Tissue-specific changes in the ability of insulin and noradrenaline to activate pyruvate dehydrogenase *in vivo* during lactation in the rat

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1. Changes are described in the total pyruvate dehydrogenase (PDH) activity, the proportion of PDH in the active state and its control by insulin and noradrenaline *in vivo*, in white adipose tissue, liver, skeletal muscle and mammary gland with pregnancy, lactation and on weaning. 2. Lactation resulted in a decrease in total PDH in white adipose tissue and an increase in the mammary gland, whereas the proportion in the active state decreased in muscle and increased in the mammary gland. 3. The ability of insulin to activate PDH of white adipose tissue was lost during lactation, whereas it was retained by the other tissues. The ability of noradrenaline to activate PDH was decreased in white adipose tissue but increased in liver during lactation. 4. These various adaptations should limit the use of glucose and lactate carbon by adipose tissue and skeletal muscle during lactation and thereby facilitate their preferential utilization by the mammary gland.

INTRODUCTION

Pregnancy and lactation result in a number of metabolic adaptations which facilitate the supply of nutrients to the foetus and then to the mammary gland to support milk production (see Williamson, 1980; Bauman & Elliot, 1983; Zammit, 1985). For example, the rate of fatty acid synthesis is decreased in white and brown adipose tissue during lactation, but is increased in the mammary gland and liver (see Vernon & Flint, 1983). In addition there is usually some use of body reserves, especially adipose-tissue lipid, during lactation, which are replenished on weaning (see Vernon & Flint, 1983, 1984).

The mechanisms underlying these tissue-specific adaptations during pregnancy and lactation are not fully elucidated. Changes in serum insulin concentration are implicated, for it increases during pregnancy and then decreases around parturition and during lactation (see Vernon & Flint, 1983). In addition, insulin resistance, as evinced by the increased serum insulin/glucose ratio and the decreased ability of insulin to increase glucose turnover *in vivo*, occurs during late pregnancy (Knopp *et al.*, 1970; Leturque *et al.*, 1980, 1984), whereas the ability of insulin to stimulate the rate of lipogenesis in white and brown adipose tissue *in vivo* is muted during lactation (Burnol *et al.*, 1983). Furthermore, a subsequent study *in vitro* (Burnol *et al.*, 1986) showed that, whereas the stimulation by insulin of glucose conversion into lactate and acylglycerol glycerol in white adipocytes was unchanged by lactation, the stimulation of glucose oxidation to CO₂ and conversion into fatty acids was decreased, suggesting that activation of PDH by insulin was impaired.

In view of its important role in the metabolism of glucose to acetyl-CoA for subsequent oxidation or biosynthesis, we have investigated the effects of pregnancy and lactation on PDH activity of white adipose

tissue, liver, skeletal muscle and mammary gland and its control by insulin and noradrenaline. We show that the ability of insulin to activate PDH of white adipose tissue, but not of the other three tissues, is indeed completely lost during lactation. Furthermore, stimulation by noradrenaline of PDH of white adipose tissue, but not liver, is also muted during lactation.

EXPERIMENTAL

Wistar rats (A. Tuck and Son, Rayleigh, Essex, U.K.) were fed on Labsure irradiated CRM diet, comprising 570 g of carbohydrate, 180 g of crude protein, 24 g of crude oil and 36 g of crude fibre per kg (Labsure, Poole, Dorset, U.K.) and water *ad libitum*. They were mated from about 10 weeks of age, and the number of pups per mother was adjusted to eight within 24 h after birth. Rats were used at various days of pregnancy and lactation, or at 2 days after litter removal following 14 days lactation; all rats, including virgin rats, were 13–16 weeks old at time of use. Rats were maintained on a 08:00–20:00 h light/dark cycle, except for one group of virgin and lactating rats which was on a reversed light/dark cycle, and were handled regularly before the experiment.

In the first experiment (results in Fig. 1) rats were anaesthetized by injection intraperitoneally with pentobarbital (60 mg/kg body wt.) with 23-gauge needles at about 10:00 h; 20 min later, samples of white (parametrial) adipose tissue and liver were removed and frozen immediately in liquid N₂ for assay of PDH. In subsequent experiments, rats were again anaesthetized at about 12:00 h as described above, except that 26-gauge needles were used and propranolol (1.5 mg/kg) was injected along with the anaesthetic (to minimize further any effects of stress), except when the rats were subsequently injected with noradrenaline. Then 10 min later rats were injected intraperitoneally with

0.15 M-NaCl, glucose (1 g/kg body wt.) either alone or with insulin (0.25 mg/rat), or noradrenaline (1.25 mg/kg body wt.), and after a further 20 min samples of white (parametrial) adipose tissue, liver, mammary gland and skeletal muscle (gastrocnemius and biceps femoris, from the hind-limb) were removed and immediately frozen in liquid N₂ for assay of PDH. Samples of blood were also taken from the heart, while additional samples of parametrial adipose tissue were taken and stored in iso-osmotic saline at 37 °C for determination of cellularity. Lactating rats were injected with a second, identical, dose of glucose 10 min after the first.

Frozen tissue samples were prepared and assayed for total PDH activity and the proportion in the active state as described by Stansbie *et al.* (1976a), except that the pyruvate and Mg²⁺ concentrations in the assay medium were increased to 3 mM and 2 mM respectively and 6 mM-pyruvate was added during the activation step. The dye *p*-(*p*-aminophenylazo)benzenesulphonic acid used in the assay was given by Dr. R. M. Denton, Department of Biochemistry, University of Bristol.

The protein content of tissue samples was determined by the method of Lowry *et al.* (1951), with bovine serum albumin as standard. The cellularity of parametrial adipose tissue was determined as described by Aitchison *et al.* (1982). Serum was prepared and stored at -20 °C before insulin assay as described by Flint *et al.* (1979) and glucose assay with an Anolox GM6 glucose analyser.

Results are expressed as means $\pm$ S.E.M. and were analysed by Student's *t* test.

RESULTS AND DISCUSSION

Changes in PDH activity during pregnancy and lactation and on weaning

Preliminary experiments showed that over 85% of the PDH of white adipose tissue was associated with adipocytes themselves; results for adipose tissue are therefore expressed per 10⁶ adipocytes, as adipocyte volume and hence number/g of tissue varies considerably during the reproductive cycle (Flint *et al.*, 1979). Total PDH activity of white adipose tissue did not change during pregnancy, but then declined during lactation, so that by 14 days of lactation the value was lower ($P < 0.02$) than that of virgin rats (Fig. 1a). The PDH_a activity of white adipose tissue mostly changed in parallel with that of total PDH (results not shown), so that there was no change in the proportion in the active state except at 7 days of pregnancy, when there was a significant increase ($P < 0.01$; Fig. 1a). A transient rise in the rate of fatty acid synthesis occurs in white adipose tissue at this stage of pregnancy (Flint *et al.*, 1983). However, the fall in the rate of fatty acid synthesis of white adipose tissue, which occurs around parturition, is much more marked and clearly precedes any change in PDH_a activity (see Vernon & Flint, 1983). In contrast with the present study, Sinnott-Smith *et al.* (1980) observed a fall in PDH_a around parturition, but in their study rats were killed by cervical dislocation; hence differences may be due to a decreased response to noradrenaline (released in response to the stress caused by killing) during lactation (see below). A fall in total PDH during lactation was confirmed in a second experiment (Table 1), which also showed that the total PDH activity had recovered ($P < 0.001$) by 2 days after

Fig. 1. Effect of pregnancy and lactation on the total pyruvate dehydrogenase (■) and the proportion of the enzyme in the active state (□) of (a) white adipose tissue and (b) liver

Rats were anaesthetized with pentobarbital at about 10:00 h, and parametrial white adipose tissue and liver were removed 20 min later. The total PDH and the proportion of the enzyme in the active state were assayed as described in the text: results are means $\pm$ S.E.M. for three to seven observations. Abbreviation: T, term.

litter removal. Weaning also induced a considerable rise ($P < 0.001$, compared with values for virgin and lactating rats) in the proportion of PDH_a (Table 2).

Hepatic total PDH did not change during pregnancy and lactation, but the proportion in the active state decreased ($P < 0.01$) around parturition and then increased ($P < 0.01$) during lactation (Fig. 1b). There is also a transient fall in the rate of hepatic fatty acid synthesis (Smith, 1973; Agius & Williamson, 1980) and in the acetyl-CoA carboxylase (Zammit & Corstorphine, 1982) activity of the liver around parturition, but then both increase during lactation to exceed the values found in virgin animals. Weaning had no effect on either the total PDH of liver or the proportion as PDH_a (Tables 1 and 2).

Total PDH activity of skeletal muscle was decreased ($P < 0.001$) by day 18 of pregnancy, but then returned to virgin values by day 14 of lactation; there was also a small but significant fall in total PDH activity on weaning ($P < 0.001$, compared with the value for lactating rats) (Table 1). The proportion in the active state, however, decreased ($P < 0.05$) by day 18 of pregnancy and further ($P < 0.01$) by day 14 of lactation; 2 days after weaning, however, the proportion in the active state had increased ($P < 0.001$) to $24.8 \pm 3.6\%$.

Table 1. Total pyruvate dehydrogenase activity of white adipose tissue, skeletal muscle, liver and mammary gland from rats at different stages of the reproductive cycle

Tissues were obtained from rats given the various treatments described in Table 2; as the various treatments had no effect on total PDH activity, values were pooled. Results are means $\pm$ S.E.M. with the numbers of observations in parentheses, and are expressed per 10^6 cells for white adipose tissue or per mg of protein for other tissues; *, **, ***, value significantly different ($P < 0.05$, $P < 0.01$, $P < 0.001$ respectively) from that for virgin rats.

Tissue	Rats . . .	Pyruvate dehydrogenase (nmol/min)			
		Virgin	Pregnant (18 days)	Lactating (14 days)	Weaned (2 days)
White adipose tissue		44.0 $\pm$ 3.7 (21)	49.3 $\pm$ 10.5 (8)	23.3 $\pm$ 2.1 (23)***	61.4 $\pm$ 7.6 (10)*
Skeletal muscle		21.0 $\pm$ 0.3 (16)	15.2 $\pm$ 0.4 (7)***	22.7 $\pm$ 0.8 (10)	18.2 $\pm$ 1.2 (10)*
Liver		9.0 $\pm$ 0.4 (16)	—	8.4 $\pm$ 0.4 (18)	9.0 $\pm$ 0.1 (6)
Mammary gland		—	4.9 $\pm$ 0.5 (8)	8.4 $\pm$ 0.3 (23)	4.0 $\pm$ 0.5 (9)

Table 2. Effects of injecting glucose, glucose plus insulin or noradrenaline on the pyruvate dehydrogenase activity of white adipose tissue, liver, skeletal muscle and mammary gland in rats at different stages of the reproductive cycle

Rats were anaesthetized through the experiment; rats receiving saline (controls), glucose or glucose plus insulin were also injected with the β -antagonist propranolol along with the anaesthetic; other agents were injected 10 min later, and tissues and blood were sampled after a further 20 min. Further details are given in the text. Results are means $\pm$ S.E.M., with the numbers of observations in parentheses; *, **, ***, value significantly different ($P < 0.05$, $P < 0.01$, $P < 0.001$ respectively) from that for rats injected with saline.

Tissue	State of rats	Injection . . .	Pyruvate dehydrogenase (% in active state)			
			Saline	Glucose	Glucose + insulin	Noradrenaline
White adipose tissue	Virgin		21.0 $\pm$ 3.1 (5)	61.9 $\pm$ 8.0 (5)***	71.2 $\pm$ 1.4 (4)***	77.5 $\pm$ 6.3 (8)***
	Pregnant (18 days)		29.9 $\pm$ 5.8 (4)	57.9 $\pm$ 5.9 (4)*	—	—
	Lactating (14 days)		27.2 $\pm$ 3.7 (6)	26.7 $\pm$ 3.4 (6)	28.5 $\pm$ 2.4 (4)	44.7 $\pm$ 5.8 (7)*
	Weaned (2 days)		58.7 $\pm$ 5.7 (5)	53.8 $\pm$ 4.8 (5)	—	—
Liver	Virgin		27.4 $\pm$ 1.6 (5)	30.9 $\pm$ 2.3 (5)	36.6 $\pm$ 3.3 (3)*	57.8 $\pm$ 3.0 (3)***
	Lactating (14 days)		32.8 $\pm$ 1.4 (4)	35.0 $\pm$ 1.0 (4)	40.5 $\pm$ 1.5 (4)*	76.3 $\pm$ 5.4 (6)***
	Weaned (2 days)		32.2 $\pm$ 1.8 (3)	30.1 $\pm$ 1.2 (3)	—	—
Skeletal muscle	Virgin		10.0 $\pm$ 0.1 (4)	27.0 $\pm$ 0.9 (4)***	70.1 $\pm$ 3.3 (3)***	10.2 $\pm$ 0.1 (5)
	Pregnant (18 days)		7.1 $\pm$ 0.5 (4)	17.1 $\pm$ 0.3 (3)***	—	—
	Lactating (14 days)		3.4 $\pm$ 0.5 (3)	8.4 $\pm$ 0.4 (4)***	60.8 $\pm$ 6.4 (3)***	—
	Weaned (2 days)		24.8 $\pm$ 3.6 (5)	22.8 $\pm$ 3.2 (5)	—	—
Mammary gland	Pregnant (18 days)		8.6 $\pm$ 0.3 (4)	21.4 $\pm$ 3.9 (4)**	—	—
	Lactating (14 days)		55.4 $\pm$ 2.1 (7)	66.7 $\pm$ 3.7 (5)**	71.6 $\pm$ 4.0 (4)**	74.4 $\pm$ 2.6 (7)***
	Weaned (2 days)		13.4 $\pm$ 3.0 (4)	30.6 $\pm$ 9.9 (5)	—	—

(Table 2), a value significantly ($P < 0.01$) greater than that of virgin rats.

In contrast with the other tissues studied, total PDH of the mammary gland (Table 1) and the proportion in the active state (Table 2) showed a substantial ($P < 0.001$) increase between day 18 of pregnancy and day 14 of lactation; both then fell on weaning ($P < 0.01$) (Tables 1 and 2). These observations are in agreement with previous reports (Coore & Field, 1974; Kankel & Reinauer, 1976).

Response to insulin

Serum insulin concentration was lower ($P < 0.01$) in lactating than in non-lactating rats, and then increased

($P < 0.001$) on weaning (Table 3), in agreement with previous studies (Robinson *et al.*, 1978; Agius *et al.*, 1979; Flint *et al.*, 1979, 1981; Burnol *et al.*, 1983).

The response of PDH to insulin was assessed by injecting a large dose of insulin plus glucose to avoid concomitant hypoglycaemia (Table 3), and secondly by injecting glucose alone to induce the release of endogenous insulin and so expose the tissue to a more physiological surge of the hormone. The amount of glucose injected was the same as that used by Stansbie *et al.* (1976b) and by Burnol *et al.* (1983). Intraperitoneal injection of glucose was used, as this leads to an increase in serum insulin, which appears to remain constant between 10 and 30 min after injection, whereas intra-

Table 3. Serum insulin and glucose concentrations in rats at different stages of the reproductive cycle after the injection of glucose, glucose plus insulin, or noradrenaline

Animals were anaesthetized throughout the experiment. Agents were injected 10 min after the anaesthetic, and blood was obtained 20 min later. Animals were the same as those used in Tables 1 and 2. Results are means $\pm$ S.E.M. with the numbers of observations in parentheses; *, **, *** indicate value is significantly different ($P < 0.05$, < 0.01 , < 0.001 respectively) from that for control rats injected with saline.

Serum component	Agent injected	State of rats			
		Virgin	Pregnant (18 days)	Lactating (14 days)	Weaned (2 days)
Insulin (ng/ml)	Saline	2.9 $\pm$ 0.6 (5)	3.3 $\pm$ 0.5 (4)	0.6 $\pm$ 0.1 (6)	3.6 $\pm$ 0.4 (4)
	Glucose	4.6 $\pm$ 0.9 (5)	10.3 $\pm$ 1.6 (4)**	1.6 $\pm$ 0.3 (6)**	5.9 $\pm$ 1.0 (5)
	Glucose plus insulin	26.6 $\pm$ 1.1 (3)***	—	24.7 $\pm$ 2.4 (4)***	—
	Noradrenaline	3.5 $\pm$ 0.6 (8)	—	2.4 $\pm$ 0.4 (7)**	—
Glucose (mm)	Saline	8.7 $\pm$ 0.8 (5)	6.6 $\pm$ 0.7 (4)	7.0 $\pm$ 0.6 (6)	7.9 $\pm$ 0.4 (4)
	Glucose	16.8 $\pm$ 2.2 (5)**	16.4 $\pm$ 2.4 (4)**	13.9 $\pm$ 3.1 (6)	13.3 $\pm$ 2.9 (5)
	Glucose plus insulin	10.5 $\pm$ 3.6 (3)	—	8.5 $\pm$ 1.6 (4)	—
	Noradrenaline	14.8 $\pm$ 1.1 (8)**	—	13.3 $\pm$ 1.6 (7)**	—

venous injection leads to a much more rapid (and larger) rise and then fall in serum insulin (Bonner-Weir *et al.*, 1983). Both serum insulin and glucose were still elevated, in general, 20 min after the glucose load (Table 3). The relatively high insulin concentration in the pregnant rats after the glucose load is in agreement with the findings of Leturque *et al.* (1980) and is probably due to the greater responsiveness of the pancreas to insulinotropic agents during pregnancy (Malaisse *et al.*, 1969; Kalkhoff & Kim, 1978). Burnol *et al.* (1983) found that serum insulin concentration declined much more rapidly after a glucose load in lactating than in non-lactating rats, probably owing to the metabolic clearance rate of glucose of lactating rats being about twice that of non-lactating rats. Hence, to minimize this, lactating rats received a second injection of glucose 10 min after the first. The time interval between injection and sampling (20 min) was based on the study of Stansbie *et al.* (1976b), which showed that the increase in PDH_a of white adipose tissue induced by an intravenous glucose load did not vary between 10 and 30 min after injection.

Injection of glucose, glucose plus insulin (or noradrenaline) had no effect on the total PDH of the four tissues studied in any of the physiological states examined; pooled mean values for each state are given in Table 1.

Injection of glucose increased ($P < 0.001$) the proportion of PDH_a of white adipose tissue of virgin and pregnant rats by 2–3-fold (Table 2); a similar effect was reported for male rats (Stansbie *et al.*, 1976b). In contrast, injection of glucose had no effect on the proportion of PDH_a in adipose tissue from lactating or weaned rats (Table 2). The lack of effect in lactating rats was not due to the lower insulin concentration, as injection of insulin plus glucose, which increased serum insulin to over 24 ng/ml (Table 3) also had no effect on the proportion of PDH_a, showing that the tissue had become completely refractory to insulin. Insulin also failed to increase the rate of lipogenesis in white adipose in lactating rats (Burnol *et al.*, 1983; Jones *et al.*, 1984).

For weaned rats, the proportion of PDH_a in rats given saline was similar to that of virgin rats after the administration of glucose, suggesting that the enzyme was already activated to the maximum extent possible in

response to insulin [administration of insulin plus glucose to virgin rats, which increased serum insulin to over 25 ng/ml (Table 3) did not have any greater effect on PDH_a than giving glucose alone (Table 2)]. Burnol *et al.* (1983) found that the rate of lipogenesis in white adipose tissue from weaned rats did not respond to insulin either, and again the value in saline-treated animals was similar to that of non-lactating rats exposed to insulin. Burnol *et al.* (1983) also showed that glucose had a greater insulinotropic effect in weaned than in non-lactating rats, so the lack of effect is not due to a lower serum insulin concentration.

In contrast with white adipose tissue, insulin activated PDH in the other three tissues examined from lactating rats. For skeletal muscle, injection of glucose provoked a 2–3-fold increase in PDH_a in virgin, pregnant and lactating rats, although this treatment again had no effect on the PDH_a of weaned rats (Table 2). The proportion of PDH_a was similar in both virgin and lactating rats after injection of insulin plus glucose (Table 2). For virgin and lactating rats the proportion of PDH_a in skeletal muscle appears to reflect the serum insulin concentration, but during pregnancy the effects of insulin appeared to be muted, for the proportion of PDH_a in both saline- and glucose-injected rats was lower ($P < 0.01$ and $P < 0.001$ respectively) than in virgin rats, despite similar or higher serum insulin concentrations in the pregnant animals. The ability of insulin to stimulate glucose utilization in the perfused rat hind-limb is also decreased during late pregnancy (Rushakoff & Kalkhoff, 1981); a diminished effect of insulin on muscle PDH would contribute to this impairment and hence to the insulin-resistance of late pregnancy referred to above.

Administration of glucose alone had no effect on hepatic PDH_a of either virgin (as noted by Stansbie *et al.*, 1976b) or lactating rats. Administration of insulin plus glucose, however, increased hepatic PDH_a in animals from both states to a similar extent (Table 2). The study provides further evidence that, although insulin can modulate hepatic PDH_a activity, the concentration required is substantially higher than that required for other tissues.

Glucose or glucose plus insulin both increased

($P < 0.01$) the proportion of mammary PDH_a to a similar extent in lactating rats (Table 2). Baxter & Coore (1978) found a similar effect with 24 h-starved lactating rats, in which the proportion of PDH_a was markedly decreased compared with that of fed lactating rats.

Response to noradrenaline

Hepatic PDH is activated by catecholamines (Assimakopoulou-Jeannet *et al.*, 1983) as well as by insulin; indeed catecholamines may be more important in the control of this enzyme in the liver than insulin, in view of the relative insensitivity of hepatic PDH to insulin. Catecholamines also activated PDH in white adipose tissue *in vivo* (Kilgour & Vernon, 1986) and *in vitro* (Smith & Saggerson, 1978; Cheng & Lerner, 1985).

Preparatory studies with virgin rats showed that effects of noradrenaline on white-adipose-tissue PDH_a were maximal between 15 and 25 min after injection. Injection of noradrenaline into virgin rats increased PDH_a in white adipose tissue ($P < 0.001$) and liver ($P < 0.001$), but had no effect on skeletal muscle (Table 2). Injection of noradrenaline into lactating rats also increased PDH_a of both tissue ($P < 0.05$ and $P < 0.001$ for white adipose tissue and liver respectively; Table 2), but the increase in white adipose tissue was less ($P < 0.05$) than that found in virgin rats, whereas in the liver the effect was greater ($P < 0.05$) than in virgin rats. Injection of noradrenaline also increased ($P < 0.001$) the proportion of PDH_a in the mammary gland (Table 2). Rat mammary epithelial cells possess β -adrenergic receptors (Clegg & Mullaney, 1985), but this effect of noradrenaline may be indirect, as there was some increase in serum insulin (Table 3).

Noradrenaline is released from nerve endings within adipose tissue. Major stress, such as cervical dislocation of conscious animals, causes a massive release of noradrenaline (see Callingham & Barrand, 1979) and also increased ($P < 0.001$) PDH_a in white adipose tissue from virgin rats to $81.4 \pm 1.2\%$ of total activity (mean $\pm$ S.E.M. for three observations), a similar value to that found on intraperitoneal injection of noradrenaline (Table 2). Cervical dislocation activated the enzyme in white adipose tissue from lactating rats to $47.8 \pm 4.9\%$ of total PDH (mean $\pm$ S.E.M. for three observations); this increase was significant ($P < 0.05$), but was significantly less ($P < 0.01$) than that found with virgin rats. The response to cervical dislocation in virgin and lactating rats thus close parallels the response to injection of noradrenaline.

Rats are most active and feed at night, which is when sympathetic activity and serum insulin are thought to be increased (see Le Magnen, 1984). In virgin rats the proportion of PDH in the active state was $40.5 \pm 3.1\%$ and $30.6 \pm 0.8\%$ at 4 h into the dark and light phase respectively (results are means of four and six observations respectively, and differ significantly; $P < 0.05$). In contrast, in lactating rats, the proportion of PDH in the active state was $26.1 \pm 2.5\%$ and $30.9 \pm 0.5\%$ at 4 h into the dark and light phases respectively (results are means of five and four values and are not significantly different). The proportion of PDH_a at 4 h into the dark phase in lactating rats was also lower than that of virgin rats ($P < 0.01$).

General discussion

The study shows that both the activity of PDH and its endocrine control exhibit tissue-specific adaptations during the reproductive cycle. One outcome is a

diminished capacity of adipose tissue and skeletal muscle to convert glucose into acetyl-CoA for oxidation or biosynthesis during lactation. Lower PDH activity would not necessarily diminish the rate of glucose utilization of the tissue, but if not, it should increase the proportion of glucose released as lactate. Burnol *et al.* (1986) found no evidence of increased lactate output *in vivo* by adipocytes from lactating rats, but lactate output was relatively high in adipocytes from virgin rats in their study. In sheep, in which PDH is also decreased in muscle and adipose tissue during lactation (Vernon *et al.*, 1984), an increased proportion of glucose utilized is released as lactate by the hind-limb *in vivo* (Pethick & Lindsay, 1982) and by adipose tissue *in vitro* (R. G. Vernon, E. Finley & E. Taylor, unpublished work) during lactation. In sheep this lactate is probably used for gluconeogenesis, which is increased during lactation (Bergman *et al.*, 1974); this may also be its fate in lactating rats, particularly during re-feeding after a period of food deprivation (Williamson *et al.*, 1985).

Curiously, the diminished PDH activity in both lactating rats and sheep (Vernon *et al.*, 1984) is due to a fall in total PDH in adipose tissue but a decreased in the proportion in the active state in skeletal muscle. Further differences are apparent in the rat at least, for, whereas the decrease in the proportion of PDH_a in skeletal muscle during lactation appears to be due primarily to a lower serum insulin concentration, adipose-tissue PDH becomes refractory to insulin.

The inability of insulin to activate PDH would account for the findings by Burnol *et al.* (1986) that insulin stimulation of glucose conversion into CO₂ and fatty acids, but not into lactate and acylglycerol glycerol, is impaired in isolated adipocytes from lactating rats, and could also be the reason for the failure of insulin to stimulate fatty acid synthesis in adipose tissue from lactating rats *in vivo* (Burnol *et al.*, 1983; Jones *et al.*, 1984), although other factors such as a lack of activation of acetyl-CoA carboxylase could be involved here.

The refractoriness of white adipose tissue to insulin, along with the decreased ability of noradrenaline to activate PDH, have further repercussions, for the increased availability of glucose on feeding, which also increases serum insulin concentration and sympathetic nervous activity (see Le Magnen, 1984; Landsberg & Young, 1985), will not result in an increased flux of glucose to acetyl-CoA in white adipose tissue. Rather, an increased lactate output by the tissue is to be expected, which may again be used for gluconeogenesis in the liver. Thus the effect of these various changes in PDH activity of adipose tissue and muscle, and its endocrine control during lactation, should limit the use of glucose and lactate carbon by adipose tissue and skeletal muscle and so should facilitate their preferential use ultimately for milk production.

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