SHORT COMMUNICATION

Presence of apolipoprotein-CII in commercially available albumin fractions

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Summary

1. Commercially available bovine serum albumin as Cohn fraction V was demonstrated to contain small amounts of apolipoprotein-CII.
2. This apolipoprotein activated lipoprotein lipase in the same way as apolipoprotein-CII purified from human very-low-density lipoproteins.

Key words: albumin, apolipoprotein-CII, Cohn fraction V.

Abbreviation: apo-CII, apolipoprotein-CII.

Introduction

Lipoprotein lipase (EC 3.1.1.34) hydrolyses triglyceride emulsions only in the presence of a certain apolipoprotein, namely apolipoprotein-CII (apo-CII) (La Rosa, Levy, Herbert, Lux & Frederickson, 1970; Östlund-Lindqvist & Iverius, 1975). Other investigators working with purified lipoprotein lipase have reported lipolytic activities in the absence of apo-CII (Hernell & Olivecrona, 1974; Kinnunen, Huttunen & Ehnholm, 1976). These differences of lipoprotein lipase activity might have several explanations. The use of different assay systems at various laboratories and the presence of impurities in the lipoprotein lipase preparations are two such possible explanations. However, in addition, different batches of serum albumin, used as the fatty acid acceptor in the lipoprotein lipase assay medium, have been reported to have an influence on the lipoprotein lipase activity (Boberg, 1969). The present investigation demonstrates that bovine serum albumin as Cohn fraction V (Sigma, St Louis, MO, U.S.A.) (Cohn, Strong, Hughes, Mulford, Ashworth, Melin & Taylor, 1946) used in the assay medium for lipoprotein lipase contains small amounts of apo-CII peptide.

Methods

Lipoprotein lipase was purified from cow's milk as described earlier (Iverius & Östlund-Lindqvist, 1976). Purified apo-CII (Brown, Levy & Frederickson, 1970) was injected into rabbits (Östlund-Lindqvist & Boberg, 1977). IgG antibodies were isolated (Levy & Sober, 1960) and coupled to cyanogen bromide-activated Sepharose 4B (Cuatrecasas, 1970). Bovine serum albumin (180 mg) was dissolved in 1 ml of Tris/HCl (0.17 mol/l)/NaCl (0.15 mol/l), pH 8.6, and passed through an anti-(apo-CII)-Sepharose column (1.5 cm x 1.5 cm) and washed with 100 ml of the buffer. The material adsorbed to the immunoabsorbent was eluted with glycine buffer (0.05 mol/l), pH 2.6, containing NaCl (0.15 mol/l). The eluted fractions (1 ml) were sampled directly into 0.15 ml of Tris/HCl (0.17 mol/l)/NaCl (0.15 mol/l), pH 8.6, and passed through an anti-(apo-CII)-Sepharose column (1.5 cm x 1.5 cm) and washed with 100 ml of the buffer. The material adsorbed to the immunoabsorbent was eluted with glycine buffer (0.05 mol/l), pH 2.6, containing NaCl (0.15 mol/l). The eluted fractions (1 ml) were sampled directly into 0.15 ml of Tris/HCl (0.17 mol/l)/NaCl (0.15 mol/l), pH 8.6, and the protein content was checked by measuring absorbance at 280 nm. The two fractions with the highest value (according to the absorbance at 280 nm) were combined and used for lipoprotein lipase-activating studies. The lipoprotein lipase assay medium was described earlier (Östlund-Lindqvist & Boberg,
Fig. 1. Effect of a protein fraction isolated from bovine serum albumin (Cohn fraction V) on lipoprotein lipase activity. Results of one representative experiment are shown. Lipoprotein lipase purified from cow's milk was incubated in assay medium containing the protein fraction isolated from bovine serum albumin in the absence (●) and in the presence (○) of antibodies against apolipoprotein-CII.

1977). Instead of serum as the activator, 0.1 ml of the two fractions from the anti-(apo-CII)– Sepharose were used.

Results and discussion

As demonstrated in Fig. 1 the protein fraction eluted from the anti-(apo-CII)– Sepharose activated lipoprotein lipase. The activation of lipoprotein lipase by this fraction is influenced by the triglyceride/apo-CII ratio in the same way as earlier described for apo-CII purified from human very-low-density lipoproteins (Östlund-Lindqvist & Iverius, 1975). In addition this activating property of the protein fraction was inhibited by antibodies raised against apo-CII. These results indicate that bovine serum albumin (Cohn fraction V) may contain apo-CII. Cohn et al. (1946) reported that 4% of the protein of fraction V has the same electrophoretic mobility as α1-globulins. Furthermore it is known that high-density lipoproteins, which contain apo-CII, also have α1-mobility. Consequently apo-CII seems to occur as high-density lipoproteins in the albumin fraction.

Thus some of the reports demonstrating lipoprotein lipase activity without addition of apo-CII might be explained by the presence of various amounts of apo-CII in the serum albumin used for the assay medium.

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References