Kidney International, Vol. 60 (2001), pp. 14–30

PERSPECTIVES IN BASIC SCIENCE

Peroxisome proliferator-activated receptors (PPARs): Novel therapeutic targets in renal disease

YouFei Guan and Matthew D. Breyer

Division of Nephrology, and Departments of Molecular Physiology and Biophysics, Veterans Administration Medical Center, and Vanderbilt University School of Medicine, Nashville, Tennessee, USA

Peroxisome proliferator-activated receptors (PPARs): Novel therapeutic targets in renal disease. Peroxisome proliferator-activated receptors (PPARs) are members of the nuclear hormone receptor superfamily of ligand-dependent transcription factors. PPARs play an important role in the general transcriptional control of numerous cellular processes, including lipid metabolism, glucose homeostasis, cell cycle progression, cell differentiation, inflammation and extracellular matrix remodeling. Three PPAR isoforms, designated PPARα, PPARβ and PPARγ, have been cloned and are differentially expressed in several tissues including the kidney. PPARα primary regulates lipid metabolism and modulates inflammation. PPARα is the molecular target of the hypolipidemic fibrates including bezafibrate and clofibrate. PPARβ participates in embryonic development, implantation and bone formation. PPARγ is a key factor in adipogenesis and also plays an important role in insulin sensitivity, cell cycle regulation and cell differentiation. Antidiabetic thiazolidinediones (TZDs) such as troglitazone and rosiglitazone are specific ligands of PPARγ, and this interaction is responsible for the insulin-sensitizing and hypoglycemic effect of these drugs. The kidney has been shown to differentially express all PPAR isoforms. PPARα is predominantly expressed in proximal tubules and medullary thick ascending limbs, while PPARγ is expressed in medullary collecting ducts, pelvic urothelium and glomerular mesangial cells. PPARβ is ubiquitously expressed at low levels in all segments of nephron. Accumulating data has begun to emerge suggesting physiological and pathophysiological roles of PPARs in several tissues including the kidney. The availability of PPAR-selective agonists and antagonists may provide a new approach to modulate the renal response to diseases including glomerulonephritis, glomerulosclerosis and diabetic nephropathy.

Peroxisome proliferators comprise a group of structurally diverse compounds including industrial chemicals and pharmaceutical agents that were originally identified as inducers of hepatic peroxisomal proliferation. In rodents, chronic exposure to peroxisome proliferators results in nongenotoxic liver tumors [1] and alters gene expression involved in lipid β-oxidation, cell differentiation and inflammation [2]. These effects are now known to be mediated through binding of peroxisome proliferators to a specific subset of nuclear receptor and transcription factor superfamily, designated peroxisome proliferator-activated receptors (PPARs). Since the identification of the first PPAR receptor in mouse [3], three isoforms—designated PPARα, PPARβ/δ and PPARγ—have been cloned and characterized by their distinct expression patterns, different ligand binding specificity and metabolic functions. In the past decade, tremendous progress has been made towards understanding the role of PPARs in whole body physiology and in many human diseases including diabetes, obesity, atherosclerosis, hypertension and cancer. PPARα plays an important role in lipid metabolism and its activation by fibrates mediates their potent hypolipidemic effects [4]. Recently, PPARβ/δ has been suggested as having an important role in cell survival and colon tumorigenesis. It has been suggested as a molecular target mediating the beneficial effect of nonsteroidal anti-inflammatory drugs (NSAIDs) on colorectal cancer [5]. Finally, activation of PPARγ by synthetic thiazolidinedione (TZD) ligands has been successfully used for treatment of patients with type II diabetes [6]. This review examines the current knowledge relating to PPARs' action, ligand binding and tissue distribution. Recent developments regarding the role of PPARs in diabetes, insulin resistance, atherosclerosis, inflammatory disease, hypertension, and cancer, with particular relevance to the kidney will also be discussed.

Key words: fibrate, thiazolidinedione, lipid metabolism, adipogenesis, atherosclerosis, diabetes, hypertension, kidney disease.

Received for publication August 11, 2000
and in revised form November 14, 2000
Accepted for publication November 22, 2000
Updated February 26, 2001

PPAR RECEPTORS: CLONING, STRUCTURE AND LIGAND BINDING

Cloning and structure of PPARs

In 1990, Isseman and Green identified the first nuclear receptor to which peroxisome proliferators bound by

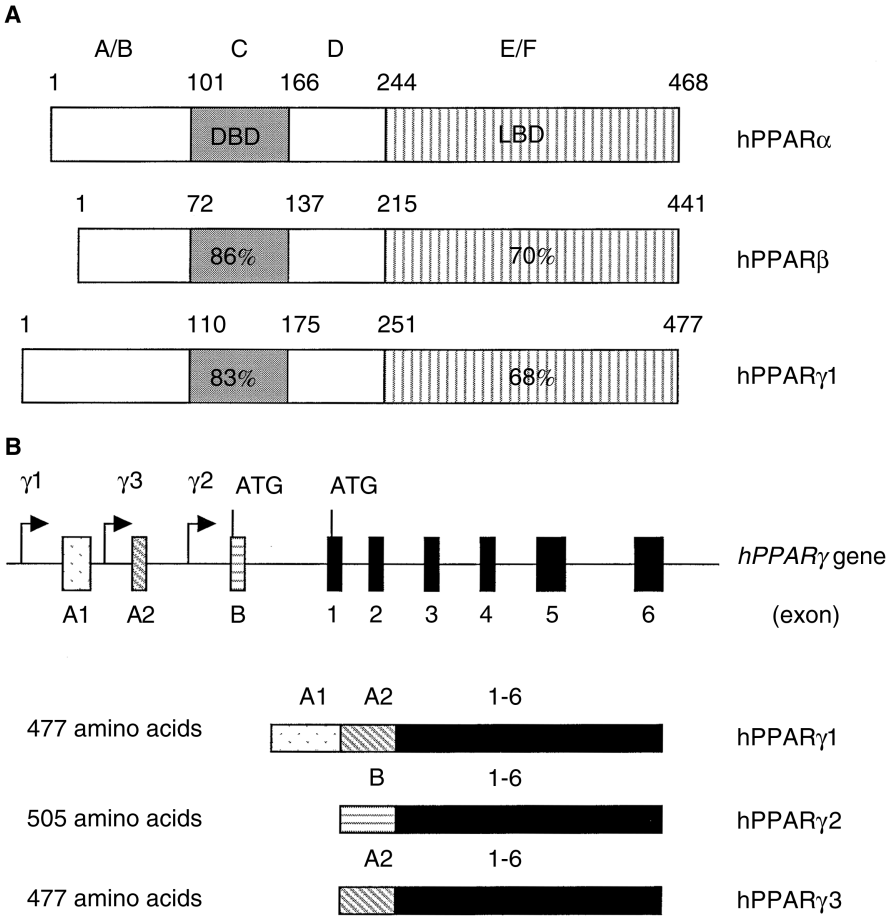

Fig. 1. (*A*) Structural and functional domains of the human peroxisome proliferator-activated receptors (PPARs). Domains are: A/B, N-terminal A/B domain containing a ligand-independent activation function (AF-1); C, DNA-binding domain (DBD); D, hinge region; and E/F, C-terminal ligand-binding domain (LBD) containing the ligand-dependent activation function (AF-2). Numbers inside the boxes represent the percentage (%) identity between the human PPAR isofoms. (*B*) The genomic organization of the human PPARγ gene. Alternative promoter usage and splicing of the three 5′ exons A1, A2, and B results in three different transcripts and two PPARγ protein products (PPARγ1 and PPARγ2). Note that transcription from the promoter γ1 and γ3 results in same protein of 477 amino acids (PPARγ1protein) and transcript using the promoter γ2 encoded a protein of 505 amino acids (PPARγ2 protein), which has additional 28 amino acids at the N-terminus compared with PPARγ1 protein.

screening of cDNA library with a probe encoding the highly conserved C domain of nuclear hormone receptors [3]. This receptor is now designated PPARα. Shortly thereafter, two additional isoforms of PPAR were identified: PPARβ/δ and PPARγ [7]. To date, no mRNA splice variant for PPARα and PPARβ/δ has been identified. In contrast, multiple splicing forms of PPARγ mRNA have been reported in many species including mouse and human [8–10]. In humans three PPARγ transcripts have been reported [9]. These three isotypes of PPARγ are derived from a single gene as a result of differential splicing and alternate promoter usage (Fig. 1). Each PPARγ transcript shares the six 3′ coding exons, differing only in the 5′ exons. The 5′ untranslated region of PPARγ1 contains exons A1 and A2, while PPARγ3 only contains exon A2. Since none of the 5′ exons distinguishing PPARγ1 and PPARγ3 possess a translation initiation site, PPARγ1 and PPARγ3 mRNAs give rise to identical protein products: PPARγ1. In contrast, PPARγ2 has a γ2-specific coding exon located between exon A2 and exon 1, yielding 30 additional N-terminal amino acids in the PPARγ2 protein [6, 11]. Due to different promoter usage, each PPARγ isoform has a distinct expression pattern [9, 10].

All three PPAR isoforms share similar structural and functional organization with other nuclear receptors (Fig. 1). Four major domains have been identified: A/B, C, D and E/F domain [reviewed in 2, 11]. The N-terminal A/B domain differs in both length and predicted amino acid sequence and contains a ligand-independent *trans*-activation domain, termed activation function 1 (AF1). This domain plays an important role in regulating PPAR activity through both phosphorylation and interdomain communication [12, 13]. The C domain is comprised of about 70 amino acids and encodes the DNA binding domain (DBD). This domain is responsible for the binding of PPAR receptor to peroxisome proliferator response element (PPRE) in the promoter region of target genes. The D hinge region is a docking domain for cofactors. The E/F C-terminal region contains two important domains. One is the ligand-binding domain (LBD), or E domain, responsible for ligand specificity and activation of PPAR binding to the peroxisomal proliferator response element (PPRE) with resultant modulation of gene expression. This region also has been found to play an important role in dimerization and nuclear localization. The other is the ligand-dependent activation domain AF-2, or F domain. AF-2 domain located in the

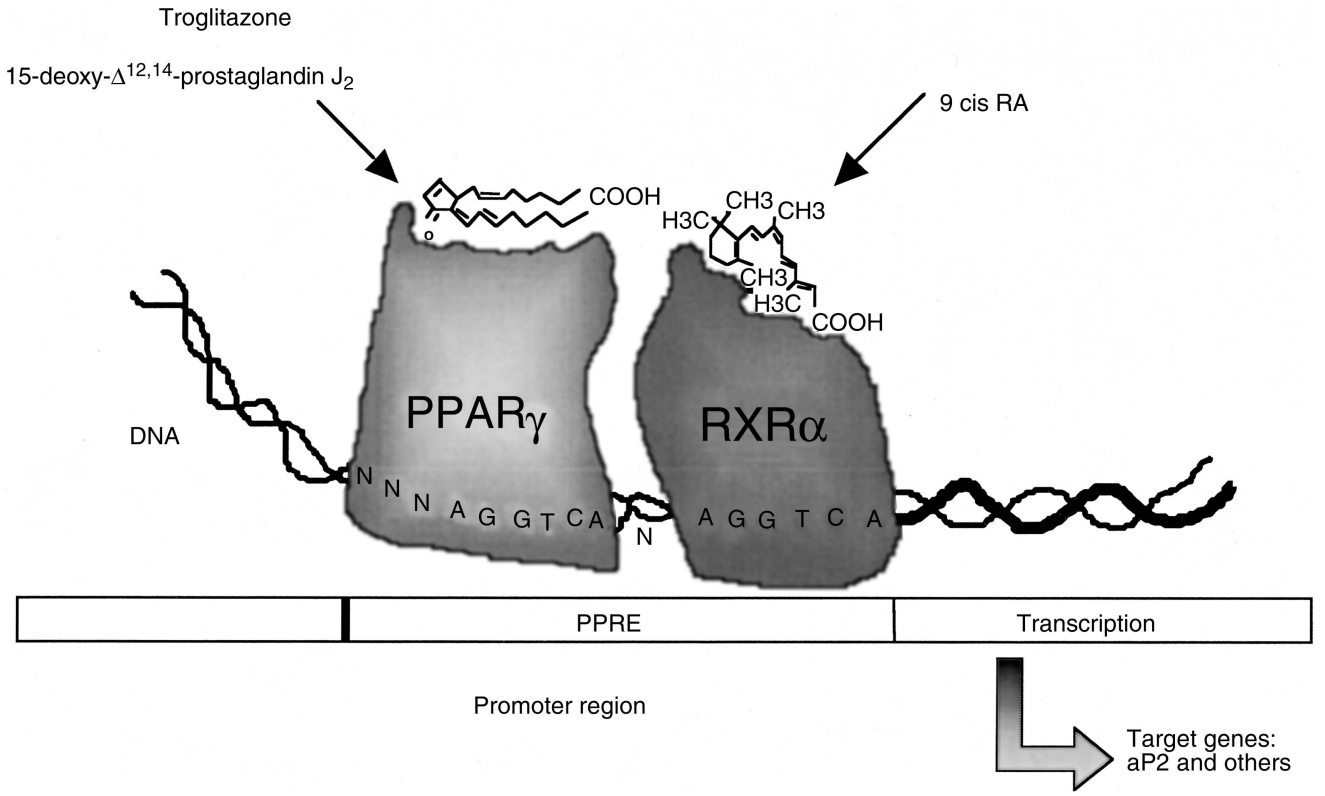

Fig. 2. Model for gene activation and repression by PPARγ.

C-terminal α-helix 12 is critical for both ligand-binding and recruitment of PPAR cofactors.

PPAR ligands

Similar to other members of the nuclear receptor family, PPARs are ligand-activated transcriptional factors, that is, modulation of target gene transcription depends on the binding of the ligands to the receptor. PPARs form heterodimers with the 9-*cis* retinoic acid receptor, RXRα. Activation of PPAR:RXRα by PPAR ligands and/or RXRα ligands results in a conformational change of these receptors, allowing the heterodimers to bind PPREs in target genes and modulate the gene transcription (Fig. 2). The divergent amino acid sequence in the LBD of the three PPAR isoforms provides the molecular basis for ligand specificity. Since PPARs have a large ligand-binding pocket (1300 Å) [14], these receptors are capable of binding several structurally diverse compounds, including industrial chemicals such as herbicides and plasticizers as well as pharmaceutical compounds including hypolipidemic fibrates (such as clofibrate) and antidiabetic thiazolidinediones (TZDs; for example, troglitazone and rosiglitazone). When assessed by *trans*activation assays, a variety of exogenous compounds have been shown to selectively activate PPAR isoforms (Table 1). Among them, several agents have been identified as PPARα activators, including hypolipidemic compounds WY-14,643 and clofibrate. Importantly, these compounds exhibit a good correlation between ability to activate PPARα and the potency to induce hepatocarcinoma in rodents, suggesting that PPARα is the major molecular target of hepatocarcinogenic peroxisome proliferators in the liver [15].

PPARγ is highly expressed in adipose tissue where it plays a critical role in adipocyte differentiation. Antidiabetic TZD including troglitazone and rosiglitazone promote adipocyte differentiation through activation of PPARγ [16–20]. In vitro, these TZDs have been shown to bind PPARγ with a kD of 30 to 10,000 nmol/L [21, 22]. A synthetic compound, L-165041, selectively activates PPARβ/δ at high affinity (kD = 6 nmol/L) with very poor affinity for PPARγ and no affinity to PPARα [23]. A search for naturally occurring ligands reveals that PPAR receptors are also activated by numerous fatty acid at physiologic concentrations [24]. PPARα is activated by a variety of long-chain polyunsaturated fatty acids including linoleic acid, branched, conjugated and oxidized fatty acids such as phytanic acid, conjugated linolenic acid and eicosanoids such as 8S-HETE and leukotriene B4 [25–28]. The endogenous prostaglandin D_2 (PGD_2) metabolite, 15-deoxy-$\Delta^{12,14}$ prostaglandin J_2 ($15dPGJ_2$) is the most potent natural ligand of PPARγ and might

Table 1. Peroxisome proliferator-activated receptor (PPAR) ligands

Ligands	PPAR α	PPAR β/δ	PPAR γ	Reference
Exogenous compounds				
WY-14,643	+++	+	++	[86, 227]
Clofibrate	+++	−	+/−	[177, 230]
ETYA	+++	−	+/−	[228]
Troglitazone	−	−	+++	[177, 230]
Rosiglitazone (BRL49653)	−	−	+++	[177, 230]
Pioglitazone	−	−	+++	[177, 230]
GW2433	−	+++	−	[231]
IL-165041	−	+++	+	[86, 227]
Indomethacin	+	−	+++	[229]
Ibuprofen	+	−	+	[229]
Endogenous activators				
Palmitic acid (16:0)	+++	++	−	[31]
Linoleic acid (18:2, n-6)	+++	++	+	[31]
Arachidonic acid (20:4, n-3)	+++	++	+++	[31]
PGA_1	+	++	+	[228]
PGD_2	++	+	++	[228]
PGJ_2	+	+/−	+++	[228]
15-Deoxy-$\Delta^{12,14}$-prostaglandin J_2	+	+/−	+++	[86]
8(S)-HETE	+++	−	−	[86, 228]
LTB_4	+/−	ND	ND	[86, 200]
9-HODE	ND	ND	+++	[29]
13-HODE	ND	ND	+++	[29]

Abbreviations are in the **Appendix**. Symbols are: +, activator; −, not an activator; +/−, possible activator; ND is not determined.

play a role in adipogenesis in vivo [21]. Interestingly, two pathophysiological oxidized metabolites of linoleic acid, 9-hydroxy and 13-hydroxy octadecadienoic acids (9-HODE and 13-HODE) present in oxidized low-density lipoprotein have also been suggested as PPARγ ligands and could play a role in atherogenesis [29, 30]. Linoleic acid, arachidonic acid, eicosapentenoic acid (EPA) and PGA_1 have been shown to bind PPARβ/δ receptor in the μmol/L range [31, 32].

PPAR ISOFORMS: TISSUE DISTRIBUTION, INTRARENAL LOCALIZATION AND FUNCTION

Tissue distribution and regulation of PPAR isoforms

Expression of the three PPAR isoforms has been examined in many species including *Xenopus*, rat, mouse, rabbit and human. In humans, PPARα is predominantly expressed in tissues exhibiting high catabolic rates of fatty acids (adipose tissue, liver, heart, muscle, and renal cortex) and it is also detected in lung, placenta, intestine, pancreas and skeletal muscle. PPARβ/δ is the most widely expressed isotype and low levels are found in almost all tissues examined. In general, PPARγ is highly expressed in adipose tissue and at lower levels in renal medulla, urinary bladder, skeletal muscle, liver, and heart [33–35]. Although all three PPARγ splice variants are expressed in adipose tissues at high levels, PPARγ2 and γ3 appear to be more restrictively expressed in fat than the γ1 isoform [9, 33, 34]. Recently, low but significant levels of PPARγ expression has also been detected in vascular smooth muscle cells, endothelial cells, hepatic Stellate cells, bone marrow stromal cells, monocytes/macrophages, and malignant epithelial cells including breast, colon, prostate and bladder cancer cells [36–46], suggesting that PPARγ may participate in many physiological and pathophysiological events in these tissues as well.

The expression of PPARα in liver is decreased by insulin and increased by glucocorticoids, respectively [47, 48]. PPARα expression cycles in parallel with the circadian rhythm of circulating glucocorticoids [49]. In hepatic cells and pancreatic islets, expression of PPARα is down-regulated with treatment of growth hormone [50], but up-regulated by fibrates or free fatty acid (FFA) [51, 52]. In humans, alternative splicing of mRNA may play a role in regulating PPARα expression. Human liver has a splice variant of PPARα transcript lacking exon 6. The resulting reading frame shift results in a premature stop codon shortly after the DNA binding domain, resulting in a truncated PPARα protein [53]. This process might diminish the hepatic expression of PPARα. In addition, a mutation at codon 162 (L162 V) of PPARα has been shown to be associated with hyperapobetalipoproteinemia in humans [54].

Regulation of PPARγ has been extensively investigated. Insulin and corticosteroids synergistically induce PPARγ expression, while tumor necrosis factor-α (TNF-α) suppresses the PPARγ level in adipocytes [55, 56]. In rodents, in vivo studies revealed that PPARγ expression in adipose tissue is significantly up-regulated by high-fat feeding and down-regulated by fasting [57–59]. In humans, increased expression of the PPARγ receptor has been observed in adipose tissue of obese humans [55] and skeletal muscle of patients with obesity or type II diabetes [55, 60, 61]. Genetic polymorphisms and mutations have been also found to play a role in PPARγ function in humans [61–63]. Pro12Ala substitution of PPARγ2 is associated with decreased receptor activity, low body mass index (BMI), and increased insulin sensitivity in nonobese subjects [62, 64]. In contrast, Pro115Gln mutation destroys the mitogen-activated protein (MAP) kinase target site in Ser112, resulting in increased activity of PPARγ and severe obesity [63]. Thus far, very little is known about the regulation of PPARβ/δ receptor. However a recent report suggests its expression may be repressed by the adenomatous polyposis coli (APC) gene through β-catenin/Tcf-4 transcription factor [65]. These data suggest a role for PPARβ/δ in the pathogenesis of colon cancer.

Intrarenal localization of PPARs

Renal expression of PPARs have been investigated in many species including mouse, rat, rabbit and humans [34, 35, 66–68]. Northern hybridization and RNase pro-

tection assay show that all three PPAR isoforms are expressed in the kidney. In situ hybridization analysis and reverse transcription-polymerase chain reaction (RT-PCR) provide concordant results for nephron expression [35, 67, 69]. PPARα is predominantly expressed in proximal tubules and medullary thick ascending limbs, while PPARγ is selectively expressed in medullary collecting ducts and pelvic urothelium. PPARβ/δ, like in other tissues, is equally expressed in renal cortex and medulla and is expressed ubiquitously in all segments of nephron tested. Recently, functional expression of PPARγ has also been reported in freshly isolated glomeruli and cultured mesangial cells in both rabbit and rat (abstract; Guan et al, *J Am Soc Nephrol* 11:A3381, 2000) [70, 71]. Renal expression of PPAR isoforms is also developmentally regulated. All three PPAR isoforms are expressed in early embryonic kidneys. PPARα and PPARγ expression is enhanced with development and remains at high level 2 weeks after birth, while PPARβ/δ expression peaks in late gestation and declines after birth [69, 72].

PHYSIOLOGICAL ROLES OF PPAR ISOFORMS

Distinct expression patterns and differential ligand-binding profiles indicate unique roles for each of the three PPAR isoforms. The target genes of PPARα participate in lipid oxidation [73], suggesting that PPARα is mainly involved in lipid catabolism. Conversely, PPARγ target genes are generally implicated in the lipogenic pathway and the storage of fatty acids in adipose tissue, consistent with a critical role of PPARγ in adipogenesis [20, 74]. Although PPARβ/δ target genes are poorly characterized, the fact that PPARβ/δ regulates acyl coenzyme A (CoA) synthase 2 expression suggests a role for this receptor in lipid metabolism [75]. In addition, gene targeting and human genetic studies provide important clues regarding the functional roles of PPAR receptors in mammalian metabolism.

PPARα in lipid catabolism

PPARα is predominantly expressed in tissues with high mitochondrial and peroxisomal β-oxidation activities, exemplified by its high levels in liver, heart and renal cortex and intestine. Its known target genes comprise a relatively homogenous group of genes involved in almost all aspects of lipid metabolism, including uptake, binding and oxidation of fatty acids, lipoprotein assembly and lipid transport [11]. PPARα activated genes include fatty acid transport protein (FATP), fatty acid translocase (FAT/CD36) and hepatic cytosolic fatty acid binding protein (L-FABP), and it may play critical roles in uptake of fatty acids in the digestive tract [76, 77]. Other genes involved in the transformation of long-chain fatty acids into triglycerides and their incorporation into chylomicrons and VLDL particles, such as acyl CoA synthase (ACS) [78] and apolipoproteins (apo) are also PPARα target genes [79, 80]. In liver, PPARα is a pivotal factor involved in every aspect of fatty acid oxidation. Triglycerides in chylomicrons are extracellularly hydrolyzed into glycerol and fatty acids by the lipoprotein lipase (LPL), another well-characterized PPARα target gene [76, 81, 82]. The resultant nonesterified fatty acids are then subsequently taken up by hepatocytes through the PPARα target gene, fatty acid transport protein (FATP) [83]. Once imported into hepatocytes, fatty acids are activated into acyl CoA thioesters by ACS. Acyl CoA may then either be esterified or, alternatively, enter the pathway of β-oxidation. Metabolizing long-chain fatty acyl CoA (>C20) via β-oxidization takes place in peroxisomes. All three enzymes involved in this process have been shown to be directly regulated by PPARα via binding to the PPRE sites in the promoter region of these genes. These genes include acyl CoA oxidase (ACO), a rate-limiting enzyme, enoyl CoA hydratase/dehydrogenase multifunctional enzyme (HD), and keto-acyl CoA thiolase [7, 84–86]. Long- (C14 to C20) and medium-chain (C8 to C12) fatty acids can be oxidized either in peroxisome or mitochondria. Mitochondrial β-oxidation is approximately twice as efficient as peroxisomal β-oxidation. The rate-limiting step in mitochondrial oxidation is entry flux of fatty acids into the mitochondria, which is controlled by a carnitine-dependent facilitated transport system. One of its critical components, carnitine palmitoyl transferase I (CPT I), is a direct target gene of PPARα [87, 88]. In addition, PPARα further regulates the mitochondrial β-oxidiation spiral by modulating the expression of the medium-chain acyl CoA dehydrogenase (MCAD) gene [89]. The acetyl CoA produced in mitochondria is converted to ketone bodies, mainly acetoacetate and 3-hydroxybutyrate, and serves as an important energy source for extrahepatic tissues such as brain, renal cortex, heart, and skeletal muscle. The main enzyme responsible for generation of ketone bodies is mitochondrial hydroxymethylglutaryl-CoA synthase (mHMG-CoAS), which is directly activated by PPARα [90]. The critical role of PPARα in fatty acid β-oxidation is confirmed by the studies in PPARα-null mice [91]. These animals are unresponsive to PPARα ligand stimulation in both peroxisomes and mitochondria, and exhibit fatty liver. Basal expression of mitochondrial enzymes are lower in PPARα knockout mice versus wild-type mice.

PPARα also plays an important role in regulating microsomal ω-oxidation of fatty acids and eicosanoids. The cytochrome P450 4 A (CYP4a) enzymes are central enzymes participating in this process. For example, ω-oxidation is the first step in the degradation of leukotriene B_4 (LTB_4), a PPARα ligand [92, 93]. To date, at least two CYP4A enzymes have been shown to be direct target genes of PPARα: 4A1 in rat and 4A6 in rabbit. Putative PPRE sites have been reported in the promoter

region of these genes and PPARα agonists are able to enhance their expression both in vivo and in vitro [94–97]. Since 4A1 and 4A6 are active arachidonate ω-hydroxylase [98], these data suggest that PPARα may actively participate in the oxidation of a variety of endogenous lipids as well as exogenous compounds in microsomes.

Additional PPARα target genes include apolipoprotein (apo) I, apo-II and apo C-III. PPARα ligand fibrates decrease apo C-III by activation of PPARα, leading to enhanced LPL-mediated catobolism of very low-density lipoprotein (VLDL) particles [99–101]. In contrast, PPARα activation increases plasma levels of HDL and its major constituents, apo A-I and A-II through PPAR/RXR heterodimers [79, 102]. These effects, together with negative regulation of apo C-III, contribute to the hypolipidemic effects of fibrates. Based on these observations, PPARα is a critical player in lipid catabolism, including peroxisomal and mitochondrial oxidation, microsomal ω-oxidation, ketogenesis, and lipid transport.

PPARβ/δ in development, fertility, bone formation and lipid metabolism

While it has been widely accepted that PPARα and PPARγ are two key players in lipid metabolism, it now appears likely that PPARβ/δ is also involved in lipid homeostasis. A recent study by Leibowitz et al showed that PPARβ/δ specific agonist L-165041 significantly increased high-density lipoprotein (HDL)-cholesterol levels without effects on blood glucose or triglycerides concentration in *db*/*db* obese mice. These results suggest that PPARβ/δ agonists could be a novel therapeutic agents in dyslipidemia [103].

Ubiquitous expression, poor availability of specific ligands and lack of a connection to clinical disorders have impeded progress in elucidating the specific function of PPARβ/δ. PPARβ/δ expression is particularly high in certain developing and adult tissues, such as central nervous system, kidney and placenta [34, 35, 66]. This expression pattern indicates a potential function of PPARβ/δ in these tissues. For example, PPARβ/δ has been found to be predominant isotype expressed in the uterus during the pre-implantation period in mice [104]. Its expression was induced in the stroma surrounding the implanting blastocyst and localized in the decidual zone postimplantation. Its obligate partner RXRα was also highly expressed in endometrial cells and decidua. In addition, PGI_2, an endogenous PPARβ/δ ligand, is generated in stromal cells at the site of implantation. More importantly, the PPARβ/δ agonists such as carbaprostacyclin or L-165041 in combination of 9-*cis* retinoic acid were able to restore implantation in the COX-2 gene knockout mice, in whom defects in implantation and decidualization were previously reported [104, 105]. These data suggest that PPARβ/δ plays an essential role in fertility. Recently, PPARβ/δ has been reported to be the predominant PPAR isoform expressed in bone osteoclasts, implicating it in regulating resorption of mineralized bone surface [106]. Activation of PPARβ/δ by carbaprostacyclin significantly induced both bone-resorbing activity and mRNA expression of the osteoclastic genes including carbonic anhydrase type II (CA II) and cathepsin K. Moreover, these effects were completely blocked by an antisense oligonucleotide of PPARβ/δ, suggesting a critical role for PPARβ/δ in modulating osteoclastic bone resorption.

PPARγ and adipogenesis

PPARγ has been the most extensively investigated PPAR isoform. The receptor has been cloned from many species and identified as the isoform most highly expressed in adipose tissues. The expression of adipocyte fatty acid binding protein (aP2), as well as many other adipose target genes including lipoprotein lipase (LPL), fatty acid translocase (FAT), fatty acid transport protein (FATP), ACS, malic enzyme (ME), insulin-dependent glucose transporter (GLUT4) and c-Cbl-associated protein (CAP), are activated by PPARγ ligands [49, 86]. This array of PPARγ target genes is consistent with the role of PPARγ in lipid accumulation. Moreover, a gain-of-function study showed that forced ectopic expression of PPARγ in fibroblasts produced a marked stimulation adipogenesis in the presence of PPARγ activators [107, 108]. This observation was confirmed further in vivo. Mutation of Pro115Gln in PPARγ destroys a MAP kinase target site in Ser112, resulting in increased activity of PPARγ receptor and severe obesity in humans [63]. Conversely, in vitro introduction of a dominant negative PPARγ mutant with a defective *trans*activation domain blocked TZD-induced terminal differentiation of human pre-adipocytes [109]. Unfortunately, loss-of-function (gene knockout) studies of adipogenesis using gene targeting have been hampered by an unexpected requirement of PPARγ for placental development and embryonic lethality [110]. Recently, several lines of evidence supported the critical role of PPARγ in adipogenesis in vivo. Study on mice chimeric for wild-type and PPARγ-null cells showed that PPARγ (−/−) embryonic stem (ES) cells fail to contribute to adipose tissues, whereas most other organs examined did not require PPARγ for proper development [18]. These data support an essential role for PPARγ in fat formation. Similar conclusions were supported in another study using PPARγ mutant mice generated by supplementing PPARγ null embryos with wild-type placenta, via aggregation with tetraploid embryos [110]. The live-born PPARγ null mice were notable for total absence of white and brown adipose tissue (lipodystrophy). Taken together, these data demonstrate that PPARγ is required for adipogenesis both in vitro and in vivo. Thus, together with a number of other transcription factor including CAAT/enhancer binding

proteins (C/EBPs), sterol regulatory element binding proteins (SREBPs) and PPARβ/δ, PPARγ promotes and maintains adipocyte function [19, 86, 111, 112].

PPARS AND RENAL DISEASE

Beyond a role in adipogenesis, PPARs have been linked to many human diseases including obesity, insulin resistance, type II diabetes, atherosclerosis, hypertension, inflammation and cancer. Ligands for these receptors have been suggested to have therapeutic potential for selected human diseases.

PPARγ, insulin resistance, and type II diabetes

Numerous studies have supported a therapeutic role for PPARγ in the treatment of type II diabetes. TZD PPARγ ligands including troglitazone (Rezulin®), pioglitazone (ACTOS®) and rosiglatazone (Avandia®) are a relatively new class of oral antidiabetic agents that improve all three primary defects in patients with type II diabetes, that is, insulin resistance, hyperinsulinemia and hyperglycemia [113]. Troglitazone significantly reduces plasma glucose, % glycosylated hemoglobin, insulin and C-peptide level in type II diabetes [113–116]. Moreover, this agent also shows beneficial effects of lipid metabolism, blood pressure and cardiac output [113, 117]. These effects are generally thought to be mediated by activation of PPARγ and enhanced insulin sensitivity in peripheral tissues [118]. In support of this, ligands of RXRα, the obligate partner of PPARγ, also exhibit a glucose- and lipid-lowering activity [119] that is additive to a PPARγ agonist [120]. Although TZDs have been successfully used in the treatment of type II diabetes, the relevant tissue targets remains undefined. Since PPARγ is abundantly expressed in adipose tissues, fat cells have been implicated as a primary target tissue for TZDs [121]. Arguing against this hypothesis is the observation that mice genetically manipulated to delete adipose tissue develop severe insulin resistance, and that troglitazone ameliorates hyperinsulinemia and hyperglycemia in this model as well [122]. This suggests that the insulin-sensitizing effects of TZD are independent of effects on adipose tissues. Another proposed target tissue for TZDs is skeletal muscle, the major site of impaired insulin action in type II diabetes. PPARγ has been found to be expressed in skeletal muscle tissue [123] and its expression is enhanced in type II diabetic patients [60, 124–126]. The activation of PPARγ increases expression of adipocyte lipid binding protein (ALBP) and muscle fatty acid binding protein (mFABP) in this tissue [125], potentially modulating lipid and fatty acid uptake. Fatty acids are key regulators of insulin sensitivity. It is well-established that fatty acids decrease glucose usage in skeletal muscle. PPARγ-induced increase in fatty acids uptake by ALBP and mFABP may be responsible, at least in part, for enhanced insulin sensitivity. TZD PPARγ ligands have been also shown to ameliorate TNF-α-induced insulin resistance, suggesting that suppression of TNF-α activation also might be involved in favorable effect of TZDs in diabetes and insulin resistance [127, 128].

Most recently, a new adipocyte-secreted hormone designated resistin (for "resistant to insulin") has been proposed as a molecular link between obesity and type II diabetes. TZD rosiglitazone markedly reduced resistin's expression in adipocytes as well as its circulating level. Resistin appears to play a role in pathogenesis of insulin resistance [129, 130], since administration of neutralizing antibody to resistin significantly improved blood glucose and insulin action in mice with diet-induced obesity, suggesting that TZD activation of PPARγ might increase insulin sensitivity by suppressing the production of resistin by adipocytes.

Despite the successful use of TZDs as antidiabetic drugs, the specific role of altered PPARγ function in the pathogenesis of insulin resistance and type II diabetes remains uncertain. It has long been known that obesity is a major risk factor in the development of type II diabetes. In obese rats, TZDs increase the number of small adipocytes and decrease the number of large adipocytes through adipocyte differentiation induced by PPARγ activation [131, 132]. Troglitazone may increase insulin sensitivity through these small adipocytes via inhibition of TNF-α expression and decreased plasma lipids [127]. In humans, loss-of-function mutations of PPARγ share elements of the insulin resistance syndrome (insulin resistance, type II diabetes, hypertension and dyslipidemia), but not obesity [133]. In contrast, patients with a gain-of-function mutation of PPARγ are obese, but have a relatively low level of insulin and increased sensitivity to insulin [63]. These data suggest that excess PPARγ activity contributes to obesity and reduced PPARγ activity elicits insulin resistance. However, studies on mice heterozygous for the PPARγ gene paint a more complex picture [134, 135]. PPARγ (+/−) mice develop normally, have a normal number and size of adipocytes and show no metabolic defect. However, the size of the adipocytes in PPARγ (+/−) mice on a high-fat diet is much smaller than those in wild-type mice, and PPARγ (+/−) mice unexpectedly exhibit enhanced insulin sensitivity and reduced weight gain as compared to wild-type mice. In this setting, PPARγ may promote small adipocyte formation, which is insulin sensitive. On the other hand, PPARγ activation can stimulate hypertrophy (large adipocytes) of pre-existing adipocytes, which leads to insulin resistance. Taken together, these data suggest a complex role for PPARγ in adiposity and insulin resistance.

PPARs and obesity

Both PPARα and PPARγ receptors have been implicated in the pathogenesis of obesity. PPARα agonist fibrate treatment has been reported to reduce weight gain in rodents [136, 137]. Moreover, PPARα null mice accumulate more body fat with aging [138, 139]. These data suggest that PPARα plays an important role in obesity. The mechanisms may involve both regulation of lipid catabolism and energy expenditure. PPARα has been shown to increase the expression of uncoupling proteins (UCP-1, UCP-2, and UCP-3), mitochondrial inner-membrane transporters responsible for heat generation through uncoupling proton entry from ATP synthesis [140, 141]. UCPs are important regulators of thermogenesis and body weight regulation. All three UCPs are abundantly expressed in brown adipose tissue (BAT), however, each UCP differs in its expression pattern in other tissues [142–147]. UCP-1 is relatively restricted to brown fat tissue [148]. In contrast, UCP-2 is expressed in most tissues at varying levels, while UCP-3 is expressed predominantly in skeletal muscle and brown adipose tissue. Given the wide tissue distribution pattern of UCP-2 and UCP-3, it was originally thought that they may contribute more to adaptive thermogenesis than UCP-1. However, increased rather than decreased expression of UCP-2 and UCP-3 has been observed after starvation, a state known to be associated with decreased energy expenditure [149, 150]. Furthermore, although transgenic mice with toxigen-mediated reduction of brown fat (UCP-DTA) develop obesity [151], UCP-1 targeted mice have a defect in adaptive thermogenesis but not obesity [142]. Thus, based on these undefined roles of UCPs in obesity, regulation of PPARα in this setting of disease requires more investigation.

The role of PPARγ in pathogenesis of obesity appears more clear. As discussed above, PPARγ plays a critical role in adipogenesis both in vitro and in vivo. Dysfunction of the PPARγ receptor has significant effects on body fat mass. This has been confirmed both by transgenic studies and human genetic studies [152]. Patients with an activating missense mutation of the PPARγ2 protein exhibit accelerated adipocyte differentiation and develop severe obesity [63]. In contrast, patients with a PPARγ2 mutation associated with decreased receptor activity exhibit lower body mass and improved insulin sensitivity [64]. Together with the previously discussed effects of PPARγ disruption on adiposity in knockout mice, these data clearly demonstrate that PPARγ plays a critical role in obesity. These effects on adipocytes may include modulation of leptin-stimulated lipolysis and glucose utilization. Thiazolidinediones inhibit leptin production [153] through PPARγ-dependent PPRE in promoter region of the leptin (*ob*) gene [154]. In addition, PPARγ has been also shown to regulate many other genes implicated in energy homeostasis, including TNF-α and UCPs [127, 155]. These data suggest that multiple mechanisms contribute to the pathogenic role of PPARγ in obesity.

PPARγ and hypertension

Hypertension in Western countries is commonly linked to obesity and insulin resistance [156, 157]. Recently thiazolidinediones (TZDs) have been found not only to sensitize tissue to insulin but also to lower blood pressure in both animals and humans [158–161]. Studies on hyperinsulinemic Watanabe heritable hyperlipidemic (WHHL) rabbits or obese Zucker rats show that the PPARγ ligand troglitazone increases insulin sensitivity and lowers blood pressure [159–161]. Similarly, troglitazone has been shown to be effective in ameliorating hypertension in patients with type II diabetes [158]. It has been suggested that the antihypertensive effect of TZDs is a result of improved insulin resistance, since enhanced insulin sensitivity has been found to be associated with lower blood pressure both in diabetic animals [162–165] and humans [166]. Direct vascular effects of PPARγ also may play a role, since recent studies have identified PPARγ in both endothelium and vascular smooth muscle cells [167–171]. Studies on one-kidney, one-clip hypertensive rats, a model not associated with insulin resistance, shows that pioglitazone significantly lowers the blood pressure [172]. It is conceivable that vascular expression of PPARγ could modulate the release of vasodilator autacoids [173]. Troglitazone has been reported to dilate peripheral blood vessels both in vitro and in vivo in animals [173, 174] and humans [117, 175]. These effects seem to depend on prostaglandin synthesis since troglitazone-induced vasorelaxation was abolished by the prostaglandin synthase inhibitor indomethacin [175], suggesting that these vasorelaxant effects are mediated by locally generated prostanoids, possibly endothelial derived prostacyclin [173]. Other studies suggest that antihypertensive effects of TZDs also involve decreased fatty acid levels, blockade of L-type calcium channels and modulation of vascular production of vasoactive peptides including type-C natriuretic peptide, endothelin, and plasminogen activator inhibitor-1 (PAI-1) [176, 177]. Alternatively, PPARγ may directly affect vascular smooth muscle cell (VSMC) tone. Troglitazone has been shown to inhibit VSMC proliferation and migration in vitro [40, 168], as well as following angioplasty [178]. Finally, PPARγ has been identified in collecting ducts and glomeruli (abstract; Guan et al, *J Am Soc Nephrol* 11:A3381, 2000) [35]. Constitutive expression of PPARγ in these segments might have important regulatory effects on salt and water absorption. These effects could contribute to the edema complicating treatment using TZDs in patients and animals [161, 179, 180]. Taken together, these data suggest that PPARγ expression con-

tributes to blood pressure regulation through multiple mechanisms.

PPARs and kidney disease

The role of PPAR isoform expression in the kidney is just now being elucidated, All three PPAR isoforms are expressed in the kidney of many species including humans [35, 69, 72]. PPARα expression predominates in the proximal tubule and thick limb, and could contribute to dietary lipid-induced gene expression of peroxisomal and mitochondrial β-oxidation in these segments [181]. The PPARα agonist clofibrate significantly induced expression of β-oxidation enzymes in renal cortex, including long-chain acyl CoA dehydrogenase (LCAD), medium-chain acyl CoA dehydrogenase (MCAD) and acyl CoA oxidase (ACO). These data suggest that renal PPARα plays a major role in triggering fatty acid utilization and the adaptive response to dietary lipids in the kidney. Notably, renal PPARα also colocalizes with P450 4 A enzymes, which are known PPARα target genes [93] that appear to be involved in blood pressure regulation [182]. Interestingly, clofibrate has been shown to reduce blood pressure in Dahl salt-sensitive rats [183]. These findings suggest that PPARα participates in blood pressure regulation through activation of P450 4 A enzymes in the proximal tubules. In addition, PPARα has also been shown to play an important role in maintaining a sustained balance of energy production/utilization in the kidney through regulating expression of genes involved in fatty acid β-oxidation [184]. This effect may contribute to the observation that PPARα ligands appear to protect kidney from ischemia/reperfusion injury in rats [184]. Mice lacking the PPARα gene exhibit more severe cortical necrosis and worse kidney function than wild-type control animals [184]. These findings help to explain the molecular mechanisms by which ischemia/reperfusion leads to suppression of fatty acid β-oxidation, and might provide a new therapeutic strategy for ischemic renal injury.

PPARγ mRNA is also highly expressed in the kidney, but in contrast to PPARα is predominantly expressed in collecting ducts, which suggests that this isoform might play a role in systemic water and sodium retention. In fact, in both rodents and humans, treatment with TZD PPARγ ligands may cause marked fluid retention, edema and hemodilution [161, 179, 180]. Although other mechanisms may be involved, the possibility that collecting duct PPARγ activation is responsible for water and sodium retention in TZD-treated diabetic patients has not been examined.

Cultured mesangial cells have also been found to express PPARγ [70]. Interestingly they also may produce a precursor of 15dPGJ2, a putative endogenous PPARγ ligand [185]. A preliminary report also demonstrates the presence of PPARγ in freshly isolated glomeruli, and suggest that a paracrine PPARγ system exists in glomerular mesangial cells (abstract; Guan et al, *J Am Soc Nephrol* 11:A3381, 2000). It is well established that mesangial cells play an important role in the progression of nephropathy. During inflammatory injury or high glucose stimulation, mesangial cells undergo a process of activation from a quiescent phenotype to a proliferative myofibroblast-like phenotype, characterized by increased α-smooth muscle actin and pro-inflammatory cytokines, as well as enhanced production of extracellular matrix proteins [186–188]. This phenotypic alteration has been considered a fundamental feature in the development of diabetic nephropathy and glomerulonephritis. Studies by Asano et al showed that the TZD PPARγ ligands, troglitazone, and endogenous PPARγ ligand, 15dPGJ2, can significantly suppress the production of α-smooth muscle actin, indicating that PPARγ activation may inhibit de-differentiation of mesangial cells [70]. In addition, in vitro studies have shown PPARγ ligands can significantly inhibit proliferation of mesangial cells in a dose-dependent manner [70]. These data suggest that PPARγ activation may reverse the phenotypic change of mesangial cells and induce cell growth arrest and reduction of extracellular matrix production. Therefore, glomerular PPARγ activation could be a therapeutic target for treating diabetic nephropathy. Indeed, troglitazone treatment has been shown to ameliorate microalbuminuria both in the streptozotocin-induced diabetic rat and type II diabetic patients [189, 190]. In the *fa/fa* rat, a genetic model of type II diabetes, intervention with troglitazone decreased proteinuria and halted the early onset and progression of mesangial expansion and glomerulosclerosis [191]. Similarly, pioglitazone and rosiglitazone markedly reduce proteinuria and slow the progression of diabetic nephropathy in genetically obese diabetic rats [161, 192].

Accumulating evidence suggests that TZDs have direct and beneficial glomerular effects. A recent study in streptozotocin-induced diabetic rat showed that troglitazone was not only able to prevent diabetic glomerular hyperfiltration and albuminuria, but also to reduce mRNA expression of extracellular matrix proteins (fibronectin and type IV collagen) and TGF-β1 in diabetic glomeruli. These beneficial effects in insulin-dependent diabetes mellitus suggest that PPARγ ligands can protect glomerular function independently of their capacity to increase glucose tolerance. Furthermore, in a nondiabetic glomerulosclerotic model, PPARγ ligands were effective in preventing the deterioration of renal function. In a 5/6 ablation model of renal failure, Ma et al recently noted that troglitazone treatment significantly reduced proteinuria, serum creatinine level and glomerulosclerosis possibly via inhibition of glomerular cell proliferation and reduction of TGF-β1 expression in glomeruli and renal tubules, suggesting a paracrine PPARγ system in glomerular mesangial cells (**Note added in proof**). This is consistent with

other studies showing a beneficial effect of troglitazone on glomeruli. Taken together, these studies suggest that TZDs may be used as possible therapeutic agents for diabetic nephropathy and glomerulosclerosis.

Finally, both PPARα and PPARγ have potent anti-inflammatory effects in macrophages [42, 193]. It is reasonable to anticipate that PPAR activation might also play a role in renal inflammation, including glomerulonephritis and interstitial nephritis; however, thus far few reports have addressed this important issue. A recent preliminary report suggested that the PPARγ ligand, troglitazone, significantly enhanced rather than repressed glomerular expression of MCP-1, a critical cytokine in recruiting monocytes in glomeruli, in a rat model of mesangioproliferative glomerulonephritis (abstract; Panzer et al., *J Am Soc Nephrol* 11:A2625, 2000). These results raise the possibility that certain PPARγ ligands might promote, rather than inhibit, the progression of glomerulonephritis. If so, PPARγ antagonists could provide a novel therapeutic approach to renal inflammatory diseases.

PPARs, inflammation and atheroscerosis

Renovascular stenosis and atheroemboli represent increasingly common disorders contributing to renal dysfunction in elderly diabetic patients [194, 195]. PPAR activation also may modify the course of atherosclerosis via effects on inflammation, modulation of lipid accumulation in the arterial wall and altered endothelial function, proliferation of vascular smooth muscle, and foam cell formation. All three PPAR isoforms may modify the progression of atherosclerosis through multiple mechanisms including modulation of plasma lipoprotein concentration, foam cell formation, inflammatory response and pro-atherosclerotic protein level.

Hypolipidemic fibrates (clofibrate and benzofibrate) are potent PPARα agonists proven to be beneficial in the prevention of atherosclerosis [196]. Formation of atherosclerotic lesions require recruitment of monocytes into the arterial wall through expression of specific adhesion molecules by endothelial cells. Not only is PPARα expressed in human endothelial cells [197], but its activation decreases the expression of the adhesion molecule VCAM-1 in these cells [198]. Moreover, PPARα has been implicated in inflammatory processes of the atherosclerotic lesion, which is believed to be responsible for disruption of atherosclerotic plaque [199]. PPARα-deficient mice show a prolonged response to inflammatory stimuli, suggesting PPARα modulates inflammation [200]. In hyperlipidemic patients, the PPARα ligand fenofibrate, significantly decreased plasma concentrations of interleukin-6 (IL-6). Anti-inflammatory effects were also observed in cultured vascular smooth muscle cells (VSMCs), which participate in plaque formation and postangioplasty re-stenosis. PPARα is functionally active in VSMCs and another ligand, Wy14643, inhibited the IL-1–stimulated release of IL-6 and prostaglandin and expression of cyclooxygenase-2 (COX-2) [199]. Data suggest that these effects were mediated by inhibition of nuclear factor-κB (NF-κB) and activator protein-1 (AP-1), two ubiquitous transcription factors that transduce the effects of many pro-atherogenic and inflammatory factors [201–203]. PPARα has also been shown to lower the plasma concentration of fibrinogen, an important risk factor for cardiovascular disease, both in rat and in human [199, 204]. In addition, PPARα ligands effectively increased HDL cholesterol level and decreased the low-density lipoprotein (LDL) cholesterol level in hyperlipidemic patients, as a result of transcriptional induction of HDL apolipoproteins, apo A-I and apo A-II, and LPL, and inhibition of hepatic apo C-III production through the PPARα receptor (see above) [4].

Little is known about the role of PPARβ/δ in atherosclerosis. Recent evidence has emerged that PPARβ/δ might be a target for treatment of dyslipidemia since a PPARβ/δ ligand increases the HDL-cholesterol level [103]. Since hyperlipidemia is a key atherogenic factor, the effect of PPARβ/δ in increasing HDL-cholesterol may be beneficial in prevention of atherosclerosis.

PPARγ has also been implicated in at least three pathologic processes in atherosclerosis: foam cell differentiation, inflammatory reaction and cell proliferation. The infiltrating monocytes in the intima of the arterial wall take up oxidized LDL via scavenger receptors (SRAI/II, SR-BI, CD36, etc.), promoting accumulation of intracellular lipids and generation of lipid-laden foam cells [205]. It has been shown that the oxidized LDL (OxLDL) scavenger receptor CD36 (fat) is under direct control of PPARγ. PPARγ ligand, in combination of RXRα agonist LG268, significantly up-regulates the expression of CD36 and the binding of OxLDL in a monocytic cell line [30]. Interestingly, OxLDL contains naturally occurring PPARγ ligands (9-HODE and 13-HODE). Furthermore, in the presence of OxLDL, PPARγ expression was markedly induced [206]. This positive feedback loop could promote atherosclerosis by stimulating the uptake of OxLDL in macrophage/foam cells. Cytokines produced by these activated macrophage/foam cells include M-CSF, IL-1, and TNF-α, and they initiate inflammation and induce proliferation of VSMCs. In contrast, other effects of PPARγ might be beneficial in the treatment of atherosclerosis. An anti-inflammatory effect of PPARγ in monocytes has been described. PPARγ has been shown to reduce cytokine (TNFα, IL-1, and IL-6) production [42], probably by inhibiting the activity of pro-inflammatory transcription factors such as NF-κB, AP-1 and STAT [207]. Other favorable effects of PPARγ include induction of apoptosis in monocytes [208], inhibition of proliferation of VSMCs [37, 178], suppression of metalloproteinase-9 expression [39], and reduction of plasma triglycerides [209]. On the basis of

these data, an important role for PPARγ in atherosclerosis remains uncertain.

Most recently, two studies provide clear evidence that TZDs treatment reduced atherosclerosis in vivo [210, 211]. In the Watanabe heritable hyperlipidemic (WHHL) rabbits, troglitazone treatment potently reduced the development of atherosclerotic lesions [210]. Similarly, rosiglitazone improved atherosclerotic lesions in LDL receptor knockout mice [211]. These data suggest that the anti-atherogenic effect of TZDs outweighs its pro-atherogenic effects in vivo. Although it is uncertain whether the reduction in atherosclerosis following TZD treatment is a direct effect of PPARγ activation in macrophages or an indirect effect secondary to improvements of insulin sensitivity, several lines of evidence demonstrated that PPARγ is not required for differentiation of macrophage lineage and the formation of foam cells [212, 213]. More importantly, PPARγ activation can promote cholesterol removal from human foam cells through stimulation of ABCA1, a protein involved in cholesterol export [212]. Additionally, PPARγ activation also results in the down-regulation of the LDL receptor scavenger SR-A in mouse macrophage [213]. Thus, macrophage is a direct target for TZD and PPARγ activation has multiple effects on the influx and efflux of cholesterol and oxidized lipid, countering their atherosclerotic effects of CD36 induction [214]. Data from a clinical trial of PPARγ ligands in dyslipidemic patients without type II diabetic patients will be required to clarify the role of PPARγ in this setting of disease.

PPARs, the cell cycle and cancer

Effects of PPARs on renal cell proliferation seem likely, but remain unexplored. All three isoforms of PPAR have been suggested to play a role in regulating the cell cycle and in carcinogenesis. PPARα, the predominant form of PPAR in the liver, is involved in the growth promoting and hepatocarcinogenic effect of peroxisome proliferators in rodents [15]. However, these rodent data do not appear relevant to humans. No carcinogenic effect of peroxisome proliferators has been shown in humans, possibly because of the much lower level of PPARα expression in human hepatocytes as a result of exon 6 splicing of PPARα transcripts [53].

Potent effects of PPARγ on cell proliferation and cell cycling have been described. PPARγ ligands can trigger cell cycle arrest in NIH3T3 cells and HIB-1B cells [215]. PPARγ ligands can also induce terminal differentiation and withdrawal of human liposarcoma cells from the cell cycle [216]. Importantly, TZD PPARγ ligands have been found to slow the progression of advanced liposarcoma in humans [217]. Given the expression of PPARγ in non-adipose tissues, the effect of PPARγ on human breast cancer, gastric cancer, prostate cancer, colon cancer and transitional cell bladder cancer have been explored. Treatment of cultured breast cancer cells with troglitazone results in cell growth arrest and promotes differentiation [44]. Troglitazone has also been shown to inhibit tumor growth and induce apoptosis in human breast cancer cells in vitro and in BNX mice [218]. Moreover, another PPARγ ligand, GW7845, has been shown to decrease tumor incidence, tumor growth and tumor burden in the NMU induced mammary carcinoma [219]. These data suggest that PPARγ ligands may be used as novel, nontoxic and selective chemotherapeutic agents for human breast cancer. A role for PPARγ activation also has been suggested in human transitional bladder cancers [46]. PPARγ is highly expressed in normal as well as malignant human transitional cell bladder cancer. Several PPARγ ligands (including troglitazone, ciglitazone and 15dPGJ2) significantly inhibit growth in cultured human bladder cancer cells, and this action is associated with induction of cyclin-dependent kinase inhibitors (p21 and p16) and reduction of cyclin D expression. Importantly, RXRα ligands 9-*cis* retinoic acid and LG100268 exhibited synergy with PPARγ ligands in inducing cell death. Anti-tumor effects of PPARγ ligands have been observed in human prostate cancer and gastric cancer as well [43, 220]. Conversely, the role of PPARγ in human colon cancer remains controversial. While PPARγ is expressed in human colon tumor and in cultured colon cell lines [221, 222], two groups recently reported that PPARγ ligands promote polyp formation in $APC^{min/+}$ mice, a murine model of the human genetic disease familial adenomatous polyposis coli (FAP) [223, 224]. These studies showed that when such mice are treated with troglitazone or with rosiglitazone, they develop an increased frequency of large but not small bowel polyps. In contrast, troglitazone reduces colon tumor xenograft volume in nude mice [222]. This corresponds to in vitro studies showing that activation of PPARγ results in colon cancer cell cycle arrest (G_1 phase) and differentiation [221], supporting the idea that PPARγ activation might be beneficial for human colon cancer as well. The different effects of troglitazone on polyp formation and cancer growth may reflect important biological differences in the two processes.

Recently, an important role for PPARβ/δ in carcinogenesis has been proposed [65]. PPARβ/δ expression appears to be increased in colorectal cancer and localized in dysplastic epithelial cells [225]. The NSAID sulindac, which suppress colorectal tumorigenesis, can antagonize PPARβ/δ-activated gene transcription in an in vitro assay system. These findings suggest that NSAIDs may inhibit tumorigenesis through inhibition of PPARβ/δ activity, thereby contributing to their chemopreventive effects [226], and raise the possibility that PPARβ/δ antagonists might have chemopreventive effects in human colorectal cancer. Specific roles of PPARs in renal cancers and cystic disease remain unexplored.

SUMMARY

In summary, PPARs are widely expressed throughout the body and are potent regulators of cellular metabolism. Activation of PPARs can be selectively achieved with specific ligands resulting in distinct biologic effects. Activation of PPARα by fibrates activates expression of genes involved in lipid catabolism. PPARγ activation by thiazolidinediones and $15dPGJ_2$ exerts many diverse biological effects on adipogenesis, inflammation, insulin sensitivity and tumorigenesis. PPARβ/δ, possibly activated by prostacyclin and other lipids, may be involved in lipid metabolism and tumorigenesis. Given the high expression of all three isoforms in the kidney, PPARs may play an important role in renal physiology and pathophysiology. The availability of PPAR-selective agonists and antagolists provide a new approach to modulate renal function in disease states including diabetic nephropathy, glomerulonephritis, glomerulosclerosis, hypertensive nephropathy, and diseases of renal epithelial proliferation. The use of these compounds in treating renal disease awaits experimental testing.

Reprint requests to YouFei Guan, M.D., Ph.D., Division of Nephrology, S-3223 MCN, Vanderbilt University Medical Center, Nashville, Tennessee 37232–2372, USA.
E-mail: youfei.guan@mcmail.vanderbilt.edu

APPENDIX

Abbreviations used in this article are: ACO, acyl CoA oxidase; ACS, acyl CoA synthase; AF1, activation function 1; ALBP, adipocyte lipid-binding protein; AP-1, activator protein-1; aP2, adipocyte fatty acid-binding protein; APC, adenomatous polyposis coli; apo, apolipoprotein; ATP, adenosine 5′-triphosphate; BAT, brown adipose tissue; BMI, body mass index; CAP, c-Cbl-associated protein; C/EBPs, CAAT/enhancer binding proteins; COX, cyclooxygenase; CYP4a, cytochrome P450 4A; DBD, DNA binding domain; EPA, eicosapentenoic acid; ES, embryonic stem; FAP, familial adenomatous polyposis coli; FAT, fatty acid translocase; FATP, fatty acid transport protein; FFA, free fatty acid; GLUT4, insulin-dependent glucose transporter; HD, enoyl-CoA hydratase/dehydrogenase multifunctional enzyme; HDL, high-density lipoprotein; 9-HODE and 13-HODE, 9-hydroxy and 13-hydroxy octadecadienoic acids; IL, interleukin; LBD, ligand-binding domain; LCAD, long-chain acyl CoA dehydrogenase; LDL, low-density lipoprotein; L-FABP, hepatic cytosolic fatty acid binding protein; LPL, lipoprotein lipase; LTB_4, leukotriene B_4; MAP, mitogen-activated protein; MCAD, medium-chain acyl CoA dehydrogenase; ME, malic enzyme; mFABP, muscle fatty acid binding protein; mHMG-CoAS, mitochondrial hydroxymethylglutaryl-CoA synthase; NF-κB, nuclear factor-κB; NSAID, nonsteroidal antiinflammatory drug; OxLDL, oxidized low-density lipoprotein; PGD_2, prostaglandin D_2; PPARs, peroxisome proliferator-activated receptors; PPRE, peroxisome proliferator response element; SREBPs, sterol regulatory element binding proteins; TGF-β, transforming growth factor-β; TNF-α, tumor necrosis factor-α; TZDs, thiazolidinediones; UCP, uncoupling proteins; VCAM, vascular cell adhesion molecule; VSMC, vascular smooth muscle cell; WHHL, Watanabe heritable hyperlipidemia.

NOTE ADDED IN PROOF

Ma L-J, Marcantoni C, Linton MF, *et al*: Peroxisome proliferator-activated receptor-γ agonist troglitazone protects against nondiabetic glomerulosclerosis in rats. *Kidney Int* 59:1899–1910, 2001

REFERENCES

1. Reddy JK, Azarnoff DL: Hypolipidemic hepatic peroxisome proliferators from a novel class of chemical carcinogens. *Nature* 283:397–398, 1980
2. Corton JC, Anderson SP, Stauber A: Central role of peroxisome proliferator-activated receptors in the actions of peroxisome proliferators. *Ann Rev Pharmacol Toxicol* 40:491–518, 2000
3. Issemann I, Green S: Activation of a member of the steroid hormone receptor superfamily by peroxisome proliferators. *Nature* 347:645–650, 1990
4. Staels B, Dallongeville J, Auwerx J, *et al:* Mechanism of action of fibrates on lipid and lipoprotein metabolism. *Circulation* 98:2088–2093, 1998
5. Wu GDA: Nuclear receptor to prevent colon cancer. *N Engl J Med* 342:651–653, 2000
6. Auwerx J: PPARγ, the ultimate thrifty gene. *Diabetetogia* 42: 1033–1049, 1999
7. Dreyer C, Krey G, Keller H, *et al:* Control of the peroxisomal β-oxidation pathway by a novel family of nuclear hormone receptor. *Cell* 68:879–887, 1992
8. Zhu Y, Qi C, Korenberg JR, *et al:* Structural organization of mouse peroxisome proliferator-activated receptor γ (mPPAR γ) gene: Alternative promotor use and different splicing yield two mPPAR γ isoforms. *Proc Natl Acad Sci USA* 92:7921–7925, 1995
9. Fajas L, Fruchart JC, Auwerx J: PPAR γ 3 mRNA: A distinct PPAR γ mRNA subtype transcribed from an independent promoter. *FEBS Lett* 438:55–60, 1998
10. Fajas L, Auboeuf D, Raspé E, *et al:* The organization, promoter analysis, and expression of the human PPARγ gene. *J Biol Chem* 272:18779–18789, 1997
11. Escher P, Wahli W: Peroxisome proliferator-activated receptors: Insight into mutiple cellular functions. *Mut Res* 448:121–138, 2000
12. Juge-Aubry CE, Hammar E, Siegrist-Kaiser C, *et al:* Regulation of the transcriptional activity of the peroxisome proliferator-activated receptor α by phosphorylation of a ligand-independent trans-activating domain. *J Biol Chem* 274:10505–10510, 1999
13. Shao D, Rangwala SM, Bailey ST, *et al:* Interdomain communication regulating ligand binding by PPARγ. *Nature* 396:377–380, 1998
14. Moras D, Gronemeyer H: The nuclear receptor ligand-binding domain: Structure and function. *Curr Opin Cell Biol* 3:384–391, 1998
15. Corton JC, Lapinskas PJ, Gonzalez FJ: Central role of PPARα in the mechanism of action of hepatocarcinogenic peroxisome proliferators. *Mut Res* 448:139–151, 2000
16. Lambe KG, Tugwood JD: A human peroxisome-proliferator-activated receptor-γ is activated by inducers of adipogenesis, including thiazolidinedione drugs. *Eur J Biochem* 239:1–7, 1996
17. Lowell BB: PPARγ: An essential regulator of adipogenesis and modulator of fat cell function. *Cell* 99:239–242, 1999
18. Rosen ED, Sarraf P, Troy AE, *et al:* PPAR gamma is required for the differentiation of adipose tissue in vivo and in vitro. *Mol Cell* 4:611–617, 1999
19. Wu Z, Rosen ED, Brun R, *et al:* Cross-regulation of C/EBPα and PPARγ controls the transcriptional pathway of adipogenesis and insulin sensitivity. *Mol Cell* 3:151–158, 1999
20. Spiegelman BM, Hu E, Kim JB, Burn R: PPARγ and the control of adipogenesis. *Biochimie* 79:111–112, 1997
21. Forman B, Tontonoz P, Chen J, *et al:* 15-Deoxy-Δ12,14-prostaglandin J2 is a ligand for the adipocyte determination factor PPAR-gamma. *Cell* 83:803–812, 1995
22. Lehmann JM, Morre LB, Smith-Oliver TA, *et al:* An antidiabetic thiazolidinedione is a high affinity ligand for peroxisome proliferator-activated receptor γ (PPARγ). *J Biol Chem* 270:12953–12956, 1995
23. Leibowitz MD, Fievet C, Hennuyer N, *et al:* Activation of PPARdelta alters lipid metabolism in db/db mice. *FEBS Lett* 473: 333–336, 2000
24. Kliewer SA, Sundseth SS, Jones SA, *et al:* Fatty acids and eicosanoids regulate gene expression through direct interactions with peroxisome proliferator-activated receptors α and γ. *Proc Natl Acad Sci USA* 94:4318–4323, 1997

25. Heuvel JPV: Peroxisome-proliferators-activated receptors: A critical link among fatty acids, gene expression and carcinogenesis. *J Nutr* 129:575S–580S, 1999
26. Ellinghaus P, Wolfrum C, Assmann G, *et al:* Phytanic acid activates the peroxisome proliferator-activated receptor alpha (PPARalpha) in sterol carrier protein-2/sterol carrier protein X-deficient mice. *J Biol Chem* 274:2766–2772, 1999
27. Moya-Camarena SY, Heuvel JPV, Blanchard SG, *et al:* Conjugated linoleic acid is a potent naturally occurring ligand and activator of PPARα. *J Lipid Res* 40:1426–1433, 1999
28. Lin Q, Ruuska SE, Shaw NS, *et al:* Ligand selectivity of peroxisome proliferator activated receptor alpha. *Biochemistry* 38:185–190, 1999
29. Nagy L, Tontonoz P, Alvarez JGA, *et al:* Oxidized LDL regulates macrophage gene expression through ligand activation of PPARγ. *Cell* 93:229–240, 1998
30. Tontonoz P, Nagy L, Alvarez JG, *et al:* PPARγ promotes monocyte/macrophage differentiation and uptake of oxidized LDL. *Cell* 93:241–252, 1998
31. Xu HE, Lambert MH, Montana VG, *et al:* Molecular recognition of fatty acids by peroxisome proliferator-activated receptors. *Mol Cell* 3:397–403, 1999
32. Forman BM, Chen J, Evans RM: Hypolipidemic drugs, polyunsaturated fatty acids, and eicosanoids are ligands for peroxisome proliferator-activated receptors α and δ. *Proc Natl Acad Sci USA* 94:4312–4317, 1997
33. Auboeuf D, Rieusset J, Fajas L, *et al:* Tissue distribution and quantification of the expression of mRNAs of peroxisome proliferator-activated receptors and liver X receptor-α in humans. No alteration in adipose tissue of obese and NIDDM patients. *Diabetes* 46:1319–1327, 1997
34. Mukherjee R, Jow L, Croston GE, Paterniti JR Jr: Identification, characterization, and tissue distribution of human peroxisome proliferator-activated receptor (PPAR) isoforms PPARγ2 *versus* PPARγ1 and activation with retinoid X receptor agonists and antagonists. *J Biol Chem* 272:8071–8076, 1997
35. Guan Y, Zhang Y, Davis L, Breyer MD: Expression of peroxisome proliferator-activated receptors in urinary tract of rabbits and humans. *Am J Physiol* 273:F1013–F1022, 1997
36. Greene ME, Blumberg B, McBride OW, *et al:* Isolation of the human peroxisome proliferator activated receptor gamma cDNA. expression in hematopoietic cells and chromosomal mapping. *Gene Expression* 4:281–299, 1995
37. Law RE, Goetze S, Xi X-P, *et al:* Expression and function of PPARγ in rat and human vascular smooth muscle cells. *Circulation* 101:1311–1318, 2000
38. Iijima K, Yoshizumi M, Ako J, *et al:* Expression of peroxisome proliferator-activated receptor γ (PPARγ) in rat aortic smooth muscle cells. *Biochem Biophys Res Commun* 247:353–356, 1998
39. Marx N, Schönbeck U, Lazar MA, *et al:* Peroxisomal proliferator-activated receptor gamma activators inhibit gene expression and migration in human vascular smooth muscle cells. *Circ Res* 83:1097–1103, 1998
40. Marx N, Bourcier T, Sukhova GK, *et al:* PPARγ activation in human endothelial cells increases plasminogen activator inhibitor type-1 expression: PPARγ as a potential mediator in vascular disease. *Arterioscler Thromb Vasc Biol* 19:546–551, 1999
41. Galli A, Crabb D, Price D, *et al:* Peroxisome proliferator-activated receptor γ transcriptional regulation is involved in platelet-derived growth factor-induced proliferation of human hepatic stallate cells. *Hepatology* 31:101–108, 2000
42. Jiang C, Ting AT, Seed B: PPARγ agonists inhibit production of monocyte inflammatory cytokines. *Nature* 391:82–86, 1998
43. Kubuta T, Koshizuka K, Williamson EA, *et al:* Ligand for peroxisome proliferator-activated receptor γ (troglitazone) has potent antitumor effect against human prostate cancer both in vitro and in vivo. *Cancer Res* 58:3344–3352, 1998
44. Mueller E, Sarraf P, Tontonoz P, *et al:* Terminal differentiation of human breast cancer through PPARγ. *Mol Cell* 1:465–470, 1998
45. Dubois RN, Gupta R, Brockman J, *et al:* The nuclear eicosanoid receptor, PPARγ, is aberrantly expressed in colonic cancers. *Carcinogenesis* 19:49–53, 1998
46. Guan Y, Zhang Y, Breyer RM, *et al:* Expression of peroxisome proliferator-activated receptor γ (PPARγ) in human transitional bladder cancer and its role in inducing cell death. *Neoplasia* 1:330–339, 1999
47. Lemberger T, Staels B, Saladin R, *et al:* Regulation of the peroxisome proliferator-activated receptor α gene by glucocorticoids. *J Biol Chem* 269:24527–24530, 1994
48. Steineger HH, Sorensen HN, Tugwood JD *et al:* Dexamethasone and insulin demonstrate marked and opposite regulation of the steady-state mRNA level of the peroxisome proliferator-activated receptor (PPAR) in hepatic cells-hormonal modulation of fatty acid-induced transcription. *Eur J Biochem* 225:967–974, 1994
49. Lemberger T, Saladin R, Vazquez M, *et al:* Expression of the peroxisome proliferator-activated receptor α gene is stimulated by stress and follows a diurnal rhythm. *J Biol Chem* 271:1764–1769, 1996
50. Yamada J, Sugiyama HW, Atanabe T, Suga T: Suppressive effect of growth hormone on the expression of peroxisome proliferator-activated receptor in cultured rat hepatocytes. *Res Commun Mol Pathol Pharmacol* 90:173–176, 1995
51. Sterchele PF, Sun H, Peterson RE, Vanden Heuvel JP: Regulation of peroxisome proliferator-activated receptor-alpha mRNA in rat liver. *Arch Biochem Biophys* 326:281–289, 1996
52. Zhou YT, Shimabukuro M, Wang MY, *et al:* Role of peroxisome proliferator-activated receptor α in disease of pancreatic β cells. *Proc Natl Acad Sci USA* 95:8898–8903, 1998
53. Palmer CNA, Hsu M-H, Griffin KJ, *et al:* Peroxisome proliferator activated receptor-α expression in human liver. *Mol Pharmacol* 53:14–22, 1998
54. Vohl MC, Lepage P, Gaudet D, *et al:* Molecular scanning of the human PPARα gene. Association of the L162V mutation with hyperapobetalipoproteinemia. *J Lipid Res* 41:945–952, 2000
55. Vidal-Puig AJ, Considine RV, Jimenez-Linan M, *et al:* Peroxisome proliferator-activated receptor gene expression in human tissues: Effect of obesity, weight loss, and regulation by insulin and glucocorticoids. *J Clin Invest* 99:2416–2422, 1997
56. Zhang B, Berger J, Hu E, *et al:* Negative regulation of peroxisome proliferator-activated receptor-γ gene expression contributes to the antiadipogenic effects of tumor necrosis factor-α. *Mol Endocrinol* 10:1457–1466, 1996
57. Shimoike T, Yanase T, Umeda F, *et al:* Subcutaneous or visceral adipose tissue expression of the PPARgamma gene is not altered in the fatty (fa/fa) Zucker rat. *Metabolism* 47:1494–1498, 1998
58. Pearson SL, Cawthorne MA, Clapham JC, *et al:* The thiazolidinedione insulin sensitizer, BRL 49653, increases the expression of PPARγ and aP2 in adipose tissue of high-fat-fed rats. *Biochem Biophys Res Commun* 229:752–757, 1996
59. Vidal-Puig A, Jimenez-Linan M, Lowell BB, *et al:* Regulation of PPAR γ gene expression by nutrition and obesity in rodents. *J Clin Invest* 97:2553–2561, 1996
60. Loviscach M, Rehman N, Carter L, *et al:* Distribution of peroxisome proliferator-activated receptors (PPARs) in human skeletal muscle and adipose tissue: Relation to insulin action. *Diabetologia* 43:304–311, 2000
61. Kruszynska YT, Mukherjee R, Jow L, *et al:* Skeletal muscle peroxisome proliferator-activated receptor-gamma expression in obesity and non-insulin-dependent diabetes mellitus. *J Clin Invest* 101:543–548, 1998
62. Yen C-J, Beamer BA, Negri C, *et al:* Molecular scanning of the human peroxisome proliferator activated receptor γ (hPPARγ) gene in diabetic Caucasians: Identification of a Pro12Ala PPARγ2 missense mutation. *Biochem Biophys Res Commun* 241:270–274, 1997
63. Ristow M, Muller-Wieland D, Pfeiffer A, *et al:* Obesity associated with a mutation in a genetic regulator of adipocyte differentiation. *N Engl J Med* 339:953–959, 1998
64. Deeb SS, Fajas L, Nemoto M, *et al:* A Pro12Ala substitution in PPARgamma2 associated with decreased receptor activity, lower body mass index and improved insulin sensitivity. *Nat Genet* 20: 284–287, 1998
65. He TC, Chan TA, Vogelstein B, Kinzler KW: PPARdelta is

an APC-regulated target of nonsteroidal anti-inflammatory drugs. *Cell* 99:335–345, 1999
66. Braissant O, Foufelle F, Scotto C, *et al:* Differential expression of peroxisome proliferator-activated receptors (PPARs): Tissue distribution of PPAR-α, β, and -γ in the adult rat. *Endocrinology* 137:354–366, 1996
67. Kliewer SA, Forman BM, Blumberg B, *et al:* Differential expression and activation of a family of murine peroxisome proliferator-activated receptors. *Proc Natl Acad Sci USA* 91:7355–7359, 1994
68. Jain S, Pulikuri S, Zhu Y, *et al:* Differential expression of the peroxisome proliferator-activated receptor γ (PPARγ) and its coactivators steroid receptor coactivator-1 and PPAR-binding protein PBP in the brown fat, urinary bladder, colon, and breast of the mouse. *Am J Pathol* 153:349–354, 1998
69. Yang T, Michele DE, Park J, *et al:* Expression of peroxisomal proliferator-activated receptors and retinoid X receptors in the kidney. *Am J Physiol* 277:F966–F973, 1999
70. Asano T, Wakisaka M, Yoshinari M, *et al:* Peroxisome proliferator-activated receptor gamma1 (PPARgamma1) expresses in rat mesangial cells and PPARgamma agonists modulate its differentiation. *Biochim Biophys Acta* 1497:148–154, 2000
71. Iwashima Y, Eto M, Horiuchi S, Sano H: Advanced glycation end product-induced peroxisome proliferator-activated receptor γ gene expression in the cultured mesangial cells. *Biochem Biophys Res Commun* 264:441–448, 1999
72. Braissant O, Wahli W: Differential expression of peroxisome proliferator-activated receptor-α-β, and -γ during rat embryonic development. *Endocrinology* 139:2748–2754, 1998
73. Kersten S, Desvergne BW, Ahli W: Roles of PPARs in health and disease. *Nature* 405:421–424, 2000
74. Kliewer SA, Willson TM: The nuclear receptor PPARγ: Bigger than fat. *Curr Opin Genet Dev* 8:576–581, 1998
75. Basu-Modak S, Braissant O, Escher P, *et al:* Peroxisome proliferator-activated receptor beta regulates acyl-CoA synthase 2 in reaggregated rat brain cell cultures. *J Biol Chem* 274:35881–35888, 1999
76. Motojima K, Passilly P, Peters JM, *et al:* Expression of putative fatty acid transporter gene are regulated by peroxisome proliferator-activated receptor α and γ activators in a tissue- and inducer-specific manner. *J Biol Chem* 273:16710–16714, 1998
77. Issemann I, Prince R, Tugwood J, Green S: A role for fatty acids and liver fatty acid binding protein in peroxisome proliferation? *Biochem Soc Trans* 20:824–827, 1992
78. Schoonjans K, Watanabe M, Suzuki H, *et al:* Induction of the acyl-coenzyme A synthetase gene by fibrates and fatty acids is mediated by a peroxisome proliferator response element in the C promoter. *J Biol Chem* 270:19269–19276, 1995
79. Vu-Dac N, Schoonjans K, Kosykh V, *et al:* Fibrates increase human apolipoprotein A-II expression through activation of the peroxisome proliferator-activated receptor. *J Clin Invest* 96:741–750, 1995
80. Staels B, Vu-Dac N, Kosykh VA, *et al:* Fibrates downregulate apolipoprotein C-III expression independent of induction of peroxisomal acyl coenzyme A oxidase. A potential mechanism for the hypolipidemic action of fibrates. *J Clin Invest* 95:705–712, 1995
81. Schoonjans K, Peinado-Onsurbe J, Lefebvre AM, *et al:* PPARalpha and PPARgamma activators direct a distinct tissue-specific transcriptional response via a PPRE in the lipoprotein lipase gene. *EMBO J* 15:5336–5348, 1996
82. Heller F, Harvengt C: Effects of clofibrate, bezafibrate, fenofibrate and probucol on plasma lipolytic enzymes in normolipaemic subjects. *Eur J Clin Pharmacol* 25:57–63, 1983
83. Martin G, Schoonjans K, Lefebvre AM, *et al:* Coordinate regulation of the expression of the fatty acid transport protein and acyl-CoA synthetase genes by PPARalpha and PPARgamma activators. *J Biol Chem* 272:28210–28217, 1997
84. Tugwood JD, Isseman I, Anderson RG, *et al:* The mouse peroxisome proliferator activated receptor recognizes a response element in the 5′-flanking sequence of the rat acyl-CoA oxidase gene. *EMBO J* 11:433–439, 1992
85. Zhang B, Marcus SL, Sajjadi FG, *et al:* Identification of a peroxisome proliferator-responsive element upstream of the gene encoding rat peroxisomal enoyl-CoA hydratase/3-hydroxyacyl-CoA dehydrogenase. *Proc Natl Acad Sci USA* 89:7541–7545, 1992
86. Desvergene B, Wahli W: Peroxisome proliferator-activated receptor: Nuclear control of metabolism. *Endocr Rev* 20:649–688, 1999
87. Mascaro C, Acosta E, Ortiz JA, *et al:* Control of human muscle-type carnitine palmitoyltransferase I gene transcription by peroxisome proliferator-activated receptor. *J Biol Chem* 273:8560–8563, 1998
88. Yu GS, Lu YC, Gulick T: Co-regulation of tissue-specific alternative human carnitine palmitoyltransferase Ibeta gene promoters by fatty acid enzyme substrate. *J Biol Chem* 273:32901–32909, 1998
89. Gulick T, Cresci S, Caira T, *et al:* The peroxisome proliferator-activated receptor regulates mitochondrial fatty acid oxidative enzyme gene expression. *Proc Natl Acad Sci USA* 91:11012–11016, 1994
90. Rodriguez JC, Gil-Gomez G, Hegardt FG, Haro D: Peroxisome proliferator-activated receptor mediates induction of the mitochondrial 3-hydroxy-3-methylglutaryl-CoA synthase gene by fatty acids. *J Biol Chem* 269:18767–18772, 1994
91. Peters JM, Hennuyer N, Staels B, *et al:* Alterations in lipoprotein metabolism in peroxisome proliferator-activated receptor alpha-deficient mice. *J Biol Chem* 272:27307–27312, 1997
92. Mayatepek E, Tiepelmann B: Defective degradation of leukotrienes in peroxisomal-deficient human hepatocytes. *Biochem Biophys Res Commun* 227:131–134, 1996
93. Johnson EF, Palmer CNA, Griffin KJ, Hsu M-H: Role of the peroxisome proliferator-activated receptor in cytochrome P450 4A gene regulation. *FASEB J* 10:1241–1248, 1996
94. Jedlitschky G, Mayatepek E, Keppler D: Peroxisomal leukotriene degradation: Biochemical and clinical implications. *Adv Enzyme Regul* 33:181–194, 1993
95. Muerhoff AS, Griffin KJ, Johnson EF: The peroxisome proliferator-activated receptor mediates the induction of CYP4A6, a cytochrome P450 fatty acid omega-hydroxylase, by clofibric acid. *J Biol Chem* 267:19051–19053, 1992
96. Aldridge TC, Tugwood JD, Green S: Identification and characterization of DNA elements implicated in the regulation of CYP4A1 transcription. *Biochem J* 306:473–479, 1995
97. Kroetz DL, Yook P, Costet P, *et al:* Peroxisome proliferator-activated receptor alpha controls the hepatic CYP4A induction adaptive response to starvation and diabetes. *J Biol Chem* 273: 31581–31589, 1998
98. Roman RJ, Alonso-Galicia M, Wilson TW: Renal P450 metabolites of arachidonic acid and the development of hypertension in Dahl salt-sensitive rats. *Am J Hypertens* 10:63S–67S, 1997
99. de Graaf J, Hendriks JC, Demacker PN, Stalenhoef AF: Identification of multiple dense LDL subfractions with enhanced susceptibility to in vitro oxidation among hypertriglyceridemic subjects. Normalization after clofibrate treatment. *Arterioscler Thromb* 13:712–719, 1993
100. Bard JM, Parra HJ, Camare R, *et al:* A multicenter comparison of the effects of simvastatin and fenofibrate therapy in severe primary hypercholesterolemia, with particular emphasis on lipoproteins defined by their apolipoprotein composition. *Metabolism* 41:498–503, 1992
101. Lussier-Cacan S, Bard JM, Boulet L, *et al:* Lipoprotein composition changes induced by fenofibrate in dysbetalipoproteinemia type III. *Atherosclerosis* 78:167–182, 1989
102. Vu-Dac N, Schoonjans K, Laine B, *et al:* Negative regulation of the human apolipoprotein A-I promoter by fibrates can be attenuated by the interaction of the peroxisome proliferator-activated receptor with its response element. *J Biol Chem* 269:31012–31018, 1994
103. Leibowitz MD, Fievetd C, Hennuyerd N, *et al:* Activation of PPARdelta alters lipid metabolism in db/db mice. *FEBS Lett* 473: 333–336, 2000
104. Lim H, Gupta RA, Ma WG, *et al:* Cyclo-oxygenase-2-derived prostacyclin mediates embryo implantation in the mouse via PPARdelta. *Genes Dev* 13:1561–1574, 1999
105. Lim H, Paria BC, Das SK, *et al:* Multiple female reproductive failures in cyclooxygenase 2-deficient mice. *Cell* 91:197–208, 1997

106. Mano H, Kimura C, Fujisawa Y, *et al:* Cloning and function of rabbit peroxisome proliferator-activated receptor delta/beta in mature osteoclasts. *J Biol Chem* 275:8126–8132, 2000
107. Tontonoz P, Hu E, Spiegelman BM: Regulation of adipocyte gene expression and differentiation by peroisome proliferator activated receptor γ. *Curr Opin Genet Dev* 5:571–576, 1995
108. Brun RP, Tontonoz P, Forman BM, *et al:* Differential activation of adipogenesis by multiple PPAR isoforms. *Gen & Dev* 10:974–984, 1996
109. Gurnell M, Wentworth JM, Agostini M, *et al:* A dominant-negative peroxisome proliferator-activated receptor gamma (PPARgamma) mutant is a constitutive repressor and inhibits PPARgamma-mediated adipogenesis. *J Biol Chem* 275:5754–5759, 2000
110. Barak Y, Nelson MC, Ong ES, *et al:* PPARγ is required for placental, cardiac, and adipose tissue development. *Mol Cell* 4: 585–595, 1999
111. Brown MS, Goldstein JL: The SREBP pathway: Regulation of cholesterol metabolism by proteolysis of a membrane-bound transcription factor. *Cell* 89:331–340, 1997
112. Kim JB, Wright HM, Wright M, Spiegelman BM: ADD1/SREBP1 activates PPARγ through the production of endogenous ligand. *Proc Natl Acad Sci USA* 95:4333–4337, 1998
113. Kumar S, Boulton AJ, Beck-Nielsen H, *et al:* Troglitazone, an insulin action enhancer, improves metabolic control in NIDDM patients. Troglitazone Study Group. *Diabetologia* 39:701–709, 1996
114. Prigeon RL, Kahn SE, Porte D Jr: Effect of troglitazone on B cell function, insulin sensitivity, and glycemic control in subjects with type 2 diabetes mellitus. *J Clin Endocrinol Metab* 83:819–823, 1998
115. Fonseca VA, Valiquett TR, Huang SM, *et al:* Troglitazone monotherapy improves glycemic control in patients with type 2 diabetes mellitus: A randomized, controlled study. The Troglitazone Study Group. *J Clin Endocrinol Metab* 83:3169–3176, 1998
116. Spencer CM, Goa KL, Gillis JC: Tacrolimus. An update of its pharmacology and clinical efficacy in the management of organ transplantation. *Drugs* 54:925–975, 1997
117. Ghazzi MN, Perez JE, Antonucci TK, *et al:* Cardiac and glycemic benefits of troglitazone treatment in NIDDM. *Diabetes* 46:433–439, 1997
118. Schwartz MW, Kahn SE: Insulin resistance and obesity. *Nature* 402:860–861, 1999
119. Mukherjee R, Davies PJ, Crombier DL, *et al:* Sensitization of diabetic and obese mice to insulin by retinoid X receptor agonists. *Nature* 386:407–414, 1997
120. Kliewer SA, Umesono K, Noonan DJ, *et al:* Convergence of 9-*cis* retinoic acid and peroxisome proliferator signalling pathways throught heterodimer formation of their receptors. *Nature* 358: 771–774, 1992
121. Chao L, Marcus-Samuels B, Mason MM, *et al:* Adipose tissue is required for the antidiabetic, but not for the hypolipidemic, effect of thiazolidinediones. *J Clin Invest* 106:1221–1228, 2000
122. Burant CF, Sreenan S, Hirano K, *et al:* Troglitazone action is independent of adipose tissue. *J Clin Invest* 100:2900–2908, 1997
123. Park KS, Ciaraldi TP, Abrams-Carter L, *et al:* PPAR-γ gene expression is elevated in skeletal muscle of obese and type II diabetic subjects. *Diabetes* 46:1230–1234, 1997
124. Zierath JR, Ryder JW, Doebber T, *et al:* Role of skeletal muscle in thiazolidinedione insulin sensitizer (PPARγ agonist) action. *Endocrinology* 139:5034–5041, 1998
125. Park KS, Ciaraldi TP, Lindgren K, *et al:* Troglitazone effects on gene expression in human skeletal muscle of type II diabetes involve up-regulation of peroxisome proliferator-activated receptor-gamma. *J Clin Endocrinol Metab* 83:2830–2835, 1998
126. Park KS, Ciaraldi TP, Abrams-Carter L, *et al:* PPAR-gamma gene expression is elevated in skeletal muscle of obese and type II diabetic subjects. *Diabetes* 46:1230–1234, 1997
127. Miles PDG, Romeo OM, Higo K, *et al:* TNF-α-induced insulin resistance in vivo and its prevention by troglitazone. *Diabetes* 46:1678–1683, 1997
128. Peraldi P, Xu M, Spiegelman BM: Thiazolidinediones block tumor necrosis factor-α-induced inhibition of insulin signaling. *J Clin Invest* 100:1863–1869, 1997
129. Steppan CM, Brown EJ, Wright CM, *et al:* A family of tissue-specific resistin-like molecules. *Proc Natl Acad Sci USA* 98:502–506, 2001
130. Steppan CM, Bailey ST, Bhat S, *et al:* The hormone resistin links obesity to diabetes. *Nature* 409:307–312, 2001
131. Hallakou S, Doaré L, Foufelle F, *et al:* Pioglitazone induces in vivo adipocyte differentiation in the Zucker fa/fa rat. *Diabetes* 46:1393–1399, 1997
132. Okuno A, Tamemoto H, Tobe K, *et al:* Troglitazone increases the number of small adipocytes without the change of white adipose tissue mass in obese Zucker rats. *J Clin Invest* 101:1354–1361, 1998
133. Barroso I, Gurnell M, Crowley VE, *et al:* Dominant negative mutations in human PPARgamma associated with severe insulin resistance, diabetes mellitus and hypertension [see comments]. *Nature* 402:880–883, 1999
134. Kubota N, Terauchi Y, Miki H, *et al:* PPAR gamma mediates high-fat diet-induced adipocyte hypertrophy and insulin resistance. *Mol Cell* 4:597–609, 1999
135. Miles PD, Barak Y, He W, *et al:* Improved insulin-sensitivity in mice heterozygous for PPAR-gamma deficiency. *J Clin Invest* 105:287–292, 2000
136. Alegret M, Cerqueda E, Ferrando R, *et al:* Selective modification of rat hepatic microsomal fatty acid chain elongation and desaturation by fibrates: Relationship with peroxisome proliferation. *Br J Pharmacol* 114:1351–1358, 1995
137. Vazquez M, Merlos M, Adzet T, Laguna JC: Decreased susceptibility to copper-induced oxidation of rat-lipoproteins after fibrate treatment: Influence of fatty acid composition. *Br J Pharmacol* 117:1155–1162, 1996
138. Gonzalez FJ: Recent update on the PPARα-null mouse. *Biochimie* 79:139–144, 1997
139. Costet P, Legendre C, More J, *et al:* Peroxisome proliferator-activated receptor alpha-isoform deficiency leads to progressive dyslipidemia with sexually dimorphic obesity and steatosis. *J Biol Chem* 273:29577–29585, 1998
140. Nicholls DG, Locke RM: Thermogenic mechanisms in brown fat. *Physiol Rev* 64:1–64, 1984
141. Klingenberg M, Huang SG: Structure and function of the uncoupling protein from brown adipose tissue. *Biochim Biophys Acta* 1415:271–296, 1999
142. Enerback S, Jacobsson A, Simpson EM, *et al:* Mice lacking mitochondrial uncoupling protein are cold-sensitive but not obese. *Nature* 387:90–94, 1997
143. Fleury C, Neverova M, Collins S, *et al:* Uncoupling protein-2: A novel gene linked to obesity and hyperinsulinemia. *Nat Genet* 15:269–272, 1997
144. Gimeno RE, Dembski M, Weng X, *et al:* Cloning and characterization of an uncoupling protein homolog: A potential molecular mediator of human thermogenesis. *Diabetes* 46:900–906, 1997
145. Boss O, Samec S, Dulloo A, *et al:* Tissue-dependent upregulation of rat uncoupling protein-2 expression in response to fasting or cold. *FEBS Lett* 412:111–114, 1997
146. Vidal-Puig A, Solanes G, Grujic D *et al:* UCP3: An uncoupling protein homologue expressed preferentially and abundantly in skeletal muscle and brown adipose tissue. *Biochem Biophys Res Commun* 235:79–82, 1997
147. Gong DW, He Y, Karas M, Reitman M: Uncoupling protein-3 is a mediator of thermogenesis regulated by thyroid hormone, beta3-adrenergic agonists, and leptin. *J Biol Chem* 272:24129–24132, 1997
148. Cassard-Doulcier AM, Gelly C, Bouillaud F, Ricquier D: A 211-bp enhancer of the rat uncoupling protein-1 (UCP-1) gene controls specific and regulated expression in brown adipose tissue. *Biochem J* 333:243–246, 1998
149. Boss O, Samec S, Dulloo A, *et al:* Tissue-dependent upregulation of rat uncoupling protein-2 expression in response to fasting or cold. *FEBS Lett* 412:111–114, 1997
150. Weigle DS, Selfridge LE, Schwartz MW, *et al:* Elevated free fatty acids induce uncoupling protein 3 expression in muscle: A

potential explanation for the effect of fasting. *Diabetes* 47:298–302, 1998
151. Lowell BBVSS, Hamann A, *et al:* Development of obesity in transgenic mice after genetic ablation of brown adipose tissue. *Nature* 366:740–742, 1993
152. Vamecq J, Latruffe N: Medical significance of peroxisome proliferator-activated receptors. *Lancet* 354:141–148, 1999
153. Saladin R, De Vos P, Guerre-Millo M, *et al:* Transient increase in obese gene expression after food intake or insulin administration. *Nature* 377:527–529, 1995
154. Hollenberg AN, Susulic VS, Madura JP, *et al:* Functional antagonism between CCAAT/Enhancer binding protein-alpha and peroxisome proliferator-activated receptor-gamma on the leptin promoter. *J Biol Chem* 272:5283–5290, 1997
155. Aubert J, Champigny O, Saint-Marc P, *et al:* Up-regulation of UCP-2 gene expression by PPAR agonists in preadipose and adipose cells. *Biochem Biophys Res Commun* 238:606–611, 1997
156. Gans ROB, Donker AJM: Insulin and blood pressure regulation. *J Intern Med* 229(Suppl 2):49–64, 1991
157. Ferrari P, Weidmann P: Insulin, insulin sensitivity and hypertension. *J Hypertens* 8:491–500, 1990
158. Inzucchi SE, Maggs DG, Spollett GR, *et al:* Efficacy and metabolic effects of metformin and troglitazone in type II diabetes mellitus. *N Engl J Med* 338:867–872, 1998
159. Ogihara T, Rakugi H, Ikegami H, *et al:* Enhancement of insulin sensitivity by troglitazone lowers blood pressure in diabetic hypertensives. *Am J Hypertens* 8:316–320, 1995
160. Saku K, Zhang B, Ohta T, Arakawa K: Troglitazone lowers blood pressure and enhances insulin sensitivity in Watanabe heritable hyperlipidemic rabbits. *Am J Hypertens* 10:1027–1033, 1997
161. Yoshimoto T, Naruse M, Nishikawa M, *et al:* Antihypertensive and vasculo- and renoprotective effects of pioglitazone in genetically obese diabetic rats. *Am J Physiol* 272:E989–E996, 1997
162. Uchida A, Nakata T, Hatta T, *et al:* Reduction of insulin resistance attenuates the development of hypertension in sucrose-fed SHR. *Life Sci* 61:455–464, 1997
163. Grinsell JW, Lardinois CK, Swislocki A, *et al:* Pioglitazone attenuates basal and postprandial insulin concentrations and blood pressure in the spontaneously hypertensive rat. *Am J Hypertens* 13:370–375, 2000
164. Yoshimoto T, Naruse M, Shizume H, *et al:* Vasculo-protective effects of insulin sensitizing agent pioglitazone in neointimal thickening and hypertensive vascular hypertrophy. *Atherosclerosis* 145:333–340, 1999
165. Walker AB, Chattington PD, Buckingham RE, Williams G: The thiazolidinedione rosiglitazone (BRL-49653) lowers blood pressure and protects against impairment of endothelial function in Zucker fatty rats. *Diabetes* 48:1448–1453, 1999
166. Sung BH, Izzo JL Jr, Dandona P, Wilson MF: Vasodilatory effects of troglitazone improve blood pressure at rest and during mental stress in type 2 diabetes mellitus. *Hypertension* 34:83–88, 1999
167. Iijima K, Yoshizumi M, Ako J, *et al:* Expression of peroxisome proliferator-activated receptor γ (PPARγ) in rat aortic smooth muscle cells. *Biochem Biophys Res Commun* 247:353–356, 1998
168. Law RE, Goetze S, Xi X-P, *et al:* Expression and function of PPARγ in rat and human vascular smooth muscle cells. *Circulation* 101:1311–1318, 2000
169. Marx N, Schönbeck U, Lazar MA, *et al:* Peroxisomal proliferator-activated receptor gamma activators inhibit gene expression and migration in human vascular smooth muscle cells. *Circ Res* 83:1097–1103, 1998
170. Marx N, Bourcier T, Sukhova GK, *et al:* PPARγ activation in human endothelial cells increases plasminogen activator inhibitor type-1 expression: PPARγ as a potential mediator in vascular disease. *Arterioscler Thromb Vasc Biol* 19:546–551, 1999
171. Satoh H, Tsukamoto Y, Hashimoto N, *et al:* Thiazolidinediones suppress endothelial-1 secretion from bovine vascular endothelial cells: A new possible role of PPARγ on vascular endothelial function. *Biochem Biophys Res Commun* 254:757–763, 1999
172. Zhang HY, Reddy SR, Kotchen TA: Antihypertensive effect of pioglitazone is not invariably associated with increased insulin sensitivity. *Hypertension* 24:106–110, 1994
173. Fujiwara T, Ohsawa T, Takahashi S, *et al:* Troglitazone, a new antidiabetic agenet possessing radical scavenging ability, improved decreased skin blood flow in diabetic rats. *Life Sci* 63:2039–2047, 1998
174. Buchanan TA, Meehan WP, Jeng YY, *et al:* Blood pressure lowering by pioglitazone. Evidence for a direct vascular effect. *J Clin Invest* 96:354–360, 1995
175. Walker AB, Naderali EK, Chattington PD, *et al:* Differential vasoactive effects of the insulin sensitizers rosiglitazone (BRL 49653) and troglitazone on human small arteries in vitro. *Diabetes* 47:810–814, 1998
176. Kurtz TW, Gardner DG: Transcription-modulating drugs: A new frontier in the treatment of essential hypertension. *Hypertension* 32:380–386, 1998
177. Willson TM, Brown PJ, Sternbach DD, Henke BR: The PPARs: From orphan receptors to drug discovery. *J Med Chem* 43:527–550, 2000
178. Law RE, Meehan WP, Xi X-P, *et al:* Troglitazone inhibits vascular smooth muscle cell growth and intimal hyperplasia. *J Clin Invest* 98:1897–1905, 1996
179. Akanuma Y, Kosaka K, Kuzuya T, *et al:* Clinical evaluation of a new oral hypoglycemic agent CS-045 in patients with non-insulin-dependent diabetes mellitus poorly controlled by sulphonylureas: A dose-finding by dose increasing method. *J Clin Ther Med* 9 (Suppl 3):39–60, 1993
180. Gorson DM: Significant weight gain with Rezulin therapy. *Arch Intern Med* 159:99, 1999
181. Ouali F, Djouadi F, Merlet-Bénichou C, Bastin J: Dietary lipids regulate β-oxidation enzyme gene expression in the developing rat kidney. *Am J Physiol* 275:F777–F784, 1998
182. Capdevila JH, Falck JR, Harris RC, Cytochrome P: 450 and arachidonic acid bioactivation. Molecular and functional properties of the arachidonate monooxygenase. *J Lipid Res* 41:163–181, 2000
183. Roman RJ, Ma YH, Frohlich B, Markham B: Clofibrate prevents the development of hypertension in Dahl salt-sensitive rats. *Hypertension* 21:985–988, 1993
184. Portilla D, Dai G, Peters JM, *et al:* Etomoxir-induced PPARalpha-modulated enzymes protect during acute renal failure. *Am J Physiol (Renal Physiol)* 278:F667–F675, 2000
185. Reilly CM, Oates JC, Cook JA, *et al:* Inhibition of mesangial cell nitric oxide in MRL/lpr mice by prostaglandin J2 and proliferator activation receptor-γ agonists. *J Immunol* 164:1498–1504, 2000
186. Makino H, Kashihara N, Sugiyama H, *et al:* Phenotypic modulation of the mesangium reflected by contractile protein in diabetes. *Diabetes* 45:488–495, 1995
187. Johnson RJ, Iida H, Alpers CE, *et al:* Expression of smooth muscle cell phenotype by rat mesangial cells in immune complex nephritis. Alpha-smooth muscle actin is a marker of mesangial cell proliferation. *J Clin Invest* 87:847–858, 1991
188. Makino H, Kashihara N, Sugiyama H, *et al:* Phenotypic changes of the mesangium in diabetic nephropathy. *J Diabetes Compl* 9: 282–284, 1995
189. Fujii M, Takemura R, Yamaguchi M, *et al:* Troglitazone (CS-045) ameliorates albuminuria in streptozotocin-induced diabetic rats. *Metabolism* 46:981–983, 1997
190. Imano E, Kanda T, Nakatani Y, *et al:* Effect of troglitazone on microalbuminuria in patients with incipient diabetic nephropathy. *Diabetes Care* 21:2135–2139, 1998
191. McCarthy KJ, Routh RE, Shaw W, *et al:* Troglitazone halts diabetic glomerulosclerosis by blockade of mesangial expansion. *Kidney Int* 58:2341–2350, 2000
192. Buckingham RE, Al-Barazanji KA, Toseland CDN, *et al:* Peroxisome proliferator-activated receptor-γ agonist, rosiglitazone, protects against nephropathy and pancreatic islet abnormalities in Zucker fatty rats. *Diabetes* 47:1326–1334, 1998
193. Ricote M, Li AC, Willson TM, *et al:* The peroxisome proliferator-activated receptor-gamma is a negative regulator of macrophage activation. *Nature* 391:79–82, 1998
194. Crook ED: The role of hypertension, obesity, and diabetes in causing renal vascular disease. *Am J Med Sci* 317:183–188, 1999
195. van Jaarsveld BC, Krijnen P, Pieterman H, *et al:* The effect of balloon angioplasty on hypertension in atherosclerotic renal-

artery stenosis. Dutch Renal Artery Stenosis Intervention Cooperative Study Group. *N Engl J Med* 342:1007–1014, 2000
196. Rubins HB, Robins SJ, Collins D, *et al:* Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. *N Engl J Med* 341:410–418, 1999
197. Inoue I, Shino K, Noji S, *et al:* Expression of peroxisome proliferator-activated receptor α (PPARα) in primary cultures of human vascular endothelial cells. *Biochem Biophys Res Commun* 246: 370–374, 1998
198. Marx N, Sukhova GK, Collins T, *et al:* PPARalpha activators inhibit cytokine-induced vascular cell adhesion molecule-1 expression in human endothelial cells. *Circulation* 99:3125–3131, 1999
199. Staels B, Koenig W, Habib A, *et al:* Activation of human aortic smooth muscle cells is inhibited by PPARα but not PPARα activators. *Nature* 393:790–793, 1998
200. Devchand P, Keller H, Peters J, *et al:* The PPARα-leukotriene B_4 pathway to inflammmation control. *Nature* 384:39–43, 1996
201. Thurberg BL, Collins T: The nuclear factor-kappa B/inhibitor of kappa B autoregulatory system and atherosclerosis. *Curr Opin Lipidol* 9:387–396, 1998
202. Poynter ME, Daynes RA: Peroxisome proliferator-activated receptor alpha activation modulates cellular redox status, represses nuclear factor-kappaB signaling, and reduces inflammatory cytokine production in aging. *J Biol Chem* 273:32833–32841, 1998
203. Delerive P, De Bosscher K, Besnard S, *et al:* Peroxisome proliferator-activated receptor alpha negatively regulates the vascular inflammatory gene response by negative cross-talk with transcription factors NF-kappaB and AP-1. *J Biol Chem* 274:32048–32054, 1999
204. Kockx M, Gervois PP, Poulain P, *et al:* Fibrates suppress fibrinogen gene expression in rodents via activation of the peroxisome proliferator-activated receptor-alpha. *Blood* 93:2991–2998, 1999
205. Sawamura T, Kume N, Aoyama T, *et al:* An endothelial receptor for oxidized low-density lipoprotein. *Nature* 386:73–77, 1997
206. Ricote M, Huang J, Fajas L, *et al:* Expression of the peroxisome proliferator-activated receptor gamma (PPARgamma) in human atherosclerosis and regulation in macrophages by colony stimulating factors and oxidized low density lipoprotein. *Proc Natl Acad Sci USA* 95:7614–7619, 1998
207. Ricote M, Li AC, Willson TM, *et al:* The peroxisome proliferator-activated receptor-γ is a negative regulator of macrophage activation. *Nature* 391:79–82, 1998
208. Chinetti G, Griglio S, Antonucci M, *et al:* Activation of proliferator-activator receptors α and γ induces apoptosis of human monocyte-derived macrophages. *J Biol Chem* 273:25573–25580, 1998
209. Murakami K, Tobe K, Ide T, *et al:* A novel insulin sensitizer acts as a coligand for peroxisome proliferator-activated receptor-alpha (PPAR-alpha) and PPAR-gamma: Effect of PPAR-alpha activation on abnormal lipid metabolism in liver of Zucker fatty rats. *Diabetes* 47:1841–1847, 1998
210. Shiomi M, Ito T, Tsukada T, *et al:* Combination treatment with troglitazone, an insulin action enhancer, and pravastatin, an inhibitor of HMG-CoA reductase, shows a synergistic effect on atherosclerosis of WHHL rabbits. *Atherosclerosis* 142:345–353, 1999
211. Li AC, Brown KK, Silvestre MJ, *et al:* Peroxisome proliferator-activated receptor gamma ligands inhibit development of atherosclerosis in LDL receptor-deficient mice. *J Clin Invest* 106:523–531, 2000
212. Chinetti G, Lestavel S, Bocher V, *et al:* PPAR-alpha and PPAR-gamma activators induce cholesterol removal from human macrophage foam cells through stimulation of the ABCA1 pathway. *Nat Med* 7:53–58, 2001
213. Moore KJ, Rosen ED, Fitzgerald ML, *et al:* The role of PPAR-gamma in macrophage differentiation and cholesterol uptake. *Nat Med* 7:41–47, 2001
214. Lazar MA: Progress in cardiovascular biology: PPAR for the course. *Nat Med* 7:23–24, 2001
215. Altiok S, Xu M, Spiegelman BM: PPARγ induces cell cycle withdrawal: Inhibition of E2F/DP DNA-binding activity via down-regulation of PP. 2A. *Genes & Dev* 11:1987–1998, 1997
216. Tontonoz P, Singer S, Forman BM, *et al:* Terminal differentiation of human liposarcoma cells induced by ligands for peroxisome proliferator-activated receptor γ and the retinoid X receptor. *Proc Natl Acad Sci USA* 94:237–241, 1997
217. Demetri GD, Fletcher CD, Mueller E, *et al:* Induction of solid tumor differentiation by the peroxisome proliferator-activated receptor-gamma ligand troglitazone in patients with liposarcoma. *Proc Natl Acad Sci USA* 96:3951–3956, 1999
218. Elstner E, Muller C, Koshizuka K, *et al:* Ligands for peroxisome proliferator-activated receptor γ and retinoic acid receptor inhibit growth and induce apoptosis of human breast cancer in vitro and in BNX mice. *Proc Natl Acad Sci USA* 95:8806–8811, 1998
219. Suh N, Wang Y, Williams CR, *et al:* A new ligand for the peroxisome proliferator-activated receptor-gamma (PPAR-gamma), GW7845, inhibits rat mammary carcinogenesis. *Cancer Res* 59: 5671–5673, 1999
220. Takahashi N, Okumura T, Motomura W, *et al:* Activation of PPARgamma inhibits cell growth and induces apoptosis in human gastric cancer cells. *FEBS Lett* 455:135–139, 1999
221. Brockman JA, Gupta RA, DuBois RN: Activation of PPARγ leads to inhibition of anchorage-independent growth of human colorectal cancer cells. *Gastroenterology* 115:1049–1055, 1998
222. Sarraf P, Muller E, Jones D, *et al:* Differentiation and reversal of malignant change in colon cancer through PPARγ. *Nature Med* 4:1046–1052, 1998
223. Lefebvre A-M, Chen I, Desreumaux P, *et al:* Acivation of the peroxisome proliferator-activated receptor γ promotes the development of colon tumors in C57BL/6J-$APC^{MIN/+}$ mice. *Nature Med* 4:1053–1057, 1998
224. Saez E, Tontonoz P, Nelson MC, *et al:* Activators of the nuclear receptor PPARgamma enhance colon polyp formation. *Nat Med* 4:1058–1061, 1998
225. Gupta RA, Tan J, Krause WF, *et al:* Prostacyclin-mediated activation of peroxisome proliferator-activated receptor delta in colorectal cancer. *Proc Natl Acad Sci USA* 97:13275–13280, 2000
226. Janne PA, Mayer RJ: Chemoprevention of colorectal cancer. *N Engl J Med* 342:1960–1968, 2000
227. Berger J, Leibowitz MD, Doebber TW, *et al:* Novel peroxisome proliferator-activated receptor (PPAR) gamma and PPARdelta ligands produce distinct biological effects. *J Biol Chem* 274:6718–6725, 1999
228. Yu K, Bayona W, Kallen CB, *et al:* Differential activation of peroxisome proliferator-activated receptors by eicosanoids. *J Biol Chem* 270:23975–23983, 1995
229. Lehmann JM, Lenhard JM, Oliver BB, *et al:* Peroxisome proliferator-activated receptors α and γ are activated by indomethacin and other non-steroidal anti-inflammatory drugs. *J Biol Chem* 272:3406–3410, 1997
230. Henke BR, Blanchard SG, Brackeen MF, *et al:* N-(2-Benzoylphenyl)-l-tyrosine PPARgamma agonists. 1. Discovery of a novel series of potent antihyperglycemic and antihyperlipidemic agents. *J Med Chem* 41:5020–5036, 1998
231. Brown PJ, Smith-Oliver TA, Charifson PS, *et al:* Identification of peroxisome proliferator-activated receptor ligands from a biased chemical library. *Chem Biol* 4:909–918, 1997