Expression of genes participating in regulation of fatty acid and glucose utilization and energy metabolism in developing rat hearts

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Lavrentyev, Eduard N., Daifen He, and George A. Cook. Expression of genes participating in regulation of fatty acid and glucose utilization and energy metabolism in developing rat hearts. *Am J Physiol Heart Circ Physiol* 287: H2035–H2042, 2004. First published June 24, 2004; doi:10.1152/ajpheart.00372.2004.—The heart is a unique organ that can use several fuels for energy production. During development, the heart undergoes changes in fuel supply, and it must be able to respond to these changes. We have examined changes in the expression of several genes that regulate fuel transport and metabolism in rat hearts during early development. At birth, there was increased expression of fatty acid transporters and enzymes of fatty acid metabolism that allow fatty acids to become the major source of energy for cardiac muscle during the first 2 wk of life. At the same time, expression of genes that control glucose transport and oxidation was downregulated. After 2 wk, expression of genes for glucose uptake and oxidation was increased, and expression of genes for fatty acid uptake and utilization was decreased. Expression of carnitine palmitoyltransferase I (CPT I) isoforms during development was different from published data obtained from rabbit hearts. CPT Iα and Iβ isoforms were both highly expressed in hearts before birth, and both increased further at birth. Only after the second week did CPT Iα expression decrease appreciably below the level of CPT Iβ expression. These results represent another example of different expression patterns of CPT I isoforms among various mammalian species. In rats, changes in gene expression followed nutrient availability during development and may render cardiac fatty acid oxidation less sensitive to factors that influence malonyl-CoA content (e.g., fluctuations in glucose concentration) and thereby favor fatty acid oxidation as an energy source for cardiomyocytes in early development.

Carnitine palmitoyltransferase; oxidation; translocase; acetyl-CoA

LONG-CHAIN FATTY ACIDS ARE IMPORTANT oxidizable substrates that are used by heart for muscle contraction. The mechanism by which fatty acids enter cardiac myocytes is not entirely clear but appears to occur predominantly by protein-mediated transport. Fatty acid translocase (FAT/CD36) and the fatty acid transporter protein family (FATP) play important roles in transmembrane trafficking of fatty acids. FATP6 is the primary FATP family member expressed in heart (20). FATP6 localizes to the sarcolemma of cardiac myocytes juxtaposed with blood vessels where it colocalizes with FAT/CD36 (11, 25). We have not found any published data concerning the expression of transporter genes during early development of the rat heart. After birth, a dramatic increase in fatty acid oxidation occurs in hearts of several species (22). The mechanisms responsible for this increase in β-oxidation are not completely understood, but increased expression of a carnitine palmitoyltransferase I (CPT I) gene is expected to play a key role (4, 14, 47). CPT I catalyzes the conversion of acyl-CoA to acylcarnitine at the outer mitochondrial membrane to allow transport across the mitochondrial inner membrane. Acyl-carnitine is reconverted to acyl-CoA by CPT II for β-oxidation in the mitochondrial matrix.

Three isoforms of CPT I have been cloned (49, 50, 65). The most widely expressed, CPT Iα (liver isoform), is found in all cells except skeletal muscle and adipocytes, and it is primarily regulated by hormone action (14, 49). The second isoform is CPT Iβ (muscle isoform), which is highly expressed in skeletal muscle, heart, and adipocytes, and it is primarily fuel regulated (14, 65). A recently discovered isoform is the so-called brain-specific isoform, which is designated CPT IC or Iγ (34, 50). Regulation of the first two isoforms is considerably different in terms of enzyme activity and gene expression. Nothing is known about the enzymatic action or regulation of CPT Iγ, but it does bind malonyl-CoA tightly (50). The heart is the only organ known to express two kinetically distinct isoforms, CPT Iα and CPT Iβ, in the same cells. In the neonatal heart, CPT Iα, which is far less sensitive to malonyl-CoA inhibition, contributes a greater fraction to the total CPT activity than it does in the adult heart (4, 13, 44, 47). Increased rates of myocardial β-oxidation during early development have been observed in several mammalian species (4, 13, 44, 47). Malonyl-CoA synthesis in the heart is carried out by two acetyl-CoA carboxylase isoforms (ACC1 and ACC2). Decreased ACC activity results in lower malonyl-CoA levels and stimulation of fatty acid oxidation (1, 26).

One constituent of the 3-hydroxy-3-methylglutaryl (HMG) CoA pathway responsible for ketone body synthesis (41, 53) is HMG-CoA synthase. It has been shown that HMG-CoA synthase mRNA levels increase in the rat liver and kidney slowly during fetal life and markedly after birth (64). The increase in transcription of the CPT I and HMG-CoA synthase genes is carried out through peroxisome proliferator-activated receptor (PPAR)α by long-chain fatty acids (10, 39, 53). PPARγ coactivator-1α (PGC-1α) is a tissue-specific coactivator that enhances the activity of many nuclear receptors and coordinates transcriptional programs important for energy metabolism and homeostasis (10, 37). So far, nothing is known about PPARα and PGC-1α expression in early heart development. The effects of fatty acids on CPT I mRNA expression in rat heart during development have not been examined. Medium-chain acyl-CoA dehydrogenase (MCAD) is accepted as an important regulatory site in the utilization of fatty acids inside...
the mitochondrion and is known to be important in the heart (42).

Glucose enters the heart via two facilitative glucose transporters, GLUT1 and GLUT4 (45). GLUT1 is a major mediator of basal cardiac glucose uptake and normally accounts for ~30% of the total cardiac glucose transporters in the adult heart (17, 35). However, the contribution of GLUT1 to total cardiac glucose transporters is even higher in late fetal life and early neonatal life when protein expression of GLUT4, which is the insulin-controlled isoform, is relatively low (59). However, the relative contribution of the two glucose transporters changes during early postnatal development when GLUT1 protein expression is downregulated but GLUT4 protein expression is upregulated and, consequently, glucose uptake by heart becomes more insulin sensitive (59, 61).

Regulation of glucose oxidation in the rat heart is limited by inhibitory phosphorylation of pyruvate dehydrogenase (PDH) by PDH kinase (PDK). PDK isoforms are activated by NADH+ and acetyl-CoA that are generated by increased rates of fatty acid β-oxidation, whereas pyruvate produced by glycolysis suppresses PDK activity. Three of the four members of the PDK family identified to date are found in adult rat hearts, namely, PDK-1, -2, and -4 (9). Furthermore, in early development, cardiac PDK-4 protein expression was more dramatically upregulated than PDK-1 (60, 61).

Given that it is apparent that there are distinct "predominantly fetal" vs. "predominantly adult" isoforms of key regulatory enzymes of cardiac fuel utilization, several important questions are raised. First, is there dissimilar CPT I isoform expression in the heart during development in different species? Second, is there distinct expression in early development of "predominantly fetal" vs. "predominantly adult" isoforms of PDK, GLUT, or CPT I? Finally, do changes of fuel sources lead to reiteration of the FAT/CD36, FATP6, CPT I, PDK, GLUT, or CPT I? Questions are raised. First, is there dissimilar CPT I isoform expression in the heart during development in different species? Second, is there distinct expression in early development of "predominantly fetal" vs. "predominantly adult" isoforms of PDK, GLUT, or CPT I? Finally, do changes of fuel sources lead to reiteration of the FAT/CD36, FATP6, CPT I, PDK, GLUT, or CPT I?

Materials and Methods

All protocols were approved by the University of Tennessee Health Science Center Animal Care and Use Committee.

Animals. Timed-pregnant Sprague-Dawley female rats (Charles River; Boston, MA) were fed Purina rodent chow ad libitum (Ralston Purina; Richmond, IN). Fetuses were removed and used 1 to 2 days before expected birth. Rat pups ≤45 days old were taken at the same time of day, and hearts were removed using the same protocol. Each age group contained 5–14 animals.

RNA extraction and cDNA synthesis. Hearts were removed and quickly homogenized in 4 ml of RNA STAT-60 (Tel-Test “B”; Friendswood, TX). RNA isolation was carried out according to the manufacturer’s protocol except that we repeated the extraction to decrease the possibility of contamination by genomic DNA. Extracted RNA was treated with DNase I (Ambion). For reverse transcription, we used 2 μg of total RNA. Synthesis of single-stranded cDNA was done using SuperScript reverse transcriptase II-RNase H (Invitrogen). Total CDNA was diluted (1:100) with deionized pyrogen-free water, and was analyzed by real-time quantitative polymerase chain reaction (RTQ-PCR) on an iCycler thermal cycler (Bio-Rad).

RTQ-PCR quantitation of cDNA. Primers were designed to have a size of ~23 bp and ~50% G/C content (Table 1). The target fragment sizes were <100 bp. RTQ-PCR was performed in a total volume of 25 μl that consisted of 12.5 μl of SYBR green PCR Master Mix (Applied Biosystems; Warrington, UK), 2.5 μl of each primer (100 nM), 2.5 μl of deionized pyrogen-free water, and 5 μl of CDNA diluted 1:100 (~50 pmol). For RTQ-PCR, this protocol was used 10 min at 95°C for denaturation, 40 cycles for amplification and temperature annealing at 62°C, and extension at 72°C. A DNA dissociation curve was obtained for all samples to ensure that all amplicons were consistent with the intended targets and to avoid secondary products, which show up as secondary peaks in a first-derivative plot. Single-stranded oligonucleotides of ~100 bp (a few nucleotides longer on each end than the primer-defined target amplicon) were used as standards. A dilution series from 10^-5 to 10^-10 pmol was established for each standard curve. The correlation coefficient and amplification efficiencies were calculated using iCycler software. The correlation coefficient was always >0.98, and PCR efficiency was between 93 and 100% in all experiments. For normalization of RTQ-PCR results, we used 18S RNA as an endogenous control.

Results

Expression of FATP6 and FAT/CD36 genes in rat heart during development. Both of these transporters were expressed in fetal hearts, but the level of expression of FAT/CD36 mRNA was >10-fold greater than that of FATP6. [Six iso-

Table 1. Primer design for real-time PCR with GenBank accession numbers

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ACC, acetyl-CoA carboxylase; CPT, carnitine palmitoyltransferase; FAT, fatty acid translocase; FATP, fatty acid transport protein; GLUT, glucose transporter; HMG, 3-hydroxy-3-methylglutaryl; MCAD, medium-chain acyl-CoA dehydrogenase; PDK, pyruvate dehydrogenase kinase; PGC, peroxisome proliferator-activated receptor coactivator.
forms of the human and mouse FATP family have been described, and FATP6 is thought to be the most abundant in adult heart (see Ref. 20). Six isoforms of rat FATP have also been cloned but not extensively studied. We found this isoform (GenBank accession no. D85100), which we have designated FATP6, to be the most abundant in the rat heart, along with FATP1, even though it is designated in UniGene as rat FATP2 (NP_113924). Expression of both genes increased immediately after birth by 2.78 ± 0.02-fold for FAT/CD36 and 2.92 ± 0.38-fold for FATP6 (Fig. 1). Expression of FATP6 returned to the fetal level by day 7, decreased to one-fifth of the fetal level by day 11, and remained depressed thereafter. It appears that FATP6 may function mostly during the fetal and early newborn periods rather than in adults. FAT/CD36 did not show any significant changes after day 3. Some increase in mRNA expression of both transporters was observed after 20 days.

**Expression of CPT Iα, Iβ, and Iγ isoforms during rat heart development.** All three isoforms of CPT I were expressed in the fetal heart, although CPT Iγ is supposedly a brain-specific isoform (Fig. 2). At birth, expression of CPT Iβ mRNA was elevated by more than twofold, whereas CPT Iα expression increased slightly. However, expression of CPT Iβ mRNA remained elevated for only 3 days. By day 4, CPT Iβ mRNA had decreased to the fetal level of expression and remained at approximately that same level through 45 days. CPT Iα expression increased slowly and reached a peak at 4 days before decreasing below the fetal level of expression by day 20 (the time of weaning). CPT Iγ expression increased only slightly and then decreased after day 9. The amount of CPT Iγ mRNA was on average one-fifth to one-fourth that of CPT Iα. From day 21 of life, when the animals were separated from their mothers, CPT I isoform expression became similar to that of adult animals, in which CPT Iβ is the predominant isoform.

Expression of ACC1 and ACC2 mRNA during rat heart development. Changes in expression levels of the two ACC isoforms were quite different and reflect the control of two opposite processes in the heart: synthesis and β-oxidation of fatty acids (Fig. 3). ACC1 was highly expressed in fetal tissues, but decreased by day 11 to one-fifth of the fetal level. Expression of the ACC2 isoform, which produces malonyl-CoA for CPT I inhibition, was very low in fetuses but quickly increased within the first 4 days of life to four times the level of fetal expression. It was interesting to see that expression of CPT Iβ (a more-sensitive CPT I isoform in the heart to malonyl-CoA inhibition) was decreased by day 4 (highest ACC2 mRNA amount). The level of expression of ACC2 mRNA significantly decreased after day 11 exactly like ACC1 expression.

Expression of MCAD and HMG-CoA synthase 2 mRNA during rat heart development. MCAD and HMG-CoA synthase 2 are located inside mitochondria and are directly involved in the pathway of mitochondrial β-oxidation of fatty acids and conversion of acetyl-CoA to ketone bodies. Increased expression of MCAD and HMG-CoA synthase 2 assumes that increased rates of β-oxidation will occur at these times. HMG-CoA synthase mRNA expression was increased by 7.7 ± 0.8-fold and MCAD was increased by 1.9 ± 0.3-fold.
compared with fetal rates of expression (Fig. 4). After day 4 of life, expression of HMG-CoA synthase was gradually reduced until it reached the fetal level again by 45 days. The increased MCAD expression dipped between days 9 and 20, and then it returned to the elevated level of expression.

Expression of nuclear receptor PPARα and coactivator PGC-1α mRNA. Long-chain fatty acids can stimulate CPT I and HMG-CoA synthase 2 gene expression through the PPARα nuclear receptor (10, 39, 53). PGC-1α plays a role as coactivator of the PPARα receptor. Expression of these genes increased gradually during the first 7–9 days of life (2.4 ± 0.6- and 1.5 ± 0.1-fold, respectively), and both decreased rapidly by day 11 (Fig. 5). It may be very significant that expression of these genes is so elevated within the first 2 wk of postnatal life. It suggests that regulation of β-oxidation in early development may operate by the same mechanisms as in adult animals.

Isoform switching of GLUT1 and GLUT4 during heart development. Our RTQ-PCR data support results published previously (61) in which protein expression of these glucose transporters was measured by Western blot analysis. We observed a similar ratio of GLUT1 to GLUT4 mRNA amounts in the fetal heart (1.2 ± 0.1). The expression of these genes demonstrated a perfect example of isoform switching from the fetal to adult heart with GLUT1 expression continuously decreasing and GLUT4 expression continuously increasing (Fig. 6). The total fold increase in GLUT4 mRNA was approximately ninefold, whereas GLUT1 mRNA expression decreased by more than a factor of 30.

PDK-1 and -4 mRNA expression during early postnatal rat heart development. PDK-1 mRNA expression decreased by one-half by day 2 and then increased back to the fetal level by 1 wk. After 2 wk (suckling-weaning transition), the amount of PDK-1 isoform reached ~2.1 ± 0.3-fold higher than was found in neonatal animals (Fig. 7). Expression of the PDK-4 isoform was quite different. We observed an increase of eightfold at birth with subsequent decreased expression that reached a minimum on day 11 and increased moderately thereafter.

**DISCUSSION**

The heart is able to utilize fatty acids, glucose, lactate, and ketone bodies for energy production. There have been opposing opinions about which energy source is preferable for heart metabolism and cardiac muscle contraction during early development (22, 36, 54). It is clear, however, that adult hearts metabolizing glucose will switch to fatty acids when fatty acids are supplied in isolated perfused hearts (51). It is also clear that during pregnancy, the fetus is supplied through the placenta with carbohydrates and amino acids but very little fat (5, 6, 63). Free fatty acids are poorly transferred across the placenta and are stored in liver and adipose tissue, but there is little fatty

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**Fig. 3.** Expression of two acetyl-CoA carboxylase (ACC) isoforms during rat heart development from fetus to 45 days: ACC1 (A), ACC2 (B), and the ratio of ACC1 to ACC2 (C). Quantities of ACC isoform mRNA are presented as in Fig. 1.

**Fig. 4.** Expression of medium-chain acyl-CoA dehydrogenase (MCAD; A) and 3-hydroxy-3-methylglutaryl (HMG)-CoA synthase 2 (B) genes in rat heart during early development. Quantities of MCAD and HMG-CoA synthase 2 mRNA are presented as in Fig. 1.
acid oxidation by fetal tissues (21, 23). Immediately after birth, the maternal supply of substrates ceases abruptly, and the newborn must withstand a brief period of starvation before being fed at intervals with milk that is high in fat and low in carbohydrate. Lactose and fat are present in a 1:10 ratio in rat milk; lactose is the predominant carbohydrate in the milk, whereas 95% of milk lipids are in the form of triacylglycerol (31). The nutrient content of milk also depends on the stage of lactation. Early milk secretion, colostrum, is characterized by lower lactose and higher fat contents than mature milk (2, 7). During the first two postnatal weeks of rat development, milk is the only food ingested, and the availability of nutrients from milk strongly correlate with their concentration in plasma and their availability for energy metabolism (5, 6, 29). After 2 wk of life, the pups begin to nibble on solid matter including bedding, feces, and food (18, 29). This period is called the suckling-weaning transition. Then, after their eyes open, the pups begin to nibble the solid adult rat diet. The typical food intake rhythm of adult rats begins at 29 days (28, 29) or at 21 days if pups are isolated from their mothers as in this study.

During the immediate postnatal period, plasma glucagon increases and plasma insulin decreases or remains very low (8, 24). In newborn rats, transient neonatal hypoglycemia is observed, which many authors consider to be the factor that triggers glucagon secretion in these animals (24, 43, 66). The large liver glycogen stores accumulated during pregnancy are rapidly mobilized and are exhausted 12 h after delivery (3, 57). After this period, the suckling rats are entirely dependent on glucose supplied by milk and the capacity for gluconeogenesis in liver to maintain the blood glucose concentration in the normal range. Similarly, newborns must develop the pathways for efficient fatty acid oxidation. Free fatty acids, which are supplied from the mobilization of fat stores or from the diet, must be used to spare glucose for tissues that are entirely dependent on this substrate to maintain their physiological function. The ability to synthesize glucose from lactate, pyruvate, and amino acids is very low in the fetal liver and develops after birth (3, 67). Plasma cortisol and corticosterone levels are another regulatory signal. These levels are high at delivery and rapidly decline during the first 24 h after birth (38). Likely this hormonal environment is linked to the type of diet consumed (27, 56). In addition to hormonal signals controlling expression of metabolic proteins, fatty acids themselves act as another regulatory signal by binding to the nuclear receptor PPARα to stimulate expression of some genes and decrease expression of others (4, 10, 16, 19, 39, 53). Glucose may act directly through sterol regulatory element binding protein (27).

During the weaning period, plasma insulin levels increase and glucagon levels decrease between 20 and 30 days, when the rats are weaned to the adult high-carbohydrate diet (8, 43). Thyroid hormone has received wide attention as a possible trigger of developmental changes in infant rats, because its circulating concentration increases significantly during the second postnatal week (33, 61, 68). The elevated expression of FATP6 and FAT/CD36 mRNA immediately after birth suggests that increased expression of these two genes is linked to a later increase in fatty acid transport across the cell membrane. Decreased expression of
fatty acid transporter mRNA after 2 wk correlates with a period of suckling-weaning transition and higher dietary glucose availability.

Previous results of McGarry’s group (13) using differential inhibition methods suggested that the relationship between CPT Iα and Iβ was an example of simple isoform switching where CPT Iα is the “fetal isoform” and CPT Iβ is the “adult isoform” for the heart. Using Northern blot analysis (14), we found similar results for CPT I mRNA isoform expression in rat hearts. Our RTQ-PCR data suggest a more complex relationship than simple isoform switching (cf. GLUT1 and GLUT4 expression in Fig. 6 with CPT I isoform expression in Fig. 2). The peak of CPT Iβ expression within the first 4 days may be linked to the contents of colostrum milk, which is characterized by lower lactose and higher fat concentrations than mature milk (2, 6, 7, 28). The stimulatory effect on CPT I gene expression by fatty acids binding to the PPARα nuclear receptor has been well documented (10, 19, 39, 53). At the same time, low insulin concentrations in the first 2 wk of life (8) may also stimulate expression of CPT Iα (14, 48).

Recently published results on expression of CPT I genes in lamb heart during postnatal development show similar changes of CPT I isoform expression including the ratio of CPT Iα to Iβ (4). In the lamb heart, CPT Iβ was constitutively expressed throughout development, whereas CPT Iα was transiently upregulated (4); however, these authors may have missed the peak of CPT Iβ expression that we found in the first week, as we did previously (14), because fewer time points were examined. These authors (4) found that fatty acid concentrations in blood increased dramatically (>50-fold), and rates of β-oxidation in newborn lambs increased 112-fold compared with fetal lambs under physiological conditions.

CPT I gene expression in the human heart is identical to our findings in the rat heart. In the fetal human heart, both CPT I isoforms were expressed, and CPT Iβ mRNA was approximately twofold higher than CPT Iα. In the adult human heart, CPT Iβ mRNA expression was about sixfold higher than CPT Iα (52).

Expression of CPT I isoforms during development of the rabbit heart was different from the results reported for lamb (4) or human (52) hearts or our RTQ-PCR data for the rat heart. In the rabbit heart, CPT Iα mRNA expression followed a similar pattern to the one seen here, but data from rabbit heart experiments did not show fetal expression, and it was not possible to compare CPT Iα and Iβ expression at day 1 (47). CPT Iβ mRNA expression was very low at day 1 and was elevated only after 10 days; it reached extremely high levels (30-fold greater than fetal expression) at 14 days (47). These differences suggest that the CPT Iβ gene in the rabbit heart has a much different mechanism of regulation compared with rat, human, or lamb hearts. Perhaps rabbits have differences in diet or hormonal changes that cause differences in regulation of CPT Iβ expression during early postnatal development. Or, this may simply be another example of a species difference in basal expression of CPT I isoforms. Rat and human adipose tissue cells express only the CPT Iβ isoform, but mouse adipocytes express only CPT Iα (12).

Our data for rat ACC isoforms also differ from data reported for rabbit hearts, where both ACC proteins are reported to decrease after birth; this could explain the increased rate of fatty acid oxidation in rabbits (37). The activity of all CPT I isoforms can be inhibited by malonyl-CoA (most likely produced by ACC2), which is colocalized to the mitochondrial outer membrane with CPT I (1, 26, 46). The CPT Iβ isoform is more sensitive to this inhibition by a factor of at least 10 (14, 26, 37, 47).

Increased fatty acid oxidation rates in early heart development are attributed to increased carnitine levels that allow a greater level of substrate saturation for the CPT I isoforms, especially CPT Iα, which has a much lower K_m value for carnitine (13). Although increased carnitine has been found in rats, it has not been found in rabbit hearts during early development. Rabbit hearts have a much higher level of carnitine present during early development (47).

Additional data that show the importance of CPT Iα in early development come from kinetic studies of heart mitochondria from 4-day-old and adult rats (30). The susceptibility to inhibition by malonyl-CoA was almost 10 times greater in adult hearts than in hearts of 4-day-old rats, which indicates a much higher level of CPT Iα in the neonatal hearts (30).

Until the present study, no information was available regarding the pattern of expression of PPARα and PGC-1α in rat hearts during early development. High expression levels of PPARα and PGC-1α mRNA in the first few days of life correlate closely with increased expression of CPT I isoforms and HMG-CoA synthase 2; this indicates the plausibility for regulation by binding of long-chain fatty acids in the heart during early development (39, 53).

We have not found any unexpected results in GLUT1 or GLUT4 expression. The switching of GLUT isoforms during

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Fig. 7. Expression of pyruvate dehydrogenase kinase (PDK) genes PDK-1 (A) and PDK-4 (B) during early rat heart development. Isoform mRNA quantities are presented as in Fig. 1.

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development seemed to be closely related to changes in insulin, thyroid hormone (33), and glucose concentrations in blood (17, 59). Another important point of these changes is that glucose uptake becomes insulin sensitive after 2 wk of life (23, 28). All changes in the expression of genes regulating glucose uptake and oxidation followed changes of nutrient supply and clearly demonstrated activation of glucose metabolism during the period of suckling-weening transition.

We also noted that expression of PDK-4 mRNA was elevated immediately after birth by 8.13 ± 0.89-fold. PDK in isolated cardiac mitochondria is activated acutely by increased NADH-to-NAD⁺ and acetyl-CoA-to-CoA concentration ratios (32). Increased expression of PDK-4 relative to PDK-1 mRNA in the heart in early neonatal life and in states associated with increased myocardial β-oxidation (diabetes, fasting, and high levels of fat feeding) has a regulatory function (61). In other words, in the rat heart, a higher level of PDK-4 mRNA in the first 9 days of life indicates a higher rate of β-oxidation immediately after birth and a decrease in glucose metabolism.

Some authors describe a “fetal gene program” (15, 55) or fetal and adult isoform switching during early postnatal development (13), but in the rat heart, an isoform switch does not seem to occur except for GLUT1 and GLUT4. Certainly, it is clear from data presented here that regulation of CPT I isoforms is more complex than a simple isoform switching. We have shown that expression of genes that encode cardiac β-oxidation enzymes and fatty acid transport is coordinately increased within the first 2 wk of life. This change in cardiac metabolic gene expression was closely related to availability of nutrients and likely acts directly on DNA transcription through nuclear receptors.

These studies should be useful for stimulating future experiments aimed at the characterization of alterations in myocardial lipid and glucose metabolism in the diabetic heart and cardiac hypertrophy in which changes in cellular fatty acid and glucose metabolism are known to exist.

GRANTS

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