Orphan Nuclear Hormone Receptor RevErbα Modulates Expression from the Promoter of the Hydratase-dehydrogenase Gene by Inhibiting Peroxisome Proliferator-activated Receptor α-Dependent Transactivation*

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Peroxisome proliferator-activated receptor α (PPARα) heterodimerizes with the 9-cis-retinoic acid receptor (RXRα) to bind to peroxisome proliferator-response elements (PPRE) present in the upstream regions of a number of genes involved in metabolic homeostasis. Among these genes are those encoding fatty acyl-CoA oxidase (AOx) and enoyl-CoA hydratase/3-hydroxyacyl-CoA dehydrogenase (HD), the first two enzymes of the peroxisomal β-oxidation pathway. Here we demonstrate that the orphan nuclear hormone receptor, RevErbα, modulates PPARα/RXRα-dependent transactivation in a response element-specific manner. In vitro binding analysis showed that RevErbα bound the HD-PPRE but not the AOx-PPRE. Determinants within the HD-PPRE required for RevErbα binding were distinct from those required for PPARα/RXRα binding. In transient transfections, RevErbα antagonized transactivation by PPARα/RXRα from an HD-PPRE luciferase reporter construct, whereas no effects were observed with an AOx-PPRE reporter construct. These data identify the HD gene as a target for RevErbα and illustrate cross-talk between the RevErbα and PPARα signaling pathways on the HD-PPRE. Our results suggest a novel role for RevErbα in regulating peroxisomal β-oxidation.

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§ The abbreviations used are: PPAR, peroxisome proliferator-activated receptor; AOx, fatty acyl-CoA oxidase; DR, direct repeat; EMSA, electrophoretic mobility shift analysis; HD, enoyl-CoA hydratase/3-hydroxyacyl-CoA dehydrogenase; PPRE, peroxisome proliferator-response element; RXRα, 9-cis-retinoic acid receptor.

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which is suggested to be the cause of its repressive effects on transcriptional activation (25, 31, 36, 38). Monomeric RevErb α binds to the TGACC(T/C) consensus half-site flanked by a 5-base A/T-rich region (23, 25, 30, 36). This consensus half-site is found in both the AOX- and HD-PPREs. Considering the similarities between RevErb α and PPARs with respect to their consensus half-sites, tissue distribution, and putative biological functions, we investigated a possible role for RevErb α as an upstream regulatory factor for the peroxisomal β-oxidation pathway.

**EXPERIMENTAL PROCEDURES**

**Plasmids—**The luciferase reporter plasmids pCPSluc, pHDIx3luc, pM2ix3luc, and pAOxix2luc, and the expression plasmids for rat PPARs and human RXRs, have been described previously (2, 18, 22, 39, 40). The expression plasmid for human RevErb α (RevErbαSG5) was constructed by excision of the human RevErb α cDNA as a BamHI fragment from the plasmid pCMXhRevErb α (a kind gift of H. P. Harding and M. A. Lazar) (23), followed by cloning of the fragment in the correct orientation into the BamHI site of the plasmid pSG5 (Promega).

**In Vitro Transcription/Translation—**Transcription/translation of cDNA encoding PPARs, RXRs, and RevErb α was performed using the TNT T7-coupled rabbit reticulocyte lysate system according to the manufacturer’s protocol (Promega). Translation products labeled with [35S]methionine were analyzed on 15% SDS-polyacrylamide gels. Synthesis of proteins for use in electrophoretic mobility shift analysis (EMSA) was carried out in parallel with unlabeled methionine.

**Electrophoretic Mobility Shift Analysis—**EMSA using in vitro translated proteins and radiolabeled oligonucleotide probes was carried out essentially as described (18). Oligonucleotides corresponding to the AOX-PPRE (5'-gatCTTTTCCCAGAACGTGATTTTGCTCCTGGTCCCC-TTTGTGCa and its complement) and to the HD-PPRE (5'-gatCCCTCTCCTTGGACCTTGGAAGTCCTACATATTTGGA and its complement) were annealed and end-labeled with the Klenow fragment of DNA polymerase I and [α-32P]dATP. The underlined sequences indicate the TGAACC(T/C) consensus RevErb α binding site.

**RESULTS**

**RevErb α Binds to the HD-PPRE—**Both the HD- and AOX-PPREs contain potential binding sites for RevErb α (Fig. 1A). We therefore examined whether RevErb α is capable of binding to the HD- and AOX-PPREs by performing EMSA with radiolabeled PPRE probes and in vitro translated receptors. As expected, PPARα and RXRα bound as a heterodimer on the HD-PPRE (Fig. 1B). RevErb α also bound the HD-PPRE, forming a complex with a mobility slightly less than that formed by the PPARα/RXRα heterodimer (Fig. 1B). Inclusion of RXRα or PPARα with RevErb α in the binding reactions had no effect on the formation of the RevErb α-HD-PPRE complex. Moreover,
HD-PPRE, RevErb conditions used. In contrast to the results obtained with the HD-PPRE could be influenced by RevErb the HD-PPRE. Conversely, when EMSA was performed with a greater amounts of RevErb co-incubated with PPARB.

Furthermore, only the characteristic PPAR 

A 

B 

FIG. 2. REVéRBA AND PPARα/RXRa bind independently to the HD-PPRE. Binding reactions contained in vitro synthesized PPARα/RXRa (0.5 μl of each) and REVéRBA (5 or 10 μl) (A), or REVéRBA (10 μl) and PPARα/RXRa (1 or 2 μl of each) (B). Radiolabeled HD-PPRE probe was added at 200, 2, or 0.2 fmol, as indicated. Lysate volumes were kept constant by addition of unprogrammed lysate. EMSA was performed on a 2.5% polyacrylamide gel. Autoradiography was for 16 h (200 fmol, constant by addition of unprogrammed lysate. EMSA was performed on a 2.5% polyacrylamide gel with in vitro synthesized PPARα/RXRa (1 μl of each) and REVéRBA (10 μl) and wild-type or mutant HD-PPRE probes, as indicated. Lysate volumes were kept constant by addition of unprogrammed lysate. Arrows indicate the positions of the PPARα/RXRa heterodimer and the REVéRBA monomer.

co-incubation of all three receptors with the HD-PPRE produced only two distinct complexes corresponding to PPARα/RXRa heterodimers and REVéRBA monomers (Fig. 1B). Therefore, the three receptors do not co-occupy the HD-PPRE in some higher order complex, and REVéRBA does not form complexes with PPARα or RXRa on this element in vitro under the EMSA conditions used. In contrast to the results obtained with the HD-PPRE, REVéRBA was unable to bind to the AOx-PPRE. Furthermore, only the characteristic PPARα/RXRa heterodimer was generated on the AOx-PPRE when REVéRBA was co-incubated with PPARα and RXRa (Fig. 1B). Binding of REVéRBA to the HD-PPRE was specific, since the radiolabeled complex was refractory to competition by nonspecific unlabeled oligonucleotide but was eliminated by addition of unlabeled HD-PPRE oligonucleotide (data not shown).

We next investigated whether binding by PPARα/RXRa to the HD-PPRE could be influenced by REVéRBA and vice versa. EMSA was first performed using radiolabeled HD-PPRE, constant amounts of PPARα/RXRa, and increasing amounts of REVéRBA (Fig. 2A). Increasing the amount of REVéRBA did not affect binding of PPARα/RXRa to the HD-PPRE, but did result in greater amounts of REVéRBA monomeric complex forming on the HD-PPRE. Conversely, when EMSA was performed with a constant amount of REVéRBA, increasing the amounts of PPARα and RXRa led to increased formation of PPARα/RXRa complexes on the HD-PPRE, while the binding of REVéRBA monomers was unaffected (Fig. 2B). Similar findings were observed over a wide range of HD-PPRE probe concentration (Fig. 2, A and B). These results suggest that REVéRBA and PPARα/RXRa bind independently to the HD-PPRE, and that REVéRBA monomers and PPARα/RXRa heterodimers do not simultaneously occupy the HD-PPRE.

REVéRBA and PPARα/RXRa Bind to Distinct Sites on the HD-PPRE—The HD-PPRE consists of four half-sites (sites I-IV) related to the consensus TGACC(T/C) hexameric half-site. Each half-site of the HD-PPRE was used in binding studies (Fig. 3). Increasing the amounts of REVéRBA did not affect binding of PPARα/RXRa to the HD-PPRE, but did result in greater amounts of REVéRBA monomeric complex forming on the HD-PPRE. Conversely, when EMSA was performed with a constant amount of REVéRBA, increasing the amounts of PPARα and RXRa led to increased formation of PPARα/RXRa complexes on the HD-PPRE, while the binding of REVéRBA monomers was unaffected (Fig. 2B). Similar findings were observed over a wide range of HD-PPRE probe concentration (Fig. 2, A and B). These results suggest that REVéRBA and PPARα/RXRa bind independently to the HD-PPRE, and that REVéRBA monomers and PPARα/RXRa heterodimers do not simultaneously occupy the HD-PPRE.

REVéRBA and PPARα/RXRa bind to distinct sites within the HD-PPRE. Sequences of the wild-type HD-PPRE and the oligonucleotides M1, M2, M3, and M4 containing mutations (underlined) in the consensus hexameric half-sites I, II, III, and IV (bold), respectively. EMSA was performed on a 2.5% polyacrylamide gel with in vitro synthesized PPARα/RXRa (1 μl of each) and REVéRBA (10 μl) and wild-type or mutant HD-PPRE probes, as indicated. Lysate volumes were kept constant by addition of unprogrammed lysate. Arrows indicate positions of bound complexes.

FIG. 3. REVéRBA and PPARα/RXRa bind to distinct sites within the HD-PPRE. Sequences of the wild-type HD-PPRE and the oligonucleotides M1, M2, M3, and M4 containing mutations (underlined) in the consensus hexameric half-sites I, II, III, and IV (bold), respectively. EMSA was performed on a 2.5% polyacrylamide gel with in vitro synthesized PPARα/RXRa (1 μl of each) and REVéRBA (10 μl) and wild-type or mutant HD-PPRE probes, as indicated. Lysate volumes were kept constant by addition of unprogrammed lysate. Arrows indicate positions of bound complexes.
RevErbα Antagonizes Transactivation by PPARα/RXRα from the HD-PPRE but Not the AOx-PPRE—To investigate the in vivo properties of RevErbα on transcriptional regulation, we carried out transient transfection assays using luciferase reporter plasmids containing either the HD-PPRE (pHD(x3)luc) or the AOx-PPRE (pAOx(x2)luc), along with expression plasmids for RevErbα, PPARα, and/or RXRα in BSC40 African monkey kidney cells. These cells were chosen because they contain low levels of endogenous PPARα and RXRα (2, 18). Co-transfection of PPARα and RXRα with the HD-PPRE reporter plasmid led to a 2-fold induction of transcription over basal levels in the absence of the peroxisome proliferator, Wy-14,643 (Fig. 4A). Addition of proliferator led to a potent induction of transcription (10–15-fold) over basal levels. Increasing amounts of RevErbα expression plasmid inhibited transcription from the HD-PPRE by PPARα/RXRα both in the presence and absence of peroxisome proliferator. Transactivation by PPARα/RXRα was reduced by 80% at the highest concentration of RevErbα expression plasmid used (2 μg). PPARα/RXRα also activated transcription of a reporter gene that contained the HD-PPRE harboring a disruption in site II; however, in this case, RevErbα-dependent inhibition was not observed (Fig. 4B). This finding is in agreement with in vitro binding data for RevErbα (see Fig. 3) and indicates that inhibition of PPARα/RXRα-mediated activation is dependent upon RevErbα binding to the HD-PPRE.

RevErbα did not significantly affect transcriptional activation by PPARα/RXRα on the AOx-PPRE, in either the absence or presence of Wy-14,643 (Fig. 4C). These data are in keeping with in vitro binding data showing that RevErbα failed to bind the AOx-PPRE (Fig. 1B). Control transfections with the parental reporter construct pCPSluc, which lacks a PPRE, showed that the presence of RevErbα, PPARα, and RXRα did not influence basal levels of luciferase activity (data not shown), demonstrating the need for a functional PPRE for receptor activity. Together, these data show that RevErbα antagonizes transactivation by PPARα/RXRα specifically from the HD-PPRE.

Increased Amounts of PPARα and RXRα Can Overcome Repression by RevErbα—We were interested in determining whether increased amounts of PPARα and RXRα could modulate the repression exerted by RevErbα on transcription from the HD-PPRE. Transient transfections with a constant amount of RevErbα expression plasmid and increasing amounts of PPARα and RXRα expression plasmids demonstrated that the repressive effects of RevErbα could be alleviated in a dose-dependent manner by increasing amounts of PPARα and RXRα, either in the presence or absence of Wy-14,643 (Fig. 5). These results suggest that the net transcriptional response from the HD-PPRE is influenced by the relative levels of PPARα, RXRα, and RevErbα in vivo.

**DISCUSSION**

A number of recent observations have pointed to possible interplay between the RevErbα and PPARα signaling pathways. First, there is strong sequence similarity between the RevErbα consensus binding site and the PPREs of the AOx and HD genes. Second, a role for RevErbα in PPARα-mediated signaling has been suggested by studies showing that the PPRE of the CYP4A6 gene binds both RevErbα and PPARα (30), and that the PPARα isoform, a key regulator of adipogenesis, may in turn be regulated by RevErbα, whose mRNA levels are dramatically increased during differentiation of preadipocytes to adipocytes (24). Third, RevErbα has been shown to be encoded on the opposite strand of thyroid hormone receptor α and to be able to bind the thyroid hormone, triiodothyronine (27), and we have previously demonstrated cross-talk between thyroid hormone receptor α and PPARα in regulating transcription from the AOx-PPRE (20). As a result of these observations, we considered the possibility of a role for RevErbα in regulating transcription from the AOx- and HD-PPREs.

We have demonstrated here that the RevErbα and PPARα signaling pathways converge and that RevErbα serves to repress transcriptional activation specifically from the HD-PPRE. Interestingly, RevErbα had no effect on PPARα/RXRα-mediated activation via the AOx-PPRE. Consistent with this observation, RevErbα was shown to bind specifically to the HD-PPRE but not the AOx-PPRE. The HD-PPRE is comprised
Cross-talk between RevErbα and PPARα/RXRα Signaling Pathways

Fig. 5. Exogenous expression of PPARα/RXRα can overcome transcriptional repression by RevErbα on the HD-PPRE. BSC40 cells were transfected with 5 μg of the luciferase reporter pHDx3/luc and expression plasmids for RevErbα (2 μg) and PPARα/RXRα (0.5–4 μg) in the absence or presence of 0.1 mM Wy-14,643. Plasmid dosage was normalized by the addition of empty expression vector. Transfections were carried out in duplicate and represent the average (± S.E.) of three independent experiments. Values presented are relative to the value obtained for cells transfected with RevErbα (2 μg) alone in the presence of Wy-14,643 (taken as 100%). DMSO, dimethyl sulfoxide.

of four hexameric direct repeats arranged as two tandem DR1 arrays separated by two nucleotides (a DR2). This complex arrangement is thought to permit the interaction of a diverse array of nuclear hormone receptors with the HD-PPRE, thereby increasing the complexity of transcriptional regulation from this PPRE. RevErbα and PPARα/RXRα target distinct half-sites on the HD-PPRE. The integrity of site II is required for RevErbα binding, while sites III/IV serve to bind RXRα/PPARα. Site II was also required for the RevErbα-dependent repressive effects on transcriptional activation by PPARα/RXRα, indicating that inhibition requires binding of RevErbα to the HD-PPRE. Although sites III/IV have been shown to be essential and sufficient for PPARα/RXRα binding and activity (22, 40), an arrangement in which PPARα/RXRα heterodimers are bound to both DR1 sites has been suggested to yield the highest level of transactivation (41). Since RevErbα occupies site II within the HD-PPRE, this may preclude binding of PPARα/RXRα to the upstream DR1 element, resulting in reduced levels of transactivation from the HD-PPRE by PPARα/RXRα.

Although RevErbα and PPARα/RXRα use distinct determinants on the HD-PPRE, we did not observe a higher order complex containing all three receptors in vitro. However, the limitation of our in vitro binding analysis does not preclude the possibility of a higher order complex forming among RevErbα, PPARα, and RXRα in vivo, perhaps through the cooperative or association of regulatory cofactors such as SRC-1, p300, N-CoR, and SMRT-1 (42–45). The involvement of such cofactors in transcriptional regulation by nuclear hormone receptors is well established. Indeed, N-CoR has been shown to interact with RevErbα in mammalian cells (38). A model can be proposed in which repression of transactivation by RevErbα is the result of a shift from active to repressive states of the receptor through its association with corepressors and dissociation from coactivators, respectively. RevErbα can also be envisioned to be subjected in vivo to post-translational modifications such as phosphorylation that could initiate its repressive state. Interestingly, the amino terminus of RevErbα contains numerous serine and threonine residues that could potentially be phosphorylated (27).

The ligand for RevErbα remains unknown. It is therefore impossible to ascertain at this time whether an endogenous ligand exists in BSC40 cells that could induce transcriptional repression upon binding RevErbα, as has been demonstrated for androstanol and the mCARβ receptor (46). It has been suggested that RevErbα lacks the AF2 transactivation domain that is responsible for ligand binding (37), thus precluding the possibility of RevErbα having any capacity for ligand-dependent activation or repression (38). Orphan receptors lacking AF2 domains could instead act as competitors for ligand-inducible receptors (37), and such a scenario has been proposed to explain the blocking of RZRo-mediated transactivation by RevErbα (25, 35). Nevertheless, the absence of an AF2 domain does not preclude the possibility that RevErbα contains undetected, and yet undefined, activation domains that could be revealed through interaction with a novel ligand (36). RevErbα could also potentially activate transcriptions by cooperative interaction with a non-AF2-dependent coactivator or, indirectly, by recruiting corepressors away from other nuclear receptors (38). TRα and PPARα have been reported to form nonbinding heterodimers in vivo (20, 47), and RevErbα could similarly form inactive, nonbinding complexes (36) with PPARα, RXRα, or other nuclear receptors, effectively sequestering these receptors and preventing them from forming heterodimers that normally potentiate transcription, leading to an overall repression of transcription. However, we consider the latter scenario unlikely, since inhibition by RevErbα required the integrity of the HD-PPRE and had no effect on PPARα/RXRα-mediated transactivation via the AOX-PPRE.

In summary, our results identify the gene encoding enoyl-CoA hydratase/3-hydroxyacyl-CoA dehydrogenase, the second enzyme of the peroxisomal β-oxidation pathway, as a target for RevErbα and demonstrate that this orphan nuclear hormone receptor serves as a negative modulator of PPARα/RXRα-mediated transactivation from the PPRE of this gene. Interestingly, Gervois et al. (48) have recently demonstrated that fibrate drugs, which are potent activators of PPARα, induce expression of the RevErbα gene through competition between RevErbα and PPARα/RXRα for binding to the autoregulatory RevDR2 site of the RevErbα gene. These results are in agreement with our findings that the repressive effects of RevErbα on the HD-PPRE can be overcome by increasing the concentration of PPARα/RXRα, and together, illustrate a convergence of the RevErbα and PPARα signaling pathways in gene regulation. Transcriptional control of peroxisomal β-oxidation involves a complex network of interacting regulatory factors that integrate a diverse array of host signaling pathways to determine a net transcriptional to a particular environmental or physiological cue. An understanding of the various transcriptional factors that control peroxisomal β-oxidation could provide for the development of pharmaceutical agents that specifically target this metabolic pathway as a means to modulate overall lipid metabolism.

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REFERENCES
1. Tugwood, J. D., Issemann, I., Anderson, R. G., Bundell, K. R., McPheat, W. L., and Green, S. (1992) EMBO J. 11, 433–439
2. Zhang, B., Marcus, S. L., Sajjadi, F. G., Alvares, K., Reddy, J. K., Subramani, S., Radchubinski, R. A., and Capone, J. P. (1992) Proc. Natl. Acad. Sci. U. S. A. 89, 7541–7545
3. Spiegelman, B. M., and Flier, J. S. (1996) Cell 87, 377–389
4. Juge-Aubrey, C., Pernin, A., Favez, T., Burger, A. G., Wahli, W., Meier, C. A., and Desvergne, B. (1997) J. Biol. Chem. 272, 25252–25259
5. Latruffe, N., and Vamecq, J. (1997) Biochimie (Paris) 79, 61–94
6. Lock, E. A., Mitchell, A. M., and Klombe, C. R. (1989) Annu. Rev. Pharmacol. Toxicol. 29, 145–163
7. Kliever, S. A., Lenhard, J. M., Wilson, Y. M., Patel, I., Morris, D. C., and Lehmann, J. M. (1995) Cell 83, 813–819
8. Lehmann, J. M., Moore, L. B., Smith-Oliver, T. A., Wilkinson, W. O., Willson, T. M., and Kliever, S. A. (1995) J. Biol. Chem. 270, 12853–12856
9. Lambe, K. G., and Tugwood, J. D. (1996) Eur. J. Biochem. 239, 1–7
10. Kliever, S. A., Sundseth, S. S., Jones, S. A., Brown, P. J., Wisely, G. B., Koble, C. S., Devechard, P., Wahli, W., Wilson, T. M., Lenhard, J. M., and
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Lehmann, J. M. (1997) Proc. Natl. Acad. Sci. U. S. A. 94, 4318–4323
11. Forman, B. M., Chen, J., and Evans, R. M. (1997) Proc. Natl. Acad. Sci. U. S. A. 94, 4312–4317
12. Kaikaus, R. M., Chan, W. K., Lyssenko, N., Ray, R., Ortiz de Montellano, P. R., and Bass, N. M. (1993) J. Biol. Chem. 268, 9593–9603
13. Besnard, P., Mallorby, A., and Carlier, H. (1993) FEBS Lett. 327, 219–223
14. Gulick, T., Creasi, S., Caia, T., Moore, D. D., and Kelly, D. P. (1994) Proc. Natl. Acad. Sci. U. S. A. 91, 11012–11016
15. Castelein, H., Gulick, T., Declercq, P. E., Mannaerts, G. P., Moore, D. D., and Eae, M. I. (1994) J. Biol. Chem. 269, 26754–26758
16. Rodriguez, J. C., Gil-Gomez, G., Hegardt, F. G., and Haro, D. (1994) J. Biol. Chem. 269, 18767–18772
17. Vu-Dac, N., Chopin-Delannoy, S., Gervois, P., Bonneyle, E., Martin, G., Fruchart, J.-C., Lautent, V., and Staels, B. (1998) J. Biol. Chem. 273, 25713–25720
18. Miyata, K. S., Zhang, B., Marcus, S. L., Capone, J. P., and Rachubinski, R. A. (1993) J. Biol. Chem. 268, 19169–19172
19. Winrow, C. J., Marcus, S. L., Miyata, K. S., Zhang, B., Capone, J. P., and Rachubinski, R. A. (1993) Gene Expr. 4, 53–62
20. Hunter, J., Kassam, A., Winrow, C. J., Rachubinski, R. A., and Capone, J. P. (1996) Mol. Cell. Endocrinol. 116, 213–221
21. Miyata, K. S., McCaw, S. E., Patel, H. V., Rachubinski, R. A., and Capone, J. P. (1996) J. Biol. Chem. 271, 9189–9192
22. Winrow, C. J., Capone, J. P., and Rachubinski, R. A. (1998) J. Biol. Chem. 273, 31442–31448
23. Harding, H. P., and Lazar, M. A. (1993) Mol. Cell. Biol. 13, 3113–3121
24. Chawla, A., and Lazar, M. A. (1993) J. Biol. Chem. 268, 16265–16269
25. Forman, B. M., Chen, J., Blumberg, B., Kiewer, S. A., Henschaw, R., Ong, E. S., and Evans, R. M. (1994) Mol. Endocrinol. 8, 1253–1261
26. Spanjaard, R. A., Nguyen, V. P., and Chin, W. W. (1994) Mol. Endocrinol. 8, 286–295
27. Miyajima, N., Horiiuchi, R., Shibuya, Y., Fukushima, S., Matsubara, K., Toyoshima, K., and Yamamoto, T. (1989) Cell 57, 31–39
28. Lazar, M. A., Jones, K. E., and Chin, W. W. (1990) DNA Cell Biol. 9, 77–83
29. Downes, M., Carozzi, A. J., and Muscat, G. E. O. (1995) Mol. Endocrinol. 9, 1666–1678
30. Hsa, M.-H., Palmer, C. N. A., Song, W., Griffin, K. J., and Johnson, E. F. (1998) J. Biol. Chem. 273, 27988–27997
31. Adelmant, G., Begue, A., Stohelin, D., and Laude, V. (1996) Proc. Natl. Acad. Sci. U. S. A. 93, 3553–3558
32. Ikeeda, Y., Lala, D. S., Luo, X., Kim, E., Moisan, M. P., and Parker, K. L. (1993) Mol. Endocrinol. 7, 852–860
33. Milbrandt, J. (1988) Neuron 1, 183–188
34. Searce, L. M., Laz, T. M., Hanel, T. G., Lau, L. F., and Taub, R. (1993) J. Biol. Chem. 268, 8855–8861
35. Retnakaran, R., Flock, G., and Giguère, V. (1994) Mol. Endocrinol. 8, 1234–1244
36. Harding, H. P., and Lazar, M. A. (1995) Mol. Cell. Biol. 15, 4791–4802
37. Durand, B., Saunders, M., Gaudon, C., Roy, B., Losson, R., and Chambon, P. (1994) EMBO J. 13, 5370–5382
38. Zamir, I., Hardin, H. P., Atkins, G. B., Horlein, A., Glass, C. K., Rosenfeld, M. G., and Lazar, M. A. (1996) Mol. Cell. Biol. 16, 5458–5465
39. Marcus, S. L., Miyata, K. S., Zhang, B., Subramani, S., Rachubinski, R. A., and Capone, J. P. (1993) Proc. Natl. Acad. Sci. U. S. A. 90, 5724–5727
40. Zhang, B., Marcus, S. L., Miyata, K. S., Subramani, S., Capone, J. P., and Rachubinski, R. A. (1993) J. Biol. Chem. 268, 12939–12945
41. Chu, R. Y., Lin, Y., Rao, M. S., and Reddy, J. K. (1995) J. Biol. Chem. 270, 29636–29639
42. Horlein, A. J., Naar, A. M., Heinz, T., Torchia, J., Gloss, B., Kurokawa, R., Ryan, A., Kamai, Y., Soderstrom, M., Glass, C. K., and Rosenfeld, M. G. (1995) Nature 377, 397–404
43. Chen, J. D., and Evans, R. M. (1995) Nature 377, 454–457
44. DiFrenzo, J., Soderstrom, M., Kurokawa, R., Ogliastro, M. H., Rice, M., Ingrey, S., Horlein, A., Rosenfeld, M. G., and Glass, C. K. (1997) Mol. Cell. Biol. 17, 2166–2176
45. Dowell, P., Ishmael, J. E., Avram, D., Peterson, V. J., Nevrivy, D. J., and Leid, M. (1997) J. Biol. Chem. 272, 33435–33443
46. Forman, B. M., Tzameli, I., Choe, H.-S., Chen, J., Simha, D., Seel, W., Evans, R. M., and Moore, D. D. (1998) Nature 395, 612–615
47. Bogazzi, F., Hudson, L. D., and Nikodem, V. (1994) J. Biol. Chem. 269, 11683–11686
48. Gervois, P., Chopin-Delannoy, S., Fadel, A., Dubois, G., Koushyk, V., Fruchart, J. C., Najib, J., Laude, V., and Staels, B. (1999) Mol. Endocrinol. 13, 400–409