Abstract: Pemafibrate is the first clinically-available selective peroxisome proliferator-activated receptor α modulator (SPPARMα) that has been shown to effectively improve hypertriglyceridemia and low high-density lipoprotein cholesterol (HDL-C) levels. Global gene expression analysis reveals that the activation of PPARα by pemafibrate induces fatty acid (FA) uptake, binding, and mitochondrial or peroxisomal oxidation as well as ketogenesis in mouse liver. Pemafibrate most profoundly induces HMGCS2 and PDK4, which regulate the rate-limiting step of ketogenesis and glucose oxidation, respectively, compared to other fatty acid metabolic genes in human hepatocytes. This suggests that PPARα plays a crucial role in nutrient flux in the human liver. Additionally, pemafibrate induces clinically favorable genes, such as ABCA1, FGF21, and VLDLR. Furthermore, pemafibrate shows anti-inflammatory effects in vascular endothelial cells. Pemafibrate is predicted to exhibit beneficial effects in patients with atherogenic dyslipidemia and diabetic microvascular complications.

Keywords: pemafibrate; SPPARMα; ketogenesis; fatty acid β-oxidation; ASCVD; EndMT

1. Introduction

Although low density lipoprotein cholesterol (LDL-C)-lowering therapy by statins has been proven to reduce the events of atherosclerotic cardiovascular disease (ASCVD) [1,2], there still remains a high residual cardiovascular risk from elevated triglycerides (TG) and low HDL cholesterol (HDL-C) levels [3–6]. Synthetic PPARα ligands and fibrates have been shown to effectively reduce plasma TG levels by 25–50% and increase HDL-C levels by 5–20% [7–10]. Therefore, theoretically, fibrates are suitable drugs to use as an add-on statin treatment to improve hypertriglyceridemia and atherogenic dyslipidemia. However, there is a lack of adequate evidence to support statin-fibrate combination
therapy for the prevention of definitive mortality rate. In addition, the use of fibrates in patients with hepatic and renal insufficiency has been limited due to adverse drug reactions (ADRs) such as plasma transaminase and creatinine elevation, as well as reduced estimated glomerular filtration rates (eGFRs) [11–14]. Under these circumstances, pemafibrate was developed as a selective peroxisome proliferator-activated receptor α modulator (SPPARMα) that enhances the beneficial effects and reduces the adverse effects of fibrates. To date, 50 papers have been published on this subject and few papers reported the effect of pemafibrate on target gene expression. Through the limited reports, we describe the pemafibrate-regulated genes and potential clinical implications.

2. Pemafibrate as a Novel SPPARMα

Pemafibrate (K-877, Parmodia®) was developed as a novel SPPARMα that enhances PPARα activity and selectivity by introducing a 2-aminobenzoxazolic ring and phenoxyalkyl chain into fibric acid (Figure 1a) [15–17]. These side-chains confer a Y-shape structure and fill the entire ligand-binding pocket of PPARα [18] (Figure 1b), thereby allosterically changing the PPARα conformation to enhance complex formation with coactivators such as peroxisome proliferative activated receptor gamma coactivator 1α (PGC1α) and exhibiting full agonistic activity. Actually, pemafibrate has greater PPARα activation potency than fenofibrate, along with a lower EC50 value (1.5 nM) and a higher degree of subtype selectivity (>2000-fold) (Figure 1c) [19]. In preclinical studies, pemafibrate exhibited a greater TG-lowering effect than fenofibrate in normolipidemic and hypertriglyceridemic rodent models [15,20,21]. In addition, in human apoA-I transgenic mice, pemafibrate treatment resulted in a greater increase in levels of plasma h-apoAI, a major component of HDL, than occurred with fenofibrate treatment [15,22]. Furthermore, pemafibrate has been shown to reduce atherosclerotic lesion areas in Ldlr-null mice [17] and western diet-fed APOE2 KI mice [22]. Although fibrates have been specifically shown to induce peroxisome proliferation and related hepatomegaly and hepatocellular carcinoma in rodents [23–25], pemafibrate causes less weight gain of the liver than fenofibrate [15]. Under the fed condition, the liver accumulated the highest concentration of pemafibrate and reached 105 nM after four weeks of treatment with a 0.0006% (w/w) pemafibrate-containing diet, which is an equivalent or higher dose than needed to demonstrate pharmacological action [22,26,27]. As indicated in Figure 1c, pemafibrate was unable to activate PPARγ or PPARδ at this concentration. In addition, the therapeutic dose of pemafibrate is 0.2–0.4 mg/day, which is equivalent to the dose of 0.004–0.008 mg/kg/day (based on a 50 kg human); therefore, it is unlikely that pemafibrate shows the other PPARs subtype-mediated pharmacological effect in clinical use.

Pemafibrate was approved in Japan 2017 for the treatment of dyslipidemia [28–38]. A phase II study showed that 0.05–0.4 mg/day pemafibrate significantly reduced plasma TG levels (−30.9% to −42.7%) and increased HDL-C levels (11.9% to 21.0%) [29]. Although the difference was not statistically significant, the improvement of these parameters was more significant with pemafibrate than fenofibrate. The incidence of adverse events (AEs) in the pemafibrate treatment group was comparable to those in the placebo and 100 mg/day fenofibrate groups. However, the incidence of ADRs in the pemafibrate treatment group was lower than those in the placebo and 100 mg/day fenofibrate groups [29,31]. In addition, when compared to placebo and fenofibrate treatment, pemafibrate significantly increased the level of plasma FGF21, which is an endocrine factor regulating glucose uptake, metabolism, and energy expenditure [39]. Therefore, pemafibrate could replace fibrates as the first clinically-available SPPARMα to improve atherogenic dyslipidemia and prevent macro- and microvascular risks.
Pemafibrate (K_d, 2 μM) has binding affinity higher than fenofibrate [15]. Pemafibrate was developed as a novel SPPARMα that enhances PPARα selectivity at molar concentration. In preclinical models [15,22], pemafibrate treatment significantly induced greater PPARα activation potency than fenofibrate. Furthermore, pemafibrate has been shown to reduce atherosclerotic lesion areas, with a greater increase in eGFRs [11–14]. Under these circumstances, pemafibrate was developed as a novel SPPARMα that enhances PPARα activity. Actually, pemafibrate has occurred in clinical use.

Figure 1. Structure and PPARα selectivity of pemafibrate. (a) Structure of pemafibrate and fenofibrate. (b) Binding mode of the ligand with human PPARα. Pemafibrate in magenta and fenofibrate in blue. The binding pocket is divided into three pharmacophore regions according to the interactions with the ligands. While fenofibrate acid occupies the magenta cavity, 2-aminobenzoxazole ring and phenoxyalkyl group of Y-shaped pemafibrate occupies the green cavity and yellow cavity, respectively. Therefore, pemafibrate fills all the areas of the ligand-binding pocket. Reprinted from Yamamoto Y, et al. with permission from Elsevier [18]. (c) Transactivation profile of pemafibrate. Transactivation curves for human PPARα, PPARβ, and PPARγ are shown. Reproduced Raza-Iqbal S., et al. with permission from authors [19].

3. Pemafibrate Regulates the Availability of FA and Glucose Oxidation

Species differences have been well documented for PPARα-regulated genes, such as those involved in peroxisome biogenesis and peroxisomal FA β-oxidation [40–42]. In addition, whether PPARα mediates gene expression regulation by pemafibrate and whether human exposure to pemafibrate regulates the same target genes as those found in mice are still a matter of debate. To predict the mode of action and untoward effects of pemafibrate in humans, we carried out microarray analyses and compared the data of pemafibrate-treated primary human hepatocytes and mouse livers [19].

Global gene expression profiling clearly demonstrated that pemafibrate regulates the entire FA catabolism in mouse liver. Pemafibrate significantly induces Vldlr, TG hydrolysis (Lpl), FA cellular uptake (Cd36/Fat, Scl27a1, and Scl27a4), FA binding (Fabp2 and Fabp4), FA activation (Acs1, Acs3, Acs5, and Aco1), FA ω-oxidation (Cyp4a14, Cyp4a31, and Aldh3a2), and peroxisomal (Abcd2, Abcbl3, Ech1, Decr2, Acox1, Ehhadh, Hsd17b4, Acaa1, Crat, Aco3, Aco4, and Aco8) and mitochondrial (Cpt1, Cpt2, Slc25a20, Acadbl, Acadn, Acads, Acadm, Acad11, Ehhadh, Hadha, Hadhb, and Decr1) FA β-oxidation, and ketogenesis (Acac1, Hmgcs2, and Hmgcl). In addition, pemafibrate induces peroxisome biogenesis genes (Pex1, Pex3, Pex11a, Pex14, and Pex19). The upregulation of these genes was not observed in the pemafibrate-treated Ppara-null mouse liver [19]. In accordance with our results, Takei et al. also reported that the effect of pemafibrate was abolished in Ppara-null mice [21]. Thus, these observations indicate that PPARα is crucial for the regulation of FA catabolic genes in mouse liver following pemafibrate treatment.

Similarly, pemafibrate induced VLDLR, FABPI, and mitochondrial FA β-oxidation gene (ACSL1, ACSL5, CPT1A, CPT2, SLC25A20, ACADVL, HADHA, HADHB, and ACAA2) expression in human
hepatocytes, as seen in the livers of pemafibrate-treated mice. However, the induction of these genes was much lower in the human hepatocytes (Figure 2). Additionally, pemafibrate did not induce almost all FA ω-oxidation, peroxisomal FA β-oxidation, and peroxisome biogenesis genes expressions. The first step of FA ω-oxidation is ω-hydroxylation, which is catalyzed by the CYP4A family. Generated products are further metabolized to dicarboxylic acid by cytosolic aldehyde dehydrogenase, which is encoded by ALDH3A2, and they are efficiently metabolized by peroxisomal FA β-oxidation [43,44]. Numerous reports clearly indicated that the CYP4A family of enzymes are regulated by PPARα in rodent livers and are shown to parallel the induction of peroxisomal fatty acid β-oxidation enzymes and peroxisome proliferation [45]. In contrast, respect to the induction of CYP4A subtype is controversial in humans. Some studies showed that fibrates induce CYP4A11 mRNA expression in primary human hepatocytes and PPARα overexpressed HepG2 cells [46,47]. However, 100 µM of fenofibric acid, a concentration which is equal with our previous study, has been reported to fail induction of CYP4A11 expression in HepG2 cells [41]. Although it is difficult to declare the possibility to induce FA ω-oxidation enzyme in humans at present, peroxisome proliferation and related liver toxicities would not occur following a clinical dose of pemafibrate treatment.

Interestingly, pemafibrate most profoundly induced PDK4 and HMGCS2 gene expression in the primary human hepatocytes. Robust induction of PDK4 indicated inactivation of pyruvate dehydrogenase (PDH) and glucose oxidation [48–50]. In contrast, HMGCS2 expression has been reported to control not only ketogenesis but also mitochondrial fatty acid oxidation in HepG2 cells [51]. In addition, this report also showed that the expression of FGF21 (another target of pemafibrate) is upregulated by HMGCS2 activity or acetoacetate, which is the oxidized form of the ketone bodies. Furthermore, the ketone body, β-hydroxybutyrate, as an inhibitor of class I histone deacetylases (HDAC), and β-hydroxybutyrate-integrated histone H3 lysine 9 (H3K9bhb) are associated with the upregulation of genes involved in the starvation-responsive pathways, including the PPAR signaling pathway [52]. Thus, PPARα activation by pemafibrate cooperatively regulates nutrient availability through the induction of the key target genes, namely PDK4 and HMGCS2, which suppress the availability of carbohydrate oxidation and enhance acyl-CoA flux. This thereby facilitates mitochondrial long-chain fatty acid β-oxidation and ketogenesis in human hepatocytes. As a result, pemafibrate reduces the availability of acetyl-CoA for de novo lipogenesis and VLDL secretion.
CYP4A subtype is controversial in humans. Some studies showed that fibrates induce CYP4A11 mRNA expression in primary human hepatocytes and PPARα overexpressed HepG2 cells [46,47]. However, 100 μM of fenofibric acid, a concentration which is equal with our previous study, has been reported to fail induction of CYP4A11 expression in HepG2 cells [41]. Although it is difficult to declare the possibility to induce FA ω-oxidation enzyme in humans at present, peroxisome proliferation and related liver toxicities would not occur following a clinical dose of pemafibrate treatment.

**Figure 2.** Effect of pemafibrate on fatty acid metabolism-related gene expression. Heat map illustrating the genes regulated by pemafibrate treatment in mouse liver and primary hepatocytes. Gray boxes represent the absence call or no probe of the genes from microarray data.

4. Pharmacologically Favorable Target Genes of Pemafibrate as a SPPARMα

As shown in Figure 3, compared to fenofibrate, pemafibrate effectively induces the expression of pharmacologically favorable genes, such as very-low-density lipoprotein receptor (VLDLR), ATP binding cassette subfamily A member 1 (ABCA1), and fibroblast growth factor 21 (FGF21), by maximizing PPARα activation [19]. VLDLR is a member of the LDL-receptor family and is expressed...
in many tissues, including skeletal muscles, heart, and adipose tissues, whereas its expression is very low in the liver, under normal conditions [53,54]. VLDLR binds TG-rich lipoproteins such as chylomicron and VLDL and mediates the uptake of TG-rich lipoproteins by peripheral tissues through LPL-dependent lipolysis or receptor-mediated endocytosis. Importantly, Gao et al. [55] reported that fenofibrate induces liver Vldlr expression in a PPARα-dependent manner and that the TG-lowering effect of fenofibrate was abolished in Vldlr-null mice. In addition, although LPL is typically not expressed in the adult liver [56], pemafibrate PPARα dependently induced the expression of Lpl in the mouse liver. Thus, pemafibrate enhances TG-rich lipoprotein hydrolysis and uptake by coordinated regulation of Vldlr, Lpl, and Cd36 expression. ABCA1, a member of the superfamily of ATP-binding cassette (ABC) transporters, regulates the formation and function of HDL by facilitating the efflux of cholesterol and phosphatidylcholine to lipid-poor apoAI [57,58]. In fact, pemafibrate significantly induced ABCA1 and ABCG1 in human primary macrophages and enhanced HDL stimulated cholesterol efflux [22]. ABCA1 not only plays an important role in the initial step of reverse cholesterol transport (RCT) but is also involved in the anti-inflammatory action to suppress the expression of pro-inflammatory factors [59,60]. Therefore, pemafibrate-mediated increased ABCA1 expression could contribute to HDL-C elevation as well as anti-inflammatory and anti-atherosclerotic activities. FGF21 is a member of the fibroblast growth factor family [39,61], and its administration has been shown to reduce fasting plasma glucose, TG, insulin, and glucagon levels in diabetic rhesus monkeys [62]. FGF21 is a direct target of PPARα [63,64], and pemafibrate increases fasting and postprandial FGF21 levels along with improving dyslipidemia in humans [65]. Interestingly, CREBH [66] and HMGCS2 [51], the liver target genes of pemafibrate, have been reported to regulate FGF21 gene expression. Moreover, similar upregulation of Abca1, Crebh, and Fgf21 was observed in pemafibrate-treated Ldlr knockout mice liver [26]. Thus, pemafibrate enhances the combination of PPARα, CREBH, and HMGCS2 for the regulation of FGF21 expression.

Beyond regulation of nutrient oxidation, pemafibrate induces mannose-binding lectin 2 (MBL2) and glutamyl aminopeptidase (ENPEP) only in human hepatocytes (Figure 4). MBL is a soluble pattern recognition molecule involved in the humoral innate immune system [67,68]. In consecutive non-diabetic men, the serum MBL concentration was reduced in obese individuals accompanied by low insulin sensitivity and increased levels of inflammatory markers [69]. ENPEP encodes aminopeptidase A (APA), a member of the M1 endopeptidase family, involved in the catabolic pathway of the renin-angiotensin-aldosterone system that converts angiotensin II to angiotensin III [70–72]. In an animal study, the loss of function of ENPEP led to hypertension, and recombinant APA reduced the systolic blood pressure (SBP) [73]. Moreover, a rare nonsense variant in ENPEP is reported to be associated with increased SBP [74]. Therefore, these additional pemafibrate targets are likely to reduce cardiovascular disease risks.
such as diabetic nephropathy and 69-8-lotinamide adenine dinucleotide phosphate NAD(P)H has been reported to induce endothelial activation and dysfunction. Experimental evidence demonstrated that pemafibrate induces mannose-6-phosphate receptor expression in human hepatocytes. Data represent ± s.e.m. * P < 0.05; ** P < 0.01. Reproduced Raza-Iqbal S., et al. with permission from authors [19].

Dysfunction and injury of vascular endothelial cells play a critical role in the pathogenesis of ASCVD and CKD share common risk factors. In particular, increased levels of aldosterone system that converts angiotensin II to angiotensin III, regulated on activation, normal T cell expressed and secreted (RANTES) and chemokine (C-C motif) ligand 2 (CCL2) only in human hepatocytes (Figure 4). MBL is a soluble lectin 2 (MBL2) that is associated with increased SBP [74]. Therefore, these additional pemafibrate targets are likely to reduce cardiovascular disease risks.

Figure 3. Pemafibrate effectively induces VLDLR, FGF21, and ABCA1 mRNA expression in primary human hepatocytes. Data represent ± s.e.m. * P < 0.05; ** P < 0.01. Reproduced Raza-Iqbal S., et al. with permission from authors [19].

Figure 4. Pemafibrate effectively induces MBL2 and ENPEP mRNA expression in primary human hepatocytes. Data represent ± s.e.m. * P < 0.05; ** P < 0.01. Reproduced Raza-Iqbal S., et al. with permission from authors [19].
Dysfunction and injury of vascular endothelial cells play a critical role in the pathogenesis of ASCVD and chronic kidney disease (CKD) [75–77]. ASCVD and CKD share common risk factors including hypertension, hyperglycemia, obesity, and dyslipidemia and are associated with endothelial activation and dysfunction. In particular, high glucose-induced reactive oxygen species (ROS) have been shown to be involved in vascular dysfunction via a diacylglycerol (DAG)-protein kinase C (PKC)-dependent activation of nicotinamide adenine dinucleotide phosphate NAD(P)H oxidase pathway. Pemafibrate has been reported to reduce Fn1, Tgfb1, Nox4, and Ncf1 expression, and reduce DAG level, PKC activity, and oxidative stress marker (urinary 8-OHdG excretion) level in kidneys of diabetic db/db mice [78]. Pemafibrate also reduces serum starvation induced monocyte chemoattractant protein-1 (MCP-1), regulated on activation, normal T cell expressed and secreted (RANTES), interleukin 6 (IL6), and interferon gamma (IFNγ) expression and secretion in human coronary endothelial cells (HCECs) [79]. Besides its role in inflammation and ROS production, we found that pemafibrate suppresses high glucose-induced endothelial-mesenchymal transition (EndMT) in human umbilical vein endothelial cells (HUVECs). EndMT has emerged as an important process in the pathobiology of valve calcification, myocardial fibrosis, macrovascular complications, and microvascular complications such as diabetic nephropathy and retinopathy [80–82]. Experimental evidence demonstrated that TGFβ and Wnt/β-catenin signaling play a role in EndMT and may further contribute to tissue fibrosis [83–85]. Interestingly, pemafibrate reduces high glucose-induced TGFB2, COL1A2, CX3CL1, VCAM1 and DKK1 expression in HUVECs (Tanaka et al. personal communication). Likewise, fenofibrate has been reported to inhibit TGFβ-induced endothelin-1 (ET-1) expression in human microvascular endothelial cells [86]. ET-1 is a major vasoactive peptide that has been implicated in organ fibrosis through stimulation of EndMT [87,88]. In addition, fenofibrate has been reported to reduce progression of albuminuria and improve diabetic retinopathy [89–91]. Therefore, pemafibrate would be expected to prevent endothelial activation and dysfunction, thereby revealing protective effects against diabetic retinopathy, nephropathy, neuropathy, and ASCVD.

5. Possible Mechanism for the Gene Expression Regulation Induced by Pemafibrate?

Finally, we will discuss a potential mechanism for transcriptional regulation of hepatic target genes via PPARα activation by pemafibrate. As described in the text, PPARα activation by pemafibrate not only activates transcription of hepatic lipid metabolism genes, but also represses transcription of pro-inflammatory and EndMT-related genes. From the numerous observations, several models have been proposed for gene transcriptional regulation induced by PPARα [92–94]. In particular, PPARα functions as obligate heterodimers with retinoid X receptor (RXR). Ligand activated PPARα-RXR heterodimer mainly binds to DR1 elements termed PPAR response elements (PPREs) and recruits numerous coactivators, including CBP/p300 and SRC/p160 family, which contain histone acetyl transferase (HAT) activity, mediators, and the transcriptional preinitiation complex (PIC) [95–98]. This mechanism explains the main PPARα-dependent transactivation because DNA binding domain (DBD) mutant of PPARα (PPARαΔDBD), which maintains heterodimerization and coactivator interaction ability, lost PPRE binding and transactivation of PPRE-driven reporter genes [99]. On the other hand, transcriptional repression by PPARα is mainly mediated through protein-protein interactions. Ligand-activated PPARα has been reported to directly interact with pro-inflammatory transcription factor p65 and c-Jun, thereby suppressing their target genes such as IL6 and TNFα [100–102]. Interestingly, transcriptional repression ability is retained in PPARαΔDBD, indicating PPARα-dependent transrepression of the pro-inflammatory signaling pathway is PPRE-independent [99]. In addition, ligand-activated PPARα binds to coactivator of GRIP1/TIF2, thereby interfering with the C/EBPβ-induced fibrinogen-β gene transcription [103]. Furthermore, several nuclear receptors such as HNF4s, COUP-TFs, and RXR homodimer bind DR1 PPREs and may modulate PPARα-regulated gene expression [104–107]. Therefore, pemafibrate-induced gene expression appears as a combination of these multiple mechanisms.
6. Conclusions

PPARα regulates many hepatic metabolic genes along with lipid and glucose metabolism during prolonged starvation at the transcription levels and produces ketone bodies to provide metabolic fuel for the extrahepatic tissues. Despite accumulating evidence of the residual cardiovascular risks resulting from elevated TGs and lower HDL-C levels, low potent synthetic PPARα agonists (fibrates) have not shown enough evidence to reduce the definitive mortality rate when combined with statin treatment, despite an improvement in dyslipidemia. To overcome this issue, pemafibrate, a more potent and subtype-selective SPPARMα, was developed. By maximizing PPARα activation, pemafibrate effectively enhances TG hydrolysis, FA uptake, FA β-oxidation, and ketogenesis and thereby stimulates plasma TG hydrolysis and reduces VLDL secretion. In addition, pemafibrate enhances ABCA1-mediated HDL neogenesis and prevents the transfer of HDL-cholesteryl esters into TG-rich lipoproteins through the TG-lowering effect of pemafibrate. Through these mechanisms, pemafibrate effectively improves hypertriglyceridemia and low HDL-C levels. Importantly, PPARα activation by pemafibrate induces not only the generation of FAs via TG hydrolysis but also the generation of ketone bodies via FA β-oxidation and ketogenesis. In turn, the FAs could further activate PPARα, and the ketone bodies could promote the transcriptional activity of PPARα. Therefore, pemafibrate is expected to exert strong pharmacological effects and novel therapeutic action through a positive feedback loop and cooperative target gene regulation (Figure 5). In fact, pemafibrate induces clinically favorable key target genes (VLDLR, FGF21, ABCA1, MBL2, and ENPEP) and thereby has the therapeutic potential to address the residual cardiovascular risk. In addition, pemafibrate enhances ABCA1-mediated HDL neogenesis and prevents the transfer of HDL-cholesteryl esters into TG-rich lipoproteins through the TG-lowering effect of pemafibrate. Through these mechanisms, pemafibrate effectively improves hypertriglyceridemia and low HDL-C levels. Importantly, PPARα activation by pemafibrate induces not only the generation of FAs via TG hydrolysis but also the generation of ketone bodies via FA β-oxidation and ketogenesis. In turn, the FAs could further activate PPARα, and the ketone bodies could promote the transcriptional activity of PPARα. Therefore, pemafibrate is expected to exert strong pharmacological effects and novel therapeutic action through a positive feedback loop and cooperative target gene regulation (Figure 5). In fact, pemafibrate induces clinically favorable key target genes (VLDLR, FGF21, ABCA1, MBL2, and ENPEP) and thereby has the therapeutic potential to address the residual cardiovascular risk. In addition, pemafibrate would expect to show vascular endothelial cell protective effects and prevent diabetic microvascular complications. Currently, a major outcome study, PROMINENT (Pemafibrate to Reduce cardiovascular OutcoMes by reducing triglycerides IN diabetic patiENTs), is underway to investigate whether pemafibrate reduces cardiovascular events in type 2 diabetic patients with atherogenic dyslipidemia [108]. This study will evaluate the role of pemafibrate in the management of residual cardiovascular risk as an add-on therapy to statins.

![Figure 5](image_url)

**Figure 5.** Overviewing pemafibrate regulated fatty acid metabolism genes in human hepatocytes. Red font and arrows indicate the upregulated genes and pathways in the expression microarray of pemafibrate-treated human hepatocytes, respectively, which are based on our microarray data and the published literature.
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Abbreviations

| Abbreviation | Description |
|--------------|-------------|
| ABCA1        | ATP binding cassette subfamily A member 1 |
| Abcd2        | ATP binding cassette subfamily D member 2 |
| ABCG1        | ATP binding cassette subfamily G member 1 |
| Acaa1        | acetyl-CoA acyltransferase 1 |
| Acadl        | acyl-Coenzyme A dehydrogenase long-chain |
| Acadm        | acyl-Coenzyme A dehydrogenase medium-chain |
| Acadl        | acyl-Coenzyme A dehydrogenase short chain |
| Acad1        | acyl-Coenzyme A dehydrogenase very long-chain |
| Acad11       | acyl-Coenzyme A dehydrogenase family member 11 |
| Acat1        | acetyl-CoA acyltransferase 1 |
| Acot3        | acyl-CoA thioesterase 3 |
| Acx1         | acyl-Coenzyme A oxidase 1 |
| Acsl1        | acyl-CoA synthetase long-chain family member 1 |
| Acsf3        | acyl-CoA synthetase family member 3 |
| ADR          | adverse drug reaction |
| Aldh3a2      | aldehyde dehydrogenase 3 family member A2 |
| APA          | aminopeptidase A |
| ASCVD        | atherosclerotic cardiovascular disease |
| CBP/p300     | cAMP-response element binding protein (CREB) binding protein |
| C/EBPβ       | CCAAT enhancer binding protein β |
| CKD          | chronic kidney disease |
| COL1A2       | collagen type I alpha 2 chain |
| COUP-TFs     | chicken ovalbumin upstream promotor-transcription factors |
| Cpt1         | carnitine palmitoyltransferase 1 |
| Crat         | carnitine acyltransferase |
| CREBH        | cAMP-responsive element-binding protein 3 like 3 |
| CX3CL1       | C-X3-C motif chemokine ligand 1 |
| Cyp4a10      | cytochrome P450, family 4, subfamily a, polypeptide 10 |
| DAG          | diacylglycerol |
| DBD          | DNA binding domain |
| Decr1        | 2,4-dienoyl-CoA reductase 1 |
| DKK1         | dickkopf WNT signaling pathway inhibitor 1 |
| DR1          | direct repeat 1 |
| Ech1         | enoyl-CoA hydratase 1 |
| eGFR         | estimated glomerular filtration rate |
| Ehhadh       | enoyl-Coenzyme A, hydratase/3-hydroxyacyl Coenzyme A dehydrogenase |
| EndMT        | endothelial-mesenchymal transition |
| ENPEP        | glutamyl aminopeptidase |
ET-1 endothelin-1
FA fatty acid
FAT fatty acid translocase
Fabp2 fatty acid-binding protein 2
FGF21 fibroblast growth factor 21
Fn1 fibronectin 1
GRIP1/TIF2 glucocorticoid receptor interacting protein1/transcriptional intermediary factor 2
Hadha hydroxyacyl-CoA dehydrogenase trifunctional multienzyme complex subunit alpha
Hadhb hydroxyacyl-CoA dehydrogenase trifunctional multienzyme complex subunit beta
HAT histone acetyl transferase
HCECs human coronary endothelial cells
HDAC histone deacetylases
HDL-C high-density lipoprotein cholesterol
Hmgcl 3-hydroxy-3-methylglutaryl-CoA lyase
HMGCS2 3-hydroxy-3-methylglutaryl-CoA synthase 2
HNF4s hepatocyte nuclear factor 4s
Hsd17b4 hydroxysteroid 17-beta dehydrogenase 4
HUVECs human umbilical vein endothelial cells
H3K9bhb β-hydroxybutyrylated histone H3 lysine 9
IFNγ interferon gamma
IL6 interleukin 6
LDL low-density lipoprotein
LDLR low-density lipoprotein receptor
Lpl lipoprotein lipase
MBL2 mannose-binding lectin 2
MCP-1 monocyte chemoattractant protein-1
NAD(P)H nicotinamide adenine dinucleotide phosphate
Ncf1 neutrophil cytosolic factor 1
Nox4 NADPH oxidase 4
8-OHdG 8-hydroxy-2’-deoxyguanosine
PDK4 pyruvate dehydrogenase kinase 4
Pexl peroxisome biogenesis factor 1
PDH pyruvate dehydrogenase
PGC1α peroxisome proliferative activated receptor gamma coactivator 1α
PIC preinitiation complex
PKC protein kinase C
PPARα peroxisome proliferator-activated receptor α
PPREs PPAR response elements
RANTES regulated on activation, normal T cell expressed and secreted
ROS reactive oxygen species
RXR retinoid X receptor
SBP systolic blood pressure
Slc27a1 solute carrier family 27 member 1
Slc25a20 solute carrier family 25 member 20
SPPARMα selective peroxisome proliferator-activated receptor α modulator
SRC/p160 steroid receptor coactivator
TG triglyceride
Tgfb1 transforming growth factor beta 1
TNFα tumor necrosis factor α
VCAM1 vascular cell adhesion molecule 1
VLDLR very-low-density lipoprotein receptor
References

1. Cholesterol Treatment Trialists’ Collaboration. Efficacy and safety of statin therapy in older people: A meta-analysis of individual participant data from 28 randomised controlled trials. *Lancet* 2019, 393, 407–415. [CrossRef]

2. Cholesterol Treatment Trialists’ (CTT) Collaboration; Fulcher, J.; O’Connell, R.; Voysey, M.; Emberson, J.; Blackwell, L.; Mihaylova, B.; Simes, J.; Collins, R.; Kirby, A.; et al. Efficacy and safety of LDL-lowering therapy among men and women: Meta-analysis of individual data from 174,000 participants in 27 randomised trials. *Lancet* 2015, 385, 1397–1405. [PubMed]

3. Fruchart, J.C.; Sacks, F.; Hermans, M.P.; Assmann, G.; Brown, W.V.; Ceska, R.; Chapman, M.J.; Dodson, P.M.; Fioretto, P.; Ginsberg, H.N.; et al. The Residual Risk Reduction Initiative: A call to action to reduce residual vascular risk in patients with dyslipidemia. *Am. J. Cardiol.* 2008, 102, 1K–34K. [CrossRef] [PubMed]

4. Alagona, P., Jr. Beyond LDL cholesterol: The role of elevated triglycerides and low HDL cholesterol in residual CVD risk remaining after statin therapy. *Am. J. Manag. Care* 2009, 15, S65–S73.

5. Reiner, Z. Managing the residual cardiovascular disease risk associated with HDL-cholesterol and triglycerides in statin-treated patients: A clinical update. *Nutr. Metab. Cardiovasc. Dis.* 2013, 23, 799–807. [CrossRef]

6. Fruchart, J.C. Peroxisome proliferator-activated receptor-γ agonists. *Cardiovasc. Diabetol.* 2012, 11, 1K–34K. [CrossRef] [PubMed]

7. Chapman, M.J.; Ginsberg, H.N.; Amarenco, P.; Andreotti, F.; Borén, J.; Catapano, A.L.; Descamps, O.S.; Fisher, E.; Kovanen, P.T.; Kuivenhoven, J.A.; et al. European Atherosclerosis Society Consensus Panel. Triglyceride-rich lipoproteins and high-density lipoprotein cholesterol in patients at high risk of cardiovascular disease: Evidence and guidance for management. *Eur. Heart J.* 2011, 32, 1345–1361. [CrossRef]

8. Vrablik, M.; Češka, R. Treatment of hypertriglyceridemia: A review of current options. *Physiol. Res.* 2015, 64, S331–S340.

9. Katsiki, N.; Nikolic, D.; Montalto, G.; Banach, M.; Mikhailidis, D.P.; Rizzo, M. The role of fibrate treatment in dyslipidemia: An overview. *Curr. Pharm. Des.* 2011, 17, 1397–1405. [PubMed]

10. McCullough, P.A.; Ahmed, A.B.; Zughaib, M.T.; Glanz, E.D.; Di Loreto, M.J. Treatment of hypertriglyceridemia with fibric acid derivatives: Impact on lipid subfractions and translation into a reduction in cardiovascular events. *Rev. Cardiovasc. Med.* 2011, 12, 173–185.

11. Nakaya, N.; Goto, Y. A retrospective meta-analysis of the efficacy and tolerability of fenofibrate 300 mg/d on high-density lipoprotein cholesterol levels in randomized, double-blind, comparative studies conducted in Japan. *Curr. Res. Clin. Exp.* 2003, 64, 634–644. [CrossRef] [PubMed]

12. Davidson, M.H.; Armani, A.; McKenney, J.M.; Jacobson, T.A. Safety considerations with fibrate therapy. *Am. J. Cardiol.* 2007, 99, 3C–18C. [CrossRef] [PubMed]

13. Ahmad, J.; Odin, J.A.; Hayashi, P.H.; Chalasani, N.; Fontana, R.J.; Barnhart, H.; Cirulli, E.T.; Kleiner, D.E.; Hoofnagle, J.H. Identification and Characterization of Fenofibrate-Induced Liver Injury. *Dig. Dis. Sci.* 2017, 62, 3596–3604. [CrossRef] [PubMed]

14. Abbas, A.; Saraf, S.; Ramachandran, S.; Raju, J.; Ramachandran, S. Fibrates and estimated glomerular filtration rate: Observations from an outpatient clinic setting and clinical implications. *Postgrad. Med. J.* 2012, 88, 503–506. [CrossRef] [PubMed]

15. Yamazaki, Y.; Abe, K.; Toma, T.; Nishikawa, M.; Ozawa, H.; Okuda, A.; Araki, T.; Oda, S.; Inoue, K.; Shibuya, K.; et al. Design and synthesis of highly potent and selective human peroxisome proliferator-activated receptor α agonists. *Bioorg. Med. Chem. Lett.* 2007, 17, 4689–4693. [CrossRef]

16. Fruchart, J.C. Peroxisome proliferator-activated receptor-α (PPARα): At the crossroads of obesity, diabetes and cardiovascular disease. *Atherosclerosis* 2009, 205, 1–8. [CrossRef]

17. Fruchart, J.C. Selective peroxisome proliferator-activated receptor α modulators (SPPARMs): The next generation of peroxisome proliferator-activated receptor α-agonists. *Cardiovasc. Diabetol.* 2013, 12, 82. [CrossRef]

18. Yamamoto, Y.; Takei, K.; Arulmozhiraja, S.; Sladek, V.; Matsuo, N.; Han, S.I.; Matsuzaka, T.; Sekiya, M.; Tokiwa, T.; Shoji, M.; et al. Molecular association model of PPARα and its new specific and efficient ligand, pemafibrate: Structural basis for SPPARMx. *Biochem. Biophys. Res. Commun.* 2018, 499, 239–245. [CrossRef]
19. Raza-Iqbal, S.; Tanaka, T.; Anai, M.; Inagaki, T.; Matsumura, Y.; Ikeda, K.; Taguchi, A.; Gonzalez, F.J.; Sakai, J.; Kodama, T. Transcriptome Analysis of K-877 (a Novel Selective PPARx Modulator (SPPARMα))-Regulated Genes in Primary Human Hepatocytes and the Mouse Liver. J. Atheroscler. Thromb. 2015, 22, 754–772. [CrossRef]

20. Fruchart, J.C. Pemafibrate (K-877), a novel selective peroxisome proliferator-activated receptor α modulator for management of atherogenic dyslipidaemia. Cardiovasc. Diabetol. 2017, 16, 124. [CrossRef]

21. Takei, K.; Han, S.I.; Murayama, Y.; Satoh, A.; Oikawa, F.; Ohno, H.; Osaki, Y.; Matsuzaka, T.; Sekiya, M.; Iwasaki, H.; et al. Selective peroxisome proliferator-activated receptor-α modulator K-877 efficiently activates the peroxisome proliferator-activated receptor-α pathway and improves lipid metabolism in mice. J. Diabetes Investig. 2017, 8, 446–452. [CrossRef] [PubMed]

22. Hennuyer, N.; Duplan, I.; Paquet, C.; Vanhoutte, J.; Woitrain, E.; Touche, V.; Colin, S.; Vallez, E.; Lestavel, S.; Lefebvre, P.; et al. The novel selective PPARα modulator (SPPARMα) pemafibrate improves dyslipidemia, enhances reverse cholesterol transport and decreases inflammation and atherosclerosis. Atherosclerosis 2016, 249, 200–208. [CrossRef] [PubMed]

23. Gibson, G.G. Peroxisome proliferators: Paradigms and prospects. Toxicol. Lett. 1993, 68, 193–201. [CrossRef]

24. Misra, P.; Viswakarma, N.; Reddy, J.K. The role of peroxisome proliferator-activated receptors in carcinogenesis. Subcell. Biochem. 2013, 69, 77–99. [PubMed]

25. Peters, J.M.; Shah, Y.M.; Gonzalez, F.J. The effects of K-877, a novel selective PPARα modulator (SPPARMα), K-877 (Pemafibrate), Attenuates Postprandial Hypertriglyceridemia in Mice. J. Atheroscler. Thromb. 2018, 25, 142–152. [CrossRef]

26. Fruchart, J.C.; Santos, R.D.; Aguilar-Salinas, C.; Aikawa, M.; Al Rasadi, K.; Amarenco, P.; Barter, P.J.; Ceska, R.; Corsini, A.; Després, J.P.; et al. The selective peroxisome proliferator-activated receptor α modulator (SPPARMα) paradigm: Conceptual framework and therapeutic potential: A consensus statement from the International Atherosclerosis Society (IAS) and the Residual Risk Reduction Initiative (R3i) Foundation. Cardiovasc. Diabetol. 2019, 18, 71.

27. Takei, K.; Nakagawa, Y.; Wang, Y.; Han, S.I.; Satoh, A.; Sekiya, M.; Matsuzaka, T.; Shimano, H. Effects of K-877, a novel selective PPARα modulator, on small intestine contribute to the amelioration of hyperlipidemia in low-density lipoprotein receptor knockout mice. J. Pharm. Sci. 2017, 133, 214–222. [CrossRef]

28. Sairyo, M.; Kobayashi, T.; Masuda, D.; Kanno, K.; Zhu, Y.; Okada, T.; Koike, M.; Ohama, T.; Nishida, M.; Sakata, Y.; et al. A Novel Selective PPARα Modulator (SPPARMα), K-877 (Pemafibrate), Attenuates Postprandial Hypertriglyceridemia in Mice. J. Atheroscler. Thromb. 2018, 25, 142–152. [CrossRef]

29. Fruchart, J.C.; Santos, R.D.; Aguilar-Salinas, C.; Aikawa, M.; Al Rasadi, K.; Amarenco, P.; Barter, P.J.; Ceska, R.; Corsini, A.; Després, J.P.; et al. The novel selective peroxisome proliferator-activated receptor α modulator (SPPARMα) paradigm: Conceptual framework and therapeutic potential: A consensus statement from the International Atherosclerosis Society (IAS) and the Residual Risk Reduction Initiative (R3i) Foundation. Cardiovasc. Diabetol. 2019, 18, 71.

30. Arai, H.; Yamashita, S.; Arai, H.; Araki, E.; Yokote, K.; Suganami, H.; Ishibashi, S.; K-877 Study Group. Efficacy and safety of K-877, a novel selective PPARα modulator (SPPARMα), in dyslipidemic patients: A randomized, double blind, active- and placebo-controlled, phase 2 trial. Atherosclerosis 2016, 249, 36–43. [CrossRef]

31. Arai, H.; Yamashita, S.; Arai, H.; Araki, E.; Sugarami, H.; Ishibashi, S.; K-877 Study Group. Efficacy and safety of K-877, a novel selective PPARα modulator (SPPARMα), in combination with statin treatment: Two randomised, double-blind, placebo-controlled clinical trials in patients with dyslipidemia. Atherosclerosis 2017, 261, 144–152.

32. Ishibashi, S.; Arai, H.; Yokote, K.; Araki, E.; Sugarami, H.; Yamashita, S.; K-877 Study Group. Efficacy and safety of pemafibrate (K-877), a selective peroxisome proliferator-activated receptor α modulator, in patients with dyslipidemia: Results from a 24-week, randomized, double blind, active-controlled, phase 3 trial. J. Clin. Lipidol. 2018, 12, 173–184. [CrossRef] [PubMed]

33. Araki, E.; Yamashita, S.; Arai, H.; Yokote, K.; Araki, E.; Sugarami, H.; Ishibashi, S.; K-877 Study Group. Efficacy and Safety of Pemafibrate Versus Fenoibrate in Patients with High Triglyceride and Low HDL Cholesterol Levels: A Multicenter, Placebo-Controlled, Double-Blind, Randomized Trial. J. Atheroscler. Thromb. 2018, 25, 521–538. [CrossRef] [PubMed]
34. Matsuba, I.; Matsuba, R.; Ishibashi, S.; Yamashita, S.; Arai, H.; Yokote, K.; Suganami, H.; Araki, E. Effects of a novel selective peroxisome proliferator-activated receptor-α modulator, pemafibrate, on hepatic and peripheral glucose uptake in patients with hypertriglyceridemia and insulin resistance. *J. Diabetes Investig.* 2018, 9, 1323–1332. [CrossRef] [PubMed]

35. Yamashita, S.; Masuda, D.; Matsuzawa, Y. Clinical Applications of a Novel Selective PPARα Modulator, Pemafibrate, in Dyslipidemia and Metabolic Diseases. *J. Atheroscler. Thromb.* 2019, 26, 389–402. [CrossRef] [PubMed]

36. Ida, S.; Kaneko, R.; Murata, K. Efficacy and safety of pemafibrate administration in patients with dyslipidemia: A systematic review and meta-analysis. *Cardiovasc. Diabetol.* 2019, 18, 38. [CrossRef]

37. Araki, E.; Yamashita, S.; Arai, H.; Yokote, K.; Satoh, J.; Inoguchi, T.; Nakamura, J.; Maegawa, H.; Yoshioka, N.; Tanizawa, Y.; et al. Efficacy and safety of pemafibrate in people with type 2 diabetes and elevated triglyceride levels: 52-week data from the PROVIDE study. *Diabetes Obes. Metab.* 2019, 21, 1737–1744. [CrossRef]

38. Yokote, K.; Yamashita, S.; Arai, H.; Araki, E.; Suganami, H.; Ishibashi, S.; K-Study Group. Long-Term Efficacy and Safety of Pemafibrate, a Novel Selective Peroxisome Proliferator-Activated Receptor-α Modulator (SPPARMα), in Dyslipidemic Patients with Renal Impairment. *Int. J. Mol. Sci.* 2019, 20, 706. [CrossRef]

39. Fisher, F.M.; Maratos-Flier, E. Understanding the Physiology of FGF21. *Annu. Rev. Physiol.* 2016, 78, 223–241. [CrossRef]

40. Holden, P.R.; Tugwood, J.D. Peroxisome proliferator-activated receptor α: Role in rodent liver cancer and species differences. *J. Mol. Endocrinol.* 1999, 22, 1–8. [CrossRef]

41. Lawrence, J.W.; Li, Y.; Chen, S.; DeLuca, J.G.; Berger, J.P.; Umbenhauer, D.R.; Moller, D.E.; Zhou, G. Differential gene regulation in human versus rodent hepatocytes by peroxisome proliferator-activated receptor (PPAR) α. PPARα fails to induce peroxisome proliferation-associated genes in human cells independently of the level of receptor expression. *J. Biol. Chem.* 2001, 276, 31521–31527. [CrossRef] [PubMed]

42. Hsu, M.H.; Savas, U.; Griffin, K.J.; Johnson, E.F. Identification of peroxisome proliferator-responsive human genes by elevated expression of the peroxisome proliferator-activated receptor α in HepG2 cells. *J. Biol. Chem.* 2001, 276, 27950–27958. [CrossRef] [PubMed]

43. Adeva-Andany, M.M.; Carneiro-Freire, N.; Seco-Filgueira, M.; Fernández-Fernández, C.; Mourinho-Bayolo, D. Mitochondrial β-oxidation of saturated fatty acids in humans. *Mitochondrion* 2019, 46, 73–90. [CrossRef] [PubMed]

44. Ferdinandusse, S.; Denis, S.; Van Roermund, C.W.; Wanders, R.J.; Daemcott, G. Identification of the peroxisomal beta-oxidation enzymes involved in the degradation of long-chain dicarboxylic acids. *J. Lipid Res.* 2004, 45, 1104–1111. [CrossRef]

45. Yeldandi, A.V.; Rao, M.S.; Reddy, J.K. Hydrogen peroxide generation in peroxisome proliferator-induced oncogenesis. *Mutat. Res.* 2000, 448, 159–177. [CrossRef]

46. Raucy, J.L.; Lasker, J.; Ozaki, K.; Zoleta, V. Regulation of CYP2E1 by ethanol and palmitic acid and CYP4A11 by clofibrate in primary cultures of human hepatocytes. *Toxicol. Sci.* 2004, 79, 233–241. [CrossRef]

47. Savas, U.; Hsu, M.H.; Johnson, E.F. Differential regulation of human CYP4A genes by peroxisome proliferators and dexamethasone. *Arch. Biochem. Biophys.* 2003, 409, 212–220. [CrossRef]

48. Pettersen, I.K.N.; Tusubira, D.; Ashrafi, H.; Dyrstad, S.E.; Hansen, L.; Liu, X.Z.; Nilsson, L.I.H.; Løvsletten, N.G.; Berge, K.; Wergedahl, H.; et al. Upregulated PDK4 expression is a sensitive marker of increased fatty acid oxidation. *Mitochondrion* 2019, 49, 97–110. [CrossRef]

49. Attia, R.R.; Sharma, P.; Janssen, R.C.; Friedman, J.E.; Deng, X.; Lee, J.S.; Elam, M.B.; Cook, G.A.; Park, E.A. Regulation of pyruvate dehydrogenase kinase 4 (PDK4) by CCAAT/enhancer-binding protein alpha (C/EBPalpha). *J. Biol. Chem.* 2011, 286, 23799–23807. [CrossRef]

50. Holness, M.J.; Bulmer, K.; Smith, N.D.; Sugden, M.C. Investigation of potential mechanisms regulating protein expression of hepatic pyruvate dehydrogenase kinase isoforms 2 and 4 by fatty acids and thyroid hormone. *Biochem. J.* 2003, 369, 687–695. [CrossRef]

51. Vilà-Brau, A.; De Sousa-Coelho, A.L.; Mayordomo, C.; Haro, D.; Marrero, P.P. Human HMGC2 regulates mitochondrial fatty acid oxidation and FGF21 expression in HepG2 cell line. *J. Biol. Chem.* 2011, 286, 20423–20430. [CrossRef]

52. Xie, Z.; Zhang, D.; Chung, D.; Tang, Z.; Huang, H.; Dai, L.; Qi, S.; Li, J.; Colak, G.; Chen, Y.; et al. Metabolic Regulation of Gene Expression by Histone Lysine β-Hydroxybutyrylation. *Mol. Cell* 2016, 62, 194–206. [CrossRef]
53. Webb, J.C.; Patel, D.D.; Jones, M.D.; Knight, B.L.; Soutar, A.K. Characterization and tissue-specific expression of the human ‘very low density lipoprotein (VLDL) receptor’ mRNA. Hum. Mol. Genet. 1994, 3, 531–537.

54. Tiebel, O.; Oka, K.; Robinson, K.; Sullivan, M.; Martínez, J.; Nakamura, M.; Ishimura-Oka, K.; Chan, L. Mouse very low-density lipoprotein receptor (VLDLr): Gene structure, tissue-specific expression and dietary and developmental regulation. Atherosclerosis 1999, 145, 239–251. [CrossRef]

55. Gao, Y.; Shen, W.; Lu, B.; Zhang, Q.; Hu, Y.; Chen, Y. Upregulation of hepatic VLDLR via PPARα is required for the triglyceride-lowering effect of fenofibrate. J. Lipid Res. 2014, 55, 1622–1633. [CrossRef]

56. Merkel, M.; Weinstock, P.H.; Chajek-Shaul, T.; Radner, H.; Yin, B.; Breslow, J.L.; Goldberg, I.J. Lipoprotein lipase expression exclusively in liver. A mouse model for metabolism in the neonatal period and during cachexia. J. Clin. Investig. 1998, 102, 893–901. [CrossRef]

57. Wang, S.; Smith, J.D. ABCA1 and nascent HDL biogenesis. Biofactors 2014, 40, 547–554. [CrossRef]

58. Babashamsi, M.M.; Koukhaloo, S.Z.; Halalkhor, S.; Salimi, A.; Babashamsi, M. ABCA1 and metabolic syndrome: a review of the ABCA1 role in HDL-VLDL production, insulin-glucose homeostasis, inflammation and obesity. Diabetes Metab. Syndr. 2019, 13, 1529–1534. [CrossRef]

59. Liu, Y.; Tang, C. Regulation of ABCA1 functions by signaling pathways. Biochim. Biophys. Acta 2012, 1821, 522–529. [CrossRef]

60. Brunham, L.R.; Singaraja, R.R.; Duong, M.; Timmins, J.M.; Fievet, C.; Bissada, N.; Kang, M.H.; Samra, A.; Fruchart, J.C.; McManus, B.; et al. Tissue-specific roles of ABCA1 influence susceptibility to atherosclerosis. Arterioscler. Thromb. Vasc. Biol. 2009, 29, 548–554. [CrossRef]

61. Kharitonenkova, A.; Shiyanova, T.L.; Koester, A.; Ford, A.M.; Micanovic, R.; Galyas, E.; Hammond, L.J.; Moyers, J.S.; Owens, R.A.; et al. FGF-21 as a novel metabolic regulator. J. Clin. Investig. 2005, 115, 1627–1635. [CrossRef]

62. Véniant, M.M.; Komorowski, R.; Chen, P.; Stanislaus, S.; Winters, K.; Hager, T.; Zhou, L.; Wada, R.; Hecht, R.; Xu, J. Long-acting FGF21 has enhanced efficacy in diet-induced obese mice and in obese rhesus monkeys. Endocrinology 2012, 153, 4192–4203. [CrossRef]

63. Inagaki, T.; Dutchak, P.; Zhao, G.; Gautron, L.; Parameswara, V.; Li, Y.; Goetz, R.; Mohammad, M.; Esser, V.; et al. Endocrine regulation of the fasting response by PPARα-mediated induction of fibroblast growth factor 21. Cell Metab. 2007, 5, 415–425. [CrossRef]

64. Lundäsen, T.; Hunt, M.C.; Nilsson, L.M.; Sanyal, S.; Angelin, B.; Alexson, S.E.; Rudling, M. PPARα is a key regulator of hepatic FGF21. Biochem. Biophys. Res. Commun. 2007, 360, 437–440. [CrossRef]

65. Yamashita, S.; Arai, H.; Yokote, K.; Araki, E.; Suganami, H.; Ishibashi, S.; K-877 Study Group. Effects of pemafibrate (K-877) on cholesterol efflux capacity and postprandial hyperlipidemia in patients with atherogenic dyslipidemia. J. Clin. Lipidol. 2018, 12, 1267–1279. [CrossRef]

66. Kim, H.; Mendez, R.; Zheng, Z.; Chang, L.; Cai, J.; Zhang, R.; Zhang, K. Liver-enriched transcription factor CREBH interacts with peroxisome proliferator-activated receptor α to regulate metabolic hormone FGF21. Endocrinology 2014, 155, 769–782. [CrossRef]

67. Ip, W.K.; Takahashi, K.; Ezekowitz, R.A.; Stuart, L.M. Mannose-binding lectin and innate immunity. Immunol. Rev. 2009, 230, 9–21.

68. Hansen, T.K. Growth hormone and mannan-binding lectin: Emerging evidence for hormonal regulation of humoral innate immunity. Minerva. Endocrinologia 2003, 28, 75–84.

69. Fernández-Real, J.M.; Straczkowski, M.; Vendrell, J.; Soriguer, F.; Pérez Del Pulgar, S.; Gallart, L.; López-Bermejo, A.; Kowalska, I.; Manco, M.; Cardona, F.; et al. Protection from inflammatory disease in insulin resistance: The role of mannan-binding lectin. Diabetologia 2006, 49, 2402–2411. [CrossRef]

70. Holmes, R.S.; Spradling-Reeves, K.D.; Cox, L.A. Mammalian Glutamyl Aminopeptidase Genes (ENPEP) and Proteins: Comparative Studies of a Major Contributor to Arterial Hypertension. J. Data Min. Genomics Proteom. 2017, 8, 2. [CrossRef]

71. Mizutani, S.; Ishii, M.; Hattori, A.; Nomura, S.; Numaguchi, Y.; Tsujimoto, M.; Kobayshi, H.; Murohara, T.; Wright, J.W. New insights into the importance of aminopeptidase A in hypertension. Heart Fail. Rev. 2008, 13, 273–284. [CrossRef]

72. Tsujimoto, M.; Goto, Y.; Maruyama, M.; Hattori, A. Biochemical and enzymatic properties of the M1 family of aminopeptidases involved in the regulation of blood pressure. Heart. Fail. Rev. 2008, 13, 285–291. [CrossRef]
73. Mitsui, T.; Nomura, S.; Okada, M.; Ohno, Y.; Kobayashi, H.; Nakashima, Y.; Murata, Y.; Takeuchi, M.; Kunoo, N.; Nagasaka, T.; et al. Hypertension and angiotensin II hypersensitivity in aminopeptidase A-deficient mice. Mol. Med. 2003, 9, 57–62. [CrossRef]

74. Surendran, P.; Drenos, F.; Young, R.; Warren, H.; Cook, J.P.; Manning, A.K.; Grarup, N.; Sim, X.; Barnes, D.R.; Witkowska, K.; et al. Trans-ancestry meta-analyses identify rare and common variants associated with blood pressure and hypertension. Nat. Genet. 2016, 48, 1151–1161. [CrossRef]

75. Rajendran, P.; Rengarajan, T.; Thangavel, J.; Nishigaki, Y.; Sakthisekaran, D.; Sethi, G.; Nishigaki, I. The vascular endothelium and human diseases. Int. J. Biol. Sci. 2013, 9, 1057–1069. [CrossRef]

76. Cheng, H.; Harris, R.C. Renal Endothelial Dysfunction in Diabetic Nephropathy. Cardiovasc. Hematol. Disord. Drug Targets 2014, 14, 22–33. [CrossRef]

77. Gimbrone, M.A., Jr.; García-Cardeña, G. Endothelial Cell Dysfunction and the Pathobiology of Atherosclerosis. Circ. Res. 2016, 118, 620–636. [CrossRef]

78. Maki, T.; Maeda, Y.; Sonoda, N.; Makimura, H.; Kimura, S.; Maeno, S.; Takayanagi, R.; Inoguchi, T. The vascular endothelium and human diseases. Int. J. Biol. Sci. 2013, 9, 1057–1069. [CrossRef]

79. Kitajima, K.; Miura, S.; Mastuo, Y.; Uehara, Y.; Saku, K. Newly developed PPAR-agonist (R)-K-13675 inhibits endothelial cell proliferation or tube formation. Atherosclerosis 2009, 203, 75–81. [CrossRef]

80. Kovacic, J.C.; Dimmeler, S.; Harvey, R.P.; Finkel, T.; Aikawa, E.; Krenning, G.; Baker, A.H. Endothelial to Mesenchymal Transition in Cardiovascular Disease: JACC State-of-the-Art Review. J. Am. Coll. Cardiol. 2019, 73, 190–209. [CrossRef]

81. Cho, J.G.; Lee, A.; Chang, W.; Lee, M.S.; Kim, J. Endothelial to Mesenchymal Transition Represents a Key Link in the Interaction between Inflammation and Endothelial Dysfunction. Front. Immunol. 2018, 9, 294. [CrossRef]

82. Thomas, A.A.; Biswas, S.; Feng, B.; Chen, S.; Gonder, J.; Chakrabarti, S. IncRNA H19 prevents endothelial-mesenchymal transition in diabetic retinopathy. Diabetologia 2019, 62, 517–530. [CrossRef]

83. Gong, H.; Lyu, X.; Wang, Q.; Hu, M.; Zhang, X. Endothelial to mesenchymal transition in the cardiovascular system. Life Sci. 2017, 184, 95–102. [CrossRef]

84. Li, Y.; Lui, K.O.; Zhou, B. Reassessing endothelial-to-mesenchymal transition in cardiovascular diseases. Nat. Rev. Cardiol. 2018, 15, 445–456. [CrossRef]

85. Cheng, S.L.; Shao, J.S.; Behrmann, A.; Krchma, K.; Towler, D.A. Dkk1 and MSX2-Wnt7b signaling reciprocally regulate the endothelial-mesenchymal transition in aortic endothelial cells. Arter. Thromb. Vasc. Biol. 2013, 33, 1679–1689. [CrossRef]

86. Glineur, C.; Gross, B.; Neve, B.; Rommens, C.; Chew, G.T.; Martin-Nizard, F.; Rodríguez-Pascual, F.; Lamas, S.; Watts, G.F.; Staels, B. Fenofibrate inhibits endothelin-1 expression by peroxisome proliferator-activated receptor α-dependent and independent mechanisms in human endothelial cells. Arter. Thromb. Vasc. Biol. 2013, 33, 621–628. [CrossRef]

87. Widiantoro, B.; Emoto, N.; Nakayama, K.; Anggrahini, D.W.; Adiarto, S.; Iwasa, N.; Yagi, K.; Miyagawa, K.; Rikitake, Y.; Suzuki, T.; et al. Endothelial cell-derived endothelin-1 promotes cardiac fibrosis in diabetic hearts through stimulation of endothelial-to-mesenchymal transition. Circulation 2010, 121, 2407–2418. [CrossRef]

88. Cipriani, P.; Di Benedetto, P.; Ruscitti, P.; Capece, D.; Zazzaroni, F.; Liakoulí, V.; Pantano, I.; Berardicurti, O.; Carubbi, F.; Pecetti, G.; et al. The Endothelial-mesenchymal Transition in Systemic Sclerosis Is Induced by Endothelin-1 and Transforming Growth Factor-β and May Be Blocked by Macitentan, a Dual Endothelin-1 Receptor Antagonist. J. Rheumatol. 2015, 42, 1808–1816. [CrossRef]

89. Keec, A.; Simes, R.J.; Barter, P.; Best, J.; Scott, R.; Taskinen, M.R.; Forder, P.; Pillai, A.; Davis, T.; Glasziou, P.; et al. FIELD study investigators. Effects of long-term fenofibrate therapy on cardiovascular events in 9975 people with type 2 diabetes mellitus (the FIELD study): Randomised controlled trial. Lancet 2005, 366, 1849–1861. [CrossRef]

90. Keec, A.C.; Mitchell, P.; Summanen, P.A.; O’Day, J.; Davis, T.M.; Moffitt, M.S.; Taskinen, M.R.; Simes, R.J.; Tse, D.; Williamson, E.; et al. FIELD study investigators. Effect of fenofibrate on the need for laser treatment for diabetic retinopathy (FIELD study): A randomised controlled trial. Lancet 2007, 370, 1687–1697. [CrossRef]
91. Chew, E.Y.; Davis, M.D.; Danis, R.P.; Lovato, J.F.; Perdue, L.H.; Greven, C.; Genth, S.; Goff, D.C.; Leiter, L.A.; Ismail-Beigi, F.; et al. Action to Control Cardiovascular Risk in Diabetes Eye Study Research Group. The effects of medical management on the progression of diabetic retinopathy in persons with type 2 diabetes: The Action to Control Cardiovascular Risk in Diabetes (ACCORD) Eye Study. Ophthalmology 2014, 121, 2443–2451.

92. Bougarne, N.; Weyers, B.; Desmet, S.J.; Deckers, J.; Ray, D.W.; Staels, B.; De Bosscher, K. Molecular Actions of PPARα in Lipid Metabolism and Inflammation. Endocr. Rev. 2018, 39, 760–802. [CrossRef] [PubMed]

93. Duncan, J.G. Peroxisome proliferator activated receptor-α (PPARα) and PPAR gamma coactivator-1α (PGC-1α) regulation of cardiac metabolism in diabetes. Pediatr. Cardiol. 2011, 32, 323–328. [CrossRef] [PubMed]

94. Pawlak, M.; Lefebvre, P.; Staels, B. Molecular mechanism of PPARα action and its impact on lipid metabolism, inflammation and fibrosis in non-alcoholic fatty liver disease. J. Hepatol. 2015, 62, 720–733. [CrossRef]

95. Viswakarma, N.; Jia, Y.; Bai, L.; Gao, Q.; Lin, B.; Zhang, X.; Misra, P.; Rana, A.; Jain, S.; Gonzalez, F.J.; et al. The Med1 subunit of the mediator complex induces liver cell proliferation and is phosphorylated by AMP kinase. J. Biol. Chem. 2013, 288, 27988–27991. [CrossRef]

96. Mukherjee, R.; Sun, S.; Santomenna, L.; Miao, B.; Walton, H.; Liao, B.; Locke, K.; Zhang, J.H.; Nguyen, S.H.; Zhang, L.T.; et al. Ligand and coactivator recruitment preferences of peroxisome proliferator activated receptor α. J. Steroid Biochem. Mol. Biol. 2002, 81, 217–225. [CrossRef]

97. Surapureddi, S.; Yu, S.; Bu, H.; Hashimoto, T.; Yeldandi, A.V.; Kashireddi, P.; Cherkaoui-Malki, M.; Qi, C.; Zhu, Y.J.; Rao, M.S.; et al. Identification of a transcriptionally active peroxisome proliferator-activated receptor α-interacting cofactor complex in rat liver and characterization of PRIC285 as a coactivator. Proc. Natl. Acad. Sci. USA 2002, 99, 11836–11841. [CrossRef]

98. Jia, Y.; Qi, C.; Kashireddi, P.; Surapureddi, S.; Zhu, Y.J.; Rao, M.S.; Le Roith, D.; Chambon, P.; Gonzalez, F.J.; Reddy, J.K. Transcription coactivator PBP , the peroxisome proliferator-activated receptor (PPAR)-binding factor, is required for PPARα-regulated gene expression in liver. J. Biol. Chem. 2004, 279, 24427–244234. [CrossRef]

99. Pawlak, M.; Baugé, E.; Bourguet, W.; De Bosscher, K.; Lalloyer, F.; Tailleux, A.; Leberherz, C.; Lefebvre, P.; Staels, B. The transrepressive activity of peroxisome proliferator-activated receptor α is necessary and sufficient to prevent liver fibrosis in mice. Hepatolgy 2014, 60, 1593–1606. [CrossRef]

100. Delerive, P.; De Bosscher, K.; Besnard, S.; Vanden Berghe, W.; Peters, J.M.; Gonzalez, F.J.; Fruchart, J.C.; Tedgui, A.; Haegeman, G.; Staels, B. Peroxisome proliferator-activated receptor α negatively regulates the vascular inflammatory gene response by negative cross-talk with transcription factors NF-κB and AP-1. J. Biol. Chem. 1999, 274, 32048–32054. [CrossRef]

101. Planavila, A.; Iglesias, R.; Giralt, M.; Villarroya, F. Sirt1 acts in association with PPARα to protect the heart from hypertrophy, metabolic dysregulation, and inflammation. Cardiovasc. Res. 2011, 90, 276–284. [CrossRef] [PubMed]

102. Bougarne, N.; Paumelle, R.; Caron, S.; Hennuyer, N.; Mansouri, R.; Gervois, P.; Staels, B.; Haegeman, G.; De Bosscher, K. PPARα blocks glucocorticoid receptor α-mediated transactivation but cooperates with the activated glucocorticoid receptor α for transrepression on NF-κB. Proc. Natl. Acad. Sci. USA 2009, 106, 7397–7402. [CrossRef] [PubMed]

103. Gervois, P.; Vu-Dac, N.; Kleemann, R.; Kockx, M.; Dubois, G.; Laine, B.; Kosyk, V.; Fruchart, J.C.; Kooistra, T.; Staels, B. Negative regulation of human fibrinogen gene expression by peroxisome proliferator-activated receptor α agonists via inhibition of CCAAT box/enhancer-binding protein β. J. Biol. Chem. 2001, 276, 33471–33477. [CrossRef] [PubMed]

104. Dongol, B.; Shah, Y.; Kim, I.; Gonzalez, F.J.; Hunt, M.C. The acyl-CoA thioesterase I is regulated by PPARx and HNF4α via a distal response element in the promoter. J. Lipid Res. 2007, 48, 1781–1791. [CrossRef]

105. Marrapodi, M.; Chiang, J.Y. Peroxisome proliferator-activated receptor α (PPARα) and agonist inhibit cholesterol 7α-hydroxylase gene (CYP7A1) transcription. J. Lipid Res. 2000, 41, 514–520.

106. Spann, N.J.; Kang, S.; Li, A.C.; Chen, A.Z.; Newberry, E.P.; Davidson, N.O.; Hui, S.T.; Davis, R.A. Coordinate transcriptional repression of liver fatty acid-binding protein and microsomal triglyceride transfer protein blocks hepatic very low density lipoprotein secretion without hepatosteatosis. J. Biol. Chem. 2006, 281, 33066–33077. [CrossRef]
107. Ilpenberg, A.; Tan, N.S.; Gelman, L.; Kersten, S.; Seydoux, J.; Xu, J.; Metzger, D.; Canaple, L.; Chambon, P.; Wahl, W.; et al. In vivo activation of PPAR target genes by RXR homodimers. *EMBO J.* 2004, 23, 2083–2091. [CrossRef]

108. Pradhan, A.D.; Paynter, N.P.; Everett, B.M.; Glynn, R.J.; Amarenco, P.; Elam, M.; Ginsberg, H.; Hiatt, W.R.; Ishibashi, S.; Koenig, W.; et al. Rationale and design of the Pemafibrate to Reduce Cardiovascular Outcomes by Reducing Triglycerides in Patients with Diabetes (PROMINENT) study. *Am. Heart J.* 2018, 206, 80–93. [CrossRef]

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