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Differential metabolic response to 48 h food deprivation at different periods of pregnancy in the rat

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Since during pregnancy the mother switches from an anabolic to a catabolic condition, the present study was addressed to determine the effect of 48 h food deprivation on days 7, 14 and 20 of pregnancy in the rat as compared to age matched virgin controls. Body weight, free of conceptus, decreased with food deprivation more in pregnant than in virgin rats, with fetal weight (day 20) also diminishing with maternal starvation. The decline of plasma glucose with food deprivation was greatest in 20 day pregnant rats. Insulin was highest in fed 14 day pregnant rats, and declined with food deprivation in all the groups, the effect being not significant in 7-day pregnant rats. Food deprivation increased plasma glycerol only in virgin and 20 day pregnant rats. Plasma NEFA and 3-hydroxybutyrate increased with food deprivation in all groups, the effect being highest in 20 day pregnant rats. Food deprivation decreased plasma triacylglycerols in 14 day pregnant rats but increased in 20 day pregnant rats. In 20-day fetuses, plasma levels of glucose, NEFA and triacylglycerols were lower than in their mothers when fed, and food deprivation caused a further decline in plasma glucose, whereas both NEFA and 3-hydroxybutyrate increased. Liver triacylglycerols concentration did not differ among the groups when fed, whereas food deprivation caused an increase in all pregnant rats and fetuses, the effect being highest in 20-day pregnant rats. Lipoprotein lipase (LPL) activity in adipose tissue was lower in 20 day pregnant rats than in any of the other groups when fed, and it decreased in all the groups with food deprivation, whereas in liver it was very low in all groups when fed and increased with food deprivation only in 20 day pregnant rats. A significant increase in liver LPL was found with food deprivation in 20 day fetuses, reaching higher values than their mothers. Thus, the response to food deprivation varies with the time of pregnancy, being lowest at mid pregnancy and greatest at late pregnancy, and although fetuses respond in the same direction as their mothers, they show a specific response in liver LPL activity.

Key words: Fasting, Pregnancy, Rat, Triacylglycerols, Liver, Lipoprotein lipase, Adipose tissue.

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During pregnancy the mother must adapt her metabolism in order to sustain the availability of substrates to the fetus, and although maternal hyperphagia is established from the beginning of pregnancy both in women (29) and rats (25), two clearly differentiated phases take place (15). At an early stage, when growth and nutrient requirements of the fetus are limited, the mother accumulates fat depots (18, 47) due to both an augmented adipose tissue lipoprotein lipase activity (19) and lipogenesis (31). However, at late pregnancy when fetal growth rate is maximal, the mother switches to a catabolic condition, as shown by both an enhanced adipose tissue lipolytic activity (26, 49) and hepatic gluconeogenesis (1, 54).

These metabolic adaptations taking place during pregnancy are driven by hormones, changes in plasma insulin levels and in insulin sensitivity. During early pregnancy the maternal pancreas increases its insulin content (28) and the sensitivity to insulinotropic stimuli is enhanced (5), whereas insulin sensitivity is either unchanged or even enhanced, in both women (8, 9) and rats (18). In contrast, an intense insulin resistant condition arises in late pregnancy (6, 28), even though the sensitivity to insulinotropic stimuli remains enhanced (11, 28).

Short periods of food deprivation have been used to study the metabolic capacity of the mother to sustain fetal growth when the availability of exogenous substrates becomes limited. The term "accelerated starvation" defines the maternal active catabolic condition to warrant an appropriate substrate availability to the fetus when late pregnant mothers are exposed to short periods of starvation (12). Since most of the studies using the fasting stratagem during pregnancy have been done during its late phase, and we have previously shown that 24 h starvation causes different metabolic changes during early versus late pregnancy in the rat (24), this work was addressed to determine the effect of 48 h of food deprivation on days 7, 14 and 20 of pregnancy in the rat as compared to age matched virgin controls.

Material and Methods

Female Sprague Dawley rats, initially weighing 180-190 g were mated and the day spermatozoids appeared in vaginal smears was considered day 0 of gestation. Since young animals were used, special attention was given to use age matched virgin controls which at the onset of the study their body weight did not differ from that of pregnant rats. Virgin rats were always studied in parallel to pregnants, and special care was taken to sacrifice some virgin animals the same days as pregnants, although their values are presented as one group. All animals were fed ad libitum a standard nonpurified diet (B&K Universal, Barcelona, Spain) and housed under controlled light and temperature (12-h light-dark cycle; 22 ± 1 °C).The experimental protocol was approved by the Animal Research Committee of the University San Pablo-CEU in Madrid, Spain. Pregnant rats were randomly placed in groups and studied on days 7, 14 and 20 of gestation, and the starved rats were deprived of food, but not of water, for 48 h. All animals were sacrificed by decapitation and trunk blood was collected in Na_2-EDTA for plasma separation. It was kept at –80°C until analyzed for 3-hydroxybutyrate (51) and glycerol (13) after deproteinization (44), and glucose, triacyglycerols (TAG), nonesterified fatty acids (NEFA) and immunoreactive insulin by commercial

kits (Boehringer-Mannheim, Germany, Menarini Diagnostics, Italy, Wako Chemical GmbH, Germany and Mercodia AB, Sweden, respectively). Liver and lumbar fat pads were immediately dissected, and aliquots of both tissues were placed into liquid nitrogen and kept at –80 °C until analysis. The conceptus was also dissected and after being weighed, fetuses were weighed and decapitated and blood from all pups of the same mother was collected and pooled into receptacles containing Na_2-EDTA. Fetus livers were also dissected, placed in liquid nitrogen, and pooled those from the same mother.

Frozen liver aliquots were used for lipid extraction (10), and aliquots of lipid extracts were quantified after image analysis and separation by one-dimensional TLC (41) using the G5-700 BIOIMAGE TLC scanner of Bio-Rad ((Hercules, CA). Spots were quantified as integrated optical densities against an internal standard of cholesteryl formate and calibration curves of triacylglycerol standards.

Lipoprotein lipase (LPL) activity (EC 3.1.1.34) was measured in acetone powders from frozen adipose tissue and liver aliquots by the method previously described (21).

Expression of the results and statistical evaluation.– Results were expressed as means ± S.E.M. The effects of day of pregnancy were analyzed by one-way ANOVA, using a computer software (Systat Version 5.03, Wilkinson, Evanston, Il., USA). When treatment effects were significantly different ($p<0.05$), specific means were tested by Tukey's test. Differences between two groups were analyzed by Student's *t* test.

Results

As shown in Table I, body weight increased as pregnancy advanced, and 48 h of food deprivation had a greater effect on body weight in pregnant than in virgin rats. Net body weight (free of the conceptus) in pregnant rats was also higher than body weight in virgin rats (day 0) from the 7th day of gestation, and although 48 h of food deprivation always decreased this variable, its effect was greater in pregnant (absolute loss of 47-59 g) than in virgin rats (loss of 24 g), independently of the day of pregnancy. Conceptus weight was relatively smaller in 7- and 14-day pregnant rats as compared to 20-day pregnant rats, and maternal starvation produced a non-significant decline in this variable at days 14 and 20 of gestation. However, in starved 20-day pregnant rats, fetal weight was significantly decreased as compared to the fed condition, whereas placental weight remained unchanged (Table I). Both liver and lumbar fat pad weights were higher in 14 and 20 day fed pregnant rats than in virgins, although the change in lumbar fat pad at day 14 of pregnancy did not reach statistical significance (Table I). Forty eight hours of food deprivation decreased these two variables in all the groups, although the change in lumbar fat pad weight was not significant in 20 day pregnant rats (Table I).

Plasma metabolite levels are summarized in Table II. In fed rats, glucose levels decreased at days 14 and 20 of gestation as compared to virgin rats, and although 48 h of food deprivation decreased this variable in all the groups, the greatest effect was found in the 20 day pregnant rats. In both fed and 48 h food deprived 20 day old rats, plasma glucose levels in fetuses were lower than in their mothers, yet maternal starvation produced a further significant

Table I. *Effect of pregnancy and 48 h of food deprivation on body, conceptus and tissue weights (g) in the rat.*

Day of pregnancy	Food deprivation	Body weight (b.w.)	Net b.w.	Conceptus	Fetus	Placenta	Liver	Lumbar fat pad
0	-	187 ± 4^{a}	187 ± 4^{a}				9.2 ± 0.2^{a}	0.92 ± 0.04^{a}
0	+	$163 \pm 8^{a*}$	$163 \pm 8^{a*}$				$5.2 \pm 0.1^{a***}$	$0.4 \pm 0.04^{a***}$
7	-	222 ± 8^{a}	221 ± 8^{b}	1.1 ± 0.16^{a}			10.2 ± 0.6^{a}	0.8 ± 0.1^{a}
7	+	$175 \pm 3^{a***}$	$174 \pm 0.4^{a***}$	1.1 ± 0.08^{a}			$6.2 \pm 0.2^{b***}$	$0.4 \pm 0.07^{a**}$
14	-	273 ± 7^{b}	261 ± 6^{bc}	11.5 ± 1.6^{a}			13.4 ± 0.4^{b}	1.2 ± 0.1^{ab}
14	+	$210 \pm 5^{b***}$	$202 \pm 6^{b***}$	8.1 ± 1.1^{a}			$7.2 \pm 0.1^{c***}$	$0.8 \pm 0.05^{b*}$
20	-	361 ± 9^{c}	282 ± 7^{c}	82.4 ± 6.7^{b}	4.2 ± 0.04	0.75 ± 0.02	15.1 ± 0.7^{b}	1.6 ± 0.18^{b}
20	+	$295 \pm 4^{c***}$	$230 \pm 5^{c***}$	65.4 ± 4^{b}	$3.5 \pm 0.05^{***}$	0.58 ± 0.02	$11.6 \pm 0.5^{d**}$	1.3 ± 0.2^{c}

Values are expressed as means ± SEM, n=7-9. Tukey's test was used to determine differences between groups after ANOVA. Different superscripts in a row indicate significant differences ($P< 0.05$) among the four groups at different day of gestation. Statistical comparisons between fed and food deprived rats for the same variable and same day of gestation is shown by astherisks (*$p < 0.05$; **$p < 0.01$; ***$p < 0.001$).

Table II. *Effect of pregnancy and 48 h of food deprivation on plasma metabolites and insulin in the rat.*

Day of pregnancy	Food deprivation	Glucose (mg/dL)	Insulin (μM)	Glycerol (μM)	NEFA (μM)	3-hydroxybutyrate (μM)	Triacylglycerols (mg/L)
0	-	164 ± 7^{a}	28 ± 3^{ab}	164 ± 8^{a}	263 ± 18^{a}	238 ± 8^{a}	8.4 ± 0.7^{a}
0	+	$98 \pm 5^{a***}$	$4.7 \pm 0.5^{a***}$	$268 \pm 21^{a***}$	$719 \pm 47^{a***}$	$3760 \pm 305^{a***}$	7.4 ± 0.9^{a}
7	-	166 ± 17^{ab}	24 ± 7^{a}	119 ± 8^{a}	235 ± 36^{a}	144 ± 14^{a}	8.4 ± 1.3^{a}
7	+	$94 \pm 5^{a**}$	9 ± 5^{ab}	200 ± 54^{a}	$609 \pm 65^{a**}$	$4690 \pm 840^{a***}$	15 ± 3.1^{a}
14	-	115 ± 13^{c}	50 ± 5^{b}	237 ± 16^{b}	242 ± 35^{a}	227 ± 36^{a}	21.5 ± 1.7^{b}
14	+	$91 \pm 7^{a*}$	$11 \pm 3^{ab***}$	229 ± 25^{a}	$511 \pm 17^{a***}$	$4492 \pm 545^{a***}$	$8.5 \pm 1.3^{a***}$
20	-	121 ± 5^{bc}	34 ± 4^{ab}	177 ± 45^{a}	388 ± 27^{a}	162 ± 45^{a}	33 ± 6.2^{c}
20	+	$56 \pm 4^{b***}$	$15 \pm 5^{b*}$	$442 \pm 81^{b*}$	$1285 \pm 119^{b***}$	$13261 \pm 1516^{b***}$	$77 \pm 14^{b*}$
Fetus (day 20)	-	$37 \pm 3^{+++}$		189 ± 40	$44 \pm 25^{+++}$	65 ± 17	$5.6 \pm 0.6^{++}$
Fetus (day 20)	+	$22 \pm 4^{+++*}$		$166 \pm 30^{+}$	$139 \pm 14^{+++*}$	$17328 \pm 1246^{***}$	$6.0 \pm 0.5^{+++}$

Statistics and symbols as in Table I. $^{++}p < 0.01$; $^{+++}p < 0.001$ *vs.* the corresponding values in mothers (20 days gestation).

decrease. Plasma insulin levels were higher in fed 14 day rats than in any of the other groups, the difference being significant only with values in the 7 day pregnant rats. Insulin levels declined in all the groups after 48 h of food deprivation, although this change did not reach statistical significance in 7 day pregnant rats. Plasma glycerol levels were also higher in fed 14 day pregnant rats than in any of the other groups, and whereas they increased with 48 h food deprivation in virgin and 20 day pregnant rats, the highest value attained in the latter group, they did not change with food deprivation in 7- and 14-day pregnant rats. In 20 day fetuses, plasma glycerol levels were similar to their mothers when fed, whereas this variable did not change in the 48 h food deprived fetuses despite the increase seen in their mothers. Plasma NEFA levels did not differ between the groups when rats were fed, and although 48 h of food deprivation increased this variable in all groups, the greatest effect appeared in pregnant rats of 20 days. In fetuses, plasma NEFA levels were much lower than in their mothers, although they increased significantly in those from 48 h food deprived mothers. Plasma 3-hydroxy butyrate levels were similarly low in all the groups of fed animals, and although 48 h of food deprivation always increased this variable, the effect was greatest in pregnant rats of 20 days. Plasma 3-hydroxy butyrate levels in 20 day fetuses mimic very closely the values seen in their mothers, with practically undetectable levels when fed and an enormous increment when food deprived. Major differences between the groups were found in plasma TAG levels. In fed animals, they progressively increased in pregnant rats of 14 and 20 days, and although 48 h of food deprivation did not modify plasma TAG levels in virgin and 7 day pregnant rats, they significantly decreased in 14 day pregnant rats whereas they were increased in 20 day pregnant rats. Plasma TAG levels in fetus were much lower than in their mothers, and 48 h fasting did not modify this variable.

Changes in plasma TAG prompted their determination in liver. As shown in Figure 1, liver TAG concentration was similar in fed virgin and 7-, 14- or 20-day pregnant rats, and whereas 48 h of food deprivation did not modify liver TAG concentration in virgin rats, they were significantly increased in all the groups of pregnant rats, whit the effect being highest in 20 day pregnant rats. Nevertheless, this

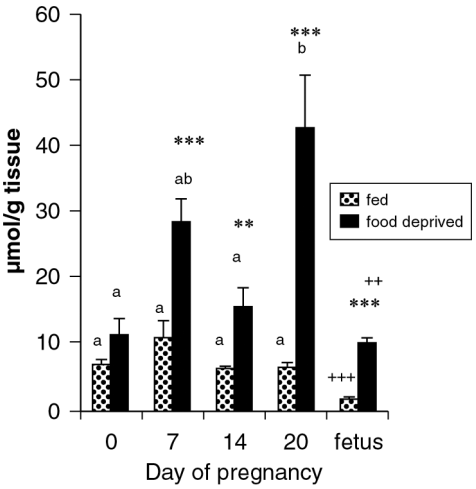

Fig. 1. *Effects of food deprivation on liver triacylglycerols concentration in virgin, pregnant rats and 20 day fetuses.*

Values are expressed as means ± SEM, n=7-9. Tukey's test was used to determine differences between groups after ANOVA. Different letters for each condition indicates significant differences ($P< 0.05$) among the four groups at different day of gestation. (**, $p < 0.01$, ***, $p < 0.001$) between fed and food deprived rats for the same variable and same day of gestation. ($^{++}$, $p < 0.01$, $^{+++}$, $p < 0.001$) between fetus and mothers (20 of gestation) for the same condition.

change in liver TAG concentration seen in 48 h food deprived pregnant rats was not significant when expressed per whole organ (data not shown) except for 20 day pregnant rats, where it was significantly higher than in the fed condition (82.1 ± 14.4 µmols/liver in fed vs. 445.2 ± 86.7 in 48 h fasted rats, $p<0.001$). As also shown in figure 1, liver TAG concentration was much lower in 20 day fetuses than in their mothers, although food deprivation caused a striking and significant increase. In fact, although liver weight was not determined in fetuses, such intense increase in liver TAG concentration in the fasted fetus cannot be justified through liver shrinkage.

Since LPL activity in adipose tissue and liver could help to understand plasma and liver TAG change patterns, these variables were secured, and values summarized in Table III. Lumbar adipose tissue LPL activity was similar in fed virgin and 7- and 14-day pregnant rats, whereas it was significantly decreased in 20-day pregnant rats. Food deprivation for 48 h greatly decreased adipose tissue LPL in all groups, attaining a similar value for all of them. As expected, liver LPL activity was almost undetectable in both fed virgin and pregnant rats, independently of the gestational time. While 48 h food deprivation produced no increases in liver LPL activity in virgin and 7- and 14-day pregnant rats, it was significantly increased in food deprived 20 day pregnant rats. Liver LPL activity was higher in fed 20 day fetuses than in their mothers, and maternal food deprivation caused a further and significant increase in this variable, attaining values that were significantly higher than both their respective mothers and the fed fetuses.

Discussion

The present study shows that the metabolic response to 48 h of food deprivation not only differs between late pregnant and virgin rats, but also between pregnant rats at different times of gestation. As expected, the increase in body weight during pregnancy does not corresponds only to the presence of fetal-placental structures, but to the own maternal structures (i.e., net body weight). The decline with 48 h of food deprivation appeared consistently

Table III. *Lipoprotein lipase (LPL) activity in lumbar adipose tissue (pKat/g) and liver (pKat/mg protein) in pregnant rats and 20 day fetuses.*

Day of pregnancy	Food deprivation	Adipose tissue	Liver
0	-	2245 ± 449^{a}	0.023 ± 0.01^{a}
0	+	$265 \pm 69^{a**}$	0.054 ± 0.02^{a}
7	-	2608 ± 326^{a}	0.01 ± 0.006^{a}
7	+	$277 \pm 45^{a***}$	0.07 ± 0.035^{ab}
14	-	2394 ± 271^{a}	0.013 ± 0.007^{a}
14	+	$425 \pm 13^{a***}$	0.056 ± 0.03^{a}
20	-	902 ± 124^{b}	0.026 ± 0.018^{a}
20	+	$338 \pm 120^{a*}$	$0.11 \pm 0.02^{b*}$
Fetus	-		$0.123 \pm 0.026^{++}$
Fetus	+		$0.304 \pm 0.062^{*++}$

Statistics and symbols as in Table II.

greater in pregnant than in age matched virgin rats. Since previous reports have shown that the increase in net maternal body weight during pregnancy mainly corresponds to fat depots (22), such decline in maternal own structures with food deprivation must correspond to a decrease in adipose tissue mass, which accordingly to present findings is greater in pregnant than in virgin rats. Although this interpretation contrasts with the similar or even smaller decline in lumbar adipose tissue mass seen here with food deprivation in pregnant than in virgin rats, we previously reported that changes in different fat depots during pregnancy are not parallel (23). In addition it is known that differences exist in the response to lipolytic stimuli depending on the fat depot source, as reported in humans (4, 27, 40).

Despite the profound breakdown of metabolic stores with 48 h of food deprivation between days 18 and 20 of pregnancy, fetal growth was impaired as shown by the decreased body weight. This finding is in contrast with the reported stable fetal body weight seen when rats are subjected to the same period of food deprivation during earlier stage of pregnancy (days 17 and 19), where fetal size is smaller and its metabolic needs are lower (20), suggesting a limited capacity of the late pregnant food deprived mother to sustain appropriate substrate availability to support fetal growth. In fact, the deep decrease in plasma glucose level seen here in fetuses of 48 h food deprived dams indicates a decline in the availability of this essential oxidative fuel which may contribute to the impaired growth.

The decline in plasma glucose seen in fed 14 day pregnant rats as compared to virgin and 7 day pregnant rats deserves special attention. This is coincident with the increment in plasma insulin, which confirms our previously reported greater increase in plasma insulin after oral glucose administration (28), which together with the unchanged insulin sensitivity (18) contributes to the anabolic condition of the mother at this stage of gestation, when fetal growth is small and his metabolic demands are still low. This is also concurrent with the small decline in plasma glucose and the lack of change in plasma glycerol with 48 h food deprivation in the 14 day pregnant rat. In fact the latter contrasts with the higher plasma glycerol levels seen in these animals when fed, which together with the unchanged plasma NEFA levels would indicate a decreased utilization of the former rather than an enhanced production due to augmented adipose tissue lipolytic activity. Since glycerol is known to be the main substrate for gluconeogenesis (53), it is proposed that the hyperinsulinemia in the fed 14 day pregnant rats would have inhibited liver gluconeogenesis and thus restraining the consumption of glycerol for this pathway. Adipose tissue lipolysis also appears to be restricted in the 48 h food deprived 14 day pregnant rat as compared to the fed animals at the same time of gestation and to the other food deprived rats studied, as suggested by the decline in plasma TAG.

The metabolic picture completely switches to a catabolic condition in the 20 day pregnant rat and is specially manifested under the 48 h food deprivation conditions, where maternal glycemia reach a clear hypoglycemic status that even induces a decline in fetal plasma glucose levels. Since the fetus cannot perform gluconeogenesis (30), its glucose levels directly depend on those crossing the placenta in a concentration dependent manner (17). Adipose tissue lipolytic activity appears highly augmented in the 48 h food

deprived pregnant rat, as indicated by the high increase in plasma glycerol and NEFA levels; although the fate of these two products differ. Glycerol crosses the placenta in very small proportion (16), which justifies the stable glycerol levels seen in the fed and the 48 h food deprived fetus. Maternal glycerol is used as a preferential substrate for gluconeogenesis (53, 54), and thus the fetus indirectly benefits from this lipolytic product, particularly under fasting conditions where maternal hypoglycemia is restraining placental transfer of glucose. The high increase in plasma NEFA levels in the fasted 20 d pregnant rat may also have important implications to the fetus. While NEFA crosses the placenta in a concentration dependent manner (16), supporting their increase in the fetus of the 48 h food deprived mothers as compared to the fed ones, circulating NEFA is partially oxidized and used as substrates for ketogenesis in the liver. The higher arrival of NEFA in the liver of the 48 h fasted 20 day pregnant rat accounts for the huge increment in circulating 3-hydroxy butyrate levels. Since the fetus cannot perform ketogenesis (7, 37, 50), the fact that 48 h food deprived fetal plasma 3-hydroxy butyrate reaches the same levels as its mother's confirms the easy transfer of maternal ketones through the placenta (16). The fetus also benefits from this high arrival of ketone bodies from maternal circulation since it is capable of using them as both oxidative and lipogenetic substrates (34, 35, 43).

Furthermore, NEFA may be subject to esterification for the synthesis of acylglycerols in maternal liver, a pathway reported to be significantly enhanced in the food deprived 20 day pregnant rat (52), therefore contributing to the increase in both maternal hypertriglyceridemia and liver TAG concentration seen here under food deprived conditions. Both of these changes may also be influenced by the changes found in LPL activity in both lumbar adipose tissue and liver, which clearly differed from those found in any of the other experimental groups studied. The decline in adipose tissue LPL activity seen in the fed 20 day pregnant rat, which is known to be caused by the insulin resistant condition (38, 39), is though to decrease the peripheral utilization of TAG and therefore contribute to their increase in plasma. Under food deprived conditions, the decline in lumbar adipose tissue LPL activity was even smaller in the 20 day pregnant rat than in any of the other groups studied, and the enhanced hypertriglyceridemia must therefore be the result of an enhanced liver production of TAG rather than a decreased utilization. Besides the enhanced release of TAG from the liver of the late pregnant rats (45, 48), the increased arrival of NEFA at their livers under food deprived conditions may also be a contributing factor for the greater increase in TAG production. The hepatic TAG accumulation in the food deprived 20 day pregnant rat is coincident with an increase in liver LPL activity, which as previously proposed, must come from extrahepatic sources (24), but would contribute to the liver hydrolysis and uptake of circulating TAG. Different arguments must be proposed to explain the observed fetal TAG changes. Fetal plasma TAG are much lower than in their mothers and do not vary with maternal food deprivation despite their increase in the maternal side, which fits with the placental impermeability for the maternal-fetal TAG transfer (16). However liver TAG was enhanced in the 48 h food deprived fetus mainly due to the greater arrival of NEFA, which are known to be

rapidly driven to the liver once being released by the placenta to fetal plasma where they are bound to a specific oncofetal protein, the alpha-fetoprotein (3, 33).

The observed increment in fetal liver LPL activity is a second positive factor for the enhanced liver TAG concentration, as it promotes both hydrolysis and uptake of circulating TAG. This augmented fetal liver LPL activity demands particular attention because during the perinatal phase there is an induction in liver LPL expression, which is maintained throughout lactation (21, 46). We have previously found that this activity was enhanced with fasting in the newborn liver (14), and this is the first time that maternal food deprivation in the 20 day pregnant rat is also shown to enhance LPL activity in fetal liver, thereby supporting that its regulatory factors are already present at this stage of intrauterine development. In neonates, liver LPL was shown to be modulated similarly to adult heart LPL (36), with hypoglycemia and subsequent hormonal changes playing a key role. Therefore, the profound hypoglycemia found here in the fetus of food deprived late pregnant rats could contribute to the increase in liver LPL activity. Yet another possibility would be the NEFA-mediated induction of liver PPARα, of which LPL is one of its target genes (2, 42). We have previously found that already in 10 day old rats, increments in plasma NEFA caused by oral intralipid administration induced PPARα expression and its target genes (32), thus it is proposed that a similar mechanism may already be active in the fetus of the food deprived late pregnant rat. Further specific experiments need to be performed to explore these possibilities.

In short, present findings indicate that during the first two thirds of gestation in the rat, and particularly at 14 day of pregnancy, the response to 48 h food deprivation is milder than in virgin rats, allowing a maximal preservation of maternal metabolic stores when fetal growth is still very slow. However, the response to 48 h food deprivation in the 20 day pregnant rats is enhanced, which despite facilitating the availability of substrates to the fetus, is unable to preserve normal fetal development. In fact, apart from modifying the levels of metabolites like glucose, ketone bodies and NEFA that cross the placenta in the same direction as in the mother, the fetus itself is able to respod to this metabolic insult, as shown by the increase in liver LPL activity.

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Puesto que durante la gestación, la madre cambia de una situación anabólica a otra catabólica, el presente trabajo ha estado dirigido a determinar el efecto de 48 h de ayuno en ratas de 7, 14 y 20 días de gestación y en vírgenes controles. El ayuno produce mayor disminución del peso corporal libre de conceptus en las ratas preñadas que en las vírgenes y el peso de los fetos de 20 días disminuye con el ayuno de sus madres. El descenso en la glucosa plasmática con el ayuno es máximo en las ratas preñadas de 20 días. La insulinemia es máxima en las ratas preñadas de 14 días y disminuye en todos los grupos con el ayuno, aunque de forma no

significativa en las preñadas de 7 días. El ayuno incrementa los niveles de glicerol solo en las ratas vírgenes y en las preñadas de 20 días. Los niveles de NEFA y 3-hidroxibutirato plasmáticos aumentan con el ayuno en todos los grupos, siendo el efecto mayor en las preñadas de 20 días. En los fetos de 20 días, los niveles de glucosa, NEFA y triglicéridos son menores que en sus madres, y el ayuno produce un mayor descenso en la glucosa plasmática, con aumento tanto de NEFA como de 3-hidroxibutirato. La concentración de triglicéridos hepáticos no varía entre los grupos de animales alimentados, mientras que el ayuno la aumenta en todas las ratas preñadas y en los fetos, siendo el efecto mayor en las preñadas de 20 días. La actividad de lipoproteína lipasa (LPL) en tejido adiposo es menor en las ratas preñadas de 20 días alimentadas que en cualquiera de los otros grupos, y el ayuno produce un descenso generalizado. En hígado, los valores de LPL son muy bajos en todos los grupos de animales alimentados, aumentando de forma significativa con el ayuno solo en las preñadas de 20 días. Un incremento significativo en la actividad LPL se observa en el hígado de los fetos de madres en ayunas, alcanzando valores superiores a éstas. Por tanto, la respuesta al ayuno varía con el tiempo de gestación, siendo mas baja a mitad de la gestación y máxima al final. A su vez, aunque los fetos responden al ayuno en la misma dirección que sus madres para la mayoría de los parámetros estudiados, presentan una respuesta específica en cuanto a la actividad de LPL hepática.

Palabras clave: Ayuno, Gestación, Rata, Triglicéridos, Hígado, Lipoproteína lipasa, Tejido adiposo.

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