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Development of insulin-sensitivity at weaning in the rat

Role of the nutritional transition

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This study was undertaken to determine the factors involved in the development of insulin-sensitivity at weaning. Glucose kinetics were studied in suckling rats and in rats weaned on to a high-carbohydrate (HC) or a high-fat (HF) diet, in the basal state and during euglycaemic–hyperinsulinaemic-clamp studies. These studies were coupled with the 2-deoxyglucose technique, allowing a measure of glucose utilization by individual tissues. In the basal state, the glycaemia was higher in HF-weaned rats (124 ± 4 mg/dl) than in suckling (109 ± 1 mg/dl) and HC-weaned rats (101 ± 3 mg/dl). Glucose turnover rates were similar in the three groups of animals (14 mg/min per kg). Nevertheless, basal metabolic glucose clearance rate was 20% lower in HF-weaned rats than in the other groups. During the euglycaemic–hyperinsulinaemic experiments, hepatic glucose production was suppressed by 90% in HC-weaned rats, whereas it remained at 40% of basal value in suckling and HF-weaned rats, indicating an insulin resistance of liver of these animals. Glucose clearance rate during the clamp was 18.3 ± 0.9 ml/min per kg in suckling rats, whereas it was 35.3 ± 1.2 ml/min per kg in HC-weaned rats and 27.8 ± 1.1 ml/min per kg in HF-weaned rats, indicating an insulin resistance of glucose utilization in suckling, and to a lower extent, in HF-weaned rats. The deoxyglucose technique showed that peripheral insulin resistance was localized in muscles and white adipose tissue of suckling and HF-weaned rats. These results indicate that the switch from milk to a HC diet is an important determinant of the development of insulin-sensitivity at weaning in the rat.

INTRODUCTION

In adult mammals, an oral or intravenous glucose load is generally followed by an increase in blood glucose and plasma insulin and by a decrease in plasma glucagon concentrations (Unger, 1981). Consequently, hepatic glucose production is inhibited and glucose utilization rate is increased, avoiding the development of an excessive hyperglycaemia. In contrast, such a regulation of glucose homoeostasis seems to be immature in newborns. During an intravenous glucose infusion, hepatic glucose production is not normally suppressed in the human baby (Cowett *et al.*, 1983) and the newborn lamb (Cowett *et al.*, 1978), dog (Varma *et al.*, 1973) and rat (Ferré *et al.*, 1985*a*; Issad *et al.*, 1987*a*). In the dog, a decreased insulin-sensitivity of liver and peripheral tissues has been reported until 55 days of age (Varma *et al.*, 1973). In 15-day-old suckling rats, we have demonstrated the presence of a marked insulin resistance at the level of hepatic glucose production as well as at the level of whole-body glucose utilization. This insulin resistance disappears in 28-day-old rats weaned on to a high-carbohydrate (HC) diet (Issad *et al.*, 1987*a*). Thus, in the rat, a large increase in insulin-sensitivity occurs over a very short period of time (less than 15 days). The mechanisms responsible for these changes of insulin-sensitivity remain unclear. The nutritional transition that occurs at weaning, from a high-fat (HF) low-carbohydrate diet (milk) to a HC low-fat diet (laboratory chow or semi-synthetic weaning diet), may be an important factor for the development of insulin-sensitivity in the young rat. Indeed, these nutritional changes are associated with large modifications in the concentrations of circulating hormones and energy substrates (Girard *et al.*, 1977; Henning, 1981), which could have an important role in the increase of insulin-sensitivity. On the other hand, the maturation of insulin-sensitive tissues may be less dependent on the nutritional conditions and more tightly linked to the stage of development of the animals. Thus, to clarify the role of nutrition in the development of insulin-sensitivity at weaning, we have studied the insulin-sensitivity of suckling rats and of rats weaned on to two different diets, one with a high fat content and one with a high carbohydrate content. In an attempt to determine which tissues were involved in the changes in whole-body insulin-sensitivity observed at weaning, euglycaemic–hyperinsulinaemic-clamp studies were coupled with the 2-deoxyglucose technique, allowing a measurement of glucose utilization by individual tissues (Ferré *et al.*, 1985*b*).

MATERIALS AND METHODS

Animals

Wistar rats bred in our laboratory were used. They were housed in plastic cages in a room in which the temperature was maintained at 24 °C, with light from 07:00 to 19:00 h. The studies were performed on 13–15-day-old suckling rats and 28–30-day-old weaned rats. The litters were standardized to eight to ten animals at birth. The rats were weaned at 19 days on to a semi-

Abbreviations used: HC diet, high-carbohydrate diet; HF diet, high-fat diet.
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synthetic HC diet [25% protein, 5% fat, 60% carbohydrate, 5% cellulose, 5% minerals (w/w)] or a HF diet [37% protein, 43% fat, 15% cellulose, 5% minerals (w/w)]. The mothers of the rats weaned on to the HC diet had free access to water and chow pellets [24% protein, 4% fat, 57% carbohydrate, 6% cellulose, 9% minerals (w/w)]. Since the suckling rats begin to nibble the food of the mother at 15 days of age (Redman & Sweney, 1976), and to avoid carbohydrate intake from chow pellets between 15 and 19 days, the mothers of rats weaned on to the HF diet were fed with the similar HF diet from the day 14 of lactation until weaning. At the time of the experiments, mean body weights were 28.5±0.1 g in suckling rats ($n = 14$), 68±1 g in rats weaned on to the HC diet ($n = 13$) and 68±2 g in rats weaned on to the HF diet ($n = 14$).

Experimental procedures

Surgery. At 90 min before anaesthesia, the food of the weaned rats was removed and the suckling rats were separated from their mother and maintained in an incubator at 37 °C. The rats were anaesthetized with sodium pentobarbital (75 mg/kg) intraperitoneally. To avoid hypothermia, the anaesthetized rats were placed on a heating blanket maintained at 37 °C. A tracheotomy was systematically performed to avoid respiratory problems during the experiments. A carotid artery was catheterized for blood sampling with a polyethylene catheter (internal diam. 0.28 mm). Infusions were performed via a butterfly needle inserted into the saphenous vein. All experiments were started 30 min after anaesthesia.

Measurement of glucose turnover in basal state. Glucose turnover rate was measured as previously described (Issad *et al.*, 1987*a*). Briefly, a primed continuous infusion of [3-^{3}H]glucose (Amersham International, Amersham, Bucks., U.K.), at a rate of 0.016 μCi/min (priming dose 0.6 μCi), dissolved in heparin (100 i.u./ml)-containing 0.9% NaCl, was performed into the saphenous vein. Three blood samples (30 μl) were withdrawn at 30, 40 and 50 min (i.e. 150, 160 and 170 min after removal of food) to determine blood glucose concentration and glucose specific radioactivity. Glucose turnover rate was calculated as the ratio rate of tracer infusion (d.p.m./min) to steady-state glucose specific radioactivity (d.p.m./mg).

Measurement of glucose utilization index in individual tissues in the basal state. A tracer dose of 2-deoxy[1-^{14}C]glucose (10 μCi; CEA, Saclay, France) in 100 μl of heparin (100 i.u./ml)/0.9% NaCl was injected as a bolus through the saphenous vein. Blood samples (15 μl in suckling and 25 μl in weaned rats) were then sampled at 1, 3, 5, 10, 20, 40 and 60 min after the 2-deoxy[1-^{14}C]glucose injection. Then a larger blood sample (800 μl) was withdrawn for subsequent determination of plasma insulin, glucagon and non-esterified fatty acids in the basal state. The rats were rapidly killed by cervical dislocation. The tissues sampled were the soleus, the extensor digitorum longus, the diaphragm, the internal white adipose tissue from the lumbar region and the interscapular brown adipose tissue. Blood and tissues were treated as previously described (Ferré *et al.*, 1985*b*). Glucose utilization by each tissue was calculated as described by Ferré *et al.* (1985*b*), except that the value found was not corrected by the lumped constant for 2-deoxyglucose in glucose metabolic pathways. Therefore the results are expressed as an index of glucose utilization rather than as an absolute value (Hom *et al.*, 1984; Kraegen *et al.*, 1985).

Euglycaemic–hyperinsulinaemic-clamp studies coupled with the 2-deoxyglucose technique. An adaptation of the euglycaemic–hyperinsulinaemic clamp technique for young animals has been previously described (Issad *et al.*, 1987*a*). Briefly, after recovery from the surgery, basal blood glucose concentration is measured with a glucose analyser (YSI 23A; Yellow Spring Instruments, Yellow Springs, OH, U.S.A.). Then an insulin infusion is performed through the saphenous vein at a constant rate of 12 munits/min per kg after a priming dose of 0.33 unit/kg. Blood glucose concentration is determined every 10 min with the glucose analyser, and a variable glucose infusion is performed through the same saphenous vein to maintain the glycaemia at the basal value. The rate of glucose utilization is determined by a primed continuous infusion of [3-^{3}H]glucose. When a steady state is reached for glycaemia and exogenous glucose infusion (usually 40 min after the beginning of the clamp), a tracer dose of 2-deoxy[1-^{14}C]glucose (10 μCi) in 100 μl of 0.9% NaCl is injected as a bolus through the other saphenous vein. Blood samples are then taken (15 μl in suckling and 25 μl in weaned rats) at 1, 3, 5, 10, 20 and 30 min after the 2-deoxyglucose injection. Then a larger blood sample (800 μl) is withdrawn for subsequent determination of plasma insulin and non-esterified fatty acids. The animals are then immediately killed by cervical dislocation and the tissues are sampled. Blood and tissue samples are treated as previously described (Ferré *et al.*, 1985*b*), and the index of glucose utilization rates by individual tissues during the euglycaemic–hyperinsulinaemic clamp is determined.

Analytical techniques

Measurement of glucose and 2-deoxyglucose specific radioactivity. Blood samples (15 μl for suckling rats and 25 μl for the weaned rats) were deproteinized with $Ba(OH)_2/ZnSO_4$ and centrifuged (2 min, 16000 ***g***). A portion of the supernatant was used for the determination of glucose concentration by a glucose oxidase method (kit from Boehringer-Mannheim, Meylan, France). Another sample was either directly counted for radioactivity in a liquid-scintillation spectrometer (Betamatic; Kontron, Montigny, France) for the determination of blood 2-deoxy[1-^{14}C]glucose, or first evaporated to dryness to remove ^3H_2O and then redissolved in 200 μl of water for the determination of blood [3-^{3}H]glucose. Since during the clamp experiments [3-^{3}H]glucose and 2-deoxy[1-^{14}C]glucose radioactivities were determined in the same experiments, the sample was counted for ^{3}H and ^{14}C radioactivity after removal of the ^3H_2O.

Measurement of hormones and substrates. A blood sample was centrifuged at 4 °C in the presence of 20 μl of the peptidase inhibitor aprotinin (Iniprol; 200000 units/ml; Laboratoire Choay, Paris, France). The plasma was kept frozen at −20 °C for the determination of pancreatic hormones. Plasma insulin was determined by radioimmunoassay as described previously (Issad *et al.*, 1987*a*).

Basal plasma pancreatic glucagon was also determined by radioimmunoassay (glucagon kit no. 10904; Serono Diagnostic S.A., Coinsins, Switzerland). Plasma non-esterified fatty acids were determined with an enzymic kit (NEFA C; Biolyon, Lyon, France).

Statistics. Results are expressed as means±S.E.M. Statistical analysis was performed by Student's *t* test for unpaired data.

RESULTS

Whole-body glucose kinetics in suckling and weaned rats

Basal state. Glucose kinetics, circulating substrates and plasma insulin concentrations in suckling rats and in rats weaned on to HC and HF diets are shown in Table 1. Basal blood glucose was similar in suckling rats and in rats weaned on to the HC diet, whereas it was significantly higher in rats weaned on to the HF diet. Plasma insulin concentrations were lower in suckling and HF-weaned rats than in HC-weaned rats. In the basal state, plasma non-esterified fatty acid concentrations were similar in the three groups of animals. Plasma glucagon concentrations in the basal state were significantly higher ($P < 0.05$) in suckling and HF-weaned rats than in HC-weaned rats: 209±29 pg/ml in suckling, 159±12 pg/ml in HF-weaned rats and 100±11 pg/ml in HC-weaned rats (all $n = 6$).

Basal glucose turnover rate was similar in the three groups of rats. Since the glycaemia was higher in HF-weaned rats, we calculated the metabolic clearance rate of glucose in each group of animals. The basal metabolic clearance rate of glucose was 30% lower in HF-weaned rats than in suckling and HC-weaned rats.

Euglycaemic–hyperinsulinaemic-clamp studies. During the euglycaemic–hyperinsulinaemic-clamp experiments, the glycaemia was maintained at the basal value in each group of animals. The plasma insulin concentrations reached were higher in suckling rats than in both groups of weaned rats, despite similar insulin infusion rates (12 munits/min per kg), indicating a lower clearance rate of insulin in suckling rats. Nevertheless, during the clamp experiment, the glucose utilization rate and the metabolic clearance rate of glucose were much lower in suckling rats than in both groups of weaned rats (Table 1). During the clamp, glucose utilization rates were similar in HC- and HF-weaned rats. However, rats weaned on to the HF diet showed a lower metabolic clearance rate of glucose than for the rats weaned on to the HC diet (Table 1).

During the clamp experiment, endogenous glucose production was suppressed by more than 90% in HC-weaned rats, whereas it was inhibited only by 40% in suckling and HF-weaned rats (Table 1).

Since non-esterified fatty acids can influence the rate of glucose utilization in peripheral tissues, we measured the plasma concentrations of these substrates during the clamp experiments in each group of animals. Plasma non-esterified fatty acid concentrations are decreased by 60% in both groups of weaned rats, but remained at basal values in suckling rats.

Insulin effect on various insulin-sensitive tissues

In an attempt to measure the insulin effect on glucose utilization by peripheral insulin-dependent tissues, the 2-deoxyglucose technique was used. Indices of glucose utilization rates were determined in several tissues, in the basal state and during the euglycaemic–hyper-insulinaemic-clamp studies. Since the glycaemia of the HF-weaned rats was higher than in other groups, we have calculated the index of glucose metabolic clearance rates by dividing the tissue glucose utilization index by the concentration of glucose in the blood during the experiment.

Muscles (Fig. 1).

(*a*) Diaphragm. Basal glucose clearance index was significantly lower in diaphragm of suckling and HF-weaned rats than in HC-weaned rats. During the clamp experiments, the glucose clearance index was much higher in HC-weaned rats than in both other groups.

(*b*) Soleus. In the basal state, the glucose clearance index was similar in the soleus of suckling and HC-

Table 1. Blood glucose, plasma insulin, plasma non-esterified fatty acids and glucose kinetics in the basal state and during euglycaemic–hyperinsulinaemic-clamp studies

Results are means±S.E.M. for six determinations: *, **, ***, differences statistically significant at $P < 0.05$, $P < 0.01$ and $P < 0.001$ respectively when compared with suckling rats under similar experimental conditions.

Rats...	Suckling		HC-weaned		HF-weaned	
Experiment...	Basal	Clamp	Basal	Clamp	Basal	Clamp
Blood glucose (mg/dl)	109±1	101±3	102±4	100±3	124±4**	130±5**
Plasma immunoreactive insulin (μunits/ml)	23±3	931±128	49±6**	366±36**	31±6	348±9**
Plasma non-esterified fatty acids (μmol/l)	410±3	440±8	480±6	180±3***	440±5	170±3***
Glucose utilization rate (mg/min per kg)	14.1±0.7	20.0±0.9	14.0±0.8	35.3±1.2***	13.0±0.9	36.7±1.5***
Glucose production rate (mg/min per kg)	14.1±0.7	8.8±0.9	14.0±0.8	1.2±0.8***	13.0±0.9	9.0±0.9
Glucose metabolic clearance rate (ml/min per kg)	12.9±0.6	18.3±0.9	13.7±0.8	35.3±1.2***	10.4±0.7*	27.8±1.1***

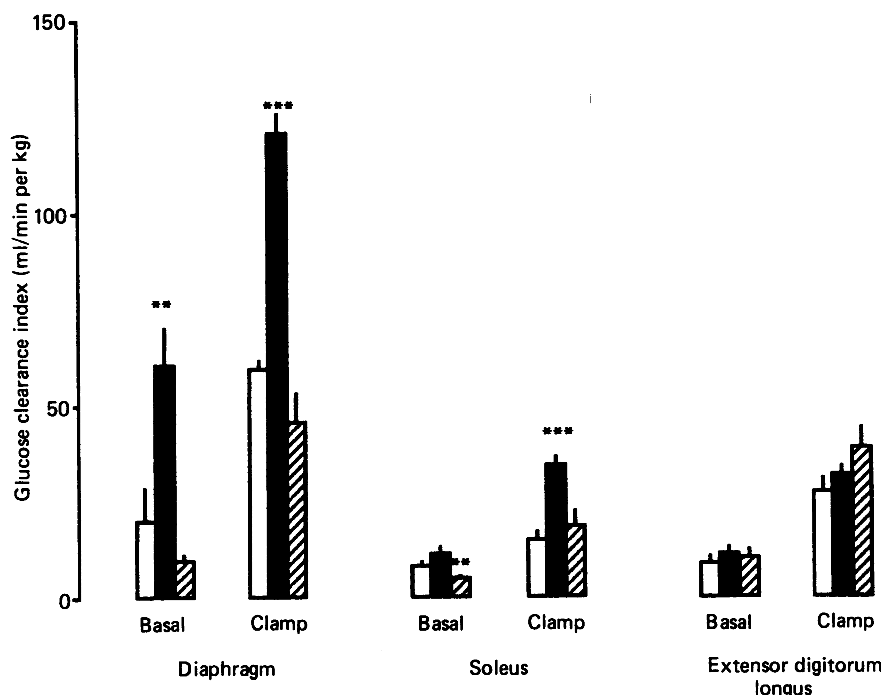

Fig. 1. Glucose clearance index in diaphragm, soleus and extensor digitorum longus, in the basal state and during euglycaemic–hyperinsulinaemic-clamp studies

Results are means ± S.E.M. for five to seven determinations: *, **, ***, differences statistically significant at $P < 0.05$, $P < 0.01$ and $P < 0.001$ when compared with suckling rats under similar experimental conditions. Rats: □, suckling; ■, HC-weaned; ▨, HF-weaned.

weaned rats, whereas it was significantly lower in HF-weaned rats. Under euglycaemic–hyperinsulinaemic conditions, the glucose clearance index was significantly lower in suckling and HF-weaned rats than in HC-weaned rats.

(*c*) Extensor digitorum longus. In the extensor digitorum longus, glucose clearance was not significantly different in the three groups of animals in the basal state and during the clamp experiments.

Adipose tissues (Fig. 2).

(*a*) White adipose tissue. In the basal state, glucose clearance index was similar in white adipose tissue of HC- and HF-weaned rats. Basal glucose clearance index was significantly lower in suckling than in HC-weaned rats. During the glucose clamp experiment, a large increase in glucose clearance index was observed in white adipose tissue of HC-weaned rats, whereas no significant increase was observed in white adipose tissue of suckling and HF-weaned rats.

(*b*) Brown adipose tissue. In the basal state, glucose clearance index was not significantly different for suckling and HC-weaned rats. It was significantly lower in HF-weaned rats than in suckling rats. During the clamp experiment, the glucose clearance indices were similar in brown adipose tissue of suckling and HC-weaned rats and significantly lower in HF-weaned rats.

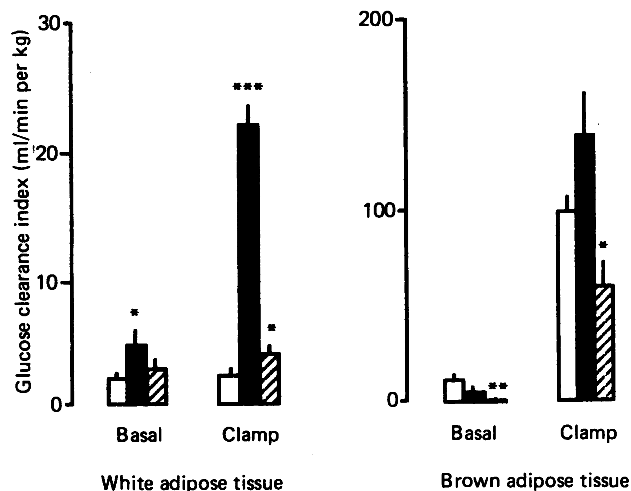

Fig. 2. Glucose clearance index in white and brown adipose tissues, in the basal state and during euglycaemic–hyperinsulinaemic-clamp studies

Results are means ± S.E.M. for six to seven determinations. Symbols are as for Fig. 1.

DISCUSSION

This study was undertaken to determine which tissues were involved in the insulin resistance observed during the suckling period in the rat and to document the role of the nutritional transition in the development of insulin-sensitivity that occurs at weaning.

Basal blood glucose was significantly higher in rats weaned on to the HF diet than in both other groups. The basal hyperglycaemia observed in rats fed on a HF diet had also been reported in adult rats by other authors (Lavau & Susini, 1975; Zaragoza & Felber, 1970). Since the glycaemia itself can influence glucose disposal by a

mass-action effect (De Fronzo & Ferrannini, 1982; Thiebaud *et al.*, 1982), a direct comparison of glucose utilization rates between HF-weaned rats and the two other groups is not possible. We have thus calculated the metabolic clearance rates of glucose in the basal state and under euglycaemic–hyperinsulinaemic conditions. For the same reasons, the indices of glucose utilization by individual tissues, as measured by the 2-deoxyglucose technique, have been expressed as glucose clearance rate index. The validity of the glucose metabolic clearance rate has been discussed at length elsewhere (Leturque *et al.*, 1984).

Insulin effect on hepatic glucose production

As in our previous study, hepatic insulin resistance is observed in suckling animals. This disappears after weaning on to the HC diet, but not after weaning on to the HF diet. This indicates that the composition of the weaning diet is an important determinant of hepatic insulin-sensitivity during development. Several factors could be involved in this insulin resistance.

(1) A decreased number or affinity of insulin receptors could theoretically be responsible for the hepatic insulin resistance observed in suckling and HF-weaned rats. It is unlikely in suckling rats, since it has been shown that the binding of insulin to liver plasma membrane did not change significantly during the suckling–weaning transition (Blazquez *et al.*, 1976).

(2) Plasma glucagon, the concentration of which is higher in suckling and HF-weaned rats than in HC-weaned rats, could exert an anti-insulin effect on the liver (Felig *et al.*, 1979; Del Prato *et al.*, 1987).

(3) Evidence (Terretaz *et al.*, 1986) obtained in adult rats fed on a standard laboratory diet suggests that the suppression of hepatic glucose production during euglycaemic–hyperinsulinaemic-clamp experiments occurs by means of an acceleration of the glycolytic flux and of lipogenesis, which is elevated under these hyperinsulinaemic conditions. Hepatic glucokinase, a key enzyme of glycolysis in the liver, has a very low activity during the suckling period, which increases considerably after weaning on to a HC diet, but not after weaning on to a HF diet (Walker & Eaton, 1967). Moreover, hepatic lipogenesis is very low during the suckling period and increases after weaning on to HC diet, but not after weaning on to HF diet (Lockwood *et al.*, 1970; Gandemer *et al.*, 1982, 1985). If the stimulation of these metabolic pathways is required for the suppression of hepatic glucose production, alteration of these pathways may be a cause of hepatic insulin resistance.

Insulin effect on glucose utilization in individual tissues

Insulin effects on muscles. In suckling rats, compared with the HC-weaned animals, insulin resistance is observed in all muscles during the hyperinsulinaemic glucose clamp except in the extensor digitorum longus. Since the insulin concentration during the clamp is much higher in suckling rats than in HC-weaned rats, one cannot exclude the possibility that the extensor digitorum longus is also insulin-resistant, although to a lower extent than other muscles studied. The results are very similar in the HF-weaned rats. Using a similar technique, Kraegen *et al.* (1986) have also observed, in adult rats fed on a HF diet, an insulin resistance in the soleus and diaphragm, but not in the extensor digitorum longus. These observations suggest that the diet, and not a stage of development, is responsible for the insulin resistance observed during the suckling period in skeletal muscles.

Since the muscle mass represents a lower percentage of body weight in suckling (16%) than in weaned rats (24%) (Issad *et al.*, 1987*a*), it could explain why, despite a similar insulin resistance in skeletal muscle, the insulin resistance at the whole-body level is milder in HF-weaned animals.

The potential reasons for muscle insulin resistance are numerous. A decreased insulin binding has been observed in soleus of adult rats fed on a HF diet (Grundleger & Thenen, 1982; Maegawa *et al.*, 1986). Although such a decrease cannot be excluded in the soleus of suckling rats, it is unlikely in diaphragm, since no modification of insulin-binding capacity has been observed *in vitro* in this tissue between 10 and 30 days of age (Wang, 1985). A decreased insulin effect on glucose transport and metabolism has been observed in isolated soleus muscle and diaphragm of adult rats fed on HF diets (Grundleger & Thenen, 1982; Maegawa *et al.*, 1986; Susini & Lavau, 1978).

Finally, in suckling rats, plasma non-esterified fatty acid concentrations were not decreased during the clamp studies, and could be involved in the lower clearance of glucose in muscles which are well equipped for their oxidation, such as soleus and diaphragm (Randle *et al.*, 1964; Rennie & Holloszy, 1977; Issad *et al.*, 1987*b*).

Insulin effect on adipose tissues

White adipose tissue. A massive insulin resistance was observed in the internal white adipose tissue of the lumbar region in suckling rats and in rats weaned on to the HF diet. This insulin resistance was also observed at other subcutaneous and internal adipose tissue sites (results not shown). The insulin resistance disappeared after weaning on to the HC diet. Studies by Tsujikawa & Kimura (1980) have shown, *in vitro*, on isolated white adipocytes of suckling rats that insulin did not stimulate the incorporation of [^{14}C]glucose into lipids, whereas a large stimulation was observed in white adipocytes of rats weaned on to a standard HC diet. When the animals are weaned on to a HF diet, insulin did not stimulate the incorporation of glucose into the lipid of white adipocytes of these animals. These authors have also shown that the activities of acetyl-CoA carboxylase and ATP citrate lyase in white adipose tissue are very low during the suckling period and increase considerably at weaning on to a HC diet, whereas they remain very low in HF-weaned rats. Since lipogenesis is one of the major pathways of glucose metabolism in white adipose tissue (Richardson & Czech, 1978), the low activities of these enzymes in suckling and HF-weaned rats could be responsible for the insulin resistance observed in the white adipose tissue of these animals, although other factors cannot be excluded.

In HC-weaned rats, the glucose utilization index for white adipose tissue under euglycaemic–hyperinsulinaemic conditions is much larger than that measured by Ferré *et al.* (1985*b*) in adult rats fed on a standard (HC) laboratory-chow diet. The reason for these differences may reside in the higher lipogenic capacity observed in white adipose tissue of young 30-day-old rats weaned on to HC diet than in adult rats raised from weaning to the adult stage on the same diet

(Tsujikawa & Kimura, 1980; Gandemer *et al.*, 1982, 1985).

Brown adipose tissue. A considerable insulin effect on glucose utilization is observed in brown adipose tissue of HC-weaned rats. Such an effect is not unexpected, since it is now well established that brown adipose tissue is extremely sensitive to insulin in animals fed on a high-carbohydrate low-fat diet (Ferré *et al.*, 1986), lipogenesis being an important pathway for glucose utilization (Isler *et al.*, 1987; Ebner *et al.*, 1987).

In suckling and HF-weaned rats, the rates of glucose utilization are stimulated by insulin, although to a lower extent than in HC-weaned rats. This might be the consequence of a lower lipogenic capacity in brown adipose tissue of suckling and HF-weaned rats (Pillay & Bailey, 1982, 1983).

Nevertheless, the brown adipose tissue of suckling animals is known to possess a very high thermogenic activity (Barnard & Skala, 1970), and high-fat feeding has been shown to stimulate thermogenesis in brown adipose tissue (Mercer & Trayhurn, 1984). Since insulin potentiates the oxidation of glucose when brown adipose tissue is activated (Isler *et al.*, 1987; Ebner *et al.*, 1987), this might explain the relatively elevated glucose utilization under insulin stimulation in suckling and HF-weaned rats, despite the low lipogenic capacity.

Conclusion

This study has shown that the large insulin resistance observed in suckling rats was localized in liver, muscles and white adipose tissue. The increase in insulin-sensitivity that occurs at weaning is, at least in part, linked to the dietary transition from a high-fat diet (milk) to a high-carbohydrate diet. Although a possible direct action of nutrient on tissue insulin-sensitivity (for instance, changes in cell membrane composition affecting membrane proteins) cannot be excluded, it is more likely that nutrients act indirectly by modulating the hormonal secretions. Among the various hormones, insulin and glucagon undergo marked changes in concentration during the suckling–weaning transition. These differences could be even more marked during the meals. One of the long-term regulatory effects of insulin and glucagon could be the modulation of the concentration of specific proteins (insulin receptors, glucose transporters) and enzymes (glycogen synthase in muscle, glucokinase in the liver, acetyl-CoA carboxylase in adipose tissue etc.) which are involved in the short-term response to insulin.

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