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Determination of the respective density distributions of low- and high-density lipoprotein particles in bovine plasma and lymph by immunoassay of apoproteins A-I and B

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In our preliminary studies dealing with bovine plasma lipoproteins, we suggested that the spectrum of particles with $d > 1.026$ g/ml could be divided into LDL (d 1.026–1.060 g/ml), light HDL (HDL_L d 1.060–1.091 g/ml) and heavy HDL (HDL_H d 1.091–1.180 g/ml) (Bauchart *et al,* 1989). However, subfractions corresponding to the d 1.040–1.090 g/ml range contain 2 types of lipoproteins with characteristics of LDL and HDL particles, respectively. Indeed, their separation by affinity chromatography on a heparin–Sepharose column showed the presence of 2 forms of HDL containing apoprotein (apo) A-I and one form of LDL containing apo B with only slight traces apo A-I (Laplaud *et al,* 1989). Therefore, we directly estimated the respective density distributions of LDL and HDL particles in bovine plasma and lymph by radial immunodiffusion (RID) assays of apo A-I and apo B. A 3 wk old preruminant calf (46 kg, 600 g/d body weight gain) was fed a milk substitute (22% tallow and 23% protein). Plasma and lymph samples were obtained from chronic catheters inserted into the portal vein and the intestinal lymph duct. Lipoproteins of d 1.006–1.210 g/ml were separated by density gradient ultracentrifugation into 25 fractions. Their respective apo A-I and apo B contents were determined by RID using specific rabbit antisera to bovine apo A-I and apo B.

Gradient distributions of apo A-I (d 1.046–1.180 g/ml; max at d 1.080 g/ml) and apo B (d 1.020–1.091 g/ml; max at d 1.050 g/ml) were similar at the 2 sites. However, apo A-I and apo B concentrations were, respectively, 3- and 2-fold higher in the plasma than in the lymph. An overlap between LDL and HDL particles occurred in the 1.040–1.091 g/ml density range, as previously reported in the calf (Bauchart *et al,* 1989). Thus, apo B was detected in HDL_L fractions to an upper density limit of d 1.091 g/ml and apo A-I was detected in LDL fractions to a lower density limit of 1.040 g/ml. In the 1.040–1.091 g/ml density range, the concentration ratio HDL/LDL calculated from compositional analysis, amounted to 2.9 (plasma) and 6.8 (lymph), which was comparable to results obtained by affinity chromatography (Laplaud *et al,* 1989). Our immunological technique could therefore be useful in studies of hepatic metabolism in the preruminant calf or in the dairy cow in which the ratio HDL/LDL (and apoA-I/B) varies markedly with the hormonal state during the gestation–lactation cycle.

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