Reduced Concentrations of Apolipoproteins B-100 and A-I in Serum from Cows with Retained Placenta

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ABSTRACT

The purpose of the present study was to evaluate apolipoprotein B-100 and A-I concentrations in cows with retained placenta. Animals used were cows with retained placenta alone (n = 10), those with both retained placenta and ketosis (n = 7), and controls (n = 10). Apolipoprotein B-100 concentrations at 2 to 4 d after parturition were significantly ($P < 0.01$) decreased in cows with retained placenta alone (mean ± SD, 0.084 ± 0.029 mg/mL of serum) when compared with those in control cows (0.154 ± 0.022 mg/mL). Apolipoprotein A-I concentrations (0.713 ± 0.177 mg/mL) were also significantly ($P < 0.05$) lower than those of controls (0.895 ± 0.159 mg/mL). The decreases were more distinct for apolipoproteins B-100 (55% of controls) than A-I concentrations (80% of controls). Concentrations of apolipoprotein B-100 (0.071 ± 0.032 mg/mL; $P < 0.01$) and A-I (0.708 ± 0.189 mg/mL; $P < 0.05$) in the cows with both retained placenta and ketosis were also reduced, when compared with values in controls. Other than apolipoproteins, cows with retained placenta alone had significantly ($P < 0.01$) higher serum nonesterified fatty acids, and lower triglyceride concentrations. Significantly ($P < 0.01$) higher nonesterified fatty acids and lower triglyceride concentrations were similarly observed in cows with both retained placenta and ketosis.

RÉSUMÉ

Le but de cette étude était d'évaluer les concentrations d'apolipoproteines B-100 et A-1 chez des vaches ayant une retention placentaire. Les animaux utilisés comprenarient des vaches avec retention placentaire seule (n = 10), d'autres evec retention placentaire et rétose (n = 7) et d'autres servant de témoins (n = 10). Les concentrations d'apolipoproteines B-100 aux jours 2 à 4 suivants la parturition étaient significativement de minimisés ($P <$ 0,01) chez les vaches avec retention placentaire seule (moyenne ± ET, 0,084 ± 0,029 mg/mL de sérum) comparées avec les vaches du groupe témoin (0,154 ± 0,022 mg/mL). Les concentrations d'apolipoproteines A-1 (0,713 ± 0,177 mg/mL) étaient également plus basses que chez les animaux du groupe témoin (0,895 ± 0,159 mg/mL). Les diminutions étaient plus margués dans les concentrations d'apolipoproteins B-100 (55 % de celles des témoins) que pour celles des A-1 (80 %). Les concentrations d'apolipoproteins B-100 et A-1 chez les vaches avec à la fois rétention placentaire et rétose étaient également réduites, lorsque comparées aux vaches du groupe témoin (0,071 ± 0,032 mg/mL, $P <$ 0,01) et (0,708 ± 0,189 mg/mL, $P <$ 0,05). En plus des apolipoproteines, les vaches avec rétention placentaire seule montraient des concentrations sériques ($P <$ 0,01) plus élevées d'aides gros non-estérifiés et plus basses de triglycerides. Des concentrations sériques significativement plus élevées d'aide gros non-estérifiés et plus basses de triglycerides ($P <$ 0,01) étaient également observées chez les vaches avec à la fois rétention placentaire et rétose.

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Dairy cows during early lactation are at particularly higher risk for reproductive, metabolic and infectious diseases, compared with other stages (1). The higher incidence of postparturient disorders is thought to arise from excess feeding during the nonlactating stage, reduced feed intake, stress near parturition, and negative energy balance attributable to initiation of lactation (2–5). These may also become the major casual factors for development of hepatic lipidosis. Fatty liver has been associated with several postparturient disorders, including ketosis, left displaced abomasum (LDA), mastitis and metritis (2,3,5–8). Impaired hepatic function and hormonal imbalance attributable to fatty infiltration are suggested to result in increased incidences of these diseases. Retained placenta (RP, see Refs. 9 and 10 for review) is one of the fatty liver-associated diseases (2,3).

Apolipoprotein B-100 (apoB-100) and apolipoprotein A-I (apoA-I) are the 2 major apoproteins in bovine lipoprotein fractions. The apoB-100 is distributed in very low-density and low-density lipoproteins, and is responsible for export of triglycerides (TG) from the liver to extrahepatic tissues. The apoA-I in high-density lipoprotein fraction has a role for the transport of cholesterol and phospholipids (PL). In cows with fatty liver, serum concentrations of apoB-100 and apoA-I are decreased (11–13). In association with decreased apoB-100 and apoA-I concentrations, cows with fatty liver show reduced levels of serum lipids (12,14,15).

We have recently found that apoB-100 and apoA-I concentrations were reduced in ketosis and LDA (16). Decreases in serum concentrations of TG, cholesterol and PL and an increase in the concentration of nonesterified fatty acids (NEFA) were also observed in the 2 fatty liver-related disorders. It is therefore conceivable that the

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decreases of both apolipoprotein concentrations, together with simultaneous changes of serum lipid concentrations, are associated with fatty liver-related disorders. The present study was undertaken to determine whether apoB-100 and apoA-I concentrations are reduced in cows with RP.

Animals used in this study (n = 27) were 2- to 6-year old lactating Holstein cows from 6 farms of Iwate Prefecture. These 6 farms had similar management styles. Cow's diet was as described previously (17), and their milk yields were approximately 8000 kg/year/cow. Of the cows, 17 cows retained the placenta from 24 to 72 h after delivery, and were designated RP cows. The remaining 10 age-matched cows expelled the placenta within 6 h after calving, and were used as controls. Cows with both RP and metritis were not included in this study. During pregnancy, all cows in both groups were apparently healthy. Twin calvings and stillbirth were not found. Serum levels of ß-carotene, vitamin A, vitamin E and selenium were not different in the 2 groups. Serum tests also revealed that all cows in both groups were free from brucellosis and tuberculosis. Of the 17 cows with RP, 7 had ketosis as detected initially by clinical signs and then by a positive urinary ketone reaction. Urinary ketone bodies were not detected in cows with RP alone and in control cows. Blood samples of RP and control cows were collected in the morning (before feeding) 2 to 4 d after parturition. Cows with RP, including ketotic cows, were not treated prior to sampling.

Serum concentrations of apoA-I (17) and apoB-100 (18) were evaluated by enzyme-linked immunosorbent assays. The serum concentration of ß-hydroxybutyrate (BHB) was determined using a commercial kit (Sanwa Kagaku Co, Nagoya, Japan). Urinary ketone bodies were detected by a kit (Fujisawa Pharmaceutical Co, Osaka, Japan). Serum concentrations of TG, total cholesterol (TC), PL and NEFA were measured using an automatic analyzer with reagent kits (Wako Pure Chemicals, Osaka, Japan). Each assay was done in duplicate. Data were analyzed, using one-way ANOVA and Student's or Welch's t-test. Values are expressed as mean ± SD.

TABLE I. Concentrations of serum lipids of controls (C), cows with retained placenta alone (RP) and cows with both retained placenta and ketosis (RP + K)

Cow	n	NEFA (mEq/L)	BHB (mM)	TG (mg/dL)	TC (mg/dL)	PL (mg/dL)
C	10	0.381 ± 0.132	0.855 ± 0.205	7.30 ± 1.90	90.2 ± 22.6	115 ± 23.5
RP	10	1.007 ± 0.366[b]	0.829 ± 0.136	4.50 ± 2.06[b]	74.3 ± 24.9	92.2 ± 26.0
RP + K	7	1.636 ± 0.500[b]	2.766 ± 0.985[b]	3.71 ± 2.25[b]	74.0 ± 42.3	91.4 ± 35.2

NEFA = nonesterified fatty acids; BHB = ß-hydroxybutyrate; TG = triglycerides; TC = total cholesterol; PL = phospholipids

[a] $P < 0.05$

[b] $P < 0.01$, compared with the respective values of controls

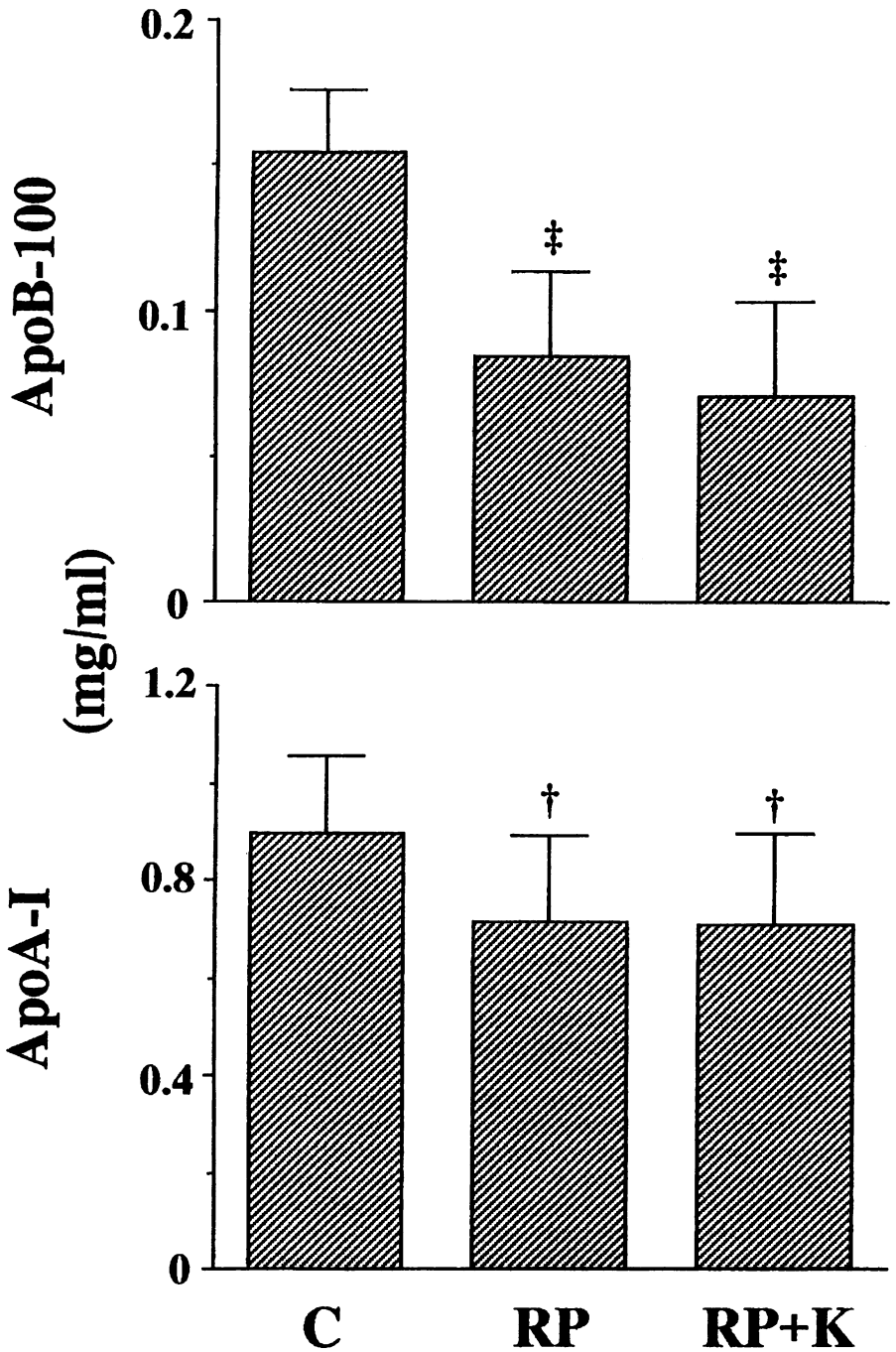

Figure 1. Apolipoprotein B-100 (apoB-100) and A-I (apoA-I) concentrations in sera from controls (C, n = 10), cows with retained placenta alone (RP, n = 10) and with both retained placenta and ketosis (RP + K, n = 7). †$P < 0.05$; ‡$P < 0.01$, when compared to the values of controls.

When compared with values in controls, significantly ($P < 0.01$) higher NEFA and lower TG concentrations were observed in cows with RP (Table I). Non-significant decreases in TC and PL concentrations were also observed. Besides the increase of BHB concentrations, ketotic RP cows showed more distinct increase of concentration of NEFA, when compared with that of nonketotic RP cows.

The control cows (0.154 ± 0.022 mg/mL) showed lower apoB-100 concentrations than cows during mid-lactation on the same farms (0.224 ± 0.028 mg/mL; $n = 22$). Compared with the value of the control cows, apoB-100 concentrations were found to be further reduced in RP cows (Fig. 1). Although not so distinct as for apoB-100, apoA-I concentration was significantly ($P < 0.05$) decreased in RP cows. Relative concentrations of apoB-100 and apo A-I, compared with those of control cows were 55% and 80%, respectively. As with the other serum analytes, ketotic RP cows showed a greater decrease of the apoB-100 concentration (46% of the control), although the difference between ketotic and nonketotic RP cows was not significant. Complication of RP with ketosis did not affect the apoA-I concentration (79% of the control).

Of the 2 apolipoproteins, the apoB-100 concentration was more distinctly reduced in cows with RP. The preferential decrease of the apoB-100 concentration was also observed in cows with fatty liver (11,12). The mode of the apolipoprotein concentration changes in cows with RP appears to be essentially identical to that of cows with fatty liver. The similarity between RP and fatty liver was also found in serum lipid concentration changes. In particular, the increased NEFA concentration observed in cows with RP is also relevant for the development of fatty liver (2,6,7). The results of concentration changes of lipids, together with those of apolipoproteins, strongly support the previous clinico-pathological findings that RP is associated with fatty liver (2,3).

Other than RP and fatty liver, apoB-100 and apoA-I concentrations are decreased in cows with ketosis and LDA (16). The association of RP with ketosis has been reported (19,20). In the present study, 7 of 17 cows with RP also suffered from ketosis. Ketosis and LDA have been shown to be associated with fatty liver (4,5,14,21,22). The serum NEFA concentrations increase in cows with ketosis and LDA (23). The decreased apoB-100 and apoA-I concentrations in cows with RP further suggest that the development of RP is closely linked to fatty liver, ketosis and LDA. Because fatty liver occurs in the prepartum period (7), RP as well as ketosis and LDA appear to be induced after development of fatty liver. A longitudinal study of measurements of serum apoB-100, apoA-I and liver TG concentrations prior to parturition and during the puerperium could provide a direct evidence for the association between the 4 conditions.

REFERENCES

1. **ERB HN, SMITH RD, HILLMAN RB, POWERS PA, SMITH MC, WHITE ME, PEARSON EG.** Rates of diagnosis of six diseases of Holstein cows during 15-day and 21-day intervals. Am J Vet Res 1984; 45: 333–335.
2. **MORROW DA.** Fat cow syndrome. J Dairy Sci 1976; 59: 1625–1629.
3. **MORROW DA, HILLMAN D, DADE AW, KITCHEN H.** Clinical investigation of a dairy herd with the fat cow syndrome. J Am Vet Med Assoc 1979; 174: 161–167.
4. **ERB HN, GROHN YT.** Epidemiology of metabolic disorders in the periparturient dairy cow. J Dairy Sci 1988; 71: 2557–2571.
5. **GRUMMER RR.** Etiology of lipid-related metabolic disorders in periparturient dairy cows. J Dairy Sci 1993; 76: 3882–3896.
6. **REID IM, ROBERTS CJ, MANSTON R.** Fatty liver and infertility in high-yielding dairy cows. Vet Rec 1979; 104: 75–76.
7. **GEROLOFF BJ, HERDT TH, EMERY RS.** Relationship of hepatic lipidosis to health and performance in dairy cattle. J Am Vet Med Assoc 1986; 188: 845–850.
8. **HERDT TH.** Fatty liver in dairy cows. Vet Clin North Am 1988; 4: 269–287.
9. **WETHERILL GD.** Retained placenta in the bovine. A brief review. Can Vet J 1965; 6: 290–294.
10. **STEVENSON JS, CALL EP.** Reproductive disorders in the periparturient dairy cows. J Dairy Sci 1988; 71: 2572–2583.
11. **MARCOS E, MAZUR A, CARDOT P, RAYSSIGUIER Y.** Serum apolipoproteins B and A-I and naturally occurring fatty liver in dairy cows. Lipids 1990; 25: 575–577.
12. **UCHIDA E, KATOH N, TAKAHASHI K.** Induction of fatty liver in cows by ethionine administration and concomitant decreases of serum apolipoproteins B-100 and A-I concentrations. Am J Vet Res 1992; 53: 2035–2042.
13. **KATOH N, MINOURA S, UCHIDA E, TAKAHASHI K.** Effect of estradiol administration and subsequent nonfeeding on liver estrogen receptor, serum apolipoprotein B-100, and serum triglycerides concentrations in steers. Am J Vet Res 1993; 54: 1476–1482.
14. **HERDT TH, LIESMAN JS, GERLOFF BJ, EMERY RS.** Reduction of serum triacylglycerol-rich lipoprotein concentrations in cows with hepatic lipidosis. Am J Vet Res 1983; 44: 293–296.
15. **RAYSSIGUIER Y, MAZUR A, GUEUX E, REID IM, ROBERTS CJ.** Plasma lipoproteins and fatty liver in dairy cows. Res Vet Sci 1988; 45: 389–393.
16. **OIKAWA S, KATOH N, KAWAWA F, ONO Y.** Decreased serum apolipoprotein B-100 and A-I concentrations in cows with ketosis and left displacement of the abomasum. Am J Vet Res 1997; 58: 121–125.
17. **OIKAWA S, KATOH N, ITOH H, MIYAMOTO T, KONNO M, KAJITA T.** Decreased serum apolipoprotein A-I concentrations in cows infected with *Salmonella* Typhimurium. Can J Vet Res 1997; 61: 182–186.
18. **YAMAMOTO O, OIKAWA S, KATOH N.** Enzyme-linked immunosorbent assay for serum apolipoprotein B-100, a major triglyceride-transport protein in dairy cows. Am J Vet Res 1995; 56: 1413–1417.
19. **MARKUSFELD O.** Factors responsible for post parturient metritis in dairy cattle. Vet Rec 1984; 114: 539–542.
20. **MARKUSFELD O.** Periparturient traits in seven high dairy herds. Incidence rates, association with parity, and interrelationship among traits. J Dairy Sci 1987; 70: 158–166.
21. **GROHN Y, LINDBERG LA, BRUSS ML, FARVER TB.** Fatty infiltration of liver in spontaneously ketotic dairy cows. J Dairy Sci 1983; 66: 2320–2328.
22. **HOLTENIUS P.** Hormonal regulation related to the development of fatty liver and ketosis. Acta Vet Scand 1993; Suppl89: 55–60.
23. **KRONFELD DS.** Plasma non-esterified fatty acid concentrations in the dairy cow: Responses to nutritional and hormonal stimuli, and significance in ketosis. Vet Rec 1965; 77: 30–34.