted to a neutral pH in an attempt to increase the possibility of detecting the virus or perhaps to reduce the toxicity of specimens to cell monolayers (Janssen et al., 1988; Popow-Kraupp and Kunz, 1988; Sorbello et al., 1988; Starr and Friedman, 1985).

The results of preliminary studies in our laboratory suggested that detection of CMV by the shell vial technique was independent of the pH of the medium (Eagle Minimal Essential Medium) in which CMV was suspended, and that medium pH values of 3 or less and 10 or more were toxic for the cells used in the shell vial assay. This communication presents results of a clinical study in which we examined the correlation of the pH of urine specimens with the detection of CMV by the shell vial assay.

The pH of 295 urine specimens (from 159 patients) submitted for the diagnosis of CMV infection was determined with a pH meter (Fisher Accumet, Model 805 MP, Fisher Scientific) between 1 and 16 hours after each sample was received in the laboratory.

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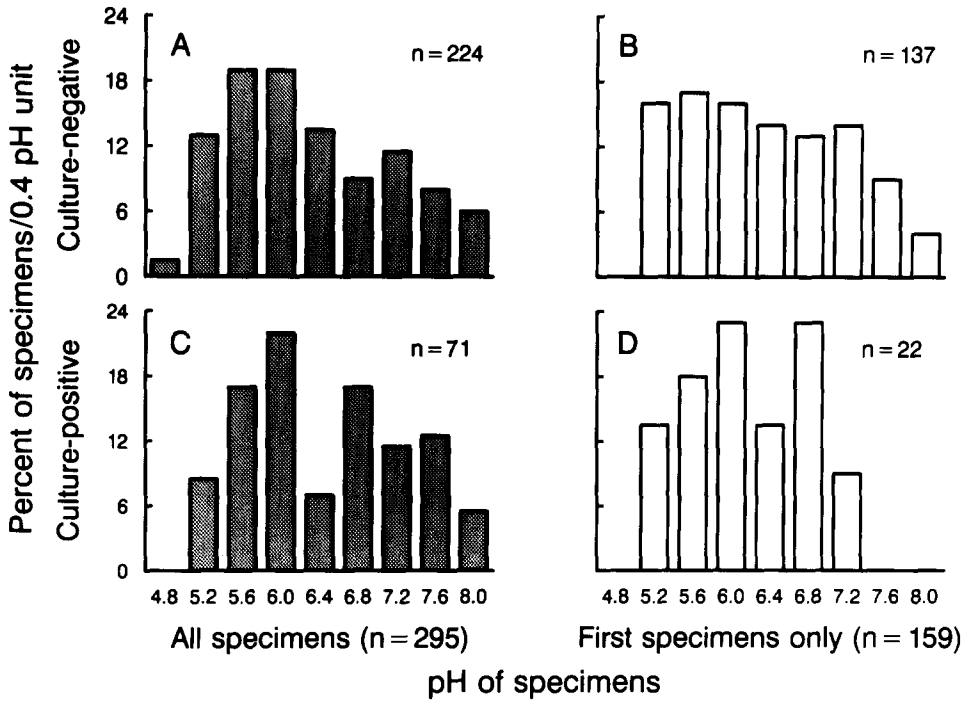

FIGURE 1 Correlation of urine specimen pH with detection of cytomegalovirus by the shell vial technique. For statistical purposes, the pH values of the specimens were grouped into nine 0.4–pH unit increments; the pH values shown are the midpoints of these increments.

The presence of CMV in each specimen was determined by the shell vial assay, which involves the use of MRC-5 cells and anti-CMV early antigen monoclonal antibody in an indirect immunofluorescence test (Gleaves et al., 1984). The pH distribution of all specimens that were culture positive for CMV was compared with the pH distribution of all specimens that were culture negative. A similar comparison was made between culture-positive and culture-negative specimens when only initial (first) specimens from each individual were examined. The distributions of the original pH values in culture-positive and culture-negative specimens were compared by using Wilcoxson rank sum tests. For statistical purposes, pH values were then grouped into nine 0.4-pH unit increments; p values of less than 0.05 were considered to be statistically significant.

The pH values of the 295 clinical urine specimens ranged from 4.7 to 8.5 (mean 6.3). This range is similar to a previously published range (4.6 to 8.0) of pH values of urine from healthy individuals (Bradley and Schumann, 1984).

When all specimens (n = 295) were examined, there was no statistically significant difference between the distribution of pH values of the culture-negative specimens and those of the culture-positive specimens (p = 0.11) (Figs. 1A and C). Similarly, when only the first urine specimen from each patient (n = 159) was examined, there was no statistically significant difference between the distribution of pH values of the culture-negative and culture-positive specimens (p = 0.65) (Figs. 1B and D). None of the 295 urine specimens appeared to be toxic for the cells used in the shell vial assay.

Our results indicated that the pH of urine specimens did not require adjustment or neutralization as a means of eliminating toxicity to cell culture monolayers or for maximal detection of CMV by the shell vial assay. We recommend inoculation of urine specimens into shell vials without adjustment of pH.

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