In: FETAL AND NEONATAL PHYSIOLOGY, R.A.Polin and W.W.Fox, eds., W.B. Saunders Co., Philadelphia, 1992.

SECTION VII

LIPID METABOLISM IN THE FETUS AND NEWBORN

MARGIT HAMOSH

This section aims to provide the reader with recently acquired information on many topics concerning lipid transport and metabolism in the perinatal period. Several chapters address specifically the fetal period, covering the subjects of placental transport of fatty acids and ketone bodies, lipid accretion in the fetus and neonate, as well as fat catabolism. Because of the importance of lipids in growth in general (as building blocks of tissue cells and as a major energy source of the newborn) and in the specific development and function of specialized organs such as the brain and retina, several chapters address these topics.

The need to maintain very low-birth weight infants on parenteral nutrition led to the development of lipid emulsions similar in structure and composition to naturally produced chylomicrons. Although, in general, these lipids are well tolerated, there are clinical complications in some infants. Therefore, two chapters of this section address this specific topic. We hope that this section will be informative as well as thought provoking, thus contributing not only new information but also stimulating further research in this interesting field.

Chapter 25

Placental Transport of Free Fatty Acids, Glycerol, and Ketone Bodies

EMILIO HERRERA, MIGUEL ANGEL LASUNCIÓN and MIRYAM ASUNCIÓN

LIPID METABOLISM IN THE MOTHER AS SOURCE OF FREE FATTY ACIDS, GLYCEROL, AND KETONE BODIES FOR THE FETUS

During the first part of gestation, there is fat accumulation in the mother[1,2] that is sustained by the combined effects of hyperphagia, enhanced lipogenesis, and unmodified or even increased extrahepatic lipoprotein lipase activity.[3,4] The tendency to accumulate fat ceases during late gestation[1,2,5,6] because the maternal lipid metabolism changes to a catabolic condition as shown by increased adipose tissue lipolysis[7,8] and reduced uptake of circulating triglycerides.[9] The reduced uptake is secondary to decreases in adipose tissue lipoprotein lipase activity,[10-13] which are unaccompanied by a change of the enzyme activity in the skeletal muscles.[14] These changes, together with hepatic overproduction of triglycerides[15-17] and the enhanced absorption of dietary lipids[18] are responsible for the marked progressive increase in maternal circulating triglycerides occurring during late gestation.[19-28] Major changes in maternal lipid metabolism are summarized in Figure 25–1. This figure demonstrates

Acknowledgments: Present work was carried out in part with a grant from the U.S.–Spain Joint Committee for Scientific and Technological Cooperation (No. CCA-8510/061).

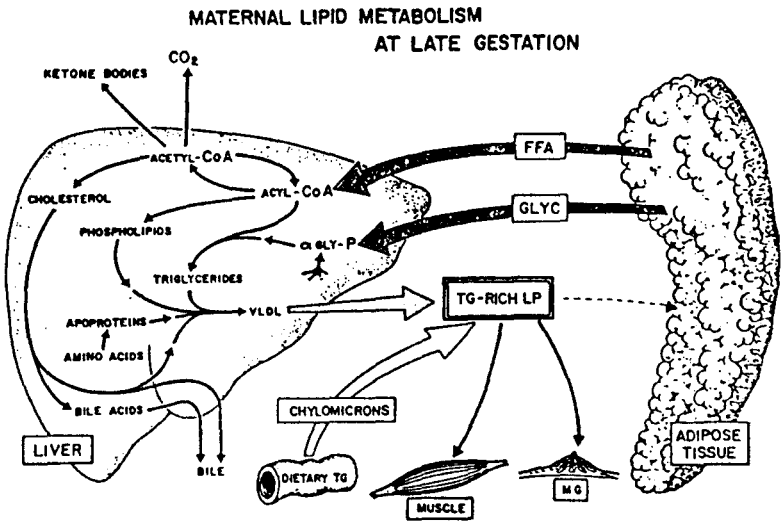

Figure 25–1. Summary of major changes in maternal lipid metabolism at late gestation. FFA = free fatty acids; TG-RICH LP = triglyceride-rich lipoproteins; TG = triglycerides; MG = mammary gland; VLDL = very low density lipoproteins; Glyc = glycerol. (Adapted from Herrera E, Gomez-Coronado D, Lasuncion MA: Lipid metabolism in pregnancy. Biol Neonate *51*:70–77, 1987. S. Karger AG, Basel.)

the changes occurring in adipose tissue, liver, and intestinal activity which are responsible for the physiologic increase in circulating free fatty acids (FFA), glycerol, and triglyceride-rich lipoproteins (VLDL and chylomicrons) during late gestation. If the pregnant woman is adequately nourished, her ketosis is not different from that in nonpregnant subjects.[25, 29]

With the exception of glycerol used in gluconeogenesis[30, 31] and the circulating triglyceride uptake by mammary gland prior to parturition,[11, 12] no part of the increase in circulating lipidic components seems to directly benefit her metabolic needs. This increase, however, may benefit the fetus since this part of the gestational period coincides with the rate of maximal fetal accretion, a time when the substrate, metabolic fuel, and essential component requirements of the fetus are greatly enhanced. The lipid component may also constitute a "floating" fuel store for both mother and fetus, easily accessible under conditions of food deprivation, and explain the well-known finding of enhanced ketogenesis in the mother under fasting conditions.[23, 25, 32, 33] This hypothesis is supported by data demonstrating an increased arrival of FFA in the liver as a result of greatly enhanced adipose tissue lipolysis[7, 8] and by studies reporting an increase in liver LPL activity[13, 34] which would facilitate the maternal liver using circulating triglycerides as ketogenic substrates.

The enhanced ketone body arrival in fasted maternal tissues allows the ketone bodies to be used as metabolic fuels and may spare other more limited and essential substrates, such as amino acids and glucose, for transport to the fetus. The fetus also receives maternal ketone bodies through the placenta, and their use plays an important role in fetal metabolic economy under conditions of maternal food deprivation. Augmented lipolytic activity also increases maternal circulating glycerol levels.[30] Glycerol can be used as a gluconeogenic substrate[31] and therefore contribute to the maintenance of fetal and maternal glucose production. Metabolic adaptations found in the mother during starvation are summarized in Figure 25–2.

Transfer of Lipidic Products to the Fetus

This section focuses on the mechanism and control of placental lipid transfer. Understanding FFA, glycerol, and ketone body placental transfer as well as their respective metabolic fates in the fetus provides an insight into the effect on the fetus of these persistent increments in maternal circulating lipid levels.

Figure 25–3 summarizes the comparison of plasma levels of these metabolites in virgin as well as 24-hour fasted late pregnant rats and their fetuses. Whereas fetal FFA and glycerol levels are present in much lower concentrations than in their mothers, ketone bodies are found in similar amounts in the fetus and mother. These maternal-fetal concentration differences reflect the efficiency or magnitude of the placental transfer process.

Maternal-fetal nutrient transfer through the placenta may be accomplished by means of different mechanisms, including facilitated diffusion, active transport, and simple diffusion.[35, 36] Simple diffusion seems to be the common and unique mechanism for the lipidic-derived moieties, although some specific and differential aspects must be considered. Simple diffusion is carried out from a high to a low concentration region, and the rate of movement is directly proportional to the concentration gradient as described by Fick's law:

$$J = D\frac{dc}{dx}$$

where J is the transfer rate, D the diffusion coefficient, and dc/dx the chemical gradient. The rate of transfer is therefore a direct function of the concentration gradient and decreases with molecular size and hydrosolubility.[37] In the specific condition of the placental transfer, there are other factors that also participate in the efficiency of nutrient transfer,[37, 41] such as the uterine and umbilical blood flows, the intrinsic placental metabolism (utilization versus production), and structural characteristics of the placental barrier. As might be expected, some of these factors, such as blood flow, affect the transfer of any nutrient crossing by passive diffusion in a similar fashion but other factors have varying effects on nutrient transport and require specific consideration.

Free Fatty Acids

The fetus not only requires essential fatty acids from the mother to support growth[44] and brain development[45] but

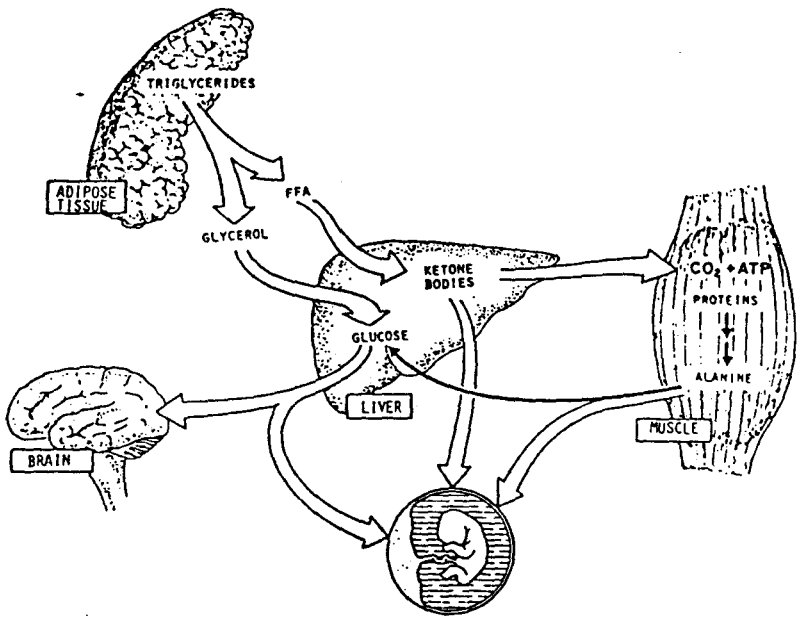

Figure 25–2. Maternal response to starvation. Enhanced adipose tissue lipolysis increases the availability in the liver of glycerol to be used as a preferential substrate for gluconeogenesis and of free fatty acids (FFA) for ketone body synthesis. By this mechanism, the mother conserves other gluconeogenic substrates, such as alanine, and ensures the adequate availability of fuels and metabolites to the fetus. (From Herrera E, Gomez-Coronado D, Lasuncion MA: Lipid metabolism in pregnancy. Biol Neonate *51*:70–77, 1987. S. Karger AG, Basel.)

also requires nonessential lipids as important substrates during early postnatal life.[46] This is especially true in species such as the guinea pig and human where body fat at term represents a substantial percentage of body weight (10 and 16%, respectively),[47] and de novo fatty acid synthesis by fetal tissues cannot fulfill fetal requirements. Using the sheep[48] and the rat,[49] early studies on the placental transfer of lipids suggested that very few fatty acids were transferred from the mother to the fetus. However, investigations in the rabbit,[50] the guinea pig,[51, 52] primates,[53] and rat[54] indicate that the amount of fatty acids crossing the placenta exceeds that needed to fulfill lipid storage requirements.[55]

Fatty acids, like other fats, are relatively insoluble in water and must be transported in the blood either as albumin-bound FFA or in their esterified form as triglycerides, phospholipids, and esterified cholesterol, which are associated with other lipids and proteins in the form of lipoproteins. Therefore, maternal FFA (esterified fatty acids that have been hydrolyzed at the placental level) and unmodified lipoproteins are the potential sources of fatty acids that cross to the fetal side.

As may be expected, there is considerable species variation in placental fatty acid transfer. In general, fatty acid transfer is low in those species with a placental barrier constitued by several maternal as well as fetal cell layers (sheep, pig, and cat), whereas in those species with a simple barrier of only fetal layers (man, rabbit, rat, and guinea pig), the net flux can be high. In these species, the fatty acid mixture entering the fetal circulation from the placenta reflects the maternal free fatty acid concentrations of different fatty acids,[56] with the common exception of arachidonic acid.

Higher proportions of arachidonic acid in fetal compared with maternal plasma have been found in both ruminant and nonruminant species.[58–61] Since significantly higher proportions of arachidonic acid have been consistently noted in the placenta when compared with the levels in maternal lipid fractions,[57] it has been suggested that placental arachidonic acid synthesis is important in the supply of this fatty

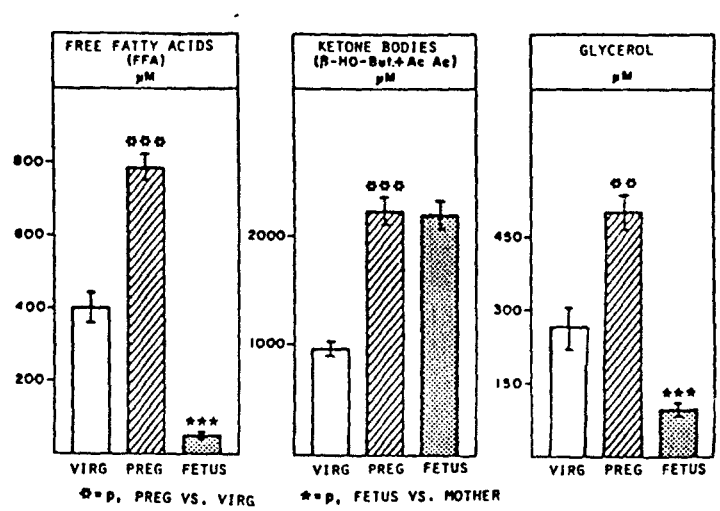

Figure 25–3. Concentration of free fatty acids, ketone bodies, and glycerol in plasma of 48-hour starved virgin rats and 48-hour starved 19-day pregnant rats and their fetuses. (From Herrera E, Gomez-Coronado D, Lasuncion MA: Lipid metabolism in pregnancy. Biol Neonate *51*:70–77, 1987. S. Karger AG, Basel.)

acid to the fetus. An active desaturation and elongation system is required to synthesize arachidonic acid from the active form of linoleic acid (linoleyl-CoA). Such a system has been described in the placenta of the sheep,[62, 63] and an active incorporation of arachidonic acid into placental phosphoglycerides has been noted in both the ovine[64] and human placenta,[65] supporting such a preferential role for the placenta in the synthesis, selective sequestration, and supply of arachidonic acid to the fetus.

Since current evidence suggests that fatty acids are not selectively transferred across the placenta, and other essential and nonessential fatty acids seem to use a common transfer mechanism, ^{14}C-palmitic acid has normally been used by most investigators to study maternal-fetal transfer of both essential and nonessential fatty acids. As indicated above, the quantity of fatty acid transferred varies considerably between species,[66] but in all instances, the system is grossly regulated by the transplacental nonesterified fatty acid gradient.

To study the mechanism of transfer of FFA across the human placenta, an *in vitro* system using perfused placenta and/or cultured trophoblast cells must be employed. A more direct approach, however, may be used in experimental animals. In the 20-day pregnant rat, we used radiolabelled palmitate to study the transfer of fatty acids *in vivo*. In this technique ^{14}C palmitate is infused through the left uterine artery for 20 minutes. The amount of label appearing in the placentae and fetuses from the left uterine horn are compared with that found in the placentae and fetuses from the right horn.[67, 71] While the left uterine horn receives the tracer directly, it reaches the right horn after dilution in the mother's circulation, so the amount of substrate transferred to the fetus can be calculated as a function of the values for the maternal FFA concentration, the difference of radioactivity in fetuses between the left and right uterine horns, and the left uterine blood flow. As shown in Figure 25–4, the estimated FFA transfer was significantly higher than zero (indicating that it was substantial), even though the absolute value of FFA transfer was lower than the level previously found for other compounds in earlier studies: glucose, 127 nmoles/min × gm fetal birth weight (bw); alanine, 23 nmoles/min × gm fetal bw; and glycerol, 1 nmol/min × gm fetal bw.[71] When the concentration of ^{14}C-lipids retained in the placentae after (1-^{14}C)-palmitate infusion was measured, it was found that their level (99±38 nmoles/gm/min) was considerably higher than in the fetuses. Of those ^{14}C-lipids incorporated into the placenta, 49±3% corresponded to esterified fatty acids, indicating that a certain proportion of the FFA that reach the placenta is actively esterified. We do not know whether fatty acid esterification participates in the FFA transfer process, but an active placental capacity to form esterified fatty acids from maternal FFA also has been described in other species[72, 73] and in man.[74, 75] The presence of an active enzymatic glyceride hydrolytic system (phospholipase and triacylglycerol lipase), ensuring a rapid triglyceride and phospholipid turnover, would point to an esterification/hydrolysis cycle in the placental cells as one type of placental FFA transport. A mechanism such as this was proposed by Szabo et al[76] and Hummel et al[77] several years ago.

Maternal circulating triglycerides have also been shown to contribute FFA to fetal circulation in the rat,[78–79] the rabbit,[80] the guinea pig,[81, 82] and man,[83] although no studies have detected the passage of intact triacylglycerol across the placenta. We applied the "in situ" uterine artery infusion technique[67] described above to test the potential transfer of 1-^{14}C-triolein incorporated into purified VLDL.[71] As also shown in Figure 25–4, we were unable to detect any significant transfer of the label into the fetus.

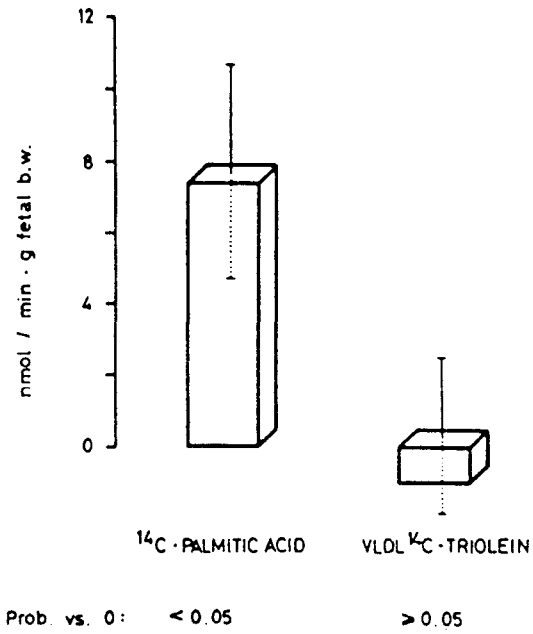

Figure 25–4. Estimation of placental transfer of palmitic acid and VLDL-triolein in the 20-day pregnant rat. Placental transfer to the fetus was determined by measuring the radioactivity appearing in fetuses after infusing each of the ^{14}C-labeled substrates through the left uterine artery and making proper correction of the data for specific activity dilution of the tracer and uterine blood flow, as previously described (see ref. 69).

Lipoprotein lipase activity has been detected in the placentae of all the species studied.[71, 84–87] Placental triglyceride hydrolysis and direct transfer of the released nonesterified fatty acids to the fetus has been proposed, but direct studies in in situ perfused guinea pig placentae have shown that this accounts for a very small percentage of all the fatty acid transferred to the fetus.[82] These data indicate that under normal conditions, the contribution of maternal triglyceride-rich lipoproteins that are broken down by lipoprotein lipase in the placenta is of minor quantitative importance as a source of fatty acids for the fetus. However, under conditions of exaggerated maternal hypertriglyceridemia, this fatty acid supply system from esterified maternal fats may be greatly enhanced in the presence of sustained placental lipoprotein lipase activity. This mechanism has been proposed to occur in streptozotocin-induced diabetic rats.[79, 88] In 20-day old pregnant diabetic rats with varying degrees of glucose intolerance, a significant linear correlation is seen when the maternal versus fetal plasma triglyceride plasma levels are compared (unpublished results). This finding agrees with the augmented levels of essential fatty acids in the circulation of fetuses from hypertrigliceridemic diabetic pregnant rats[79] and supports either the contribution of maternal triglycerides to fetal lipids or the augmented placental transfer of FFA owing to the increased maternal-fetal FFA gradient, or both.[79] These changes may therefore contribute to the well-known fat accumulation in newborns with diabetic mothers.

Figure 25–5 outlines the placental role in maternal-fetal fatty acid transport under normal conditions during late

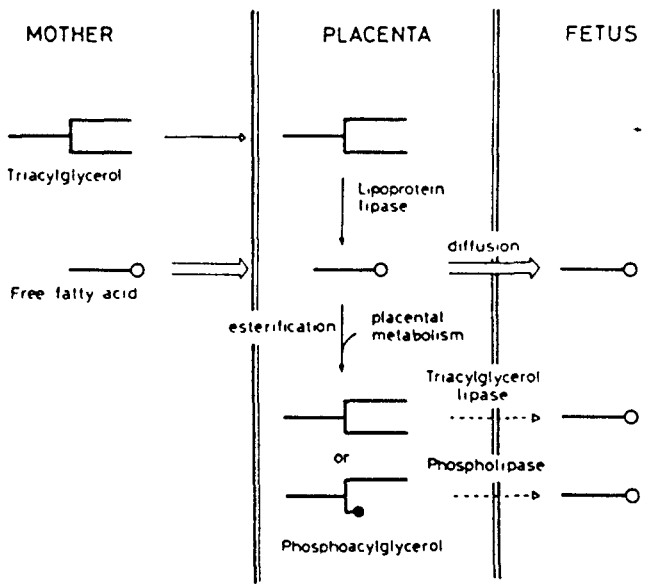

Figure 25–5. Schematic representation of the placental transfer of fatty acids to the fetus.

gestation, where simple direct diffusion constitutes the major process. No attention has been given to modifications of this picture according to each species' duration of gestation (which would modify the degree of maternal hyperlipidemia and placental structural and functional maturation) and to differences caused by pathologic conditions.

Glycerol

As a result of the active lipolytic activity of maternal adipose tissue, plasmatic glycerol levels are consistently elevated during late gestation.[90, 91] The values for plasma glycerol concentration are normally higher than in the fetus (see Fig. 25–3), but there are some interspecies differences. The maternal-fetal glycerol gradient is greater in those species with epitheliochorial placenta (ruminants)[90, 91] than in those with a hemochorial placenta.[91, 93]

There is little experimental data on placental transfer of glycerol in any species. Although the molecular characteristics of glycerol are adequate for an easy placental transfer (low weight and uncharged molecule), glycerol transfer is notably lower than for other metabolites with similar molecular characteristics such as glucose or L-alanine.[71, 94] In contrast with the carrier-mediated process used for these two metabolites, the placental glycerol transfer is accomplished by simple diffusion.[89, 95] In the sheep fetus, the umbilical glycerol balance indicates that fetal uptake is very low, accounting for no more than 1.5% of the total oxygen consumption of the fetus.[90] In man it has not been possible to detect a transfer of glycerol from mother to fetus in spite of its favorable gradient.[91] When comparing different substrates and by using the in situ infused placental technique in the rat, we have found that the transfer of glycerol is much lower than that of glucose and alanine, but higher than FFA.[71] We have also found that the fetal-placental unit converts glycerol into glyceride glycerol,[92] and this utilization may actively contribute to maintenance of the high glycerol gradient consistently found between maternal and fetal blood.[33, 91, 94]

Accelerated turnover of maternal glycerol seems to be influenced by the high liver glycerolkinase activity that facilitates its rapid phosphorylation and subsequent conversion into glucose.[30, 31] Although this mechanism indirectly benefits the fetus by providing glucose from this product of maternal adipose tissue breakdown (see Fig. 25–2), it may limit the availability of enough glycerol molecules to permit transfer to the fetus. Results summarized in Figure 25–6 support this hypothesis. Hepatectomy normally produces an increase in the plasma levels of glycerol because of a reduction in glycerol utilization secondary to absence of the liver, the major receptor organ for this metabolite.[96] As shown in Figure 25–6, in the case of pregnant rats, hepatectomy and nephrectomy cause a smaller increase in plasma glycerol levels than in nonpregnant animals. This difference cannot be interpreted as reduced lipolytic activity in the pregnant rat since plasma FFA, the other lipolytic product, increases more than in nonpregnant animals. It might, however, be interpreted as the result of an augmented transfer of glycerol to the fetus since glycerol levels in fetal plasma increase significantly under these conditions of maternal hepatectomy and nephrectomy.[96]

Consequently, placental glycerol transfer seems to be limited by the effective and rapid utilization of this substrate for gluconeogenesis by the liver and the kidney cortex of the mother. Although the fetal-placental unit actively uses glycerol, which helps to maintain a favorable transfer gradient, its quantitative and physiologic role in the fetus,

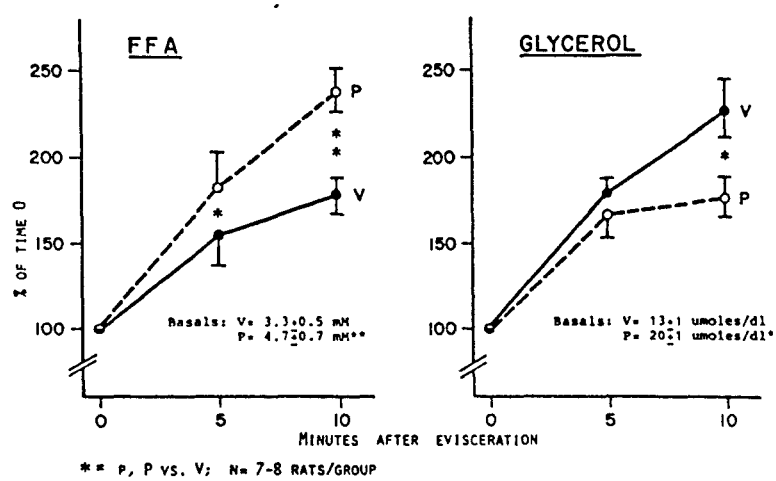

Figure 25–6. Effect of hepatectomy-nephrectomy on plasma free fatty acid and glycerol in virgin (V) and 20-day pregnant rats (P). Experimental details are as indicated in ref. 96.

except as a preferential substrate for fetal liver glyceride glycerol synthesis,[92] seems to be limited under normal conditions. However, under conditions of markedly elevated maternal glycerol levels, the placental transfer of glycerol could become much more relevant in the supply of substrates to the fetus.

Ketone Bodies

Although under physiologic conditions the plasma levels of ketone bodies in the fed late pregnant mother are unchanged, under fasting[23, 25, 32, 98, 102] or diabetic[103, 104] conditions, they are greatly increased, coincident with enhanced delivery of FFA to the liver. As noted earlier, when the supply of glucose is limited (e.g., hypoglycemia and/or reduced insulin levels or sensitivity), ketone bodies are used by some maternal tissues (for example, skeletal muscle) as alternative substrates. Ketone bodies can also cross the placental barrier and be used as energetic fuels and lipogenic substrates by the fetus.[105, 109]

With regards to its physiologic role, maternal ketonemia in the poorly controlled pregnant diabetic patient, with or without acidosis, has been associated with an increased still birth rate, an increased incidence of congenital anomalies, and impaired neurophysiologic development in the infant.[104, 109-111] These effects are thought to be secondary to placental transfer of maternal ketones to the fetus.[112]

In addition to size and lipid solubility, molecular charge has an important effect on placental membrane permeability. At pH 7.4, most of the molecules of the two main ketone bodies, β-hydroxybutyrate and acetoacetate, are present in dissociated or ionized form, retarding their diffusion across the placenta. In spite of this, in all species studied (man[92, 101, 113-115] rat,[23, 32, 116] and sheep[100, 112]), increments in maternal ketone bodies are accompanied by increments in fetal plasma levels, indicating efficient placental transfer because fetal liver ketogenesis is practically negligible.[117]

Placental transfer of ketone bodies is always carried out by simple diffusion and has a high unspecific component,[89] but its efficiency varies between species. Whereas the maternal-fetal gradient for ketone bodies is above 10 in the sheep,[100, 112] in man it is about 2,[92] and in the rat, it is close to 1[23, 33, 116] (see Fig. 25–3). This indicates that the amount of ketone bodies crossing the placenta is much lower in ruminant than in nonruminant species. It has even been proposed that in the fasted sheep, the contribution of ketone bodies to the fetal oxidative metabolism accounts for only 2 to 3% of the total oxygen consumption;[100, 118] however, based on experiments using ^{14}C-labelled substrates, it has been shown that β-hydroxybutyrate adequately replaces the glucose deficit in the placenta, fetal brain, and liver during fasting hypoglycemia in the rat.[106] This suggests a much greater contribution of ketone bodies to the fetal oxidative metabolism in the fasted nonruminant.

Key enzymes for ketone-body utilization, namely 3-hydroxybutyrate dehydrogenase (E.C. 1.1.1.30), 3-oxoacid-CoA transferase (E.C. 2.8.3.5), and acetyl-CoA acetyltransferase (E.C. 2.3.1.9), have been found in the brain and other tissues in both the human and rat fetus.[107, 119, 121] *In vitro*, both the human[109] and rat brain[105, 106] oxidize β-hydroxybutyrate in a form that is dependent on substrate concentration and not on the maternal nutritional state. Other fetal tissues known to oxidize ketone bodies are kidney, heart, liver, and placenta,[107, 122] and some are even known to use ketone bodies as substrates for fatty acid and cholesterol synthesis, as has been shown in the rat brain, liver, placenta, and lung after the *in vivo* administration of ^{14}C-β-hydroxybutyrate to pregnant animals.[123] Conditions of maternal hyperketonemia, such as starvation during the last days of gestation[124] or high fat feeding,[121] increase the activity of these ketone-body metabolism enzymes in fetal tissues (brain, liver, and kidney). Such a change is especially evident in the fetal brains from starved late pregnant rats[124] and may represent an important fetal adaptation to guarantee brain development under these conditions through ketone body consumption since fetal brain weight is better preserved than other fetal organ weights.[124]

References

1. Hytten FE, Leitch I: The Physiology of Human Pregnancy, 2nd ed. Oxford, Blackwell Scientific, 1971.
2. Beaton GH, et al: Protein metabolism in the pregnant rat. J Nutr *54*:291, 1954.
3. Knopp RH, et al: Lipid metabolism in pregnancy. II. Postheparin lipolytic activity and hypertriglyceridemia in the pregnant rat. Metabolism *24*:481, 1975.
4. Kinnunen PK, et al: Activities of post-heparin plasma lipoprotein lipase and hepatic lipase during pregnancy and lactation. Eur J Clin Invest *10*:469, 1980.
5. López-Luna P, et al: Body fat in pregnant rats at mid and late gestation. Life Sci *39*:1389, 1986.
6. Freinkel N: Metabolic changes in pregnancy. *In* Wilson JD, Foster DW (eds): Williams' Textbook of Endocrinology, 7th ed. Philadelphia, WB Saunders, 1985, pp 438–451.
7. Knopp RH, et al: Carbohydrate metabolism in pregnancy. VIII. Metabolism of adipose tissue isolated from fed and fasted pregnant rats during late gestation. J Clin Invest *49*:1438, 1970.
8. Chaves JM, Herrera E: In vitro glycerol metabolism in adipose tissue from fasted pregnant rats. Biochem Biophys Res Commun *85*:1299, 1978.
9. Herrera E, et al: Lipid metabolism in pregnancy. Biol Neonate *51*:70, 1987.
10. Otway S, Robinson DS: Significance of changes in tissue clearing-factor lipase activity in relation to the lipemia of pregnancy. Biochem J *106*:677, 1968.
11. Hamosh M, et al: Lipoprotein lipase activity in adipose tissue and mammary tissue and plasma triglyceride in pregnant and lactating rats. Biochim Biophys Acta *210*:473, 1970.
12. Ramírez I, et al: Circulating triacylglycerols, lipoproteins and tissue lipoprotein lipase activities in rat mothers and offspring during the perinatal period: effect of postmaturity. Metabolism *32*:333, 1983.
13. Herrera E, et al: Role of lipoprotein lipase activity on lipoprotein metabolism and the fate of circulating triglycerides in pregnancy. Am J Obstet Gynecol *158*:1575, 1988.
14. Champigny O, Hitier Y: Lipoprotein lipase activity in skeletal muscle and brown adipose tissue of pregnant and lactating rats. J Nutr *117*:349, 1987.
15. Humphrey JL, et al: Lipid metabolism in pregnancy. VII. Kinetics of chylomicron triglyceride removal in the fed pregnant rat. Am J Physiol *239*:E81, 1980.
16. Kalkhoff RK, et al: Influence of pregnancy and sex steroids on hepatic triglyceride biosynthesis. Diabetes *21*(Suppl 1):365, 1972.
17. Wasfi I, et al: Hepatic metabolism of (1-14C) oleate in pregnancy. Biochim Biophys Acta *619*:471, 1980.
18. Argilés J, Herrera E: Appearance of circulating and tissue 14C-lipids after oral 14C-tripalmitate in the late pregnant rat. Metabolism *38*:104, 1989.
19. Stemberg L, Dagenais-Perusse P, Dreyfuss M: Serum proteins in parturient mother and newborn: an electrophoretic study. Can Med Ass J *74*:49, 1956.
20. Knopp RH, et al: Carbohydrate and lipid metabolism in normal pregnancy. Food and nutrition board: Laboratory indices of

nutritional status in pregnancy, pp 35–88 National Academy of Sciences, Washington, DC 1978.
21. Russ M, et al: Protein-lipid relationships in human plasma. III. In pregnancy and the newborn. J Clin Invest *33*:1662, 1954.
22. Konttinen AT, et al: Serum lipid pattern in normal pregnancy and preeclampsia. J Obstet Gynaecol Br Cwth *71*:453, 1964.
23. Scow RO, et al: Hyperlipemia and ketosis in the pregnant rat. Am J Physiol *206*:796, 1964.
24. Desoye G, et al: Correlation of hormones with lipid and lipoprotein levels during normal pregnancy and postpartum. J Clin Endocrinol Metab *64*:704, 1987.
25. Herrera E, et al: Carbohydrate metabolism in pregnancy. VI. Plasma fuels, insulin, liver composition, gluconeogenesis and nitrogen metabolism during late gestation in the fed and fasted rat. J Clin Invest *48*:2260, 1969.
26. Knopp RH, et al: Two phases of adipose tissue metabolism in pregnancy: maternal adaptations for fetal growth. Endocrinology *92*:984, 1973.
27. Knopp RH: Cardiovascular effects of endogenous and exogenous sex hormones over a woman's lifetime. Am J Obstet Gynecol *158*:1630, 1988.
28. Argilés J, Herrera E: Lipids and lipoproteins in maternal and fetus plasma in the rat. Biol Neonate *39*:37, 1981.
29. Rémésy CH, Demigné CH: Adaptation of hepatic gluconeogenesis and ketogenesis to altered supply of substrates during late pregnancy in the rat. J Develop Physiol *8*:195, 1966.
30. Chaves JM, Herrera E: "In vivo" glycerol metabolism in the pregnant rat. Biol Neonate *37*:172, 1980.
31. Zorzano A, et al: Role of availability of substrates on hepatic and renal gluconeogenesis in the fasted late pregnant rat. Metabolism *35*:297, 1986.
32. Scow RO, et al: Ketosis in the rat fetus. Proc Soc Exp Biol Med *98*:833, 1958.
33. Girard J, et al: Fetal metabolic response to maternal fasting in the rat. Am J Physiol 232:E456, 1977.
34. Testar X, et al: Increase with starvation in the pregnant rat of the liver lipoprotein lipase activity. Biochem Soc Transac *13*:134, 1985.
35. Dancis J, Schneider H: Physiology of the placenta. *In* Falkner, Tanner (eds): Human Growth, Vol 1. New York, Plenum Publishing, 1978, pp 355–378.
36. Munro HN, et al: The placenta in nutrition. Annu Rev Nutr 3:97, 1983.
37. Palacín M, et al: Transporte de metabolitos a través de la placenta. Rev Esp Pediatr *40*(3):163, 1984.
38. Yudilevich DL, Sweiry JH: Transport of amino acids in the placenta. Biochim Biophys Acta *822*:169, 1985.
39. Hill EP, Longo LD: Dynamics of maternal-fetal nutrient transfer. Fed Proc 39:239, 1980.
40. Baur R: Morphometric data and questions concerning placental transfer. Placenta suppl 2:35, 1981.
41. Fox H: The correlation between placental structure and transfer function. *In* Chamberlain GVP, Wilkinson AW (eds): Placental Transfer. London, Pitman Medical Ed, 1979, pp 15–33.
42. Morris FH Jr: Placental factors conditioning fetal nutrition and growth. Am J Clin Nutr 34:760, 1981.
43. Meschia G: Substrate availability and fetal growth. *In* Naftolin F (ed): Abnormal Fetal Growth: Biological Bases and Consequences. Berlin, Dahlem Knoferenzen, 1978, pp 221–228.
44. Burr GO, Burr MM: On the nature and role of the fatty acids essential in nutrition. J Biol Chem *82*:345, 1929.
45. Hassam AG, Crawford MA: The differential incorporation of labelled linoleic, Γ-linolenic, dihomo-Γ-linolenic and arachidonic acids into the developing brain. J Neurochem 27:967, 1976.
46. Hull D: Total fat metabolism. *In* Beard, NG (ed): Fetal Physiology and Medicine. London, WB Saunders, 1976, pp 105–120.
47. Widdowson EM: Chemical composition of newly born mammals. Nature (Lond) *166*:626, 1950.
48. James E, et al: A-V differences of free fatty acids and glycerol in the ovine umbilical circulation. Proc Soc Exp Biol Med *138*:823, 1971.
49. Koren Z, Shafrir E: Placental transfer of free fatty acids in the pregnant rat. Proc Soc Exp Biol Med *116*:411, 1964.
50. Elphick MC, et al: Transfer of fatty acids across the rabbit placenta. J Physiol *252*:29, 1975.
51. Hershfield MS, Nemeth AM: Placental transport of free palmitic and linoleic acids in the guinea pig. J Lipid Res *9*:460, 1968.
52. Bohmer T, Havel RJ: Genesis of fatty liver and hyperlipemia in the fetal guinea pig. J Lipid Res *16*:454, 1975.
53. Portman OW, et al: Transfer of free fatty acid across the primate placenta. Am J Physiol *216*:143, 1969.
54. Hummel L, et al: Studies on the lipid metabolism using 14C-1-palmitate in fetal rats. Biol Neonate *24*:298, 1974.
55. Jones CT: Lipid metabolism and mobilization in the guinea pig during pregnancy. Biochem J *156*:357, 1976.
56. Hull D, Stammers JP: Placental transfer of fatty acids. Biochem Soc Trans *13*:821, 1985.
57. Noble RC, et al: The role of the placenta in the supply of essential fatty acids to the fetal sheep: studies of lipid compositions at term. Placenta 3:287, 1982.
58. Portman OW, et al: Transfer of free fatty acids across the primate placenta. Am J Physiol *216*:143, 1969.
59. Satomi S, Matsuda I: Microsomal desaturation of linoleic into Γ-linolenic acid in livers of fetal, suckling and pregnant rats. Biol Neonate 22:1, 1973.
60. Phanteliadis C, Troll U: Fatty acid pattern of the serum lipids (normal values in mothers, placentae and infants). Z Ernahrungswiss *15*:305, 1976.
61. Elphic MC, Hull D: The transfer of free fatty acids across the rabbit placenta. J Physiol *264*:751, 1977.
62. Shand JH, Noble RC: Ag and A6-desaturase activities of the ovine placenta and their role in the supply of fatty acids to the foetus. Biol Neonate *36*:298, 1979.
63. Shand JH, Noble RC: The metabolism of 18:0 and 18:2 (n-6) by the ovine placenta at 120 and 180 days of gestation. Lipids *16*:68, 1981.
64. Shand JH, Noble RC: Incorporation of linoleic and arachidonic acids into ovine placental phospholipids in vitro. Biol Neonate *48*:299, 1985.
65. Kuhn DC, Crawford M: Placental essential fatty acid transport and prostaglandin synthesis. Prog Lipid Res 25(1–4):345, 1986.
66. Hull D, Elphick M: Transfer of fatty acids. *In* Chamberlain GUP and Wilkinson AW (eds): Placental Transfer. London, Pitman Medical, 1979, pp 159–165.
67. Lasunción MA, et al: Method for the study of metabolite transfer from rat mother to fetus. Biol Neonate *44*:85, 1983.
68. Palacín M, et al: Placental formation of lactate from transferred L-alanine and its impairment by aminooxyacetate in the late-pregnant rat. Biochim Biophys Acta *841*:90, 1985.
69. Herrera E, et al: Relationship between maternal and fetal fuels and placental glucose transfer in rats with maternal diabetes of varying severity. Diabetes *34*(suppl 2):42, 1985.
70. Palacín M, et al: Decreased uterine blood flow in the diabetic pregnant rat does not modify the augmented glucose transfer to the fetus. Biol Neonate *48*:197, 1985.
71. Lasunción MA, et al: Maternal factors modulating nutrient transfer to fetus. Biol Neonate *51*:86, 1987.
72. Elphick MC, Hull D: Incorporation "in vivo" of 1-14C-palmitic acid into plancental and fetal liver lipids of the rabbit. Biol Neonate 32:24, 1977.
73. Thomas ChR: Placental transfer of non-esterified fatty acids in normal and diabetic pregnancy. Biol Neonate *51*:94, 1987.
74. Coleman RA: Placental metabolism and transport of lipid. Fed Proc *45*(10):2519, 1986.
75. Coleman RA, Haynes EB: Synthesis and release of fatty acids by human trophoblast cells in culture. J Lipid Res *28*:1335, 1987.
76. Szabo AJ, et al: Triglyceride synthesis by the human placenta. I. Incorporation of labelled palmitate into plancental triglycerides. Am J Obstet Gynecol *115*:257, 1973.
77. Hummel L, et al: Quantitative studies on the metabolism of placental triglycerides and phospholipids in the rat. Acta Biol Med Germ 32:311, 1974.

78. Hummel L, et al: Maternal plasma triglycerides as a source of fetal fatty acids. Acta Biol Med Germ 35:1635, 1976.
79. Shafrir E, Barash V: Placental function in maternal-fetal fat transport in diabetes. Biol Neonate *51*:102, 1987.
80. Elphick MC, Hull D: Rabbit placental clearing-factor lipase and transfer to the fetus of fatty acids derived from triglycerides injected into the mother. J Physiol *273*:475, 1977.
81. Thomas CR, Lowy C: The clearance and placental transfer of free fatty acids and triglycerides in the pregnant guinea pig. J Develop Physiol *4*:163, 1982.
82. Thomas CR, Lowy C: The interrelationships between circulating maternal esterified and non-esterified fatty acids in pregnant guinea pigs, and their relative contributions to the fetal circulation. J Devel Physiol *9*:203, 1987.
83. Elphick MC, et al: The passage of fat emulsion across the human placenta. Br J Obstet Gynaecol *85*:610, 1978.
84. Brenneman DE, Spector AA: Utilization of ascites plasma very low density lipoprotein triglycerides by Ehrlich cells. J Lipid Res *15*:309, 1974.
85. Clegg RA: Placental lipoprotein lipase activity in the rabbit, rat and sheep. Comp Biochem Physiol *69B*:585, 1981.
86. Thomas CR, et al: Studies on the placental hydrolysis and transfer of lipids to the fetal guinea pig. Miller RK, Thied HA (eds): *In* Fetal Nutrition, Metabolism and Immunology: Role of the Placenta. New York, Plenum Press, 1984, pp 135–148.
87. Rotherwell JE, Elphick MC: Lipoprotein lipase activity in human and guinea pig placenta. J Devel Physiol *4*:153, 1982.
88. Knopp RH, et al: Lipoprotein metabolism in pregnancy, fat transport to the fetus, and the effects of diabetes. Biol Neonate *50*:297, 1986.
89. Seeds AE, et al: Comparison of human and sheep chorion laeve permeability to glucose, beta-hydroxybutyrate and glycerol. Am J Obstet Gynecol *138*:604, 1980.
90. James E, et al: A-V differences of free fatty acids and glycerol in the ovine umbilical circulation. Proc Soc Exp Biol Med *138*:823, 1971.
91. Sabata V, et al: The role of free fatty acids, glycerol, ketone bodies and glucose in the energy metabolism of the mother and fetus during delivery. Biol Neonate *13*:7, 1968.
92. Palacín M, et al: Lactate production and absence of gluconeogenesis from placental transferred substrates in fetuses from fed and 48-h starved rats. Pediatr Res *22*:6, 1987.
93. Gilbert M: Origin and metabolic fate of plasma glycerol in the rat and rabbit fetus. Pediatr Res *11*:95, 1977.
94. Battaglia FC, Meschia C: Principal substrates of fetal metabolism. Physiol Rev *58*:499, 1978.
95. Bissonette JM: Studies "in vivo" of glucose transfer across the guinea-pig placenta. Placenta Suppl 2:155, 1981.
96. Mampel T, et al: Hepatectomy-nephrectomy effects in the pregnant rat and fetus. Biochem Biophys Res Commun *131*:1219, 1985.
97. Mampel T, et al: Changes in circulating glycerol, free fatty acids and glucose levels following liver transplant in the pig. Arch Int Physiol Biochim *89*:195, 1981.
98. Mackay EM, Barnes RH: Fasting ketosis in the pregnant rat as influenced by adrenalectomy. Proc Soc Exp Biol Med *34*:682, 1936.
99. Williamson DH: Regulation of the utilization of glucose and ketone bodies by brain in the perinatal period. *In* Camerini-Davalos RA, Cole HS (eds): Early Diabetes in Early Life. New York, Academic Press, 1975, pp 195–202.
100. Morris FH, et al: Umbilical V-A differences of acetoacetate and beta hydroxybutyrate in fed and starved ewes. Proc Soc Exp Biol Med *145*:879, 1974.
101. Kim YJ, Felig P: Maternal and amniotic fluid substrate levels during caloric deprivation in human pregnancy. Metabolism *21*:507, 1971.
102. Felig P, Lynch V: Starvation in human pregnancy: hypoglycemia, hypoinsulinemia, and hyperketonemia. Science *170*:990, 1970.
103. Persson B, Lunell NO: Metabolic control of diabetic pregnancy. Variations in plasma concentrations of glucose, free fatty acids, glycerol, ketone bodies, insulin and human chorionic somatomammotropin during the last trimester. Am J Obstet Gynecol *122*:737, 1975.
104. Drury MI, et al: Pregnancy complicated by clinical diabetes mellitus. Obstet Gynecol *49*:519, 1977.
105. Shambaugh GE III, et al: Fetal fuels I: Utilization of ketones by isolated tissues at various stages of maturation and maternal nutrition during late gestation. Metabolism *26*:623, 1977.
106. Shambaugh GE III, et al: Fetal fuels II: contributions of selected carbon fuels to oxidative metabolism in rat conceptus. Am J Physiol 233:E457, 1977.
107. Shambaugh GE III, et al: Fetal fuels III: ketone utilization by fetal hepatocytes. Am J Physiol 235(3):E330, 1978.
108. Page MA, Williamson DH: Enzymes of ketone-body utilisation in human brain. Lancet *2*:66, 1971.
109. Adam PAJ, et al: Oxidation of glucose and D-hydroxybutyrate by the early human fetal brain. Acta Paediatr Scand *64*:17, 1975.
110. Drew JH, et al: Congenital malformations, abnormal glucose tolerance, and estriol excretion in pregnancy. Obstet Gynecol *51*:129, 1978.
111. Churchill JA, et al: Neuropsychological deficits in children of diabetic mothers. Am J Obstet Gynecol *105*:257, 1969.
112. Miodovnik M, et al: Effect of maternal ketoacidemia on the pregnant ewe and the fetus. Am J Obstet Gynecol *144*:585, 1982.
113. Paterson P, et al: Maternal and fetal ketone concentrations in plasma and urine. Lancet *1*:862, 1967.
114. Smith AL, Scanlon J: Amniotic fluid D(-)-β-hydroxybutyrate and the dysmature newborn infant. Am J Obstet Gynecol *115*:569, 1973.
115. Felig P, Lynch V: Starvation in human pregnancy: hypoglycemia, hypoinsulinemia, and hyperketonemia. Science *170*:990, 1970.
116. Arola LI, et al: Effects of 24 hour starvation on plasma composition in 19 and 21 day pregnant rats and their foetuses. Horm Metab Res *14*:364, 1982.
117. Lee LPK, Fritz IB: Hepatic ketogenesis during development. Can J Biochem *49*:599, 1971.
118. Boyd RD, et al: Growth of glucose and oxygen uptakes by fetuses of fed and starved ewes. Am J Physiol *225*:897, 1973.
119. Page MA, Williamson DH: Enzymes of ketone body utilization in human brain. Lancet *2*:66, 1971.
120. Patel MS, et al: The metabolism of ketone bodies in developing human brain: Development of ketone-body utilizing enzymes and ketone bodies as precursors for lipid synthesis. J Neurochem *25*:905, 1975.
121. Dierks-Ventling Ch: Prenatal induction of ketone-body enzymes in the rat. Biol Neonate *19*:426, 1971.
122. Williamson DH: Ketone body metabolism and the fetus. *In* Van Assche FA, Robertson WB, Renaer MC (eds): Fetal Growth Retardation. Edinburgh, Churchill Livingstone, 1981, pp 29–34.
123. Seccombe DW, et al: Fetal utilization of maternally derived ketone bodies for lipogenesis in the rat. Biochim Biophys Acta *438*:402, 1977.
124. Thaler MM: Effects of starvation on normal development of beta-hydroxybutyrate dehydrogenase activity in fetal and newborn rat brain. Nature New Biol 236:140, 1972.