Abstract

Objective: To compare a standardized recommended procedure for drawing blood to measure blood lipid and lipoprotein levels with the procedure commonly used in clinical practice. The aim was to see if hemoconcentration and spuriously elevated lipid levels could be avoided.

Design: An open randomized crossover clinical trial.

Setting: The University of Calgary.

Patients: Twenty-five patients with dyslipidemia.

Interventions: Blood drawing using a standardized procedure in which the patient remained seated for 5 minutes before blood collection and tourniquet use was minimized or avoided.

Main outcome measures: Differences in lipid levels between the usual clinical procedure and the recommended procedure for drawing blood.

Results: Prior to drawing blood, laboratories have sat patients for an average of 1.4 minutes (95% CI, 0.9 to 1.9) and used a tourniquet in every patient. In the standardized procedure, patients rested for an average of 5.6 minutes (95% CI 5.0 to 6.2), and a tourniquet was used briefly in only 3 of 23 patients. There were no differences in lipid and lipoprotein values and no clinically significant difference in hemoglobin or albumin levels or in the calculation of hemoconcentration.

Conclusions: Efforts to rest patients and avoid tourniquet use when drawing blood for assessment of lipid levels are unlikely to be useful.

Résumé

Objectif : Comparer une procédure recommandée normalisée pour prélever du sang afin de mesurer les taux sanguins de lipides et de lipoprotéines à la procédure utilisée couramment en pratique clinique. On voulait déterminer s’il était possible d’éviter l’hémoconcentration et les taux de lipides élevés transitoires.

Conception : Étude clinique croisée randomisée ouverte.

Contexte : Université de Calgary.

Patients : Vingt-cinq patients atteints de dyslipidémie.

Interventions : Prélèvement de sang au moyen d’une procédure normalisée au cours de laquelle le patient est demeuré assis 5 minutes avant le prélèvement du sang et l’on a réduit au minimum ou évité l’utilisation du tourniquet.

Principales mesures de résultats : Différences des taux
Introduction

Dyslipidemias are strong reversible risk factors for cardiovascular disease. Various guidelines advocate periodic screening of lipid levels for healthy adults and for those with established cardiovascular disease.1–5 Risk classification and therapeutic monitoring require accurate assessment of lipid and lipoprotein levels. Unfortunately lipid levels in clinical assessments vary substantially.6–10 It is estimated that in up to 40% of patients the risk classification changes on retesting.11 Although some of the variability associated with assessing lipids is related to laboratory procedures, the large proportion is related to biological variation (i.e., before the blood sample reaches the laboratory).6,12,13 The method of drawing blood has been reported to affect the lipid value. In particular, the use of a tourniquet and the time that the patient remains seated before blood is drawn are reported to be important factors causing lipid levels to be artificially elevated.14–17 Interestingly, only one of the recent major trials19 examining the benefits of lipid-lowering agents indicated compliance with recommendations for standardized blood drawing.18–23

We compared the effect of usual clinic phlebotomy to a standardized, recommended procedure (resting patients 5 minutes before blood sampling and avoiding tourniquet use) on measured blood lipid levels. Further, we examined the compliance with guidelines not to use a tourniquet and to seat a patient for 5 minutes before drawing blood for serum lipid measurement in the Calgary Regional Health Authority laboratories.

Methods

Design

An open, randomized, controlled, crossover design was used. It was not possible for investigators or patients to be blinded to the allocated procedures. The laboratory technicians determining lipid values were blinded to the allocated procedure, and those drawing blood routinely in the clinical laboratories were unaware of the study design. The nurse performing the standardized procedure was aware of the study design and its objectives. The random sequence for blood drawing was computer generated with the allocations enclosed in sealed envelopes.

Patients

Patients were selected from those referred to a lipid clinic for either nurse-dietician counselling or for subspecialty lipid consultation by an endocrinologist. Inclusion criteria included patients with dyslipidemia on no treatment or on stable lifestyle modification or pharmacotherapy for more than 3 months. Exclusion criteria were unstable medical conditions or medical conditions that in the opinion of the investigators would influence the results or impede the ability of the patient to complete the study (diabetes mellitus on pharmacotherapy, elevated serum creatinine concentration, diuretic therapy, diseases of the pulmonary, cardiovascular, gastrointestinal or central nervous system that interfere with the ability to perform activities of daily living). Patients with stable medical conditions and medical therapy were not excluded. Twenty-five patients (15 women, 10 men), participated in the study The mean and (standard deviation) age of the group was 58.8 (8.7) years, and the body mass index was 30.1 (5). Two patients dropped out: in one venous access was lacking, and the other would not return for the second visit. In addition to dyslipidemia, associated conditions included ischemic heart disease (3 patients), depression (3), and hypothyroidism, prostate hypertrophy, aortic
stenosis, asthma, migraine, hepatitis B, fibromyalgia and unspecified arthritis (1 each). Medications being taken included pravastatin (3 patients), amlodipine (3), flouxetine (3), and micronized fenofibrate, simvastatin, salbutamol, thyroxine, finasteride, metoprolol, acetylsalicylic acid, omeprazole (1 each).

**Blood collection**

The fasting procedure for all patients before blood drawing was that of the National Cholesterol Education Program. A research nurse performed the experimental standardized blood collection: the patients were all seated for 5 minutes before blood collection and whenever possible tourniquet use was avoided or minimized. For the usual blood drawing, patients were able to select any laboratory in the Calgary Regional Health Authority. Patients were requested to record the duration of time in the seated position before blood drawing and whether or not a tourniquet was used. The blood drawings were 1 to 4 weeks apart. Patients were advised not to change diet or activity during the study and their medications were not changed. The patients had blood drawn at 12 different sites.

**Handling of blood samples**

Blood samples were drawn into 8-mL SST (clot) tubes. The samples were centrifuged and frozen at −18 °C for up to 28 weeks. Samples from the same patients were measured at the same time to avoid interassay variation.

**Statistical analysis**

The order of the blood collection method was randomized according to a crossover design. The resultant data were first inspected descriptively for distributional properties. Subsequently the effects of 2 methods of measurement were compared by the method of Hills–Armitage. Statistical computation was performed with the use of S-Plus.

**Results**

The mean length of time that patients remained seated before blood collection was 1.4 minutes (95% CI, 0.9 to 1.9) in the usual clinical procedure compared with 5.6 minutes (95% CI 5.0 to 6.2) in the standardized procedure. Tourniquets were used in all of the patients for the usual clinical procedure but were required in only 3 patients for the standardized collections.

There were no significant differences in serum lipid levels between the standardized and the usual clinical method (Table 1). In particular, the trial ruled out an increase of greater than 0.2 mmol/L total cholesterol, 0.2 mmol/L high-density lipoprotein cholesterol, 0 mmol/L low-density lipoprotein cholesterol and 0.4 mmol/L triglycerides. There were minor changes consistent with hemoconcentration of albumin, hemoglobin and hematocrit when the clinical laboratory drew blood. This was reflected by a decrease in vascular volume as calculated by the formula of Strauss and colleagues, which is based on hemoglobin and hematocrit.

**Comment**

The usual clinical method of blood collection does not follow standardized recommendations. However, our study had the power to rule out (with at least 95% certainty) increases in lipid or lipoprotein concentrations greater than 0.2 mmol/L (triglycerides greater than 0.4 mmol/L), associated with the lack of standardized technique. The changes in lipids and

### Table 1: Percentage changes (and 95% CI) in blood and serum levels from a standardized procedure when blood was drawn by a routine laboratory procedure

<table>
<thead>
<tr>
<th>Measurement</th>
<th>After standardized procedure</th>
<th>% change from standardized procedure</th>
<th>95% CI of % change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemoglobin, g/L</td>
<td>143</td>
<td>+3.6</td>
<td>1.2, 6.0</td>
</tr>
<tr>
<td>Calculated vascular volume, %</td>
<td>100</td>
<td>−2.0</td>
<td>−3.2, −0.8</td>
</tr>
<tr>
<td>Total protein, g/L</td>
<td>69</td>
<td>+1.1</td>
<td>−0.1, 2.2</td>
</tr>
<tr>
<td>Albumin, g/L</td>
<td>40</td>
<td>+0.9</td>
<td>0.1, 1.7</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L</td>
<td>5.7</td>
<td>−0.1</td>
<td>−0.3, 0.2</td>
</tr>
<tr>
<td>HDL cholesterol, mmol/L</td>
<td>1.2</td>
<td>+0.1</td>
<td>−0.05, 0.2</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L</td>
<td>3.6</td>
<td>−0.2</td>
<td>−0.4, 0.0</td>
</tr>
<tr>
<td>Triglycerides, mmol/L</td>
<td>2.2</td>
<td>0</td>
<td>−0.4, 0.4</td>
</tr>
</tbody>
</table>

HDL = high-density lipoprotein, LDL = low-density lipoprotein.
lipoproteins of this magnitude are unlikely to affect clinical decisions and therefore efforts to standardize the method of blood drawing in lipid assessments are not necessary.

There are several potential reasons why we did not find an effect of tourniquet use or rest in the seated position whereas other, well-conducted studies found substantial effects. Ours was an effectiveness study that examined usual clinical practice whereas other studies examined the extremes of clinical practice (efficacy studies). Patients in usual clinical practice tend to wait in the seated position in the laboratory until called for blood drawing. Research practice tends to wait in the seated position in the laboratory (efficacy studies). Patients in usual clinical practice tend to wait in the seated position in the laboratory until called for blood drawing. Research conducted on the necessity of sitting for 5 to 10 minutes had patients standing before blood collection as a comparison.16,17 Additionally, the duration of use and pressure exerted by the tourniquet before blood sampling may be factors determining the degree of hemoconcentration. In this study a tourniquet was used by all clinical laboratories during blood sampling but in only 3 of the experimental blood collections. The duration and pressure exerted by tourniquet use are likely less in the clinical laboratories than in research studies that demonstrated hemoconcentration due to tourniquet use.14,15 Tourniquet use and lack of standardized rest have similar effects on blood lipid levels. The absence of any discernible lipid effect in clinical practice, where both the lack of a standardized rest and the use of a tourniquet were common, suggests that expending resources to deter tourniquet use or to standardize time in the seated position will not be beneficial. However, there may be unusual circumstances (e.g., patients with difficult venous access) in which patient position and tourniquet use may cause hemoconcentration. Technicians drawing blood will need to use judgement, and clinicians and laboratory scientists need to be aware of the potential for spurious results.

References


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