Eur. J. Clin. Chem. Clin. Biochem.
Vol. 31, 1993, pp. 869—874

TECHNICAL NOTE

Automated Turbidimetry of Serum Lipoprotein(a)

By *L. Borque, C. Maside* and *A. Iglesias*

Laboratorio Central, Hospital San Millan, Logroño, Spain

(Received April 6/September 27, 1993)

Summary: We describe a simple immunoturbidimetric method for quantifying lipoprotein(a) in serum based on latex-enhanced particle agglutination technology. Carboxylated latex particles (diameter 240 nm) covalently coated with $F(ab')_2$ fragments of anti-lipoprotein(a) antibodies are incubated with the sample for 5 min at 37 °C, and the resulting agglutination is quantified by measuring the change of turbidity produced at 700 nm. The assay is rapid, precise and fully automated on the Hitachi 911 analyser. The assay range is about 0.03—0.9 g/l. Average analytical recovery was 97.8%. Precision (CV) ranged from 1.9 to 3.1% at different lipoprotein(a) values. There was no interference from bilirubin, Intralipid®, haemoglobin, plasminogen or apolipoprotein B. Comparisons with a latex nephelometric assay carried out on the Behring nephelometer analyser, and with three commercially available methods, a radioimmunoassay and two ELISA assays, gave good correlations ($r > 0.95$), although a large among-method variation in lipoprotein(a) values was found. We conclude that the proposed latex turbidimetric immunoassay method is suitable for routine use in clinical laboratories.

Introduction

Lipoprotein(a) [Lp(a)] was initially though to be a genetic variant of low density lipoprotein (LDL) (1). Lp(a) is a low density lipoprotein-like particle containing apolipoprotein B-100 disulphide-linked to one (or two) large glycoprotein called apolipoprotein(a) (M_r 300 000—700 000) (2). Apolipoprotein(a) has been shown to have a considerable degree of homology with human plasminogen (3). The characteristic feature of lipoprotein(a) is that it is distinct from all other serum proteins and apolipoproteins. This protein is believed to be inherited as an autosomal dominant trait and appears to be insensitive to either diet, lifestyle or most hypolipidaemic drugs (4, 5).

Since its discovery by *Berg* in 1963 (1), there has been a considerable rise in interest, not only in specialized research centres but also in clinical routine laboratories, in the accurate measurement of lipoprotein(a) in blood. This interest was stimulated by reports indicating that levels above 0.2—0.3 g/l, present in approximately 25% of the population, are associated with an increased risk of coronary heart disease (6, 7). Many investigators have confirmed that a high lipoprotein(a) concentration represents an indicator of risk for cardiovascular disease, especially when other serum lipidic quantities differ significantly from the values for healthy subjects (8, 9).

These findings generated the need for a serum lipoprotein(a) assay in the clinical chemistry laboratory. Various quantitative immunological assays for lipoprotein(a) determination, including electrophoretic techniques (10), radial immunodiffusion (11), enzyme immunoassays (12), and radio immunoassays (13) have been developed. However, these procedures generally require specialized reagents and equipment, long reaction times, and multiple washing steps, and they are not easily automated. Recently, in an effort to overcome these problems several automated fluid-phase immunoprecipitation procedures have been described (14, 15).

In this study, we present a turbidimetric method which uses particle-enhanced immunoassay technology and is carried out on a last generation clinical chemistry analyser. This turbidimetric method has the advantage of being precise, rapid, easy to perform, and suffers no interference from apolipoprotein B, plasminogen, or from endogenous lipids, haemoglobin, or bilirubin. Here we describe the characteristics and performance of the method.

Materials and Methods

Apparatus

We used an Hitachi 911 analysis system (Boehringer Mannheim, D-68305 Mannheim, Germany) for all the determinations.

Samples

Blood was taken in red-top vacutainer tubes (Becton Dickinson, Rutherford, NJ), allowed to clot, and the serum removed for immediate assay or for storage at −30 °C if the analysis was to be carried out later. Unselected serum specimens used for comparative studies were obtained from the lipid laboratory of our hospital.

Antibody

Rabbit polyclonal antiserum directed against human lipoprotein(a) was manufactured by Dakopatts A/S (Glostrup, Denmark). It showed no cross-reaction with apolipoprotein B or plasminogen and was stored at 4 °C. (Lot No. 012, 3.7 g/l of protein.)