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REVIEW

The peroxisome proliferator-activated receptors in cardiovascular diseases: experimental benefits and clinical challenges

Wai San Cheang[1], Xiao Yu Tian[2], Wing Tak Wong[2] and Yu Huang[1]

[1]*Shenzhen Research Institute, Institute of Vascular Medicine and Li Ka Shing Institute of Health Sciences, Chinese University of Hong Kong, Hong Kong, China, and* [2]*Department of Cardiovascular Sciences, Houston Methodist Research Institute, Houston, TX, USA*

Correspondence
Yu Huang, School of Biomedical Sciences, Chinese University of Hong Kong, Hong Kong, China.
E-mail: yu-huang@cuhk.edu.hk

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The peroxisome proliferator-activated receptors, PPARα, PPARβ/δ and PPARγ, are ligand-activated transcriptional factors belonging to the nuclear receptors superfamily and they are known to play important roles in glucose and lipid metabolism. Experimental studies in animal models of metabolic diseases have also revealed that activation of PPARs protects against the vascular complications of diabetes, hypertension, atherosclerosis, myocardial infarction and stroke, through exerting their anti-inflammatory, anti-atherogenic and antioxidant effects. In clinical trials and post-market surveillance, agonists of PPARs have been shown to effectively prevent cardiovascular events. However, adverse effects, particularly for PPARγ agonists, are also observed with the use of investigational PPAR agonists and even some approved drugs. Further exploration of underlying mechanisms is needed to develop novel ways of PPAR activation without causing serious side effects. This article reviews the cardiovascular effects of PPARs, with emphasis on the therapeutic potential of PPAR agonists in combating metabolic vascular diseases.

LINKED ARTICLES

This article is part of a themed section on Chinese Innovation in Cardiovascular Drug Discovery. To view the other articles in this section visit http://dx.doi.org/10.1111/bph.2015.172.issue-23

Abbreviations

Ang II, angiotensin II; EC, endothelial cell; RXR, retinoid X receptor; TCM, traditional Chinese medicine; VSMC, vascular smooth muscle cell

Tables of Links

TARGETS
Nuclear hormone receptors[a]
PPARα, NR1C1
PPARβ/δ, NR1C2
PPARγ, NR1C3
RXR, retinoid X receptor
Enzymes[b]
Akt
Casein kinase 2
Cdk5, cyclin-dependent kinase 5
Endothelial NOS
Glycogen synthase kinase-3
GPCR[c]
5-HT_{2B} receptor

LIGANDS	
15-deoxy-Δ12,14-PGJ_2	GW2331
5-hydroxy-L-tryptophan	Imiglitazar
9-HODE, 9-hydroxyoctadecadienoic acid	LTB_4
Aleglitazar	LY465608
Angiotensin II	Metformin
Bezafibrate	MK-767
Ciprofibrate	PGD_2
Clofibrate	Pioglitazone
Endothelin-1	Prostacyclin
Fenofibrate	Ragaglitazar
Gemfibrozil	Rosiglitazone
GW0742	Troglitazone
GW1516	VEGF

These Tables list key protein targets and ligands in this article which are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY (Pawson *et al.*, 2014) and are permanently archived in the Concise Guide to PHARMACOLOGY 2013/14 ([a,b,c]Alexander *et al.*, 2013a,b,c).

Introduction

The physiological and pathophysiological roles of the nuclear receptors, PPARα, PPARβ/δ and PPARγ, have been much less studied in terms of their effects on the vascular system, compared with their functions in glucose and lipid metabolism. PPARs are expressed in endothelial cells (ECs), vascular smooth muscle cells (VSMCs) and macrophages. Activation of PPARs in the vasculature is mostly anti-inflammatory and antioxidant. It is therefore protective against endothelial dysfunction, atherosclerosis and vascular remodelling *in vivo*. The ongoing clinical use or trials of synthetic PPAR agonists support the predicted vascular benefits of PPAR activation although some adverse effects have been reported. This review will focus on how PPARα, PPARβ/δ and PPARγ regulate vascular function through different targets in ECs, VSMCs and macrophages. In particular, we wish to provide an overview of the clinically relevant aspects and controversies of the use of PPAR agonists for the treatment of cardiovascular complications associated with metabolic disorders. This information increases the potential therapeutic value of PPAR agonists against vascular dysfunction and justifies further development of novel PPAR agonists.

Transcriptional regulation and physiological functions of PPARs

PPARs heterodimerize with retinoid X receptors (RXR) and bind to specific DNA regions of target genes (AGGTCAXAGGTCA, with X being a random nucleotide) termed as peroxisome proliferator hormone response elements (Gearing *et al.*, 1993). PPAR-RXR heterodimers remain bound with co-repressor protein complex in the absence of ligand but ligand activation triggers conformational changes of PPAR-RXR which releases the co-repressor complex and, subsequently, the heterodimer assembles with co-activators to activate the transcription of target genes (DiRenzo *et al.*, 1997). Furthermore, activated PPARs can interact with other transcription factors, such as AP-1 and NF-κB, resulting in transcriptional repression (Delerive *et al.*, 1999). These transcriptional regulation and key functions of PPARs are summarized in Figure 1.

The endogenous ligands for PPARs are fatty acids and eicosanoids. Both PPARα and PPARδ are activated by a wide variety of saturated and unsaturated fatty acids while PPARγ interacts only with polyunsaturated fatty acids (Xu *et al.*, 1999). Differences also exist between eicosanoids as activators of PPARs. For instance, LTB_4 and 8(S)-hydroxyeicosa-5,8,11,13-tetraenoic acid (8(S)-HETE) are PPARα ligands; 9- and 13-hydroxyoctadecadienoic acids (9- and 13-HODEs), and 15-deoxy-$\Delta^{12,14}$-PGJ_2 are agonists for PPARγ whereas prostacyclin, PGA_1, and PGD_2 are agonists for PPARδ (Forman *et al.*, 1997; Kliewer *et al.*, 1997).

PPARs have different levels of expression in different tissues. PPARα is predominantly expressed in cells with high rates of fatty acid catabolism such as liver, heart, kidney and skeletal muscle (Braissant *et al.*, 1996) to regulate fatty acid oxidation systems. PPARγ is mainly associated with adipose tissue to regulate adipocyte differentiation and thus, lipid and energy storage (Rosen and Spiegelman, 2001). PPARδ is ubiquitously expressed at much higher levels than PPARγ and PPARα, markedly in brain, adipose tissue and skin (Kliewer *et al.*, 1992). It is implicated in fatty acid oxidation and energy dissipation, improving lipid profiles and reducing adiposity (Wang *et al.*, 2003).

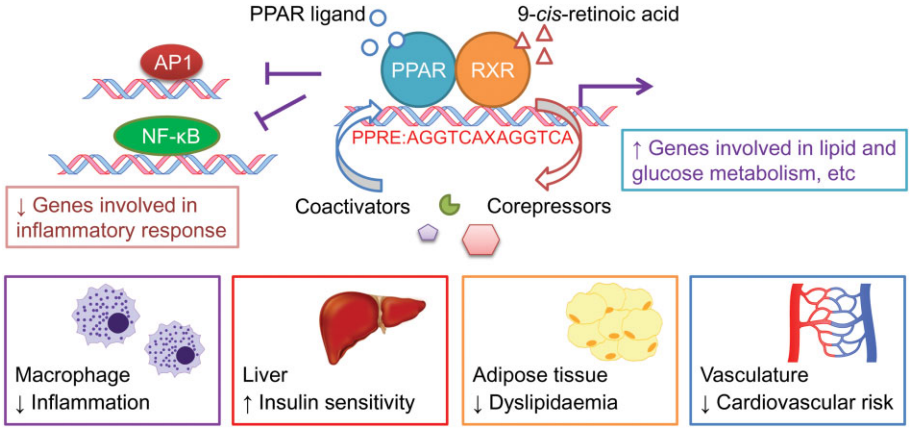

Figure 1

Transcriptional regulation and key functions of PPAR family. Ligand binding to PPAR recruits coactivators in replacement of corepressors to induce transcription and inhibits other transcriptional factors such as AP-1 and NF-κB, regulating different functions.

Cardiovascular effects of PPARα activation and deletion

PPARα activation limits inflammatory responses in ECs and VSMCs (Plutzky, 2011) as well as macrophages (Neve *et al.*, 2001) by inhibiting NF-κB and AP-1 and improves lipid profile, thus contributing to the anti-atherogenic action of PPARα agonists. PPARα inhibits the activities of NF-κB and AP-1 through direct interactions with p65 and c-Jun subunits, respectively (Delerive *et al.*, 1999), or through induction of IκBα which sequesters NF-κB in the cytoplasm (Delerive *et al.*, 2000). PPARα agonists were developed to treat dyslipidaemia. For example, fibric acid derivatives (fibrates), including clofibrate, ciprofibrate, fenofibrate and gemfibrozil, reduce triglyceride and raise high-density lipoprotein (HDL) with modest effect to lower low-density lipoprotein (Remick *et al.*, 2008). Fenofibrate retards the development of atherosclerosis in ApoE$^{-/-}$ and LDLR$^{-/-}$ mice (Duez *et al.*, 2002; Srivastava *et al.*, 2006). However, atherosclerosis develops more slowly in PPARα$^{-/-}$ mice (Tordjman *et al.*, 2007), while another study shows that PPARα deficiency in macrophages increases atherosclerosis (Babaev *et al.*, 2007). Such conflicting data may raise concerns over the relevance of the choice of mouse models used to reveal the roles of molecular targets in the development of atherosclerosis. Using tissue-specific gene deletion, mice are probably better than global knockout mice as more selective activity can be investigated with less effects on the whole body. Deletion of PPARα further increased the plasma level of triglyceride in ApoE$^{-/-}$ mice which might cause increased atherosclerosis (Babaev *et al.*, 2007). Interestingly, fenofibrate exerts some PPARα-independent actions in human microvascular ECs, reducing endothelin-1 expression via inhibition of glycogen synthase kinase-3 (Glineur *et al.*, 2013).

Clinical perspective of PPARα agonists

Several clinical studies evaluated the vascular effects of fibrates. In these trials, gemfibrozil alone reduces cardiovascular events such as coronary heart disease, myocardial infarction and stroke in type 2 diabetic patients without any statin use (Asztalos *et al.*, 2008). By contrast, the more potent PPARα agonist, fenofibrate, shows no overall benefit in reducing macrovascular events in diabetic patients (Group *et al.*, 2010). On the other hand, fenofibrate prevents microvascular complications such as diabetic retinopathy (Chen *et al.*, 2013). This agonist also ameliorates coronary flow velocity reverse and arterial stiffness in patients with hypertriglyceridaemia (Wang *et al.*, 2013). It appears that fenofibrate may preferentially regulate those cellular targets which control for microvascular dysfunction. In particular, many clinical trials have found that fibrates are more effective in patients with high triglyceride and low-HDL cholesterol (Fruchart *et al.*, 1998; Tenkanen *et al.*, 2006), both of which are risk factors for cardiovascular diseases, implying a higher effectiveness to protect vascular function in such patients. Patients with high triglyceride and low-HDL cholesterol have not been the basis of an entire clinical trial, so the vasoprotective effect of fibrates may have been underestimated in the trials so far (Tenkanen *et al.*, 2006; Scott *et al.*, 2009).

Four fibrates are available for clinical application to treat hyperlipidaemia. The US Food and Drug Administration (FDA) approved fenofibrate and gemfibrozil but not ciprofibrate and bezafibrate while the latter two are available elsewhere except in the United States. Treating mice and rats with fibrates increases incidence of hepatocellular carcinogenesis but no significant toxic or carcinogenic effects have been reported in patients (Gonzalez and Shah, 2008). The underlying mechanism attributed to this differential effect among species is currently unclear but the different levels of PPARα expression might play a role. Notably, mouse liver expresses about 10-fold more PPARα than human liver, implying that PPARα signalling might be much stronger in mice and thus recruit more molecular targets to cause more severe toxicity. Furthermore, the hepatocarcinogenicity of fibrates in rodents may be independent of their binding to PPAR. Fibrates are known to affect NADH cytochrome c reductase activity, disrupting mitochondrial electron respiratory chain (Scatena *et al.*, 2003). Because peroxisomal

β-oxidation is more active in rodents than in humans (Perrone *et al.*, 1998), this process may be a factor in the higher level of hepatocarcinogenicity in rodents. Identifying the mechanisms behind adverse drug effects is inherently difficult but new synthetic agonists should be modified to be more targeted, with minimal side effects. Correct dosing is also a critical factor but it is important to achieve the beneficial effects without severe side effects. More potent and specific PPARα agonists including AVE8134, DRF10945, GW590735 and LY518674 have been developed and currently under investigation in phase II trials.

PPARγ linking to cardiovascular complications in diabetes and obesity

Extensive studies support that PPARγ activation by thiazolidinediones (TZDs), such as troglitazone, rosiglitazone and pioglitazone, confers vascular benefits in diabetic animals. Pioglitazone (Huang *et al.*, 2008) and rosiglitazone (Wong *et al.*, 2011) improve endothelial function in diabetic and obese mice by stimulating NO production and lowering oxidative stress. Existing *in vivo* data indicate that PPARγ activation by both pharmacological ligands (Chen *et al.*, 2001) and genetic overexpression (Hu *et al.*, 2010) attenuates the occurrence of atherosclerosis in ApoE$^{-/-}$ mice. PPARγ activation in monocytes/macrophages is anti-inflammatory by reducing inflammatory molecules and by inducing the generation of IL-1 receptor antagonist (Jiang *et al.*, 1998; Meier *et al.*, 2002). VSMC-specific PPARγ knockout aggravates angiotensin II (Ang II)-induced vascular remodelling and impairment of endothelium-dependent relaxations (Marchesi *et al.*, 2012), atherosclerosis (Chang *et al.*, 2012) and vascular calcification (Woldt *et al.*, 2012) in mice, thus revealing a vasoprotective property of PPARγ agonists.

TZDs suppress the vasoconstrictor effects of endothelin-1 (Tian *et al.*, 2010), Ang II (Yuen *et al.*, 2011) and 5-HT$_{2B}$ receptor agonists (Liu *et al.*, 2012), suggesting a promising strategy to delay the development of hypertension and vascular dysfunction. However, contradictory findings on BP regulation are reported in animals with VSMC-selective PPARγ deletion. Hypertension [systolic BP: ~130 vs. 120 mmHg (day) and ~145 vs. 135 mmHg (night) for transgenic vs. control mice] (Halabi *et al.*, 2008) was accompanied by increased RhoA and Rho kinase activity (Pelham *et al.*, 2012). However, in another study, hypotension (systolic: 96 ± 27 vs. 123 ± 14 mmHg and diastolic: 84 ± 15 vs. 102 ± 14 mmHg for knockout vs. control mice in daytime) was described, accompanied with reduced vasoconstriction and increased vasodilatation in aortas (Chang *et al.*, 2009). These contradictory results between nuclear receptor deficiency and agonists might be due to compensation mechanisms and/or the absence of active repression of gene transcription by unliganded nuclear receptors. Apart from these considerations, the discrepancies between findings of genetic deletion and pharmacological activation may be due to the PPAR-independent activities of PPAR ligands, summarized in Table 1. Pioglitazone restores blood flow recovery in ischaemic muscle of diabetic mice via the Akt-VEGF pathway (Biscetti *et al.*, 2009). Troglitazone causes dephosphorylation of endothelial NOS at Ser^{116} to increase endothelial NO (Cho *et al.*, 2004), inhibits ECs proliferation by suppressing casein kinase 2 (Lee *et al.*, 2006b) and triggers apoptosis of VSMCs by activating the Gadd45 and p53 pathway (Okura *et al.*, 2000).

Rosiglitazone increases angiogenesis and functional recovery after focal cerebral ischaemia (Chu *et al.*, 2006) and pioglitazone enhances angiogenesis and blood flow recovery in hind limb ischaemia in diabetic mice through activating endothelial NO synthesis (Huang *et al.*, 2008). Besides, PPARγ activation favours angiogenesis through increasing endothelial progenitor cell (EPC) formation (Tousoulis *et al.*, 2008) and inhibiting EPC apoptosis (Zhang *et al.*, 2013). Of interest, EC-specific PPARγ$^{-/-}$ mice show dyslipidaemia and rosiglitazone therapy is unable to lower serum levels of free fatty acid and triglyceride (Kanda *et al.*, 2009), implying that the PPARγ in ECs plays a significant role in lipid metabolism which may in turn affect vascular functions; and thus endothelial PPARγ could be a therapeutic target.

The promising data on vasoprotective effects of TZDs in animal studies led to several clinical trials in patients. A meta-analysis confirmed the prevention of diabetes-

Table 1

PPAR-independent effects of clinically used PPAR ligands in the vascular system

Agonist	Model	Pathway	Effect	Reference
PPARα				
Fenofibrate	Human microvascular ECs (HMEC-1)	Inhibit glycogen synthase kinase-3	Reduce endothelin-1 expression	Glineur *et al.*, 2013
PPARγ				
Pioglitazone	Streptozotocin-diabetic mice with hind limb ischaemia	Stimulate Akt-VEGF pathway	Improve blood flow recovery in ischaemic muscle	Biscetti *et al.*, 2009
Troglitazone	Bovine aortic endothelial cells	Induce endothelial NOS dephosphorylation at Ser^{116}	Increase NO production	Cho *et al.*, 2004
	HUVECs	Suppress casein kinase 2 activity	Inhibit proliferation of ECs	Lee *et al.*, 2006b
	Rat aortic VSMCs	Activate Gadd45 and p53 pathway	Induce apoptosis of VSMCs	Okura *et al.*, 2000

associated vascular complications by pioglitazone (Lincoff *et al.*, 2007) and another clinical study also shows that pioglitazone promoted endothelial function in non-diabetic patients at high cardiovascular risk (Campia *et al.*, 2006). In the PROactive clinical trial, pioglitazone reduced non-fatal myocardial infarction and stroke, but it did not affect the risk of coronary revascularization while increasing leg revascularization in type 2 diabetic patients (Wilcox *et al.*, 2008). Pioglitazone also improved aortic elasticity and decreased inflammation and disease progression, with minimal safety concerns, in patients with rheumatoid arthritis (Marder *et al.*, 2013). A number of clinical studies now show that TZDs reduce BP independent of their insulin-sensitizing properties, suggesting a direct vascular benefit (Giles and Sander, 2007). In addition, PPARγ agonists inhibit atherosclerosis in diabetic patients (Nissen *et al.*, 2008).

Deciphering the adverse effects of PPARγ agonists

TZDs were launched in the late 1990s, as an effective means of restoring insulin sensitivity in type 2 diabetes. However, troglitazone was removed from the market in 2000 because of liver failure in diabetic patients (Kohlroser *et al.*, 2000). Rosiglitazone and pioglitazone currently remain marketed but their application is seriously questioned as adverse effects have been reported. Meta-analyses from randomized trials and Medicare databases show that rosiglitazone was associated with increased risk of stroke, myocardial infarction, heart failure and death (Nissen and Wolski, 2007; Graham *et al.*, 2010). As a result, rosiglitazone has also been withdrawn from Europe by the European Medicines Agency. However, the FDA removed restrictions in November 2013, based on all available data including the re-adjudicated RECORD trial showing no increase of cardiovascular incidence with rosiglitazone therapy (Mahaffey *et al.*, 2013). Likewise, pioglitazone has been associated with bone fracture, heart failure and bladder cancer (Vallarino *et al.*, 2013). Meanwhile, new selective PPARγ modulators are being synthesized and tested for potential therapeutic efficacy and safety. For example, a PPARγ ligand SR1664 improved insulin sensitivity without causing significant adverse effects on fluid retention or bone formation (Choi *et al.*, 2011). SR1664, unlike classical PPARγ agonists, has a unique mode of binding to PPARγ with high affinity, blocking the Cdk5-mediated phosphorylation of PPARγ but not acting as a transcriptional agonist. This difference in action might explain the fewer side effects of this drug. It may therefore be possible to develop a new class of antidiabetic drug with fewer side effects by specifically targeting the Cdk-mediated phosphorylation of PPARγ. Additionally, the development of tissue-specific agonists may enhance the therapeutic benefits and reduce deleterious effects.

PPARδ in vascular dysfunction

Several PPARδ agonists consistently correct dyslipidaemia and improve insulin and glucose tolerance in animal models of obesity and diabetes (Wang *et al.*, 2003; Lee *et al.*, 2006a). Dyslipidaemia and inflammation in macrophage are common contributors to the development of atherosclerosis. PPARδ agonists favourably modulate lipid metabolism and attenuate inflammation, thus reducing the susceptibility to atherosclerosis in both ApoE$^{-/-}$ and LDLR$^{-/-}$ mice (Barish *et al.*, 2008; Bojic *et al.*, 2014). Conversely, PPARδ$^{-/-}$ bone marrow transplants reduce atherosclerotic lesion area in LDLR$^{-/-}$ mice because of the elevated availability of inflammatory suppressor Bcl-6 (Lee *et al.*, 2003). This discrepancy is probably related to the different actions of PPARδ activation in different cell types.

PPARδ agonists directly induce endothelium-dependent relaxation when used at higher concentrations (Jimenez *et al.*, 2010) and can restore endothelial function in animal models of both type 1 (Quintela *et al.*, 2012) and type 2 (Tian *et al.*, 2012) diabetes through increasing NO bioavailability and suppressing generation of NADPH oxidase-derived superoxide anions. In hypertensive rats, GW0742 prevented the development of endothelial dysfunction and hypertension by enhancing endothelial NOS activity, and expression of antioxidant genes and regulators of G protein-coupled signalling proteins (RGS) 5, limiting NADPH oxidase activity and reducing expressions of proinflammatory and pro-atherogenic genes (Zarzuelo *et al.*, 2011; 2013). A recent study also suggested that PPARδ plays a crucial rule for vasoprotective effect of the commonly used antidiabetic drug metformin in obese mice (Cheang *et al.*, 2014).

Clinical translation of PPARδ agonists

Clinical trials on selected PPARδ agonists have assessed both metabolic and vascular outcomes. No severe side effects have been so far reported, except for GW1516 which induced cancer in several organs in rodents (Gupta *et al.*, 2004; Pollock *et al.*, 2010) and its development has been stopped. MBX-8025, another PPARδ agonist, normalized lipid profiles and C-reactive protein levels in patients with the metabolic syndrome and has completed a phase II clinical trial (Choi *et al.*, 2012). More long-term clinical studies are needed to further evaluate the safety profile before any of these PPARδ ligands can be marketed for clinical treatment of metabolic and vascular complications. To conclude, protective effects of PPARδ agonists against metabolic and vascular diseases are strongly indicated by resulst from animal models but, unlike the other isoforms, none of the PPARδ agonists has been approved for clinical use. Furthermore, PPARδ is ubiquitously expressed and any differential mechanism of PPARδ action in different tissues should be explored for developing new PPARδ agonists with improved efficacy and safety. Developing such agonists with high tissue specificity may reduce adverse effects.

Dual PPAR agonism

Because both fibrates and TZDs exerted favourable effects on lipid and glucose metabolism as well as inhibitory effects on

vascular inflammation, interest has grown to develop and explore the clinical outcomes of dual PPARα/γ agonists. Indeed, dual PPARα/γ agonists such as ragaglitazar (Chakrabarti *et al.*, 2003) and muraglitazar (Mittra *et al.*, 2007) provide better glycaemic control than conventional TZDs. Unfortunately, the development of diverse dual PPARα/γ agonists has not met with the anticipated success. Their development was halted in late-phase trials because of reported side effects such as increased cardiovascular risk (muraglitazar), carcinogenicity (ragaglitazar and MK-767), liver toxicity (imiglitazar) and renal injury (tesaglitazar) (Fievet *et al.*, 2006). The effect of dual PPARα/γ agonists on atherogenesis is now also being debated. For instance, GW2331 (Claudel *et al.*, 2001), LY465608 (Zuckerman *et al.*, 2002) and tesaglitazar (Zadelaar *et al.*, 2006) attenuated atherosclerosis in ApoE$^{-/-}$ mice however, treatment with compound 3q ((*S*)-3-(4-(2-carbazol(phenoxazin)-9-yl-ethoxy) phenyl)-2-ethoxy-propionic acid) increases atherosclerotic plaque formation associated with an increased oxidative stress in these mice (Calkin *et al.*, 2007). The reasons for these disparate findings are unclear, possibly related to the different potency of these drugs to activate PPARα and PPARγ together with differential transcriptional outcomes. For instance, muraglitazar and tesaglitazar preferentially act on PPARγ, possibly accounting for some of the adverse effects. Therefore, a balanced PPARα/γ agonist, aleglitazar, was developed, but disappointingly again, its development is terminated at phase III AleCardio trial because of safety concerns with increased risk for bone fractures, heart failure and gastrointestinal bleeding (Lincoff *et al.*, 2013). This outcome has strongly discouraged the further development of safer dual PPARα/γ ligands. Dual PPARα/γ agonists primarily acting on PPARα may provide another option in the reduction of side effects. On the other hand, dual PPARα/δ agonists are less investigated with GFT505 being shown to favourably modulate lipid and glucose metabolism in patients with metabolic syndrome (Cariou *et al.*, 2011). However, it has only completed a phase II clinical trial, so its safety profile remains to be evaluated.

Pan-PPAR activator

Bezafibrate is a pan-PPAR activator activating all three PPAR subtypes. It has been approved by the FDA for the treatment of hyperlipidaemia, despite the reports, in some countries, of renal and hepatobiliary disorders. With beneficial effects on glucose and insulin tolerance, bezafibrate decreased the incidence of and delayed the onset of type 2 diabetes in patients with impaired fasting glucose concentrations (Tenenbaum *et al.*, 2004) and in obese patients (Tenenbaum *et al.*, 2005a). Moreover, bezafibrate decreased the risk of myocardial infarction and cardiac mortality in patients with metabolic syndrome and with a history of recent myocardial infarction and/or stable angina (Tenenbaum *et al.*, 2005b). The benefits of bezafibrate appear to justify the development of newer pharmacological agents targeting more than one PPAR isoform. Consequently, several pan-PPAR agonists were synthesized. However, the majority of them (DRL 11605, GW-625019, indeglitazar, netoglitazone, sipoglitazar and sodelglitazar) have been terminated because of serious safety concerns.

Traditional Chinese compounds acting as PPAR ligands

Traditional Chinese medicine (TCM) compounds have long been used against metabolic disease and the related cardiovascular complications. TCM is an attractive resource in the design of new PPAR agonists to reduce cardiovascular risks. Screening the TCM database containing more than 30 000 candidates, two TCM compounds, (S)-tryptophan-betaxanthin and berberrubine, have been identified as potential lead compounds targeting more than one PPAR (Chen *et al.*, 2012). By *in silico* identification, two other TCM candidates, 5-hydroxy-L-tryptophan and abrine, were found to bind to PPARγ (Chen and Chen, 2014). In addition, honokiol from TCM Magnolia bark was also *in silico* predicted to bind to the ligand-binding domain of PPARγ and prevent ed hyperglycaemia in diabetic mice (Atanasov *et al.*, 2013). Formononetin, the predominant component of San-ao decoction, which is one of the most popular TCM formulae for asthma, was demonstrated to activate PPARγ (Zhou *et al.*, 2009). Through their action on PPARs, such TCM compounds may prevent inflammatory diseases, hyperglycaemia and cardiovascular diseases, apart from their originally known therapeutic functions. They warrant further investigation as novel therapeutic agents. More importantly, TCM is deployed as many different combinations of compounds to treat human diseases and the investigation of the active components together with mechanisms of action is crucial for drug development. The efficacy and safety of identified compounds can be ensured and improved using modern analytical and chemical techniques.

Conclusion

The results of many animal studies taken together have demonstrated that activation of PPARα, PPARγ and PPARδ with their corresponding agonists produced clear vasoprotective effects, including inhibition of vascular inflammation, oxidative stress and atherogenic development. These findings stimulated intense interest in using PPAR agonists as another therapeutic approach to vascular dysfunction. Disappointingly, a number of drugs targeting PPARs have failed clinical trials because of safety concerns. Only drugs that activate PPARα (ciprofibrate, fenofibrate and gemfibrozil) and PPARγ (pioglitazone and rosiglitazone) and one pan-PPAR agonist (bezafibrate) have reached the market, so far. PPARα agonists and bezafibrate are available for the treatment of hyperlipidaemia associated with metabolic syndrome while PPARγ agonists are used to correct hyperglycaemia in type 2 diabetic patients. Clinical studies reveal that several PPAR agonists have not achieved the expected levels of cardiovascular benefits or that the benefits are significantly offset by unwanted outcomes such as malignancy, liver and renal toxicity and increased cardiovascular risk. The cardiovascular benefits and side effects are summarized in Figure 2. Despite these

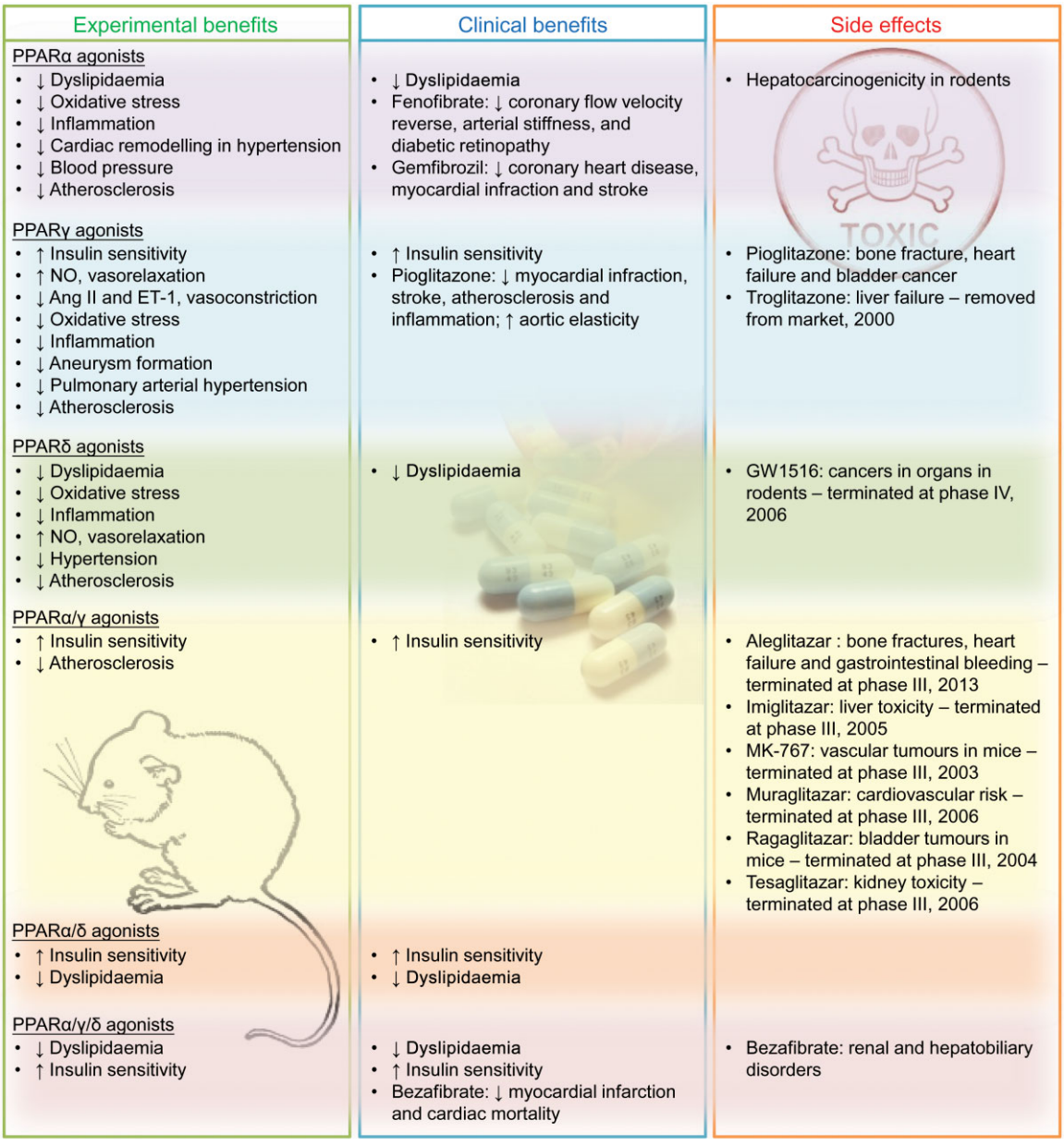

Figure 2

Cardiovascular effects of PPAR agonists in animal and clinical studies. The list shows the cardiovascular benefits shown in rodents (experimental benefits) and in patients with metabolic syndrome (clinical benefits) and the adverse effects of agonists for PPARs.

disadvantages, PPARs are important players in the control of lipid and glucose homeostasis. Therefore, new PPAR ligands are being developed, in the hope that these new drugs will produce fewer side effects. With deeper understanding of the regulatory mechanisms of PPAR activation, for example, co-repressor/co-activators of PPARs, epigenetic modifiers and intermediate metabolites that may activate certain enzymes or induce post-translational modification, new insights could be useful in developing novel or tissue-targeted PPAR agonists with more tolerable side effects for better prevention and management of cardiovascular events associated with metabolic diseases.

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Conflict of interest

None declared.

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