Blood Cholesterol Concentration: Fingerstick Plasma vs Venous Serum Sampling

Philip Greenland,1 Nancy L. Bowley,1 Barbara Melklejohn,1 Kathryn L. Doane,1 and Charles E. Sparks2

To assess whether venous and fingerstick blood samples yield similar cholesterol concentrations, we obtained both types of samples simultaneously in 188 volunteers participating in a cholesterol screening program. All samples were analyzed by the same enzymatic method in a standardized laboratory, and pairs of simultaneous samples were measured in the same laboratory run. Cholesterol concentrations in fingerstick-derived plasma were consistently higher than in the venous serum (P <0.0001), by a positive bias averaging 3.6%. Cholesterol values in fingerstick plasma also were higher than cholesterol results for venous serum placed in a capillary collection tube (average bias +2.4%). The positive bias of fingerstick plasma vs venous serum results appears to be at least partly due to specimen handling, although a true physiological difference between venous and fingerstick cholesterol concentrations is probably also involved. If a positive bias of this magnitude from fingerstick blood sampling is left unadjusted, substantial numbers of people will be labeled “at risk” and referred to physicians when their true values were actually within the acceptable range.

Additional Keyphrases: variation, source of • screening • physician’s office testing

The measurement of cholesterol concentrations in blood can be affected by many factors. Some are physiological, such as the changes in cholesterol due to dietary manipulations (1), to postural changes (2), or to seasonal variations (3). Variability of repeated cholesterol determinations can also be methodological in origin, such as differences noted between serum and plasma cholesterol measurements in some studies (4) and systematic differences (bias) between analytical methods or laboratories (5, 6).

Since 1984, the treatment of increased concentrations of blood cholesterol has focused on diagnostic and therapeutic actions based on very distinct target cholesterol values (7). The specimen and type of analysis used to derive the recommended diagnostic and therapeutic cutoff values specified in the 1984 Consensus Conference Report (8) and in the 1988 Adult Treatment Panel Guidelines (7) are a venous serum sample analyzed in a laboratory in which standardization is traceable to the Centers for Disease Control (CDC) (7). Recently, the availability of portable blood analyzers that are capable of measuring cholesterol in small volumes of serum or plasma has led to the appearance of cholesterol screening programs that often determine cholesterol concentration in blood derived from a fingerstick sample (9–11). If systematic differences exist between fingerstick and venous concentrations of cholesterol, adjustment for these differences may be advisable when making diagnostic or therapeutic decisions. Because of uncertainty about the equivalence of venous vs fingerstick blood cholesterol determinations and because of the importance of any differences for cholesterol screening, we undertook this study, using samples from a community-based cholesterol screening program.

Materials and Methods

A nonfasting venous blood sample was obtained at the same time as a fingerstick sample from 188 volunteers. All samples were analyzed on the CDC-standardized Eastman Kodak Ektachem 700 blood analyzer in the clinical laboratory at The University of Rochester Medical Center. The enzymatic method for total cholesterol in this instrument is identical to that used in the Kodak Ektachem DT-60 portable analyzer. Paired samples (i.e., fingerstick and venous) were analyzed in the same laboratory run, although samples were analyzed in multiple analytical runs. The order of analysis of samples was scrambled, to avoid any possible systematic analytical bias. All samples were obtained by trained laboratory technologists according to standard blood-collection protocols. Tourniquet times were minimized during the venous samplings, and “milking” the fingertips was avoided in the fingerstick collection procedure.

The fingerstick sample was collected in a plastic-walled capillary tube (cat. no. 18443.300; Sarstedt, Princeton, NJ) coated with lithium heparin anticoagulant. This collection tube is commonly used in cholesterol screenings involving the Kodak DT-60 analyzer. The fingerstick samples were centrifuged within 5 min and placed on ice until analyzed (within 3 h). We designated these samples “fingerstick plasma” samples. The venous sample was collected directly into a glass-walled tube containing no anticoagulant (cat. no. 6430; Becton Dickinson and Co., Rutherford, NJ) and allowed to clot at room temperature for 30 min. The resulting serum was promptly separated by centrifugation and placed on ice until analysis (within 3 h). An aliquot of the venous serum, hereafter called the “venous-capillary sample,” was placed in a second Sarstedt tube and analyzed as described earlier for the fingerstick specimen.

The venous serum samples served as the reference (“gold standard”) against which the fingerstick plasma and venous-capillary values were compared. Regression equations were generated by using an IBM-PC computer and the SAS software package. The average bias of the fingerstick plasma results was determined by subtracting the mean of the venous serum values from the mean of the plasma cholesterol results, then dividing the difference by the mean venous serum value. Multiplying the result by 100 yielded an average percentage difference between the two observations. Bias plots were developed by plotting the individual percentage differences against the corresponding venous cholesterol results. Three venous-capillary samples were unacceptable for analysis because too little sample volume was available. All 188 fingerstick and venous serum samples were acceptable for analysis.
Results

Table 1 summarizes the regression equations comparing the three pairs of simultaneous samples. As noted, correlation coefficients are nearly equal to 1.0 for each regression line, and linear slopes were also nearly equal to 1.0 for each regression.

Although these correlations between paired samples were quite good, more than 90% of the paired cholesterol determinations showed a higher cholesterol concentration in the fingerstick plasma sample than in the venous serum (gold-standard) determination (Figure 1). The average bias of the fingerstick plasma vs venous serum cholesterol values was +3.6%. Similarly, more than 80% of the fingerstick plasma cholesterol values exceeded the simultaneous venous-capillary result, yielding an average bias of +2.4% (Figure 1). The differences between both sets of means for the venous-fingerstick comparisons were highly significant (P < 0.0001, paired t-test). Figure 1 also shows that comparing the venous-capillary concentrations with the venous-serum values resulted in a mean bias of +1.1%, also significant (P < 0.0001, by paired t-test).

Whereas the bias of the fingerstick-plasma vs venous-serum cholesterol concentrations averaged +3.6%, the bias expected from the regression equations was inversely proportional to the true cholesterol concentration, the linear slope being slightly less than 1.0. Therefore, at a venous cholesterol concentration of 1990 mg/L, the bias is actually +4.6%, while at 2390 mg/L, the bias decreases to 3.4%.

Discussion

Recent recommendations from the National Cholesterol Education Program (NCEP) have specified distinct cholesterol cutpoints for diagnostic and therapeutic actions (7). These cutpoints refer to venous serum samples. Because EDTA-anticoagulated venous plasma yields cholesterol measurements approximately 3% lower than venous serum results (4), the NCEP advised increasing venous plasma results by 3% to allow comparisons with the NCEP cutpoints (7). In this study, cholesterol measurements in fingerstick plasma samples collected in lithium-heparinized tubes were found to be systematically higher than cholesterol concentrations in simultaneously collected venous serum. The average bias in the fingerstick result was 3.6%, a magnitude similar to that previously of concern to the NCEP in formulating the Adult Treatment Panel's Guidelines (7).

Previous studies comparing cholesterol concentrations in blood samples simultaneously collected from fingerstick and vein have produced inconsistent results. One study found fingerstick cholesterol values to be identical to venous cholesterol (12). Another study showed fingerstick results to be lower than venous concentration by an average of 8.7% (13), and yet another study reported a positive bias, similar to that noted here (14). To explain a difference of a similar magnitude that we had found between venous-serum results and fingerstick-plasma results in an earlier study (15), we performed the present study, comparing

<table>
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<th>x</th>
<th>y</th>
<th>n</th>
<th>Slope</th>
<th>Intercept, mg/L</th>
<th>r²</th>
<th>Sres, mg/L</th>
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<td>Venous serum</td>
<td>Fingerstick plasma</td>
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<td>+139.9</td>
<td>0.95</td>
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<tr>
<td>Venous capillary</td>
<td>Fingerstick plasma</td>
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<td>0.975</td>
<td>+121.6</td>
<td>0.95</td>
<td>71.9</td>
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<tr>
<td>Venous serum</td>
<td>Venous capillary</td>
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<td>0.994</td>
<td>+32.1</td>
<td>0.98</td>
<td>44.2</td>
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Fig. 1. Bias plots comparing (top) the bias of individual fingerstick-plasma cholesterol concentrations vs the venous-serum cholesterol concentration; (middle) the bias of individual fingerstick-plasma cholesterol concentrations vs the venous-capillary cholesterol concentration; and (bottom) the bias of individual venous-capillary cholesterol concentrations vs the venous-serum cholesterol concentration.
fingerstick cholesterol measurements in plasma against both a venous serum sample and a venous serum sample placed in the same kind of heparin-coated anticoagulated collection tube as we used for the fingerstick blood. We performed all cholesterol measurements in an identical manner. Therefore, the difference we noted here, averaging an increase of 2.4% for the fingerstick-plasma result vs the venous-capillary result, suggests that a true biological difference exists between cholesterol concentration in the two kinds of samples (venous compared with fingerstick). The difference cannot be accounted for by dilution of capillary blood with lymphatic fluid or interstitial fluid in the fingerstick sample because the fingerstick results were, on average, higher rather than lower than the cholesterol measurements in venous specimens. It is also unlikely that lymphatic or interstitial fluids increased the cholesterol concentration of the fingerstick sample because total cholesterol concentration of peripheral lymph is only about 10% of that in plasma (16). Whatever the exact reasons for the consistent differences seen, these differences cannot be explained entirely by artifact, specimen-handling differences, or effects of the collection tube and its anticoagulant alone.

All the determinations in this study utilized an enzymatic method for cholesterol, and all were performed on a CDC-standardized laboratory instrument for which CVs are highly acceptable (<3.0%) according to current laboratory standards. The enzymatic method we used is identical to that used in one of the popular methods for cholesterol screening (Ektachem DT-60, Eastman Kodak) (11). Perhaps the biases we observed here are unique to this method of cholesterol assessment. Perhaps lymphatic fluid derived during fingerstick sampling causes a matrix effect and produces the positive bias we observed, as did Alzofon et al. (14), who also used an enzymatic cholesterol analysis. However, enzymatic methods are common to several analytical systems used in cholesterol screenings, and the biases we noted here are, therefore, very likely to be found by others, especially in cholesterol screening settings.

It is worthwhile to consider the implications of the results we observed here for screening programs. In the cholesterol screening program in Rochester, had we used an upper cutoff of 190 mg/L (5.15 mmol/L) for physician referral, a 4.6% positive bias would have inappropriately referred an additional 8% of the screened population to physicians. Similarly, for a cutoff of 2390 mg/L (6.20 mmol/L), an additional 7% of screenees would have been referred, given the bias of +3.4% at this cholesterol concentration. A bias as large as we found here, therefore, should not be ignored when developing a protocol for cholesterol screening.

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References