Transcriptional regulation of nuclear encoded mitochondrial proteins is dependent on nuclear transcription factors that act on genes encoding key components of mitochondrial transcription, replication, and heme biosynthetic machinery. Cellular factors that target expression of proteins to the heart have been well characterized with respect to excitation-contraction coupling. No information currently exists that examines whether parallel transcriptional mechanisms regulate nuclear encoded expression of heart-specific mitochondrial isoforms. The muscle CPT-Iβ isoform in heart is a TATA-less gene that uses Sp1 proteins to support basal expression. The rat cardiac fatty acid response element (−301/−289), previously characterized in the human gene, is responsive to oleic acid following serum deprivation. Deletion and mutational analysis of the 5′-flanking sequence of the carnitine palmitoyltransferase Iβ (CPT-Iβ) gene defines regulatory regions in the −391/+80 promoter luciferase construct. When deleted or mutated constructs were individually transfected into cardiac myocytes, CPT-I/luciferase reporter gene expression was significantly depressed at sites involving a putative MEF2 sequence downstream from the fatty acid response element and a cluster of heart-specific regulatory regions flanked by two Sp1 elements. Each site demonstrated binding to cardiac nuclear proteins and competition specificity (or supershifts) with oligonucleotides and antibodies. Individual expression vectors for Nkx2.5, serum response factor (SRF), and GATA4 enhanced CPT-I reporter gene expression 4–36-fold in CV-1 cells. Although cotransfection of Nkx and SRF produced additive luciferase expression, the combination of SRF and GATA4 cotransfection resulted in synergistic activation of CPT-Iβ. The results demonstrate that SRF and the tissue-restricted isoform, GATA4, drive robust gene transcription of a mitochondrial protein highly expressed in heart.

Expression of nuclear and mitochondrial encoded expression of respiratory chain subunits occurs despite physical separation of transcriptional events within separate genomes. Stimulatory factor; RMNE, rat myocyte nuclear extract; TNT, transcription/translation.

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onstrated the presence of a fatty acid response element (FARE) in the human CPT-Iβ promoter (7). After exposing neonatal rat cardiac myocytes to serum-free conditions, addition of exogenous oleate increases expression of the human CPT-Iβ reporter gene 8–20-fold and CPT-Iβ mRNA levels rise 4–5-fold (7). Peroxisome proliferator-activated receptor α and the retinoid X receptor act to activate CPT-Iβ through the FARE site. Peroxisome proliferator-activated receptor α may also play a pivotal role in the expression of enzymes of β-oxidation (8). The physiological impact of fatty acid induction of CPT-Iβ in heart is less certain since cardiac CPT-I is resistant to fasting, a condition that enhances serum fatty acid concentrations (9, 10).

We hypothesized that factors that increase expression of tissue specific proteins involved in contractility and energy-utilizing reactions in heart would also be important in regulating the expression of an enzyme involved in transformation of its major energy substrate, long chain fatty acids. Our studies demonstrate for the first time that CPT-Iβ is regulated by the muscle-specific factor, GATA-4, and by combinatorial interactions between GATA-4 and the nuclear factor, serum response factor (SRF). Interaction between MADS box and C4 zinc finger proteins represents a novel coregulator mechanism of the cardiac actin promoter (11). SRF is especially abundant in embryonic and adult cardiac, skeletal and smooth muscle cells (12–15). The recent homologous recombination knockout of the murine SRF gene locus demonstrated that SRF is absolutely necessary for regulation of cardiac myocytes to serum-free conditions, addition of exogenous oleate increases expression of the human CPT-Iβ receptor act to activate CPT-Iβ in heart.

EXPERIMENTAL PROCEDURES

Cell Culture and Transfections—Primary cultures of neonatal rat cardiac myocytes were prepared as described previously (16). Cells were plated at a density of 6 × 10⁴ cells/well in six-well plates (Primera, Fisher), and maintained in Dulbecco’s modified Eagle’s medium (DMEM, CellGro, Fisher) with 1% penicillin/streptomycin (Life Technologies, Inc.) and 10% calf serum (HyClone Laboratories, Inc.). The cells were incubated at 37°C in the presence of 95% O₂ and 5% CO₂ for 36 h before transfection. Myocytes were transfected using calcium phosphate precipitation in the presence of serum as described previously (18). The calcium phosphate precipitate contained 1.0 μg of CPT-Iβ firefly luciferase vector and 0.25 μg of a CMV-driven Renilla luciferase expression vector (Promega) as a control for transfection efficiency. Six hours following transfection, the myocytes were washed twice with phosphate-buffered saline and maintained in DMEM with serum for an additional 48 h. For treatment with oleic acid, the myocytes were transfected with the CPT-Iβ promoter (–318/+80) reporter gene and maintained in DMEM + serum for 12 h, washed with phosphate-buffered saline and incubated in serum-free DMEM + 0.5 mM oleate (2:1 molar ratio oleate:bovine serum albumin) for an additional 24–30 h. CV-1 monkey kidney fibroblasts (ATCC no. CCL-70) were maintained as above but were incubated in serum-free DMEM (CellGro, Fisher) and maintained in Dulbecco’s modified Eagle’s medium (DMEM, CellGro, Fisher) with 1% penicillin/streptomycin (Life Technologies, Inc.). CV-1 cells were transfected using Lipo-fectAMINE Plus reagent system (Life Technologies, Inc.) in serum-free medium. After 3 h, the transfection medium was replaced with fresh serum-containing medium for 48 h. Cotransfections included 1.0 μg of the wild-type, truncated, or mutant CPT-Iβ firefly luciferase reporter constructs and various combinations of the following CMV expression vectors: 0.4 μg of Nkx2.5, 0.4 μg of GATA-4, 0.1 μg of SRF, 0.1 μg of SRF-ΔC, and 0.1 μg of SRF-Fpm. Total DNA for each transfection was corrected to a final concentration of 2.0 μg by addition of empty CMV vector. Total protein in each well was measured using the BCA protein assay reagent kit (Pierce). CPT-Iβ firefly luciferase was corrected for protein and normalized to that of the CMV/Renilla expression for each separate experiment.

Electrophoretic Mobility Shift Assays (EMSAs)—Nuclear extracts from primary rat neonatal cardiac myocytes were prepared as described previously (17). Double-stranded DNA probes containing sequences from the rat CPT-Iβ promoter were synthesized by Operon Technolo-
progressively deleted from the 5’ end and analyzed for luciferase expression following transfection into neonatal cardiac myocytes (Fig. 1). Compared with the full-length −391/+80, deletion of 85 bp to −366/+80 produced minimal changes in luciferase expression. Deletion of the MEF2F site within the −306/+80 reporter gene resulted in an overall decrease in luciferase expression. Deletion of the MEF2 site within the promoter gene construct produces additive effects on luciferase expression following transfection into neonatal cardiac myocytes (Fig. 3). Serum withdrawal for 44 h dramatically decreases the mRNA concentration of CPT-I gene activation. GATA-4 transfection alone stimulated CPT-I expression 36-fold, making GATA-4 the most potent tissue-specific regulatory element thus far described for this gene. Cotransfection of GATA-4 and Nkx 2.5 with the CPT-Iβ reporter construct diminished the response of the CPT-Iβ gene to this combination of factors (Fig. 5).

Although homeodomain factors achieve specificity via protein binding to DNA, Nkx, SRF, and GATA-4 have been demonstrated to exert regulatory control over cardiac gene expression via protein-protein interactions (11, 22, 23). To study biological activity of these proteins independent of DNA binding, Nkx, GATA-4, and/or SRF expression vectors were coexpressed into CV-1 cells with the full-length CPT-Iβ reporter gene construct. Transfection of Nkx 2.5 or SRF alone produces modest expression of CPT-Iβ/luciferase in CV-1 cells (−4-fold activation, Fig. 5). Cotransfection of Nkx and SRF with the promoter gene construct produces additive effects on luciferase expression (Fig. 5A), suggesting that both factors contribute independently to CPT-Iβ gene activation. GATA-4 transfection alone stimulated CPT-Iβ 36-fold, making GATA-4 the most potent tissue-specific regulatory element thus far described for this gene. Cotransfection of GATA-4 and Nkx 2.5 with the CPT-Iβ reporter construct diminished the response of the CPT-Iβ gene to this combination of factors (Fig. 5B). An additive effect would be predicted if both factors were acting independently. It is possible that Nkx/GATA4 protein-protein interactions (22–24) decreased the amount of GATA4 (and Nkx) available for DNA binding to GATA elements on the CPT-Iβ gene.

### Table I

**Oligonucleotides for EMSA and reporter gene analysis**

| SITE (position) | Gel shift oligonucleotides | EMSA mt | Quick change primers (forward) |
|----------------|--------------------------|--------|--------------------------------|
| Sp1 (−68 to −74) | ctacgcaggacctggctggccaggggctg | gTTCggg | ccacgttagagcagcAcaAGTggccaggtcgtgc |
| GATA (−93 to −96) | ctacgactgtgggtgactgggggggctg | CTTc | gggagatggggCCTcaagttccagctgaag |
| CArG (−104 to −112) | ctacgtaggatgttttcccagttgggctg | AcgTctCAa | ctgtagtcccccaccacggCtCggaggtttatcctggtaac |
| GATA (−126 to −129) | ctacgcagggctgtagtacttttcccag | Ggtc | cccgcctagcgttttctctccc |
| Ebox (−247 to −252) | ctacgacgtgcaagttgaaacctggctg | CgCcCA | atgcacacagtgcGgcCagaaacctccc |
| MEF2 (−271 to −280) | ctacgctacatttttgaaggcctctgctgctg | CtgGttaAc | cccataactctagctGtttacactgtgacccacGtac |
| FARE (−303 to −289) | ctacgacattcctgtagccctcctcctcttccctaacatcagc | tggCcgcAAttcCttccGt | aacctctgcggAatCttccGtacacctccg |

Double-stranded oligonucleotides were synthesized for use in DNA EMSAs. The consensus sequences for factor binding are underlined and the potential Nkx site (−96/−93), respectively), whereas mutation of both sites (double GATA mutant) causes a slightly greater fall in luciferase activity (55%); the binding of SRF to the SRE (−112/−104) and GATA-4 to the two potential SRE sites at −96/−93 and −129/−126 was confirmed by EMSA (Fig. 2, D and E). Mutation of the Nkx site (−94/−88) has no effect on reporter gene expression in cardiac myocytes (Fig. 3).
gene (25). In contrast to the GATA-4/Nkx cotransfections, co-expression of GATA-4 and SRF produced a synergistic response of the CPT-I<sub>b</sub> gene (Fig. 5C). This combination of factors resulted in a synergism that was significantly different from the predicted value if the actions of these two proteins were independent. These data provide the first example of a nuclear encoded, cardiac mitochondrial gene where SRF and GATA-4 act as mutual coregulators (11).

To confirm the role of the predicted regulatory domains and protein/DNA interactions in reporter gene expression, point mutations in SRE, the two GATA-4 sites (double mutation) and Nkx2.5 in the 2391/180 CPT-I<sub>b</sub> construct were cotransfected with SRF or GATA-4 alone or in combination. Mutation of the SRE (and the Nkx), but not of the GATA sites, significantly reduced CPT-I<sub>b</sub> induction by SRF (Fig. 6A). The induction of CPT-I<sub>b</sub> reporter expression by GATA-4 alone was significantly.
Fig. 3. Effects of individual site mutations on promoter activity. A model of the protein binding sites indicates the location of the mutated regions. Each region is represented by an X on a labeled line to the left of the results from each separate transfection. Mutated promoter constructs were transfected into rat neonatal cardiac myocytes as described under “Experimental Procedures.” Luciferase activity is compared with the wild-type −391/+80 promoter after normalization to protein content and Renilla expression. Bars represent three to five experiments in triplicate reported as means ± S.E. Except for the NKE mutation, all transfections of mutated constructs were significantly different from wild type (10−3 > p > 10−6).

Fig. 4. Quantitative RT-PCR of CPT-Iβ transcripts in response to serum deprivation and restoration. Total RNA from rat neonatal cardiac myocytes was isolated and analyzed by quantitative RT-PCR. ♦, freshly isolated myocytes were maintained in DMEM with 10% serum for 12 h. The medium was then removed and replaced with serum-free DMEM for an additional 8, 32, or 44 h. •, freshly isolated myocytes were placed in serum-free medium immediately after plating for 12, 20, or 44 h. Fresh DMEM with 10% serum was added, and the cells were maintained for an additional 44, 36, or 12 h for a total of 56 h/treatment. Results represent CPT-Iβ transcripts normalized to cyclophilin mRNA and are presented as mean ± S.E. of triplicate determinations.

reduced by ~50% by all three point mutations (Fig. 6B). The point mutation in the Nkx site (−94/−88) adjacent and overlapping the GATA site likely represents interruption of GATA-4 binding at −96/−93. The remaining reporter gene activity in the presence of the GATA double mutation may represent physical association of the transfected GATA-4 with endogenous factors or basal transcriptional complexes in the CV-1 fibroblasts (11, 25). The combinatorial effects of SRF and GATA4 on the CPT-Iβ reporter gene were abolished by mutation of either the SRE or the two GATA sites (Fig. 6C). These results reinforce the SRE dependence of these coregulators for synergism (11). Moreover, the results again demonstrate that dramatic induction in CPT-Iβ gene expression is also regulated in large part by GATA-4 interactions alone, some of which appear independent of binding to traditional GATA sites. The drop in synergism due to the point mutation in the Nkx site (Fig. 6C) appears to reflect a requirement for appropriate flanking sequences to facilitate GATA-4 interaction with the DNA at −96/−93.

Transfection of a DNA binding mutant SRFpm abolishes the synergistic response between GATA-4 and SRF (Fig. 7). This finding is consistent with requirement for SRF binding to the SRE to produce cardiac α actin promoter coactivation (11). Induction of the GATA-dominated CPT-Iβ reporter gene expression remains elevated in the presence of transfected GATA-4 and SRFpm. A small reduction in the normal GATA-4 up-regulation may reflect altered affinities of the gene for the SRFpm/GATA-4 complex, including direct binding of the complex to the GATA site(s). A deletion of the C-terminal activation domain of SRF (SRFΔC) inhibits the combinatorial action of SRF and GATA-4 and dramatically reduces the induction of gene expression due to GATA-4 (Fig. 7). The promoter activation that remains (13-fold) is on the order of induction suggestive of independent GATA binding to the DNA/protein complexes (Fig. 6B, 15-fold induction). The data confirm coactivation of CPT-Iβ by GATA-4 and SRF that augments the enhancement of gene expression seen in the presence of GATA-4 alone.

DISCUSSION

Basal expression of the CPT-Iβ gene is analogous to the CPT-Iα gene in that it contains no TATA box and uses Sp proteins 1 and 3 to drive expression (26). The factors that identify and separate the tissue location of these two isoforms are of particular interest to cardiac muscle where the muscle isoform is dominant, but the liver isoform (CPT-Iα) is also expressed (6). We have identified a region within −391 base pairs of the transcription start site of the muscle isoform that is sufficient to drive CPT-Iβ transcription and that contains MEF2 and E box elements as well as a cluster of other heart-specific sites. This cluster of CPT-Iβ regulatory elements is located between −137 and −68 bp of the proximal promoter and includes GATA, Nkx2.5, and SRF DNA-binding sites. Identification of these important cis-trans regulatory domains suggests a multiplicity of pathways that may be expressed and are potentially interactive depending on the CPT-Iβ gene context and the developmental environment of the myocyte.

The GATA family of transcription factors, represented by heart-specific GATA-4, are potent transactivators of several cardiac promoters. These include atrial natriuretic factor (27), cardiac sarcolemmal Na+/Ca2+ exchanger (28), B-type natri-
uretic peptide (29), and cardiac troponin I (30). GATA-4 (and GATA-6) colocalize in postnatal cardiomyocytes and are believed to act in the differential control of various cellular processes (29). The present results are the first to identify a heart-specific protein, carnitine palmitoyltransferase Iβ, as a downstream mitochondrial target of GATA-4. Two GATA binding sites are present in this gene although transcriptional activation by GATA does not always require DNA binding (25). Together with Nkx 2.5, GATA-4 has been demonstrated to be a powerful transcriptional coactivator of the ANF promoter (31) and the cardiac α actin promoter (23). In the context of the CPT-1β gene, however, coexistence of GATA-4 and Nkx 2.5 decreases reporter gene expression in CV-1 cells. Compared with a significant, but small, transactivation by Nkx 2.5, GATA-4 expression in CPT-1β-transfected CV-1 cells produces a greater than 30-fold increase in CPT-1β gene reporter activity, indicating GATA domination of the CPT-1β promoter. Nkx 2.5 may act to sequester GATA factors away from GATA-binding sites in GATA-dependent promoters (25). A DNA-independent interaction between GATA and Nkx 2.5 could therefore remove GATA-4 from its interaction sites in CPT-1β, resulting in the decreased reporter gene expression observed.

Fig. 5. Combinatorial interactions of Nkx2.5, SRF, and GATA4. Subconfluent CV-1 cells were transfected as described with 1 µg of the −391/+80 CPT-1β promoter luciferase reporter construct. Total DNA concentrations were adjusted to 2 µg with empty pcDNA vector. A, transfection reactions included 0.4 µg of Nkx2.5 or 0.1 µg of SRF CMV-driven expression vectors alone or in combination. B, the CMV-Nkx2.5 (0.4 µg) and CMV-GATA4 (0.4 µg) expression constructs were used alone or in combination to influence the −391/+80 reporter. C, CMV-driven SRF and GATA4 expression were combined to regulate luciferase expression. Cells were maintained in DMEM with 10% serum for 48 h after transfection and then harvested. Luciferase activity was measured and normalized to total protein concentration and Renilla luciferase. Results are reported as mean ± S.E. of three to five experiments performed in triplicate. Cotransfection of all expression vectors with CPT-1β reporter gene produced significant changes compared with wild type alone. GATA-4 + SRF compared with GATA-4 alone (*, p < 0.002).

Fig. 6. Induction and synergistic activation of the CPT-1 promoter by SRF and GATA4 is reduced with DNA binding site mutations. Subconfluent CV-1 cells were transfected with 1 µg of wild-type or mutant reporter constructs with expression vectors for SRF and GATA4 alone or in combination. Mutated promoters are schematically represented to the left. Total DNA concentrations were adjusted to 2 µg with empty pcDNA3 vector. Cells were harvested 48 h after transfection, and luciferase activity was recorded and normalized to protein content and Renilla expression. Bars represent mean ± S.E. of three to five experiments in triplicate. Except for the SRF induction of the CPT-1β-GATA double mutant, all results are significantly decreased compared with wild-type reporter activity (0.01 > p > 10−4).
Regulation of Carnitine Palmitoyltransferase Iβ Transcription

In cardiac myocytes, the double GATA4 site mutation reduces reporter gene expression by greater than 50%, supporting the importance of DNA binding in the cumulative GATA effects. Cotransfection of the reporter gene with the double GATA4 mutant into CV-1 cells also reduces reporter gene expression by GATA4 by 43%, but significant gene induction is still retained. These data again suggest that GATA4 may also influence CPT-Iβ gene expression by protein-protein interactions, interacting with endogenous levels of factors that affect basal transcription (25). Among these factors, SRF has recently been shown to be a mutual coregulator with GATA4 in numerous myogenic SRE-dependent promoters (11). In the absence of GATA-DNA binding, SRF binds to the CArG box sequence and physically associates with GATA-4 through the MADS box of GATA-DNA binding, SRF binds to the CArG box sequence and physically associates with GATA-4 through the MADS box of GATA-4 interacting physically and functionally to increase the transcriptional responses (32). These pathways gain additional significance in the heart where cardiac hypertrophy and failure is linked to down-regulation of the enzymes of the β-oxidation pathway (33), including CPT-Iβ (34). The CPT-Iβ promoter contains a MEF2/DNA binding site, and mutational analysis reveals a 46% depression in CPT-Iβ gene expression when the MEF2 site is mutated. Nuclear protein extracts of the neonatal cardiac myocytes demonstrate binding of both MEF2A and MEF2C in electrophoretic mobility shift assays. The ability of the MEF2 proteins to activate transcription in vivo depends on the dimer composition of the binding complex and the cellular context.

Twenty base pairs downstream of the MEF2 binding element, we have identified an E box that acts as a suppressor of CPT-Iβ in the promoter deletion analysis. The consensus E box binds basic helix-loop-helix regulatory proteins and are contained in the regulatory regions of most developmentally controlled, muscle-specific genes. In the CPT-Iβ gene, we have identified the E box-binding proteins as the upstream stimulatory factors, USF1 and USF2. Although a role for rat USF1 has been suggested in contractile-mediated activation of α myosin heavy chain gene (35), USF can either positively or negatively regulate promoter activity via independent cis regulatory elements (36). E boxes are also frequently associated with adjacent MEF2 sites with a spacing that promotes protein-protein interaction between E box and MEF2 basic helix-loop-helix factors bound to the DNA. The interaction of this E box site with MEF2 and adjacent sites in the promoter is currently under investigation.

Finally, the presence of a FARE site in the rat CPT-Iβ promoter has been confirmed by these studies. The activity of this site in our hands produces a small induction of CPT-Iβ gene expression (2–3-fold) at high physiological concentrations of oleate (2:1 molar ratio) in the cell medium. This fold induction is less than previously reported at a 7:1 oleate to albumin molar ratio (8–20-fold) (7). Although a role for fatty acids in the induction of CPT-Iβ gene transcription is an attractive regulatory mechanism physiologically, other studies have not been able to demonstrate a role for elevated serum fatty acids in altering the cardiac content of CPT-Iβ mRNA and protein (9, 10). These studies suggest that, like other heart-specific proteins, CPT-Iβ contains the same pattern of muscle-specific control regions. Its expression in the heart is likely GATA-4-dominated and is subject to protein-protein interactions that can regulate its expression in a manner that is context-dependent. Interestingly, these studies also verify that gene expression of a major controlling enzyme in mitochondrial oxidative metabolism is coordinate with expression of proteins important in contractile function and energy consumption.

REFERENCES

1. Virbasius, C. A., Virbasius, J. V., and Scarpulla, R. C. (1993) Genes Dev. 7, 2431–2445
2. Virbasius, J. V., Virbasius, C. A., and Scarpulla, R. C. (1993) Genes Dev. 7, 380–392
3. Xia, Y., Buja, L. M., Scarpulla, R. C., and McMillin, J. B. (1997) Proc. Natl. Acad. Sci. U. S. A. 94, 11399–11404
4. Lenka, N., Vijayasankathy, C., Mullick, J., and Avadhani, N. G. (1998) Prog. Nucleic Acids Res. Mol. Biol. 61, 309–344
5. Xia, Y., McMillin, J., Lewis, A., Moore, M., Zhu, W. G., Williams, R. S., and Kellemen, R. E. (2000) J. Biol. Chem. 275, 1855–1863
6. Esser, V., Brown, N. F., Covan, A. T., Foster, D. W., and McGarry, J. D. (1999) J. Biol. Chem. 274, 6972–6977
7. Brandt, J. M., Djuadi, F., and Kelly, D. P. (1998) J. Biol. Chem. 273, 23786–23792
8. Djuadi, F., Brandt, J. M., Weinheimer, C. J., Leone, T. C., Gonzalez, F. J., and Kelly, D. P. (1999) Prostaglandins Leukotrienes Essent. Fatty Acids 60, 329–343
9. Mynatt, R. L., Lappi, M. D., and Cook, G. A. (1992) Biochim. Biophys. Acta 1128, 105–111
10. Cook, G. A. (1984) J. Biol. Chem. 259, 12030–12033
11. Belaguli, N. S., Sepulveda, J. L., Nigam, V., Charron, F., Nemer, M., and Schwartz, R. J. (2000) Mol. Cell. Biol. 20, 7550–7558
12. Arsenian, S., Weinhold, M., Oelgeschlager, M., Ruther, U., and Nordheim, A. (1998) EMBO J. 17, 6289–6299
13. Belaguli, N. S., Schildmeyer, L. A., and Schwartz, R. J. (1997) J. Biol. Chem. 272, 18222–18231
14. Brons, J. H., Clevers, H., and van Oeveren, W. (1993) Eur. J. Biochem. 218, 37–44
15. Belaguli, N. S., Sepulveda, J. L., Nigam, V., Charron, F., Nemer, M., and Schwartz, R. J. (2000) Mol. Cell. Biol. 20, 7550–7558
16. McMillin, J. B., Hudson, R. K., and Buja, L. M. (1993) Methods Enzymol. 2, 175–200
17. Muller, M. M., Schreiber, E., Schaffer, W., and Matthius, P. (1989) *Nucleic Acids Res.* **17**, 6420
18. Wang, D., Harrison, W., Buja, L. M., Elder, F. F. B., and McMillin, J. B. (1998) *Genomics* **48**, 314–323
19. Depre, C., Shipley, G. L., Chen, W., Han, Q., Doentos, T., Moore, M. Stepkowski, S., Davies, P. J. A., and Taegeht Meyer, H. T. (1998) *Nat. Med.* **11**, 1269–1275
20. Sack, M. N., Dierck, D. L., Rockham, H., and Kelly, D. P. (1997) *Proc. Natl. Acad. Sci. U. S. A.* **94**, 6438–6443
21. Zhao, J. L., Austen, K. L., and Lam, B. K. (2000) *J. Biol. Chem.* **275**, 8903–8910
22. Chen, C. Y., and Schwartz, R. J. (1996) *Mol. Cell Biol.* **16**, 6372–6384
23. Sepulveda, J. L., Belaguli, N., Nigam, V., Chen, C.-H., Nemer, M., and Schwartz, R. J. (1998) *Mol. Cell Biol.* **18**, 3405–3415
24. Durocher, D., and Nemer, M. (1998) *Dev. Genet.* **22**, 250–262
25. Shiojima, I., Komuro, I., Oka, T., Hiroi, Y., Mizuno, T., Takimoto, E., Monzen, K., Aikawa, R., Akazawa, H., Yamazaki, T., Kudoh, S., and Yazaki, Y. (1999) *J. Biol. Chem.* **274**, 8231–8239
26. Steffen, M. L., Harrison, W. R., Elder, F. F. B., Cook, G. A., and Park, E. A. (1999) *Biochem. J.* **340**, 425–432
27. Lee, Y., Shiota, T., Kasahara, H., Jobe, S. M., Wiese, R. J., Markham, B. E., and Izumo, S. (1998) *Mol. Cell Biol.* **18**, 3120–3129
28. Nicholas, S. B., and Philipson, K. D. (1999) *Am. J. Physiol.* **277**, H324–H330
29. Charron, F., Paradis, F., Brezchajou, O., Nemer, G., and Nemer, M. (1999) *Mol. Cell Biol.* **19**, 4355–5365
30. Bhavsar, P. K., Dellow, K. A., Yaacoub, M. H., Brand, N. J., and Barton, P. J. (2000) *J. Mol. Cell. Cardiol.* **32**, 95–108
31. Durocher, D., Charron, F., Warren, R., Schwartz, R. J., and Nemer, M. (1997) *EMBO J.* **16**, 5687–5696
32. Lu, J., McKinsey, T. A., Nicol, R. L., and Olson, E. N. (2000) *Proc. Natl. Acad. Sci. U. S. A.* **97**, 4070–4075
33. Sack, M. N., Rader, T. A., Park, S., Bastin, J., McCune, S. A., and Kelly, D. A. (1996) *Circulation* **94**, 2837–2842
34. Leone, T. C., Weinheimer, C. J., and Kelly, D. P. (1999) *Proc. Natl. Acad. Sci. U. S. A.* **96**, 7473–7478
35. Xiao, Q., and Ojamae, K. (1998) *J. Mol. Cell. Cardiol.* **30**, 87–95
36. Navankasattusas, S., Sawadogo, M., van Bilsen, M., Dang, C. V., and Chien, K. R. (1994) *Mol. Cell. Biol.* **14**, 7331–7339
GATA-4 and Serum Response Factor Regulate Transcription of the Muscle-specific Carnitine Palmitoyltransferase I β in Rat Heart
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