Published in final edited form as:

Immunol Rev. 2012 September ; 249(1): 72–83. doi:10.1111/j.1600-065X.2012.01153.x.

LXR and PPAR as integrators of lipid homeostasis and immunity

Yoko Kidani^{1,2} and Steven J Bensinger^{*,1,2,3}

¹Institute for Molecular Medicine, David Geffen School of Medicine, University of California, Los Angeles, 675 Charles E. Young Drive, Los Angeles, California 90095

²Department of Pathology and Laboratory Medicine, David Geffen School of Medicine, University of California, Los Angeles, 675 Charles E. Young Drive, Los Angeles, California 90095

³Department of Molecular and Medical Pharmacology, David Geffen School of Medicine, University of California, Los Angeles, 675 Charles E. Young Drive, Los Angeles, California 90095

Summary

Lipid metabolism has emerged as an important modulator of innate and adaptive immune cell fate and function. The lipid-activated transcription factors PPAR (PPAR α , β/δ , γ) and LXR are members of the nuclear receptor superfamily that have a well-defined role in regulating lipid homeostasis and metabolic diseases. Accumulated evidence over the last decade indicates that PPAR and LXR signaling also influences multiple facets of inflammation and immunity, thereby providing important crosstalk between metabolism and immune system. Herein, we provide a brief introduction to LXR and PPAR biology and review recent discoveries highlighting the importance of PPAR and LXR signaling in the modulation of normal and pathologic states of immunity. We also examine advances in our mechanistic understanding of how nuclear receptors impact immune system function and homeostasis. Finally, we discuss whether LXRs and PPARs could be pharmacologically manipulated to provide novel therapeutic approaches for modulation of the immune system under pathologic inflammation or in the context of allergic and autoimmune disease.

Keywords

LXR; PPAR; lipids; lupus; immunity

Introduction

Nuclear receptors (NR) are transcription factors that regulate gene expression in response to a broad array of physiological stimuli (1). In humans, forty-eight nuclear receptors have been identified, and their function has been implicated in nearly every biologic process, including development, homeostasis, metabolism, circadian rhythms, endocrine function, reproduction, inflammation and immunity (2-4). The structure of nuclear receptors is highly conserved, composed of an amino-terminal activation domain (AF1), a zinc-finger DNA-

*corresponding author sbensinger@mednet.ucla.edu.

binding domain, a carboxy-terminal domain that binds to ligand, and second activation domain at the c-terminus (AF2) (5, 6).

The NR superfamily can be divided into three distinct subclasses (7). The first group is the prototypic hormone-driven receptors, examples of which include the estrogen receptor, progesterone receptor, glucocorticoid receptor, androgen receptor, and mineralocorticoid receptor. These receptors are cytoplasmic, bound to chaperone proteins, and translocate from cytoplasm to nucleus upon ligand binding. Once in the nucleus, these NRs bind to their respective response elements where they positively or negatively regulate gene transcription. The second class is the orphan receptor family for which the endogenous ligands have not yet been identified. These receptors have ligand binding domains, but it remains unclear if these NRs require ligand binding in the pocket to activate transcription. Indeed, many of these receptors rely on other physiologic stimuli, such as growth factor signaling, for activation. Members of this subclass that are of particular interest to the immunologic community include Nurr77 and ROR γ t. The third group of NRs are composed of ligand-activated transcription factors (also known as the TR/RAR/PPAR/VDR-like receptors) that predominately form obligate heterodimers with the Retinoid X Receptor (RXR). In the absence of ligand, most RXR heterodimers are bound to their respective DNA response elements in association with co-repressors, histone deacetylases and chromatin-modifying factors to maintain target genes in a repressed state. Ligand binding initiates a conformational change in the receptor, the exchange of co-repressors for co-activators, and the initiation of target gene transcription. In addition to their ability to transactivate target genes, several RXR heterodimers have the capacity to repress gene expression in a signal- and gene-specific manner, the details of which will be discussed below. NRs have been extensively discussed in the literature and the aim of this review is to briefly introduce the nuclear receptor superfamily and focus on the function of two members, the Liver X Receptor (LXR α and β) and Peroxisome Proliferator- Activated Receptors (PPAR α , β/δ , γ) in the context of the immune system. For a more detailed review of basic NR biology we direct the readers to an outstanding resource, the Nuclear Receptor Signaling Axis (NURSA) (8).

LXRs: regulators of cholesterol homeostasis

LXR α and LXR β (NR1H3 and NR1H2, respectively) were identified in the mid-1990s based on sequence homology with other nuclear receptors (9-11). LXR α and LXR β proteins have considerable sequence homology (~77% identity in DNA and ligand binding domains) and are activated by the same ligands. The endogenous ligands for LXR are sterol metabolites, including 22(R)-hydroxycholesterol, 24(S)-hydroxycholesterol, 27-hydroxycholesterol and 24(S), 25-epoxycholesterol (12-14). Activation of LXR α or LXR β drives nearly identical transcriptomes, and only the anti-apoptosis gene AIM (also designated Api6/Spa/CD5L) has been validated as an LXR α -specific target. In distinction, there is considerable difference in the tissue distribution of these transcription factors. LXR β is ubiquitously expressed, whereas LXR α expression is restricted to liver, adipose tissue, adrenal glands, intestines, lungs, kidneys, and myeloid origin cells (8), although there are reports of LXR α expression in other immune cell types (15, 16). The mechanisms regulating LXR expression in a tissue specific manner are poorly understood, and it is possible that as

immune cells differentiate into distinct effector or helper subtypes, they alter their LXR expression pattern. This could explain some of the differences reported in the literature regarding the expression pattern of LXR isoforms in immune cells (15, 17).

The clearest function of LXRs is to maintain whole animal sterol homeostasis. To achieve this task, LXRs coordinate gene expression programs in tissue-specific fashion. In tissue macrophages, LXR activation induces the expression of cholesterol efflux transporters ABCA1 and ABCG1, the lipoprotein remodeling enzyme PLTP, and Apo lipoproteins ApoC and ApoE (18). Upregulation of these proteins facilitates the efflux of intracellular sterols onto serum apolipoproteins for return to the liver as HDL in a process termed reverse cholesterol transport. In the liver, LXR activation regulates genes involved in cholesterol processing and excretion including cytochrome P450 7a1 (Cyp7A1), the rate-limiting enzyme for the conversion of cholesterol to bile acid (19). Intestinal LXR regulates the reabsorption of cholesterol through the expression of cholesterol transporters, ABCG5 and ABCG8 (20, 21), to inhibit cholesterol reabsorption and modulate enterohepatic recirculation of cholesterol. In combination, the LXR transcriptome regulates a gene program that preserves whole animal sterol homeostasis that serves a protective function for diet-induced atherosclerosis.

A new wrinkle in our understanding of how LXRs transcriptionally controls lipid homeostasis was added when Tontonoz and colleagues identified the Inducible Degradator of LDLR (IDOL, also known as MYLIP) as a novel LXR target gene that has a critical role in the regulation of circulating lipoprotein levels (22). Mechanistic studies demonstrated that IDOL is an E3 ubiquitin ligase that degrades the LDL and LDL-related receptors. The ability of IDOL to modulate LDL uptake further reinforces the importance of the LXR pathway in preserving cellular lipid homeostasis under conditions of sterol accumulation (18, 22). The intrinsic influence of IDOL on immune cell metabolism and function has yet to be evaluated, and it will be of interest to determine if IDOL helps to explain facets of LXRs immunomodulatory activity (detailed below).

PPARs: regulators of metabolic homeostasis

The PPAR subfamily is comprised of three related proteins, PPAR α , PPAR β/δ , and PPAR γ (also known as NR1C1, NR1C2, and NR1C3, respectively). PPAR α was the first member identified (23) with PPAR β/δ and PPAR γ subsequently discovered based on sequence homology and their influence on adipose tissue biology (24, 25). The endogenous ligands of PPARs appear to be lipids, including unsaturated fatty acids, eicosanoids, components of oxidized low-density lipoproteins (LDL), very low-density lipoproteins (VLDL), and derivatives of linoleic acid (5, 6). Work performed over the past two decades have implicated the PPARs in nearly every facet of fatty acid metabolism. Because of PPARs influence on lipid metabolism and their emerging anti-inflammatory function (detailed briefly below and more extensively reviewed in (26)), the PPARs are of considerable therapeutic interest. Not surprisingly, there is a broad range of synthetic ligands that have been generated and are used extensively in research and clinical arenas.

PPAR α biology

PPAR α has a well-defined role in promoting fatty acid catabolism, gluconeogenesis, ketogenesis, and lipoprotein assembly (27-30). PPAR α was initially cloned from rodent liver studies examining transcriptional changes in response to xenobiotics known to induce peroxisome proliferation (23). PPAR α is highly expressed in metabolic tissues including liver, brown adipose tissue, heart, skeletal muscle, and kidney (31), and can be expressed in an array of immune cells albeit at much lower levels than in metabolic tissues. In the absence of PPAR α , animals are more susceptible to hepatic steatosis and have perturbations in circulating free fatty acids, plasma glucose and circulating ketone bodies (29, 32, 33). This indicates a critical role for PPAR α signaling in the control of metabolic homeostasis. Perhaps not surprisingly, the PPAR α transcriptome includes genes involved in mitochondrial function, fatty acid uptake, β -oxidation, apolipoprotein expression and triglyceride metabolism (34).

PPAR α is the molecular target of the fibrates (e.g., gemfibrozil, clofibrate, and fenofibrate), a class of drugs important in the management of hypertriglyceremia and cardiovascular disease. The molecular action of fibrates is presumably through its effects on fatty acid and lipoprotein metabolism; however, PPAR α can also have significant anti-inflammatory activities that likely contribute to their protective actions in coronary disease. The molecular and cellular mechanisms by which PPAR α exhibits anti-inflammatory and immunomodulatory activity have yet to be fully elucidated, but are clearly multifactorial given the pleiotropic effects of PPAR α on immune cells (some of these are detailed below). Indeed, recent studies have highlighted an important role for the PPAR α target gene CPT1 in the differentiation and function of memory T cells (35). We refer readers to the companion manuscript by Pearce and colleagues for more detailed information.

PPAR β/δ biology

PPAR β/δ was initially cloned from xenopus by Walhli and colleagues and subsequently identified from rodents and human cDNA by sequence homology with PPAR α . PPAR β/δ is ubiquitously expressed and has been implicated in development, reproduction, inflammation, immunity, neoplasia and metabolism (36). Germline ablation of PPAR β/δ influences placentation, gut development, myelination, adiposity, inflammatory responses, and autoimmunity (37-41). Analogous to PPAR α , PPAR β/δ signaling is also involved in the control of energy homeostasis by stimulating genes involved in fatty acid catabolism, mitochondrial respiration, and thermogenesis. In addition, PPAR β/δ has been implicated in the control of cellular proliferation, differentiation, survival, and wound healing (42-44). Endogenous PPAR β/δ ligands are likely fatty acids and highly selective pharmacologic agonists have been developed that are protective from diet-induced obesity and peripheral insulin resistance (39). Similar to other lipid activated NRs, PPAR β/δ can have anti-inflammatory effects (45). Mechanistic studies indicate that unliganded PPAR β/δ and the transcriptional repressor BCL-6 interact, resulting in sequestration of BCL-6 away from the promoters of inflammatory genes. Activation of PPAR β/δ releases BCL-6 resulting in repression of inflammatory genes. PPAR β/δ signaling has also been implicated in multiple facets of innate and adaptive immunity, including phagocytosis, macrophage and T helper

differentiation, and the development of systemic autoimmune disease (some of which are discussed below and (46)).

PPAR γ biology

PPAR γ has two isoforms, PPAR γ 1 and PPAR γ 2, that are splice variants of the PPAR γ gene through an alternative promoter. PPAR γ is expressed in both brown and white adipose tissue, colon, myeloid cells, and placenta (47). More recently, increased expression of PPAR γ has been correlated with T cell differentiation, suggesting that immune tissues that do not express PPAR γ initially can acquire PPAR γ function. The PPAR γ transcriptome regulates genes involved in lipid metabolism, energetics, and adipocyte differentiation, including adipocyte fatty acid binding protein (aP2), phosphoenolpyruvate carboxykinase (PEPCK), lipoprotein lipase (LPL), the uncoupling protein (UCP1), the scavenger receptor CD36 and the nuclear receptor LXR α (6). Germline loss of PPAR γ is embryonic lethal and, similar to PPAR β / δ , PPAR γ appears to be critical for placentation (48). PPAR γ has also been implicated in bone homeostasis (49). An array of naturally-occurring ligands can activate PPAR γ , including unsaturated fatty acids, eicosinoids, and oxidized LDL (5, 6). However, the affinity of the receptor for many of these ligands is fairly low and the relevance of endogenous ligands in PPAR γ function remains unclear. PPAR γ has been identified as the molecular target of thiazolidinediones (TZDs, e.g., Rosiglitazone, Pioglitazone, Troglitazone). This class of drugs was in wide clinical use as insulin sensitizers and has been shown to have broad anti-diabetic effects on liver, adipose tissue, and skeletal muscle (50). The mechanism underlying the insulin-sensitizing effects of TZDs has not been definitively determined. Recently, TZDs have been placed under restricted use or withdrawn from the marketplace due to potential health complications.

Transcriptional regulation of gene expression by LXRs and PPARs

LXR and PPAR influence gene expression through three modes of transcriptional regulation: activation, repression, and transrepression (Fig.1). Both LXR and PPAR are constitutively nuclear and form obligate heterodimers with RXR. The LXR response element (LXRE) contains the hormone response element sequence (AGGTCA) in replicate separated by four nucleotides (DR4). The PPAR response element (PPRE) is a direct repeat of the hormone response element sequence separated by one base pair (DR1). In the unliganded state, these NR heterodimers are bound to their respective response elements where they repress gene expression via interactions with the co-repressor protein Nuclear Receptor Co-Repressor (NCOR) or the Silencing Mediator of Retinoic Acid and Thyroid Hormone Receptor (SMRT) in association with histone deacetylases. In the absence of LXR or PPAR, target gene expression can be modestly increased in a tissue and gene specific manner (also known as derepression). Upon ligand binding, these NRs undergo conformational changes that lead to the release of co-repressors and the recruitment of co-activators to the transcriptional complex resulting in target gene expression. LXR and PPAR also negatively influence gene expression via a process termed transrepression. In contrast to direct repression, transrepression by NRs does not appear to require recognition of DNA, although there is a requirement for TF activation. As such, NR transrepression operates in signal-specific, but sequence-independent manner.

The molecular mechanism(s) underlying transrepression by LXR and PPAR have yet to be fully elucidated and we direct the readers for a more in-depth treatment of this subject to a number of excellent reviews (7, 51). However, advances in our understanding of the transrepression process have been made over the last several years and we highlight a few of these findings. Studies by Glass and colleagues using PPAR γ provided an initial model for understanding how NRs can repress inflammatory gene expression in a sequence-independent manner (52). Under non-inflammatory conditions, NF- κ B target genes (e.g., INOS or CCL2) are held in a repressed state by co-repressor complexes assembled at the promoters. Upon inflammatory signaling (e.g., TLR signals), the co-repressor complexes are ubiquitinated and subsequently degraded by 19S proteasome in an Ubiquitin-Conjugating Enzyme 5 (UBC5)-dependent manner. TLR signaling also activates NF- κ B, resulting in nuclear translocation, binding to inflammatory gene promoters and target gene expression. Activation of PPAR γ during this process blocks NF- κ B-mediated gene expression by maintaining co-repressor occupancy on the promoters of NF- κ B target genes. In this model, binding of ligand to PPAR γ results in conformational changes that allows for SUMOylation at lysine 365 of the ligand-binding domain in an E2 UBC9- and PIAS1-dependent manner. SUMOylated PPAR γ subsequently binds to the co-repressor complex and prevents the degradation of NCOR by the 19S proteasome through an unknown mechanism, thereby preserving the repressed state. Extending on this model, Glass and colleagues demonstrated that LXR-mediated transrepression operates in a parallel pathway involving SUMOylation of LXR by SUMO2 or SUMO3 and HDAC4 as the E3-ligase (53).

In the proposed models of transrepression, how corepressors (e.g., NCOR) are maintained at the promoters of NF- κ B target genes was difficult to understand. However, recent work indicates that SUMOylated LXR inhibits clearance of NCOR in a Coronin 2A-dependent manner (54). Coronin 2A (CORO2A) is a component of co-repressor complexes of previously unknown function. Mechanistic studies indicate that SUMOylated LXR inhibits clearance of NCOR by binding to a SUMO2/SUMO3 interaction motif on CORO2A. This action prevents actin-dependent NCOR exchange, thereby preserving inflammatory genes in a repressed state. Interestingly, they also demonstrate that LXR-mediated transrepression can be inactivated by inflammatory signals that induce calcium/calmodulin-dependent protein kinase II γ (CaMKII γ) and phosphorylation of LXR. Phosphorylation of serine 427 on LXR β results in deSUMOylation by the SUMO protease SENP3 and the release from CORO2A. These results provide an important mechanistic insight into the molecular events underlying the reciprocal relationship between TLR and LXR activity (detailed below). Future studies will be required to address whether this mechanism is operative *in vivo*.

LXRs and PPAR β/δ : mediators of apoptotic cell clearance and SLE

A significant conceptual advance in our understanding of the importance of LXR and PPAR signaling pathways in immunity has come from the realization that these nuclear receptors influence self-tolerance and the development of systemic autoimmune disease. Lipid metabolism has been implicated as an accelerating factor in autoimmunity (55-58); however, a strong mechanistic explanation for these findings has remained elusive. Recently, Castrillo and colleagues demonstrated that mice deficient in LXR α and β spontaneously develop lupus-like disease characterized by the presence of circulating autoantibodies,

immunoglobulin deposition in tissues and immune cell infiltrates resulting in organ immunopathology, such as glomerulonephritis (59).

A key observation in these studies is that LXR-deficient APCs have decreased capacity to engulf apoptotic cells (Fig. 2). This deficiency in phagocytosis was not a generalized effect since LXR-deficient macrophages retain their ability to engulf bacteria and beads. An important mechanistic insight was the revelation that LXR transactivates the Mer receptor tyrosine kinase (MERTK), a phagocytic receptor that has long been implicated in self-tolerance and the pathogenesis of lupus. Importantly, these studies demonstrated that the sterol content of engulfed apoptotic cells, presumably from the plasma membrane, activates a LXR gene program that facilitates efferocytosis and tolerance in APCs. First, activation of LXR increases expression of MERTK, thereby increasing the efficiency of phagocytosis and reinforcing the anti-inflammatory signals that are intrinsic MERTK receptor function (see review by Lemke and colleagues (60) for a more detailed discussion). Second, LXR transactivates a gene program that increases lipid efflux (e.g., ABCA1, ABCG1, ApoE) and decreases lipoprotein uptake (e.g., IDOL), thus avoiding excessive lipid accumulation and ensuing lipotoxicity. Third, LXR modulates inflammatory gene expression through transrepression (detailed above) to preserve a tolerogenic cytokine environment. In this capacity, LXR signaling is a central mediator of an interconnected metabolic and inflammatory network in APCs.

These data also help to explain the intimate link between TLR signaling and LXR activity. While it is widely understood that LXR activity can repress NF- κ B-dependent inflammatory gene expression, it is less well appreciated that TLR signaling can repress LXR as well (detailed above). As to why a reciprocal relationship between TLR and LXR function should exist remains puzzling. However, the observation that LXR serves as a critical transducer of metabolic and inflammatory information in the context of apoptotic cell clearance provides some clarity. Under non-inflammatory conditions, LXR signaling would coordinate phagocytosis with a tolerogenic program in APCs. In contrast, infection and recognition of pathogens by TLRs would inhibit LXR signaling, thereby selectively down-regulating the activity of the MERTK mediated phagocytic program and relieving the “brakes” on the anti-inflammatory function of LXR. In this context, the reciprocal crosstalk between TLRs and LXRs provides host cells with the ability to reinforce immune and tolerogenic programs depending on environmental cues.

Interestingly, Chawla and colleagues have also identified PPAR β/δ as a transcriptional regulator of apoptotic cell clearance and self-tolerance (41). In gene profiling studies, they demonstrated that PPAR β/δ expression is induced and activated in response to phagocytosis of apoptotic cells, presumably by polyunsaturated fatty acid ligands from apoptotic cells. Importantly, PPAR β/δ activity drives the expression of several opsonins that mark apoptotic cells for phagocytic clearance, such as C1q and MGFE8. Genetic ablation of PPAR β/δ resulted in decreased clearance of apoptotic cells by macrophage and the associated reduction in anti-inflammatory gene expression. A consequence of the deficiency in apoptotic cell clearance is that PPAR β/δ null mice develop lupus-like disease characterized by autoantibodies, immunoglobulin deposition in tissues and glomerulonephritis. Interestingly, there appear to be distinct differences in the spectrum of inflammatory and

phagocytic genes regulated by PPAR β/δ and LXR during efferocytosis. Given that the loss of PPAR β/δ or LXR results in lupus-like disease, we conclude that these pathways are operating in a parallel and non-redundant fashion. One caveat to this interpretation is that these studies were performed on animals with germline deficiencies. Because LXRs and PPARs have pleiotropic effects on immune cell biology (discussed below), attributing the loss of tolerance solely to the observed defects in phagocyte function is unlikely. Nevertheless, one clear implication of these studies is that metabolic manipulation may represent a novel therapeutic approach for SLE.

LXR, phagocytosis and neutrophil homeostasis

Extending on these studies, we have recently identified the LXR signaling pathway as an important modulator of peripheral neutrophil homeostasis (61). Estimates of neutrophils production suggest that approximately 1×10^9 neutrophils/kg of body tissue are produced daily under non-inflammatory conditions. With a half-life of approximately 12-24 hours, an equivalent number of neutrophils must be cleared by tissue phagocytes to preserve homeostasis. The molecular network underlying the peripheral clearance of senescent neutrophils remains poorly understood. Ley and colleagues have delineated an elegant model where clearance of senescent neutrophils in peripheral lymphoid tissues plays a critical role in regulating the size of the circulating neutrophil pool through an IL-23/IL-17-dependent axis (62, 63). Based on these findings, and the work of Castrillo and colleagues delineated above, we reasoned that LXR might be involved in this homeostatic feedback loop. Indeed, BrdU labeling and adoptive transfer studies revealed a defect in peripheral neutrophil turnover in LXR-deficient mice. In contrast, systemic administration of LXR ligand GW3965 decreased the number of circulating neutrophils in a receptor-dependent manner. Mechanistic studies indicate that phagocytosis of aged neutrophils activates the LXR pathway resulting in the upregulation of MERTK and concomitant repression of the IL-23/IL-17 signaling axis. Loss of MERTK or LXR activity perturbed senescent neutrophil clearance by phagocytes and enhanced IL-23/IL-17 expression. Given the emerging importance of neutrophil homeostasis in the pathogenesis of rheumatic diseases such as lupus, these studies could provide further molecular insights into the underlying events. In combination, these studies reinforce the notion that metabolic dysregulation of immune cells could be a critical event in the loss of self-tolerance and the development of autoimmune disease.

LXR and immune cell proliferation

Nearly four decades ago, Kandutsch and colleagues made the striking observation that addition of oxysterols (in particular 25-hydroxycholesterol) to polyclonally activated T cells inhibited cell cycle progression (64). The molecular mechanism(s) underlying these observations are still poorly understood; however, a number of recent studies have begun to shed light on the relationship between intracellular sterols and proliferative capacity. Since 25-hydroxycholesterol (25HC) is an endogenous LXR agonist (discussed above), we reasoned that LXR signaling could be mediating the anti-proliferative effect of 25HC. Indeed, we observed that treating primary human and murine T lymphocytes with an array of LXR agonists decreased mitogen-driven proliferation in an LXR-dependent manner (17).

Analysis of cell cycle proteins and DNA content indicated that enforced LXR activation arrested cells in G1 of cell cycle. In contrast, we have found that LXR null T cells have a proliferative advantage over their LXR-sufficient counterparts both in vitro and in vivo. Importantly, these data demonstrate that activation of LXRs by endogenous ligands negatively regulate T cell proliferation in the context of antigen driven clonal expansion and homeostatic proliferation. Valledor and colleagues also recently reported that LXR signaling decreased the expansion of M-CSF stimulated macrophage (65). Work from a number of other laboratories have also concluded that pharmacologic activation of LXR can negatively influence the proliferative capacity of normal and neoplastic cells (66-69). It should be noted that some studies have found that LXR has little effect on proliferation or has been found to positively regulate proliferative capacity. Of particular interest, studies by Worm and colleagues reported that LXR signaling did not influence the proliferation of polyclonally stimulated human and murine B cells in vitro (16). Likewise, studies on neoplastic cells identified a number of lines that are refractory to the anti-proliferative effects of LXR activation (68). Moreover, Tall and colleagues recently reported that LXR signaling increased proliferation of murine hematopoietic stem cells through an ApoE-dependent mechanism (70).

As to “how” and “why” LXR signaling influences proliferative capacity has yet to be fully defined, but appears to be multifactorial. Work from our laboratory demonstrated that TCR signaling quickly represses LXR target gene expression, suggesting that repression of the cholesterol efflux pathway is an important component of optimal T cell growth (17). Given that LXR target genes are involved in cholesterol movement out of the cell, we hypothesize that ectopic activation of LXR serves to drain cholesterol from growing T cells, thereby inhibiting proliferation. Consistent with this notion, genetic deletion of ABCG1 in T cells provides a considerable measure of rescue from the anti-proliferative effects of enforced LXR signaling. Moreover, ABCG1 null T cells are found to have a proliferative advantage over wild type T cells (71). ABCG1 is located within the cell and appears to move sterols between intracellular compartments rather than across the plasma membrane (72). Thus, we conclude that intracellular redistribution of sterols, likely away from critical organelles such as the ER, can induce a G1/S cell cycle arrest. Alternative proposed mechanisms include alterations in fatty acid metabolism through the regulation of the LXR target gene SREBP1c (sterol regulatory element binding protein 1c) and the direct regulation of G1 cell cycle proteins. Notably, studies by Valledor and colleagues found that genetic deletion of most known LXR target genes (e.g., ABCA1, ABCG1, SREBP1c, and APOE) failed to rescue macrophage from the anti-proliferative effects of LXR agonists (65). The authors were able to correlate decreases in G1 associated cell cycle proteins with LXR signaling; however, a direct relationship between LXR activity and repression of cell cycle machinery was not well defined.

Thus, a unifying mechanism as to how LXR influences cell cycle progression remains elusive. We propose a model where acute perturbations in intracellular lipid content influences cell cycle progression and viability via a generalized ER stress response. It has been shown that changes in intracellular lipid content can induce significant ER stress (73). Given the pleiotropic effects of ER stress signals on protein expression, cell viability, growth, and proliferation, it is not difficult to envision this pathway as a potential

explanation. Moreover, the ability of a cell type to handle acute changes in intracellular lipid content, or a culture system that provides some measure of lipid buffering, would be refractory to LXR-mediated effects on proliferation. Future experiments will be required to determine if this model holds true.

LXR, PPAR and CD4 T helper cell differentiation

Another emerging theme is the influence of PPAR and LXR signaling on CD4 T helper cell differentiation (Fig. 3). Upon antigenic stimulation, naïve CD4 T cells initiate a complex developmental program characterized by robust clonal expansion and effector differentiation. The current paradigm in T helper differentiation includes 5 distinct subsets predicated on their ability to produce canonical effector cytokines or suppress immune responses (74, 75). In general, T_H1 CD4 T cells play a critical role in protecting hosts from intracellular pathogens, while T_H2 CD4 T cells are required for protective immunity from extracellular infections. Tregs and T_H17 have been shown to play critical roles in regulating autoimmunity and inflammatory neutrophilic responses to pathogens. T_{FH} cells provide critical B cell help and facilitate antibody production for humoral responses. While these T helper cell subsets are generally thought to be distinct, considerable plasticity in effector function has been observed in a context-specific manner. Given that metabolic programs can influence cellular fate and function in numerous models, perhaps it is not surprising that metabolism is emerging as an impactful regulator of T helper differentiation.

Evidence that PPAR signaling influences T helper cell differentiation and function can be found in a variety of disease models. The observation that administration of TZDs ameliorates the pathogenesis of autoimmunity in murine models suggested that PPAR signaling could influence the development or function of T helper cells. Indeed, a number of studies have demonstrated that PPAR signaling directly and indirectly impacts the differentiation of T helper cells. PPARs have a well-defined role in mediating inflammatory gene expression in APCs (76, 77), thereby indirectly influencing the differentiation of responding CD4 T cells during cognate interactions. Likewise, mechanistic studies indicate that PPARs intrinsically influence T helper differentiation and function. As mentioned above, pharmacologic activation of PPAR γ impairs T cell proliferation through an IL-2 dependent mechanism involving repression of NFAT activity (78, 79). Given the critical importance of proliferation in T helper differentiation, it is not difficult to envision that alterations in cytokine driven proliferation by PPARs would influence T helper cell biology in the context of autoimmune disease or infection.

More recently, PPARs have been shown to intrinsically regulate T_H17 differentiation and T_H17-dependent diseases, such as EAE, IBD, and collagen-induced arthritis (77, 80, 81). Studies by Knolle and colleagues demonstrated that PPAR γ signaling represses the development of T_H17 cells by blocking the induction of ROR γ t in response to TGF- β and IL-6. In a model highly analogous put forth by Glass and colleagues (detailed above), they found that the corepressor SMRT (Silencing Mediator of Retinoid and Thyroid hormone receptors) occupied the ROR γ t promoter. Addition of T_H17 differentiation cytokines TGF- β and IL-6 resulted in the clearance of SMRT from the ROR γ t promoter and T_H17 differentiation. Activation of PPAR γ maintained SMRT at the ROR γ t promoter and

inhibited ROR γ t gene expression. The PPAR γ -mediated repression appears to be highly specific in that the differentiation of other T helper subsets was not observed. As to why PPAR γ should specifically regulate T_H17 inflammatory responses remains unclear at this time. Nevertheless, the observation that PPAR γ null T cells are more aggressive in EAE clearly indicates that physiologic activation of PPAR γ does occur in the context of autoimmunity and serves to moderate pathogenicity of self-reactive T cells.

PPAR α has also emerged as a regulator of autoimmune disease and T helper cell development. Using EAE models, Steinman and colleagues found that ablation of PPAR α in T cells also resulted in more aggressive disease (82). In contrast to the PPAR γ studies described above, they observed that PPAR α deficient T cells were predisposed to a T_H1 response at the expense of T_H2 function. Interestingly, an impact on IL-17 was not observed, suggesting a distinct mechanism from PPAR γ signaling in EAE. Indeed, mechanistic studies revealed that PPAR α modulates NF- κ B and c-Jun function, thereby modulating the production of T_H1-associated pro-inflammatory cytokines. Intriguingly, the authors also demonstrated that T cells from male mice have increased expression of PPAR α when compared to their female counterparts, and show that loss of PPAR α more severely affected EAE disease progression in male mice. These data provide a molecular explanation for the long-standing observation that male mice are protected from induction of T_H1 mediated autoimmune disease.

As introduced above, PPAR β/δ has also been implicated in the pathogenesis of autoimmune diseases and appears to influence T helper cell differentiation. Similar to other studies using PPAR agonist ligands, systemic administration of PPAR β/δ ligands provided a protective effect in EAE models. Conversely, genetic ablation of PPAR β/δ resulted in a more severe clinical course in response to EAE induction, establishing a receptor dependent effect. Phenotypic and histologic studies indicate PPAR β/δ signaling down modulates pathologic damage to CNS tissue and inflammatory gene expression (IFN- γ and T_H17) by immune cell infiltrates. Defining the CD4 T cell intrinsic effects of PPAR β/δ signaling on EAE pathogenesis is complicated by the lack of mouse models with selective ablation of PPAR β/δ in the T cell compartment. Indeed, the anti-inflammatory effects of PPAR β/δ on APC are well established and likely contribute to disease pathogenesis in both gain-and loss-of-function studies. Nevertheless, a combination of bone marrow chimeric studies and *in vitro* reductive approaches provide strong evidence that PPAR β/δ signaling can intrinsically regulate T helper biology by altering proliferative capacity and expression of IFN- γ and T_H17. In contrast to PPAR γ studies, PPAR β/δ does not appear to regulate ROR γ t expression. However, a higher level of the canonical T_H1 transcription factor T-bet was noted in T helper differentiation assays performed with loss-of-function cells. As to why three closely related receptors, such as PPAR α , PPAR β/δ , and PPAR γ , should influence the differentiation of T helper cells in distinct manners remains poorly understood, but might be explained by the availability of distinct PPAR ligands or differences in receptor expression during the course of immune responses.

Recent studies have also demonstrated a role for LXR signaling in the regulation of T_H17 cells and neutrophilic immune responses (83). Similar to the PPAR ligand studies described above, systemic administration of LXR ligands GW3965 and T0901317 can ameliorate the

clinical course of EAE. In contrast, genetic ablation of LXR renders mice more susceptible to EAE induction. The molecular mechanism by which LXR influences T_H17 differentiation and EAE pathogenesis appears to be multifactorial. As delineated above, LXRs can interfere with mitogen driven proliferation, thereby influencing acquisition of T helper programs. Likewise, LXRs have a well-defined role in repressing inflammatory gene expression in APCs, including key T helper differentiation cytokines IL-6 and IL-23. *In vitro* differentiation assays provide evidence for a T cell intrinsic role of LXR signaling in the negative regulation of T_H17 differentiation. In addition to LXRs transrepressive capacity, mechanistic studies by Cui and colleagues demonstrate a novel mechanism where the LXR target gene SREBP1 antagonizes AHR at the IL-17 promoter, thereby reducing positive regulation of T_H17 cells (84). Given the essential role that SREBPs play in cellular lipid homeostasis, these results indicate that significant crosstalk exists between lipid metabolism and T_H17 inflammatory responses. As to why SREBPs would negatively regulate IL-17 expression has yet to be determined; however, an interesting implication of these data is that the dyslipidemia associated with many metabolic diseases may skew the host T helper cell response.

A final perspective on metabolism and therapeutics in the immune system

The cellular and molecular complexity of the immune system presents a substantial hurdle for effective therapeutic intervention or immunomodulation. While this pleiotropy is essential for host defense and self-tolerance, the very nature of these complex interactions makes it unlikely that targeting a single immune pathway will provide a cure. Indeed, most of the effective therapeutic approaches in use are focused on identifying the effector pathway(s) and decreasing immunopathology through blunting of effector function. Using this strategy, one can only hope to ameliorate autoimmune disease pathogenesis rather than cure the individual. Thus, it is clear that alternative therapeutic approaches for immunomodulation are required if we are to effectively reverse the course of autoimmunity in an individual. We and others have provided preliminary evidence that metabolism can be an important modulator of immune cell fate and function. A logical prediction is that pharmacologic manipulation of host metabolism could be used to reprogram deranged cellular and molecular immune responses. Indeed, recent studies demonstrating that pharmacologic or genetic alterations in lipid metabolism and energetics influence the differentiation of T cell subsets serve as important proof-of-concepts.

The ability of PPARs and LXRs to integrate metabolic and inflammatory signaling makes them attractive targets for pharmacologic intervention in metabolic and autoimmune diseases. It is worth noting that a review of the literature indicates that pharmacologic activation of PPAR or LXR has protective effects in nearly every inflammatory and autoimmune disease model. One could simply dismiss these numerous studies as either “off target effects” of the drugs or pharmacologic phenomenology. However, we would argue that these numerous reports support the notion that dysregulation of lipid metabolism is a requirement for development of pathologic inflammation and autoimmunity. Thus, enforcing “normal” metabolic states on the host through pharmacologic manipulation of PPAR or LXR would prevent immune cells from acquiring pathologic effector functions. Moreover, we hypothesize that “resetting” the metabolism of a host through manipulation of

these NRs could halt or reverse the clinical course of autoimmunity. Of course, much future work is required to test this prediction and fully evaluate the therapeutic potential of metabolic manipulation. Nevertheless, we are enthused by the gathering momentum in the field of immunometabolism and optimistic that studies will continue to yield important insights as to how metabolism and immunity are integrated.

References

1. Venteclef N, Jakobsson T, Steffensen KR, Treuter E. Metabolic nuclear receptor signaling and the inflammatory acute phase response. *Trends in endocrinology and metabolism*: TEM. 2011; 22:333–343. [PubMed: 21646028]
2. Evans RM. The steroid and thyroid hormone receptor superfamily. *Science*. 1988; 240:889–895. [PubMed: 3283939]
3. Mangelsdorf DJ, Evans RM. The RXR heterodimers and orphan receptors. *Cell*. 1995; 83:841–850. [PubMed: 8521508]
4. Mangelsdorf DJ, et al. The nuclear receptor superfamily: the second decade. *Cell*. 1995; 83:835–839. [PubMed: 8521507]
5. Giguere V. Orphan nuclear receptors: from gene to function. *Endocrine reviews*. 1999; 20:689–725. [PubMed: 10529899]
6. Castrillo A, Tontonoz P. Nuclear receptors in macrophage biology: at the crossroads of lipid metabolism and inflammation. *Annu Rev Cell Dev Biol*. 2004; 20:455–480. [PubMed: 15473848]
7. Glass CK, Ogawa S. Combinatorial roles of nuclear receptors in inflammation and immunity. *Nature reviews Immunology*. 2006; 6:44–55.
8. NURSA. <http://www.nursa.org>
9. Willy PJ, Umesono K, Ong ES, Evans RM, Heyman RA, Mangelsdorf DJ. LXR, a nuclear receptor that defines a distinct retinoid response pathway. *Genes Dev*. 1995; 9:1033–1045. [PubMed: 7744246]
10. Song C, Kokontis JM, Hiipakka RA, Liao S. Ubiquitous receptor: a receptor that modulates gene activation by retinoic acid and thyroid hormone receptors. *Proceedings of the National Academy of Sciences of the United States of America*. 1994; 91:10809–10813. [PubMed: 7971966]
11. Shinar DM, Endo N, Rutledge SJ, Vogel R, Rodan GA, Schmidt A. NER, a new member of the gene family encoding the human steroid hormone nuclear receptor. *Gene*. 1994; 147:273–276. [PubMed: 7926814]
12. Janowski BA, Willy PJ, Devi TR, Falck JR, Mangelsdorf DJ. An oxysterol signalling pathway mediated by the nuclear receptor LXR alpha. *Nature*. 1996; 383:728–731. [PubMed: 8878485]
13. Lehmann JM, et al. Activation of the nuclear receptor LXR by oxysterols defines a new hormone response pathway. *The Journal of biological chemistry*. 1997; 272:3137–3140. [PubMed: 9013544]
14. Fu X, et al. 27-hydroxycholesterol is an endogenous ligand for liver X receptor in cholesterol-loaded cells. *The Journal of biological chemistry*. 2001; 276:38378–38387. [PubMed: 11504730]
15. Walcher D, et al. LXR activation reduces proinflammatory cytokine expression in human CD4-positive lymphocytes. *Arteriosclerosis, thrombosis, and vascular biology*. 2006; 26:1022–1028.
16. Heine G, et al. Liver X receptors control IgE expression in B cells. *J Immunol*. 2009; 182:5276–5282. [PubMed: 19380774]
17. Bensinger SJ, et al. LXR signaling couples sterol metabolism to proliferation in the acquired immune response. *Cell*. 2008; 134:97–111. [PubMed: 18614014]
18. Calkin AC, Tontonoz P. Transcriptional integration of metabolism by the nuclear sterol-activated receptors LXR and FXR. *Nature reviews Molecular cell biology*. 2012
19. Peet DJ, et al. Cholesterol and bile acid metabolism are impaired in mice lacking the nuclear oxysterol receptor LXR alpha. *Cell*. 1998; 93:693–704. [PubMed: 9630215]
20. Repa JJ, et al. Regulation of absorption and ABC1-mediated efflux of cholesterol by RXR heterodimers. *Science*. 2000; 289:1524–1529. [PubMed: 10968783]

21. Repa JJ, Berge KE, Pomajzl C, Richardson JA, Hobbs H, Mangelsdorf DJ. Regulation of ATP-binding cassette sterol transporters ABCG5 and ABCG8 by the liver X receptors alpha and beta. *The Journal of biological chemistry*. 2002; 277:18793–18800. [PubMed: 11901146]
22. Zelcer N, Hong C, Boyadjian R, Tontonoz P. LXR regulates cholesterol uptake through Idol-dependent ubiquitination of the LDL receptor. *Science*. 2009; 325:100–104. [PubMed: 19520913]
23. Sher T, Yi HF, McBride OW, Gonzalez FJ. cDNA cloning, chromosomal mapping, and functional characterization of the human peroxisome proliferator activated receptor. *Biochemistry*. 1993; 32:5598–5604. [PubMed: 7684926]
24. Kliewer SA, et al. Differential expression and activation of a family of murine peroxisome proliferator-activated receptors. *Proceedings of the National Academy of Sciences of the United States of America*. 1994; 91:7355–7359. [PubMed: 8041794]
25. Tontonoz P, Hu E, Graves RA, Budavari AI, Spiegelman BM. mPPAR gamma 2: tissue-specific regulator of an adipocyte enhancer. *Genes & development*. 1994; 8:1224–1234. [PubMed: 7926726]
26. Bensinger SJ, Tontonoz P. Integration of metabolism and inflammation by lipid-activated nuclear receptors. *Nature*. 2008; 454:470–477. [PubMed: 18650918]
27. Staels B, 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. *The Journal of clinical investigation*. 1995; 95:705–712. [PubMed: 7860752]
28. Vu-Dac N, et al. Fibrates increase human apolipoprotein A-II expression through activation of the peroxisome proliferator-activated receptor. *The Journal of clinical investigation*. 1995; 96:741–750. [PubMed: 7635967]
29. Kersten S, Seydoux J, Peters JM, Gonzalez FJ, Desvergne B, Wahli W. Peroxisome proliferator-activated receptor alpha mediates the adaptive response to fasting. *The Journal of clinical investigation*. 1999; 103:1489–1498. [PubMed: 10359558]
30. Reddy JK, Hashimoto T. Peroxisomal beta-oxidation and peroxisome proliferator-activated receptor alpha: an adaptive metabolic system. *Annual review of nutrition*. 2001; 21:193–230.
31. Desvergne B, Wahli W. Peroxisome proliferator-activated receptors: nuclear control of metabolism. *Endocrine reviews*. 1999; 20:649–688. [PubMed: 10529898]
32. Hashimoto T, Cook WS, Qi C, Yeldandi AV, Reddy JK, Rao MS. Defect in peroxisome proliferator-activated receptor alpha-inducible fatty acid oxidation determines the severity of hepatic steatosis in response to fasting. *The Journal of biological chemistry*. 2000; 275:28918–28928. [PubMed: 10844002]
33. Leone TC, Weinheimer CJ, Kelly DP. A critical role for the peroxisome proliferator-activated receptor alpha (PPARalpha) in the cellular fasting response: the PPARalpha-null mouse as a model of fatty acid oxidation disorders. *Proceedings of the National Academy of Sciences of the United States of America*. 1999; 96:7473–7478. [PubMed: 10377439]
34. Dreyer C, Keller H, Mahfoudi A, Laudet V, Krey G, Wahli W. Positive regulation of the peroxisomal beta-oxidation pathway by fatty acids through activation of peroxisome proliferator-activated receptors (PPAR). *Biology of the cell / under the auspices of the European Cell Biology Organization*. 1993; 77:67–76. [PubMed: 8390886]
35. van der Windt GJ, et al. Mitochondrial respiratory capacity is a critical regulator of CD8+ T cell memory development. *Immunity*. 2012; 36:68–78. [PubMed: 22206904]
36. Michalik L, et al. International Union of Pharmacology. LXI Peroxisome proliferator-activated receptors. *Pharmacological reviews*. 2006; 58:726–741. [PubMed: 17132851]
37. Peters JM, et al. Growth, adipose, brain, and skin alterations resulting from targeted disruption of the mouse peroxisome proliferator-activated receptor beta(delta). *Molecular and cellular biology*. 2000; 20:5119–5128. [PubMed: 10866668]
38. Barak Y, et al. Effects of peroxisome proliferator-activated receptor delta on placental, adiposity, and colorectal cancer. *Proceedings of the National Academy of Sciences of the United States of America*. 2002; 99:303–308. [PubMed: 11756685]
39. Wang YX, et al. Peroxisome-proliferator-activated receptor delta activates fat metabolism to prevent obesity. *Cell*. 2003; 113:159–170. [PubMed: 12705865]

40. Nadra K, et al. Differentiation of trophoblast giant cells and their metabolic functions are dependent on peroxisome proliferator-activated receptor beta/delta. *Molecular and cellular biology*. 2006; 26:3266–3281. [PubMed: 16581799]
41. Mukundan L, et al. PPAR-delta senses and orchestrates clearance of apoptotic cells to promote tolerance. *Nature medicine*. 2009; 15:1266–1272.
42. Tan NS, et al. Critical roles of PPAR beta/delta in keratinocyte response to inflammation. *Genes & development*. 2001; 15:3263–3277. [PubMed: 11751632]
43. Di-Poi N, Tan NS, Michalik L, Wahli W, Desvergne B. Antiapoptotic role of PPARbeta in keratinocytes via transcriptional control of the Akt1 signaling pathway. *Molecular cell*. 2002; 10:721–733. [PubMed: 12419217]
44. Michalik L, Wahli W. Involvement of PPAR nuclear receptors in tissue injury and wound repair. *The Journal of clinical investigation*. 2006; 116:598–606. [PubMed: 16511592]
45. Takata Y, et al. PPARdelta-mediated antiinflammatory mechanisms inhibit angiotensin II-accelerated atherosclerosis. *Proceedings of the National Academy of Sciences of the United States of America*. 2008; 105:4277–4282. [PubMed: 18337495]
46. Chawla A. Control of macrophage activation and function by PPARs. *Circulation research*. 2010; 106:1559–1569. [PubMed: 20508200]
47. Willson TM, Lambert MH, Kliewer SA. Peroxisome proliferator-activated receptor gamma and metabolic disease. *Annual review of biochemistry*. 2001; 70:341–367.
48. Barak Y, et al. PPAR gamma is required for placental, cardiac, and adipose tissue development. *Molecular cell*. 1999; 4:585–595. [PubMed: 10549290]
49. Wahli W. PPAR gamma: ally and foe in bone metabolism. *Cell metabolism*. 2008; 7:188–190. [PubMed: 18316021]
50. Lehmann JM, Moore LB, Smith-Oliver TA, Wilkison WO, Willson TM, Kliewer SA. An antidiabetic thiazolidinedione is a high affinity ligand for peroxisome proliferator-activated receptor gamma (PPAR gamma). *The Journal of biological chemistry*. 1995; 270:12953–12956. [PubMed: 7768881]
51. Glass CK, Saijo K. Nuclear receptor transrepression pathways that regulate inflammation in macrophages and T cells. *Nature reviews Immunology*. 2010; 10:365–376.
52. Pascual G, et al. A SUMOylation-dependent pathway mediates transrepression of inflammatory response genes by PPAR-gamma. *Nature*. 2005; 437:759–763. [PubMed: 16127449]
53. Ghisletti S, et al. Parallel SUMOylation-dependent pathways mediate gene- and signal-specific transrepression by LXRs and PPARgamma. *Mol Cell*. 2007; 25:57–70. [PubMed: 17218271]
54. Huang W, et al. Coronin 2A mediates actin-dependent de-repression of inflammatory response genes. *Nature*. 2011; 470:414–418. [PubMed: 21331046]
55. Aprahamian T, et al. Impaired clearance of apoptotic cells promotes synergy between atherogenesis and autoimmune disease. *J Exp Med*. 2004; 199:1121–1131. [PubMed: 15096538]
56. Stanic AK, et al. Immune dysregulation accelerates atherosclerosis and modulates plaque composition in systemic lupus erythematosus. *Proc Natl Acad Sci U S A*. 2006; 103:7018–7023. [PubMed: 16636270]
57. Gautier EL, et al. Enhanced immune system activation and arterial inflammation accelerates atherosclerosis in lupus-prone mice. *Arterioscler Thromb Vasc Biol*. 2007; 27:1625–1631. [PubMed: 17446440]
58. Feng X, et al. ApoE^{-/-}Fas^{-/-} C57BL/6 mice: a novel murine model simultaneously exhibits lupus nephritis, atherosclerosis, and osteopenia. *J Lipid Res*. 2007; 48:794–805. [PubMed: 17259598]
59. A-Gonzalez N, et al. Apoptotic cells promote their own clearance and immune tolerance through activation of the nuclear receptor LXR. *Immunity*. 2009; 31:245–258. [PubMed: 19646905]
60. Lemke G, Rothlin CV. Immunobiology of the TAM receptors. *Nature reviews Immunology*. 2008; 8:327–336.
61. Hong C, et al. Coordinate regulation of neutrophil homeostasis by liver X receptors in mice. *The Journal of clinical investigation*. 2012; 122:337–347. [PubMed: 22156197]
62. Ley K, Smith E, Stark MA. IL-17A-producing neutrophil-regulatory Tn lymphocytes. *Immunologic research*. 2006; 34:229–242. [PubMed: 16891673]

63. Smith E, et al. IL-23 is required for neutrophil homeostasis in normal and neutrophilic mice. *J Immunol.* 2007; 179:8274–8279. [PubMed: 18056371]
64. Chen HW, Heiniger HJ, Kandutsch AA. Relationship between sterol synthesis and DNA synthesis in phytohemagglutinin-stimulated mouse lymphocytes. *Proc Natl Acad Sci U S A.* 1975; 72:1950–1954. [PubMed: 1057774]
65. Valledor AF, Hsu LC, Ogawa S, Sawka-Verhelle D, Karin M, Glass CK. Activation of liver X receptors and retinoid X receptors prevents bacterial-induced macrophage apoptosis. *Proc Natl Acad Sci U S A.* 2004; 101:17813–17818. [PubMed: 15601766]
66. Geyeregger R, et al. Liver X receptors regulate dendritic cell phenotype and function through blocked induction of the actin-bundling protein fascin. *Blood.* 2007; 109:4288–4295. [PubMed: 17255360]
67. Blaschke F, et al. Liver X receptor agonists suppress vascular smooth muscle cell proliferation and inhibit neointima formation in balloon-injured rat carotid arteries. *Circulation research.* 2004; 95:e110–123. [PubMed: 15539633]
68. Kim KH, Lee GY, Kim JI, Ham M, Won Lee J, Kim JB. Inhibitory effect of LXR activation on cell proliferation and cell cycle progression through lipogenic activity. *Journal of lipid research.* 2010; 51:3425–3433. [PubMed: 20847297]
69. Vedin LL, Lewandowski SA, Parini P, Gustafsson JA, Steffensen KR. The oxysterol receptor LXR inhibits proliferation of human breast cancer cells. *Carcinogenesis.* 2009; 30:575–579. [PubMed: 19168586]
70. Murphy AJ, et al. ApoE regulates hematopoietic stem cell proliferation, monocytosis, and monocyte accumulation in atherosclerotic lesions in mice. *The Journal of clinical investigation.* 2011; 121:4138–4149. [PubMed: 21968112]
71. Armstrong AJ, Gebre AK, Parks JS, Hedrick CC. ATP-binding cassette transporter G1 negatively regulates thymocyte and peripheral lymphocyte proliferation. *J Immunol.* 2010; 184:173–183. [PubMed: 19949102]
72. Tarling EJ, Edwards PA. ATP binding cassette transporter G1 (ABCG1) is an intracellular sterol transporter. *Proceedings of the National Academy of Sciences of the United States of America.* 2011; 108:19719–19724. [PubMed: 22095132]
73. Tabas I. The role of endoplasmic reticulum stress in the progression of atherosclerosis. *Circulation research.* 2010; 107:839–850. [PubMed: 20884885]
74. Reiner SL. Development in motion: helper T cells at work. *Cell.* 2007; 129:33–36. [PubMed: 17418783]
75. Fazilleau N, Mark L, McHeyzer-Williams LJ, McHeyzer-Williams MG. Follicular helper T cells: lineage and location. *Immunity.* 2009; 30:324–335. [PubMed: 19303387]
76. Jiang C, Ting AT, Seed B. PPAR-gamma agonists inhibit production of monocyte inflammatory cytokines. *Nature.* 1998; 391:82–86. [PubMed: 9422509]
77. Straus DS, Glass CK. Anti-inflammatory actions of PPAR ligands: new insights on cellular and molecular mechanisms. *Trends in immunology.* 2007; 28:551–558. [PubMed: 17981503]
78. Clark RB, Bishop-Bailey D, Estrada-Hernandez T, Hla T, Puddington L, Padula SJ. The nuclear receptor PPAR gamma and immunoregulation: PPAR gamma mediates inhibition of helper T cell responses. *Journal of immunology.* 2000; 164:1364–1371.
79. Yang XY, et al. Activation of human T lymphocytes is inhibited by peroxisome proliferator-activated receptor gamma (PPARgamma) agonists. PPARgamma co-association with transcription factor NFAT. *The Journal of biological chemistry.* 2000; 275:4541–4544. [PubMed: 10671476]
80. Klotz L, et al. The nuclear receptor PPAR gamma selectively inhibits Th17 differentiation in a T cell-intrinsic fashion and suppresses CNS autoimmunity. *The Journal of experimental medicine.* 2009; 206:2079–2089. [PubMed: 19737866]
81. Bassaganya-Riera J, et al. Activation of PPAR gamma and delta by conjugated linoleic acid mediates protection from experimental inflammatory bowel disease. *Gastroenterology.* 2004; 127:777–791. [PubMed: 15362034]
82. Dunn SE, et al. Peroxisome proliferator-activated receptor (PPAR)alpha expression in T cells mediates gender differences in development of T cell-mediated autoimmunity. *The Journal of experimental medicine.* 2007; 204:321–330. [PubMed: 17261635]

83. Korf H, et al. Liver X receptors contribute to the protective immune response against *Mycobacterium tuberculosis* in mice. *J Clin Invest*. 2009; 119:1626–1637. [PubMed: 19436111]
84. Cui G, et al. Liver X receptor (LXR) mediates negative regulation of mouse and human Th17 differentiation. *The Journal of clinical investigation*. 2011; 121:658–670. [PubMed: 21266776]

A. Repression

B. Activation

C. Transrepression

Figure 1. Mechanism of transcriptional control by LXR and PPAR

(A) Repression: In the unliganded state, LXR/RXR and PPAR/RXR heterodimers are bound to DNA response elements in association with co-repressor complexes resulting in repression of target genes. **(B) Activation:** Ligand binding to LXR or PPAR induces conformational changes leading to release of co-repressor complexes and recruitment of co-activator complexes and transcription of target genes. **(C) Transrepression:** Activation of PPAR or LXR represses inflammatory gene expression by maintaining co-repressors on the promoters of NF- κ B target genes. In quiescent cells, inflammatory genes are held in a repressed state by co-repressor complexes. Upon TLR signaling, co-repressor complexes are ubiquitinated and subsequently degraded by 19S proteasome in an Ubiquitin-Conjugating Enzyme 5 (UBC5)-dependent manner. TLR signaling also activates NF- κ B, resulting in nuclear translocation and binding to inflammatory gene promoters in association with co-activators resulting in target gene expression. Activation of PPAR γ or LXR preserves corepressors on the promoters of NF- κ B target genes thereby preventing inflammatory gene expression.

Figure 2. LXR and PPAR β/δ drive a program facilitating the clearance of apoptotic cells and maintenance of self-tolerance
 LXR and PPAR β/δ play critical roles in coordinating the efficient clearance of apoptotic cells and preserving a tolerogenic program in APCs. Lipids (sterols and fatty acids) from engulfed apoptotic cells activate LXRs and PPAR β/δ in phagocytes resulting in the transactivation of genes involved in receptor-mediated phagocytosis. LXR transactivates the phagocytosis receptor MERTK, whereas PPAR β/δ drives the expression of opsonins (e.g., C1q, MGFE8) coat apoptotic cells to facilitate phagocytosis. Activation of these NRs also results in the transrepression of proinflammatory genes as detailed in the text. Genetic ablation of these NRs results in the accumulation of cellular debris in vivo and the loss of self-tolerance.

Figure 3. The influence of PPAR and LXR on T helper differentiation

LXR and PPAR impacts T helper differentiation through indirect and direct mechanisms. Activation of LXR and PPAR can inhibit inflammatory cytokine expression from APCs thereby influencing T helper cell differentiation. Likewise, activation of NRs can directly influence T helper differentiation through distinct mechanisms (detailed in the text). Activation of PPAR γ , PPAR β/δ and LXR negatively influence T_H17 differentiation via the regulation of ROR γ t, AHR, and expression of IL-17. PPAR α and PPAR β/δ skew the balance of T_H1 and T_H2 cells. PPAR α and PPAR γ agonists have also been shown to activate Foxp3 transcription and the differentiation of Tregs. To date, the importance of these NRs in T follicular cells has not been addressed.