Trends in Immunology

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Opinion

Primary cilia and their effects on immune cell functions and metabolism: a model

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Primary cilia are hair-like protrusions of the plasma membrane that function as cellular antennae and are present on most cells in the human body. Primary cilia dysfunction leads to severe diseases, commonly termed 'ciliopathies'. A significant symptom of certain ciliopathies is obesity, and current research aims to identify contributing mechanisms of obesity development in these patients. Western lifestyle-associated factors can trigger chronic inflammation, or metaflammation, which can also attribute to obesity-associated metabolic disorders. However, obese individuals can also be 'metabolically healthy', as discussed for a subset of patients with obesity and ciliopathy. Here, we propose that primary cilia signaling might modulate specific immune cell phenotypes, behaviors, and functions, which might impact inflammatory responses in the context of ciliopathies and beyond.

Highlights

A Western lifestyle triggers a subclinical immune response, also called metaflammation in humans and mice. Metaflammation is now thought to underlie the development of certain noncommunicable diseases associated with obesity.

One of the major symptoms of the Bardet–Biedl Syndrome (BBS) ciliopathy in humans and mice is obesity; however, depending on the BBS mutation and age, the severity of metabolic syndrome varies.

Primary cilia evoke cell-autonomous (in cytotoxic $CD8^+$ T cells) and noncell-autonomous responses (in B lymphocytes and macrophages) that can control certain immune cell functions in humans and mice.

Primary cilia dysfunction leading to ciliopathies can evoke tissue-specific responses in immune cells in humans and mice.

Primary cilia: sensory organelles bridging metabolic and immune cell functions?

Primary cilia (see Glossary) are membrane protrusions located at the surface of almost all vertebrate cells. They are cellular antennae that receive information from the environment and locally transmit this information into a cellular response [1,2]. **Bardet–Biedl syndrome** (*BBS*) genes encode proteins that are part of, or are connected to, a larger protein complex called the **BBSome** (Box 1) [3]. The BBSome is an important regulator of the ciliary protein composition because it enables the export of **G-protein-coupled receptors** (GPCRs) out of the cilium [4,5]. A precisely controlled ciliary protein composition is crucial for the cilia sensing function and the control of cellular signaling and function (Figure 1).

Mutations in genes encoding ciliary proteins can lead to **ciliopathies** (Box 2). As of 2019, at least 190 ciliopathy-associated genes had been identified [6], encoding proteins that control primary cilia structure or function. Among those ciliopathy genes are the *BBS* genes. However, how mutations in *BBS* genes affect cellular functions is not well understood. Therefore, studying primary cilia signaling and function in different cell types is necessary to better understand the pathomechanisms underlying ciliopathies (see Clinician's corner). Moreover, because some ciliopathies can cause metabolic disorders, such as obesity, deciphering the molecular details of cilia function is also relevant to uncover disease mechanisms in syndromic obesity.

According to the WHO, obesity has tripled since 1975 and was recently labeled a pandemic[i]. Obesity is a significant risk factor for noncommunicable diseases (NCDs), such as cardiovascular diseases and metabolic disorders (i.e., type 2 diabetes mellitus; T2DM) (Figure 2A). Obesity is caused by an imbalance between energy uptake and expenditure. These lifestyle-associated factors (including a Western-type diet) can trigger a subclinical, chronic inflammatory response termed **metaflammation**, which is evoked by dysfunction of metabolic cells that generate a local, proinflammatory environment [7–10]. Metaflammation is now thought to underlie the development of such obesity-associated NCDs (Figure 2B) [8].

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Box 1. The BBSome and BBS proteins

The BBSome is a protein complex that controls ciliary protein composition [5].

The BBSome comprises eight BBS proteins (BBS1, BBS2, BBS4, BBS5, BBS7, BBS8, BBS9, and BBS18) [67,68].

So far, 26 *BBS* genes have been identified that are mutated in patients with BBS, all of which encode proteins that are involved in ciliary signaling and function [13].

However, obesity does not necessarily result in immune and metabolic dysfunction. Up to 30% of obese patients are 'metabolically healthy obese' because they display normal fasting blood glucose concentrations, **normotension**, and high insulin sensitivity, and do not exhibit pronounced systemic inflammation [11]. A striking example of metabolically healthy obese individuals includes patients with BBS, who carry a mutation in one of the *BBS* genes and are obese [12,13]. Some patients with BBS have been reported to display normal glucose tolerance [14] and, despite greater visceral adiposity, have shown the same prevalence for developing T2DM as body mass index (BMI)-matched control patients [15]. However, other reports indicate a worse metabolic outcome in patients with BBS compared with BMI-matched control patients [16]. We discuss these controversial findings in the following sections and also consider experimental evidence from mouse models.

Given that metaflammation has been identified as a common cause underlying the development of metabolic dysfunction during obesity, one might wonder whether immune cell function is altered in patients with BBS. Although primary cilia are present on most cells in the human body, they appear to be absent in hematopoietic cells [17]. In fact, so far, only one group has shown the presence of primary cilia on human hematopoietic cells [18]. Thus, we propose that primary cilia on non-immune cells might indirectly or directly impact nonciliated immune cell behaviors and functions [14,19].

From another angle, cytotoxic $CD8^+$ T cells display the so-called 'frustrated' cilium during immunological synapse formation, which relies on ciliary signaling components [20]. However, their role in metaflammation remains to be rigorously studied.

In this opinion article, we propose a model whereby primary cilia might act as either a 'friend' or a 'foe' in modulating immune cell behaviors and functions. We first review the already-established role of ciliary proteins in immunological synapse formation. We then argue that primary cilia might impact non-immune and immune cell crosstalk in a tissue-specific manner in white adipose tissue (WAT), liver, and kidney, with a focus on macrophages and B cell development in the bone marrow and spleen.

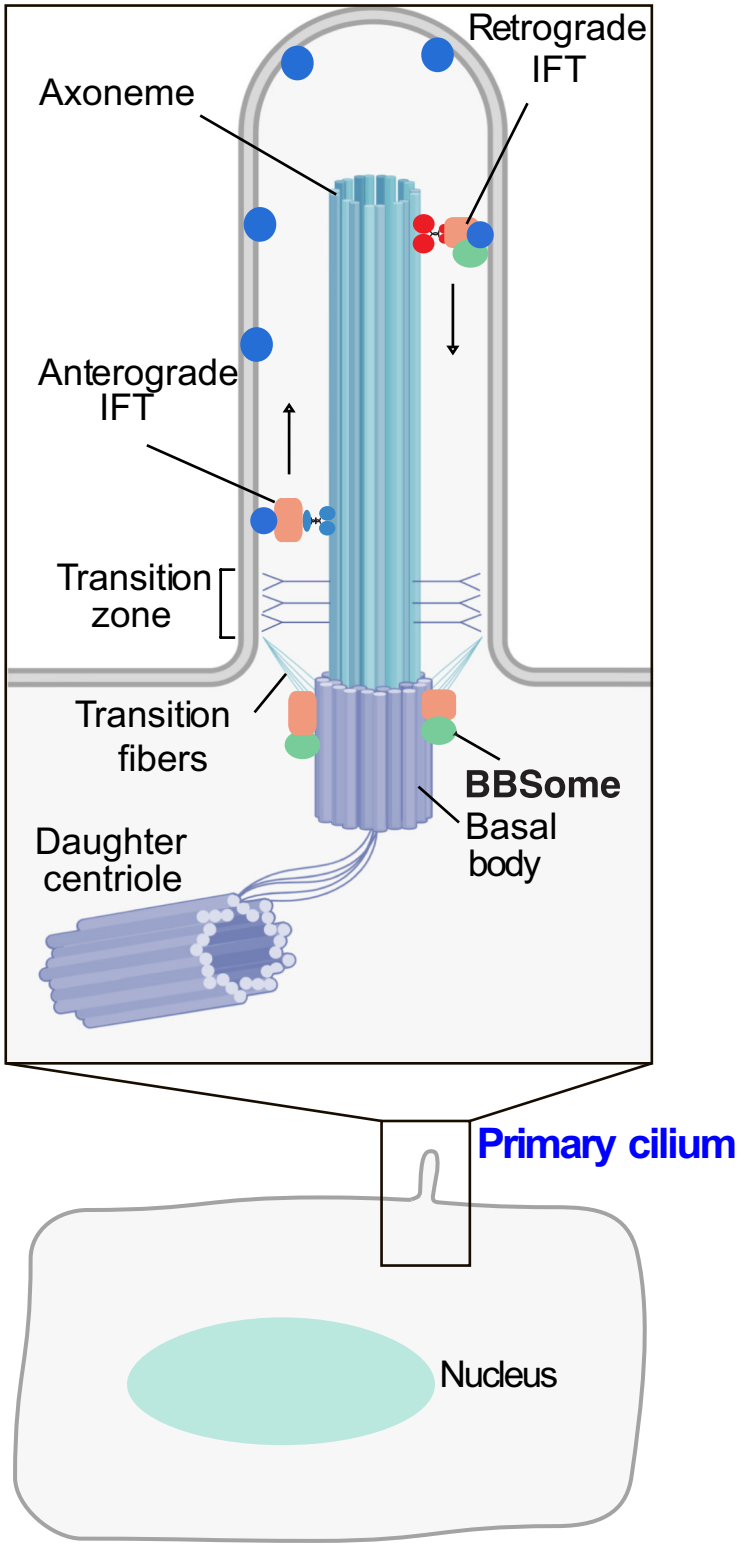
Primary cilia signaling in immune cells

Cell-autonomous role of primary cilia signaling in T cells

The first direct connection between primary cilia and immune cells was reported to occur via the centrosome [21]. The centrosome constitutes a hybrid organelle, which serves as a plasma membrane-associated primary cilium organizer and a juxtanuclear microtubule-organizing center [22]. The centrosome is essential for $CD8^+$ cytotoxic T lymphocyte (CTL) function; it directs the secretion of cytotoxic granules at the **immunological synapse** (IS) and, in turn, allows CTLs to destroy virally infected or tumorigenic cells [21]. However, the exact sequence of events underlying microtubule polymerization, IS formation, and the transport and release of lytic granules has been controversial [23]; nevertheless, the centrosome was recently demonstrated to be crucial for efficient CTL-mediated killing in primary murine CTLs [24–26], despite being dispensable for lytic granule release from CTLs or for CTL killing efficiency in human CTLs [23]. Centrosomes are required for efficient granule packaging and IS organization [24]. They also rely on Arp2/3-

Glossary

Adipokines: signaling proteins secreted by adipocytes.
Atopic dermatitis (AD): chronic skin inflammation.
Autosomal dominant polycystic kidney disease (ADPKD): mutation in one allele of *PKD1* or *PKD2* causes cyst development in the kidney.
Bardet-Biedl syndrome (BBS): develops due to mutations in one of 26 different *BBS* genes that cause primary cilia dysfunction.
BBSome: protein complex at the base of the primary cilium, comprising eight BBS proteins that control protein localization in the primary cilium.
Ciliary signaling: signaling in primary cilia.
Ciliopathies: diseases that develop due to primary cilia dysfunction.
G-protein-coupled receptors (GPCRs): transmembrane proteins that sense external stimuli and transduce this information via G proteins and second messenger signaling into a cellular response.
Immunological synapse (IS): formed between an antigen-presenting cell and a lymphocyte; here, we refer to the IS in cytotoxic $CD8^+$ T cells.
Intraflagellar transport machinery: transport machinery that moves proteins in and out of the cilium along microtubule tracks.
Metaflammation: a low-grade, chronic inflammation that can result in metabolic dysfunction.
Normotension: normal blood pressure.
Primary cilia: hair-like, solitary membrane protrusions found on most vertebrate cells; function as cellular antennae.
Recessive cystic kidney disease nephronophthisis: mutation in both alleles of an NPHP gene causes cyst development in the kidney.
Regulatory T cells: $CD4^+$ T cell subset known to suppress the immune response.
Severe biliary hyperplasia: increase in the bile duct size and dilated bile duct lumen.



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Box 2. Human ciliopathies

Ciliopathies are caused by primary cilia dysfunction [69] and are due to mutations in single genes [69].

The most prominent ciliopathy is autosomal polycystic kidney disease (ADPKD), which occurs with a prevalence of 1:1000 in Europe and the USA [70].

Other ciliopathies include BBS, Joubert syndrome, Meckel syndrome, NPHP, and retinitis pigmentosa [69].

Primary cilia dysfunction affects nearly all tissues and organs, resulting in a variety of different symptoms, including renal and liver cysts, retinal dystrophy, mental retardation, polydactyly, and obesity [71].

dependent actin nucleation to detach the centrosome from the nucleus, polarize it to the plasma membrane, and form the IS, as evidenced from murine lymphocytes using knockdown and pharmacological approaches [27].

Centrosome docking at the plasma membrane occurs during primary cilia and IS formation [21]. However, lymphocytes do not morphologically form primary cilia [28]. Nevertheless, the organization of signaling modules, such as the **intraflagellar transport machinery** (IFT), in prototypic primary cilia from mammals and the IS in CTL from mice and humans, appear to be conserved, at least partially [28,29]. First, protein transport to the IS involves the IFT machinery [29] and is key for ciliary protein transport [29]. Second, canonical Hedgehog (Hh) signaling in vertebrates relies on primary cilia [30], although compelling experimental evidence has also demonstrated a role for Hh signaling in controlling murine and human T cell functions [28]. In canonical Hh signaling, the Hh ligand binds to the receptor Patched (Ptch1) in the ciliary membrane, whereby Smoothened (Smo) enters the cilium and, in turn, activates Gli transcription factors that drive Gli-mediated transcription in vertebrates [31]. In primary murine CTLs, T cell receptor (TCR) activation not only causes IS formation and cytotoxic granule transport to the IS for secretory release, but also activates Hh signaling [28]; this activates Rac1 signaling and, in turn, controls actin dynamics, which are required for centrosome polarization and cytotoxic granule release [28]. Thus, Hh signaling 'pre-arms' CTLs to allow a rapid killing response when encountering a target. However, in contrast to the primary cilium, the interaction of the ligand with Ptch1 in murine CTLs occurs intracellularly, which promotes the accumulation of Smo at the IS [28]. These findings have created the term 'frustrated cilium' in the IS, on the one hand highlighting the commonalities of the primary cilium and IS in terms of signaling components (e.g., IFT or Hh signaling components), but, on the other hand, also alluding to differences in these structures, because the IS does not morphologically assemble a cilium [20] (Figure 3A, Key figure).

Based on these identified roles of primary cilia signaling components in controlling IS formation in CTLs, a question remained on whether lymphocyte function might also be affected in patients with ciliopathies. Recent reports have begun to partly address this question by demonstrating that canonical Hh signaling, which usually relies on a primary cilium, is also an important determinant of γδ T cell effector differentiation: activation of Hh signaling promotes murine fetal γδ T cell production *in vitro,* as well as the development and increased numbers of γδ T cells within the adult murine thymus [32]. Furthermore, Hh signaling can modulate the severity of **atopic dermatitis (AD)** in mice: specifically, a report indicated that Hh signaling was activated in skin $CD4^+$ and $CD8^+$

Figure 1. The primary cilium. Primary cilia in vertebrates elongate from the basal body, which originates from the mother centriole. The cytoskeletal core structure of the primary cilium is the axoneme, which comprises microtubules. The intraflagellar transport (IFT) machinery provides protein transport to the ciliary tip (anterograde) or to the ciliary base (retrograde) along the axoneme. Protein diffusion into the cilium is prevented by the transition zone, formed by transition fibers, at the ciliary base. Together with the transition zone and the IFT, the BBSome regulates the ciliary protein content [1–3]. Figure partially created using BioRender (https://biorender.com/).

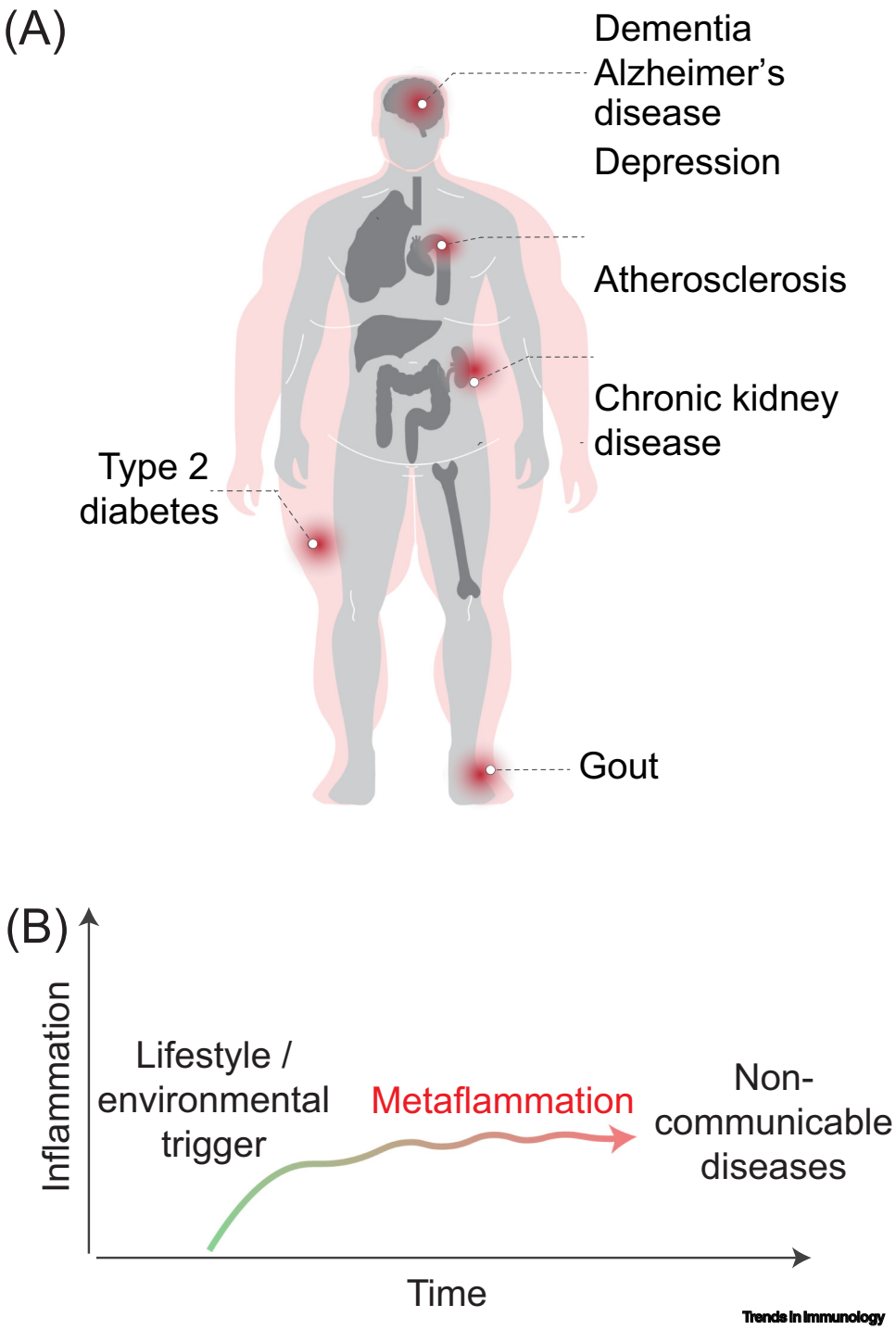
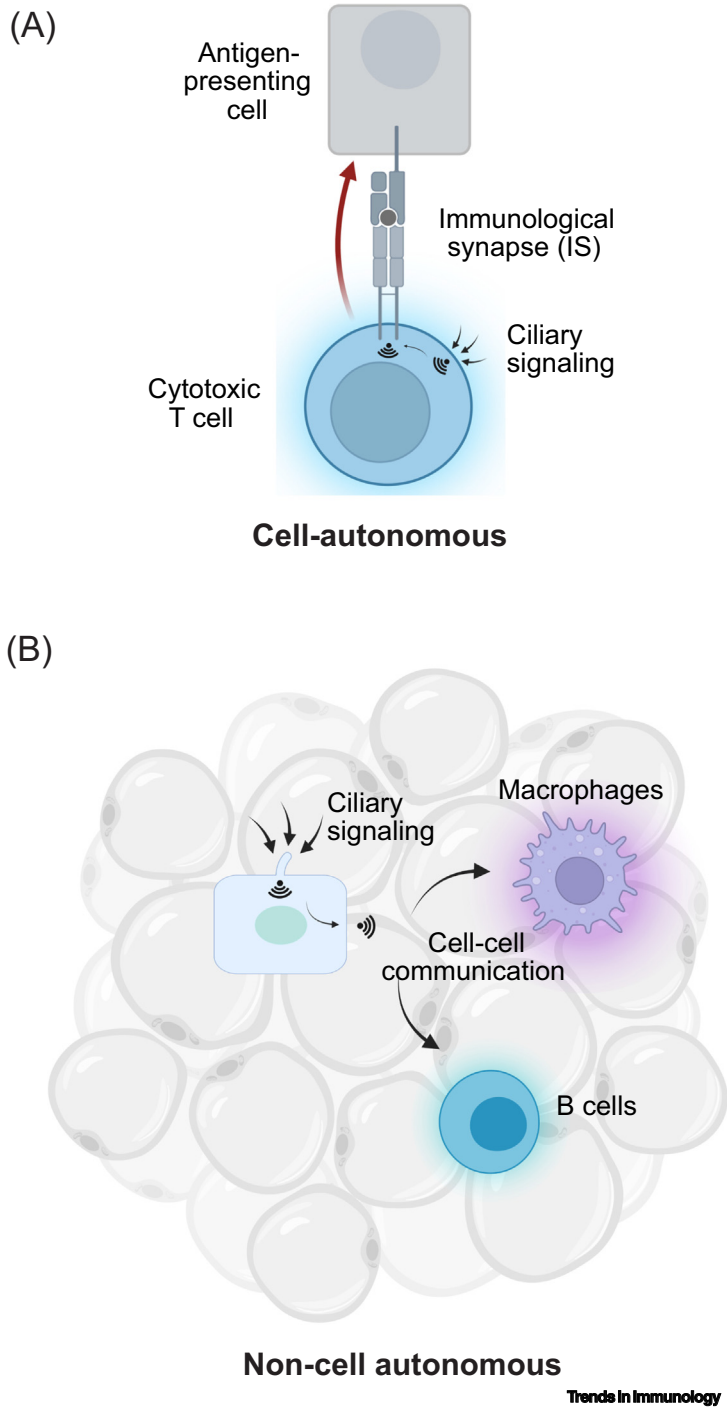


Figure 2. Metaflammation. (A) Examples of noncommunicable diseases (NCDs) associated with obesity. These include dementia, Alzheimer's disease, depression, cardiovascular diseases (such as atherosclerosis), metabolic diseases (such as type 2 diabetes mellitus), as well as chronic kidney disease or gout. (B) Metaflammation describes a low-grade, chronic inflammation triggered by lifestyle or environmental factors that can lead to the development of NCDs [7,9].

Key figure

Model of cell-autonomous versus noncell-autonomous effects of primary cilia on immune cells in mice and/or humans



T cells upon AD induction, as shown by mRNA expression analysis of Hh signaling components (i.e., Smo) [33]. In turn, pharmacological Smo inhibition in mice exacerbated the AD phenotype. After AD induction, Hh-mediated transcription of immunoregulatory genes via Gli2 in skin $CD4^+$ and $CD8^+$ T cells increased the numbers of **regulatory T cells** (Tregs) in wild-type mice compared with mice lacking functional Gli2 expression (conditional Gli2ΔN2 mice), as shown, in part, via flow cytometry; this led to Treg cell-mediated immunosuppression, thus limiting AD [33].

In addition, BBS1, the core component of the BBSome complex (*BBS1* is the most frequently mutated gene in BBS [16]), has been shown to control centrosome polarization by clearing centrosomal F-actin and its positive regulator WASH1, which is implicated in the control of IS formation [34].

However, do patients with BBS display defects in CTL function? This has not been directly tested, although BBS mouse models might provide a clue. These have been widely used to mimic human BBS disease. For example, Bbs4-knockout mice (*Bbs4*$^{-/-}$-KO), which lack Bbs4, one of the eight central BBSome components, and display major features of the human phenotype (i.e., obesity and retinopathy [35]), exhibit normal T cell development and function [19], suggesting that Bbs4 in mice does not control T cell homeostasis. However, a detailed analysis of CTL function in other BBS mouse models is missing thus far.

In sum, there are remarkable similarities between IS formation and primary cilia (i.e., both use the same ciliary Hh and IFT signaling components), but how ciliary proteins and **ciliary signaling** modulate the formation and/or function of the IS versus primary cilia appears somewhat different because the IS does not require the formation of a morphological cilium to orchestrate signaling, whereas the primary cilium relies on the formation of the ciliary axoneme; indeed, the molecular mechanisms of these signaling pathways remain ill-defined, as do their impacts on larger T cell effector functions.

A putative noncell autonomous role of primary cilia signaling in modulating immune cell numbers and functions?

A key question is whether primary cilia can control immune cell numbers, phenotypes, and functions via noncell-autonomous mechanisms; that is, via communication between a ciliated non-immune cell with a nonciliated immune cell, such as macrophages or B cells (Figure 3B) [19,36,37]. However, the molecular mechanisms underlying such presumed interactions are largely unknown.

Primary cilia signaling can affect B cell development

A recent report aimed to shed light on how ciliary signaling affected immune cell signaling and function and revealed that B cell functions might be altered in a tissue-specific context and in a primary cilia-dependent manner [19]. Specifically, analysis of *Bbs4*$^{-/-}$-KO mice revealed that primary cilia dysfunction (due to the loss of Bbs4) altered B cell development in both the bone marrow and the periphery relative to wild-type mice: IgD^- IgM^- B cell precursor numbers in the bone marrow as well as late mature B cells (IgD^+ IgM^-) in the spleen and lymph node were increased, whereas numbers of splenic marginal zone (MZ) B cells were reduced compared

Figure 3. (A) Primary cilia signaling components are also involved in the formation of the immunological synapse (IS) in cytotoxic $CD8^+$ T cells, although these cells do not form a morphological cilium [28]. This has created the term 'frustrated cilium' [20]. Thus, they control T cell function in a cell-autonomous fashion. (B) Primary cilia can control the function of several different non-immune cells, such as epithelial or adipocyte precursors in the adipose tissue. In turn, they can control certain immune cell functions in a noncell-autonomous fashion (e.g., macrophages or B cells). Figure partially created using BioRender (https://biorender.com/).

with wild-type mice [19]. Of note, primary cilia dysfunction did not alter antigen-specific T or B cell responses [19], suggesting that Bbs4-dependent changes in B cell numbers during development did not affect B cell function. Moreover, the molecular cause of the defect in B cell development was extrinsic to cells of the hematopoietic lineage or endothelial cells, as evidenced by conditional *Bbs4*$^{-/-}$-KO mice. Instead, the defect occurred in bone marrow stromal cells because Bbs4 controlled CXCL12 expression via canonical Wnt signaling in mesenchymal stem cells (MSCs) [19]. Given that CXCL12 promotes B cell development [38], reduced CXCL12 signaling in *Bbs4*$^{-/-}$-KO mice contributed to impaired B cell development relative to controls [19], but the functional implications of this developmental defect remain unknown. Furthermore, which cell types in the stroma can be affected by primary cilia dysfunction remains to be assessed. Of note, in zebrafish, primary cilia signaling in endothelial cells has been reported to control hematopoietic stem cell (HSC) maintenance in the hemogenic epithelium via Notch-dependent signaling [39], as evidenced by genetic and pharmacological approaches *in vivo* in zebrafish embryos; this suggested that primary cilia signaling in endothelial cells controls immune cell development in zebrafish. Altogether, we posit that these data support a model in which primary cilia signaling in ciliated, non-immune cells (e.g., endothelial cells) can promote immune cell development, which warrants further investigation in other species.

Primary cilia signaling can modulate innate immune cell phenotypes and functions

The noncell autonomous role of primary cilia signaling in modulating immune cell function becomes particularly evident in a tissue context: tissue-resident cells of the innate immune system (e.g., macrophages) are prime examples. Environmental factors, such as diet, can directly reprogram macrophages [7,40], but macrophages can also react in response to molecular clues received from other cell types in the tissue (e.g., endothelial cells) [41]. As shown in mice, every tissue harbors tissue-resident macrophages predominantly originating from fetal erythroid–myeloid progenitors, in addition to monocyte-derived macrophages originating from HSCs [42]. Furthermore, in mammals, macrophages colonize distinct subtissue niches during steady-state [43]. We argue that different macrophage populations are also associated with tissue inflammation in ciliopathies, as discussed below for different tissues (adipose tissue, liver, and kidney).

Primary cilia signaling in adipose tissue homeostasis, inflammation, and metabolism

Mammalian WAT is a highly plastic and dynamic organ comprising different cell types (e.g., adipocytes, preadipocytes, immune cells, endothelial cells, and fibroblasts). The primary function of WAT is to maintain the systemic energy balance: it stores and releases free fatty acids and secretes **adipokines** that communicate with other organs [44]. In turn, this regulates energy intake and expenditure as well as other metabolic processes [44]. Where and how WAT expands is an essential determinant of metabolic health: in humans, expansion of subcutaneous WAT (scWAT), which is located under the dermis, is considered protective against metabolic diseases, whereas expansion of visceral WAT (vWAT), which engulfs the organs in the abdomen, leads to central obesity and has been associated with metabolic dysfunction [45]. Expansion of WAT by hyperplasia (i.e., *de novo* adipogenesis) has been associated with metabolically healthy obesity in rodents and humans [14,46,47]. By contrast, the uptake of lipids into existing adipocytes (hypertrophy) can lead to the development of hypoxia and has been associated with metaflammation and metabolic dysfunction in humans; this has been evidenced by the direct correlation between hypertrophy and defects in lipid metabolism, T2DM, and cardiovascular disorders [46]. Thus, mechanisms that drive adipogenesis are considered prime targets for controlling inflammatory responses in the adipose tissue during obesity [48,49].

Relevant to our discussion, primary cilia are also considered key targets for regulating adipogenesis because primary cilia signaling in preadipocytes promotes adipogenesis [50,51]. Accordingly, in

the WAT of rodents and humans, MSCs and committed preadipocytes, but not mature adipocytes, display primary cilia [50,51]. Furthermore, abolishing primary cilia formation in preadipocytes can impair WAT expansion, whereas stimulation of primary cilia signaling in preadipocytes using omega-3 free fatty acids can promote adipogenesis and WAT expansion, as evidenced from genetic mouse models and pharmacological approaches [51]. Moreover, patients with obesity and BBS have been reported to display higher gene expression of adipogenic genes, such as *PPARG* or *SREBP1C*, in scWAT compared with BMI-matched control patients [42]. Furthermore, patients with BBS can also express lower amounts of proinflammatory genes, such as tumor necrosis factor alpha (*TNFA*) or inducible nitric oxide synthase (*INOS*) [14]. By contrast, the expression of anti-inflammatory genes, such as those encoding adiponectin, endothelial nitric oxide synthase (*ENOS*), and interleukin-10 (*IL10,*) has been shown to be increased in scWAT compared with BMI-matched control patients [14]; this suggests that patients with BBS, despite being obese, do not display inflammatory parameters such as these in adipose tissue. Of note, *BBS12* deletion in healthy human MSCs *in vitro* mimicked a proadipogenic expression profile, displaying increased gene expression of, for example, *PPARG* and promoting adipogenesis in culture, as evidenced from increased lipid uptake relative to controls [14]. Compared with controls, the adipogenic phenotype was recapitulated in *Bbs12*$^{-/-}$-KO mice, which displayed adipose expansion by hyperplasia and no systemic inflammation, shown via histological staining of adipose tissue sections and *in vitro* adipocyte differentiation assays, and by measuring circulating inflammatory mediators [14]. In addition, this study also reported that patients with BBS and *Bbs12*$^{-/-}$-KO mice retained normal glucose tolerance and insulin sensitivity compared with control mice (as defined by the WHO criteria) [14]. These findings suggest that primary cilia dysfunction, as seen in patients with BBS, is not detrimental for metabolic health, disconnecting obesity from inflammation, although this hypothesis remains to be rigorously assessed.

Indeed, two different cohort studies supported these findings. One study was performed in adult patients with BBS without corresponding controls [52], and another study compared 50 young patients with BBS (average age: 15 years) with 100 obese, non-BBS controls, who were BMI, but not age or sex matched [15]. These studies demonstrated that, despite harboring greater visceral adiposity than obese controls, patients with BBS showed superior glucose tolerance, accompanied by low prevalence of T2DM [15,52]. Initially, this suggested that primary cilia dysfunction, as observed in patients with BBS, is protective for metabolic health during obesity.

By contrast, another study comprising 152 adult patients with BBS, as well as sex-, age-, and BMI-matched controls, reported that patients with BBS overall displayed metabolic syndrome, as determined by higher fasting blood glucose and triglyceride concentrations, and overall higher insulin resistance compared with matched controls [16]. Moreover, a high prevalence of T2DM was described in another cohort study of 46 patients with BBS without controls [53]. These studies suggested that obesity in BBS was accompanied by the same metabolic complications as diet-induced obesity. However, one caveat of both of these latter studies [16,53] was that some patients were classified as BBS based only on their clinical features alone, likely leading to potential misdiagnoses because of shared phenotypes with Alström syndrome ciliopathy, which causes severe insulin resistance and T2DM [54].

In summary, based on the studies mentioned above, it remains controversial whether primary cilia dysfunction in patients with BBS acts as a friend or foe in modulating metabolic health during obesity; therefore, rigorous studies in humans and mice will need to investigate these discrepancies in molecular detail. Moreover, none of these studies analyzed immune cell functions that might be connected to primary cilia defects; therefore, a question to robustly address is whether various

immune cells are differentially programmed in the context of BBS compared with diet-induced obesity in humans and mice.

Mutation-specific differences have been determined across different metabolic phenotypes [16]. For instance, insulin resistance and metabolic syndrome appear to occur predominantly in patients carrying mutations in *BBS10* compared with patients carrying mutations in *BBS1* [16]. However, whether different metabolic outcomes in BBS are due to differences in primary cilia signaling resulting from different BBS mutations and/or BBS protein functions remains unknown. Likewise, whether different *BBS* mutations can contribute to differences in adipocyte fates and functions remains to be investigated. Furthermore, how cilia-dependent changes in adipocyte fates and functions might modulate immune cells and inflammatory responses in the tissue niche is also enigmatic, but represents a fruitful area for future investigation.

Primary cilia can modulate liver macrophage populations and functions

The adipose tissue and liver have central metabolic roles and also store lipids upon chronic overnutrition. Furthermore, the liver and kidney are the two organs targeted by ciliopathies that lead to cyst formation, including BBS [55]. Cysts develop due to abnormal proliferation of ciliated epithelial cells accompanied by fluid secretion, leading to liver fibrosis in patients with ciliopathies [56]. Primary cilia dysfunction has been reported to differentially affect the monocyte-derived and tissue-resident macrophage compartments in the congenital IFT88^{Orpk} ciliopathy mouse model, which carries a mutation in *Ift88* and presents with kidney and liver cysts, as well as fibrosis within the first 4 weeks after birth [36]. Such deficiency caused an accumulation of infiltrating Ly6c^{high} monocytes in the liver, as evidenced by flow cytometry [36]. These Ly6c^{high} monocytes differentiated into macrophages even before the onset of **severe biliary hyperplasia** and fibrosis in mutant mice [36]. By contrast, the population of resident macrophages in the liver decreased upon primary cilia dysfunction, suggesting that cyst development and liver fibrosis led to the specific recruitment of Ly6c^{high} monocytes in the liver, which might have contributed to disease progression [36]. Along with the accumulation of these myeloid cell populations in the liver, the expression of proinflammatory transcripts, including those encoding TGF-β, TNF-α, IL-1β, and chemokine (C-C) motif ligand 2 (CCL2), was increased in the liver of IFT88^{Orpk} mice compared with control mice [36]. The primary cause of liver cyst formation and fibrosis was primary cilia dysfunction in epithelial cells [36]; therefore, these results suggest that primary cilia signaling in non-immune cells, namely epithelial cells, modulates the function on nonciliated immune cells, such as monocytes/macrophages, contributing to an inflammatory response and promoting cyst formation in the liver, at least in mice.

Primary cilia signaling might modulate renal macrophage function in the kidney

Inflammation is a key feature of cystic kidney disease. Both ciliopathies, **autosomal dominant polycystic kidney disease (ADPKD)** and **recessive cystic kidney disease nephronophthisis** (NPHP), share an inflammatory phenotype, characterized by an increased expression of inflammatory cytokines and immune cell recruitment into the kidney [57,58]. Furthermore, both pathologies are characterized by fibrosis during disease progression in humans and rodents [59,60]. Renal macrophages are mainly derived from fetal progenitors and are self-maintained throughout life, with only minor contributions from peripheral monocytes in mice [61,62]. Using ADPKD mouse models and chemical macrophage ablation *in vivo*, renal macrophage infiltration was shown to occur at an early stage of the disease and to promote cyst enlargement [63]. Detailed analysis using two different PKD mouse models combined with flow cytometry and immunofluorescent/histological analyses as well as renal injury revealed that, postnatally, renal resident macrophages changed from CD11c^{lo} (characteristic of tissue-resident macrophages) to CD11c^{hi} (characteristic of monocyte-derived infiltrating macrophages), which coincided with a change from rapid to slow cyst formation in these ciliopathy models [64]. Numbers of

Clinician's corner

Primary cilia are cellular antennae that perceive external stimuli and transduce them into intracellular responses; primary cilia have important roles in development, sensory reception, and homeostatic maintenance [2].

Primary cilia dysfunction causes a broad spectrum of syndromes named ciliopathies. Obesity is a cardinal feature in a subgroup of ciliopathies, but the metabolic parameters differ from nonsyndromic obesity [71].

Approaches to treat obesity are mostly based on lifestyle modifications with poor results, and therapies to tackle major pathological processes, such as metaflammation, are lacking [72].

Clinicians could treat obesity more effectively by understanding the molecular mechanisms. The study of primary cilia and its role in controlling immune system homeostasis may help to find potential targets to improve the management of obesity and associated diseases.

CD11c^{lo} resident macrophages were increased in renal tissue before and during cyst formation, suggesting that CD11c^{lo} resident macrophages contributed to rapid cyst formation [64]. Moreover, epithelial cells in the kidney appeared to communicate with CD11c^{lo} resident macrophages via membrane-bound colony stimulation factor 1 (CSF1) and its receptor CSF1R in PKD mouse models, as evidenced by pharmacological inhibition of CSF1/CSF1R signaling [64,65]; this suggested that the communication between epithelial cells and resident macrophages in the kidney contributes to disease progression. Indeed, an abdominal window on the kidney coupled to two-photon imaging was used to visualize specific cellular interactions, which allowed the analysis of primary cilia signaling and immune cell movement in longitudinal studies *in vivo* [66]. Functional studies are underway, but this methodology is now in place as a first step for examining the molecular, spatiotemporal interactions between ciliated non-immune cells and nonciliated immune cells *in vivo* under physiological and pathological conditions in rodents. In summary, the kidney appears to be a prime tissue that can be assessed to reveal the intricate relationship between primary cilia signaling in non-immune cells and their communication with immune cells under such contexts.

Concluding remarks

Primary cilia control many cellular functions and are essential for intercellular communication. Loss-of-function mutations in ciliary genes can trigger ciliopathies that present with different symptoms, including cystic and fibrotic liver and kidney, as well as the development of obesity. Those symptoms have been associated with tissue and/or systemic inflammation in rodents and humans. However, because prototypic primary cilia are absent from most hematopoietic cells, a role for primary cilia in the communication between ciliated non-immune and immune cells is conceivable. However, these studies are still early and are limited to animal models. Therefore, many questions remain to be rigorously addressed experimentally (see Outstanding questions). Indeed, from the findings outlined in this emerging field, there is much to be learned regarding this tiny organelle and to understand its putative contribution to immune responses and metaflammation in ciliopathies, obesity, and other common NCD Western lifestyle-associated diseases.

Outstanding questions

When and how does primary cilia dysfunction protect from metabolic dysfunction? Answering these questions can reveal whether primary cilia signaling could be targeted in metabolic diseases.

Are ciliary and immune cell defects specific to BBS, or are they also observed in other ciliopathies? As more than 190 genes are affected in the different ciliopathies, addressing these questions is essential for understanding the molecular signaling pathways involved in BBS.

Which ciliated cell types communicate with nonciliated immune cells via primary cilia signaling, and what is variety of immune cells involved? Answering these questions may help to understand cell–cell communication in a primary cilia-dependent manner.

Why is primary cilia dysfunction eliciting a proinflammatory response in the kidney in humans and mice and not, at least in some cases, in adipose tissue? To address this question, it will be vital to understand the molecular mechanisms underlying tissue inflammation in ciliopathies and beyond.

How can tissue- and species-specific differences in the described findings be reconciled?

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Declaration of interest

The authors declare no conflicting interest.

Resources

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References

1. Wachten, D. and Mick, D.U. (2021) Signal transduction in primary cilia - analyzing and manipulating GPCR and second messenger signaling. *Pharmacol. Ther.* 224, 107836
2. Pazour, G.J. and Witman, G.B. (2003) The vertebrate primary cilium is a sensory organelle. *Curr. Opin. Cell Biol.* 15, 105–110
3. Nachury, M.V. *et al.* (2007) A core complex of BBS proteins cooperates with the GTPase Rab8 to promote ciliary membrane biogenesis. *Cell* 129, 1201–1213
4. Ye, F. *et al.* (2018) BBSome trains remove activated GPCRs from cilia by enabling passage through the transition zone. *J. Cell Biol.* 217, 1847–1868
5. Nachury, M.V. (2018) The molecular machines that traffic signaling receptors into and out of cilia. *Curr. Opin. Cell Biol.* 51, 124–131
6. Wheway, G. *et al.* (2019) Opportunities and challenges for molecular understanding of ciliopathies-The 100,000 Genomes Project. *Front. Genet.* 10, 127

7. Christ, A. *et al.* (2019) Western diet and the immune system: an inflammatory connection. *Immunity* 51, 794–811
8. Hotamisligil, G.S. (2017) Inflammation, metaflammation and immunometabolic disorders. *Nature* 542, 177–185
9. Christ, A. and Latz, E. (2019) The Western lifestyle has lasting effects on metaflammation. *Nat. Rev. Immunol.* 19, 267–268
10. Gregor, M.F. and Hotamisligil, G.S. (2011) Inflammatory mechanisms in obesity. *Annu. Rev. Immunol.* 29, 415–445
11. Blüher, M. (2010) The distinction of metabolically 'healthy' from 'unhealthy' obese individuals. *Curr. Opin. Lipidol.* 21, 38–43
12. Vaisse, C. *et al.* (2017) Cilia and obesity. *Cold Spring Harb. Perspect. Biol.* 9, a028217
13. Forsyth, R.L. and Gunay-Aygun, M. (2003) Bardet-Biedl syndrome overview. In *GeneReviews* (Adam, M.P. *et al.*, eds), University of Washington
14. Marion, V. *et al.* (2012) BBS-induced ciliary defect enhances adipogenesis, causing paradoxical higher-insulin sensitivity, glucose usage, and decreased inflammatory response. *Cell Metab.* 16, 363–377
15. Feuillan, P.P. *et al.* (2011) Patients with Bardet-Biedl syndrome have hyperleptinemia suggestive of leptin resistance. *J. Clin. Endocrinol. Metab.* 96, E528–E535
16. Mujahid, S. *et al.* (2018) The endocrine and metabolic characteristics of a large Bardet-Biedl syndrome clinic population. *J. Clin. Endocrinol. Metab.* 103, 1834–1841
17. Wheatley, D.N. (1995) Primary cilia in normal and pathological tissues. *Pathobiology* 63, 222–238
18. Singh, M. *et al.* (2016) Primary cilia are present on human blood and bone marrow cells and mediate Hedgehog signaling. *Exp. Hematol.* 44, 1181–1187
19. Tsyklauri, O. *et al.* (2021) Bardet-Biedl Syndrome ciliopathy is linked to altered hematopoiesis and dysregulated self-tolerance. *EMBO Rep.* 22, e50785
20. Griffiths, G.M. *et al.* (2010) The immunological synapse: a focal point for endocytosis and exocytosis. *J. Cell Biol.* 189, 399–406
21. Stinchcombe, J.C. *et al.* (2006) Centrosome polarization delivers secretory granules to the immunological synapse. *Nature* 443, 462–465
22. Joukov, V. and De Nicolo, A. (2019) The centrosome and the primary cilium: the Yin and Yang of a hybrid organelle. *Cells* 8, 701
23. Bertrand, F. *et al.* (2013) An initial and rapid step of lytic granule secretion precedes microtubule organizing center polarization at the cytotoxic T lymphocyte/target cell synapse. *Proc. Natl. Acad. Sci. U. S. A.* 110, 6073–6078
24. Tamzalit, F. *et al.* (2020) Centrioles control the capacity, but not the specificity, of cytotoxic T cell killing. *Proc. Natl. Acad. Sci. U. S. A.* 117, 4310–4319
25. Stinchcombe, J.C. *et al.* (2015) Mother centriole distal appendages mediate centrosome docking at the immunological synapse and reveal mechanistic parallels with ciliogenesis. *Curr. Biol.* 25, 3239–3244
26. Boding, L. *et al.* (2014) Midline 1 directs lytic granule exocytosis and cytotoxicity of mouse killer T cells. *Eur. J. Immunol.* 44, 3109–3118
27. Obino, D. *et al.* (2016) Actin nucleation at the centrosome controls lymphocyte polarity. *Nat. Commun.* 7, 10969
28. de la Roche, M. *et al.* (2013) Hedgehog signaling controls T cell killing at the immunological synapse. *Science* 342, 1247–1250
29. Finetti, F. *et al.* (2009) Intraflagellar transport is required for polarized recycling of the TCR/CD3 complex to the immune synapse. *Nat. Cell Biol.* 11, 1332–1339
30. Huangfu, D. and Anderson, K.V. (2005) Cilia and Hedgehog responsiveness in the mouse. *Proc. Natl. Acad. Sci. U. S. A.* 102, 11325–11330
31. Briscoe, J. and Therond, P.P. (2013) The mechanisms of Hedgehog signalling and its roles in development and disease. *Nat. Rev. Mol. Cell Biol.* 14, 416–429
32. Mengrelis, K. *et al.* (2019) Sonic Hedgehog is a determinant of gammadelta T-Cell differentiation in the thymus. *Front. Immunol.* 10, 1629
33. Papaioannou, E. *et al.* (2019) Sonic Hedgehog signaling limits atopic dermatitis via Gli2-driven immune regulation. *J. Clin. Invest.* 129, 3153–3170
34. Cassioli, C. *et al.* (2021) The Bardet-Biedl syndrome complex component BBS1 controls T cell polarity during immune synapse assembly. *J. Cell Sci.* 134, jcs258462
35. Mykytyn, K. *et al.* (2004) Bardet-Biedl syndrome type 4 (BBS4)-null mice implicate Bbs4 in flagella formation but not global cilia assembly. *Proc. Natl. Acad. Sci. U. S. A.* 101, 8664–8669
36. Zimmerman, K.A. *et al.* (2018) Primary cilia disruption differentially affects the infiltrating and resident macrophage compartment in the liver. *Am. J. Physiol. Gastrointest. Liver Physiol.* 314, G677–G689
37. Hilgendorf, K.I. (2021) Primary cilia are critical regulators of white adipose tissue expansion. *Front. Physiol.* 12, 769367
38. Zehentmeier, S. and Pereira, J.P. (2019) Cell circuits and niches controlling B cell development. *Immunol. Rev.* 289, 142–157
39. Liu, Z. *et al.* (2019) Primary cilia regulate hematopoietic stem and progenitor cell specification through Notch signaling in zebrafish. *Nat. Commun.* 10, 1839
40. Christ, A. *et al.* (2018) Western diet triggers NLRP3-dependent innate immune reprogramming. *Cell* 172, 162–175
41. Hernandez, G.E. and Iruela-Arispe, M.L. (2020) The many flavors of monocyte/macrophage–endothelial cell interactions. *Curr. Opin. Hematol.* 27, 181–189
42. Mass, E. (2018) Delineating the origins, developmental programs and homeostatic functions of tissue-resident macrophages. *Int. Immunol.* 30, 493–501
43. Guilliams, M. *et al.* (2020) Establishment and maintenance of the macrophage niche. *Immunity* 52, 434–451
44. Rosen, E.D. and Spiegelman, B.M. (2014) What we talk about when we talk about fat. *Cell* 156, 20–44
45. Caputo, T. *et al.* (2017) From chronic overnutrition to metaflammation and insulin resistance: adipose tissue and liver contributions. *FEBS Lett.* 591, 3061–3088
46. Ye, R.Z. *et al.* (2022) Fat cell size: measurement methods, pathophysiological origins, and relationships with metabolic dysregulations. *Endocr. Rev.* 43, 35–60
47. Vishvanath, L. and Gupta, R.K. (2019) Contribution of adipogenesis to healthy adipose tissue expansion in obesity. *J. Clin. Invest.* 129, 4022–4031
48. Jiang, N. *et al.* (2019) Cytokines and inflammation in adipogenesis: an updated review. *Front. Med.* 13, 314–329
49. Stuttgen, G.M. and Sahoo, D. (2021) FFAR4: a new player in cardiometabolic disease? *Endocrinology* 162, bqab111
50. Marion, V. *et al.* (2009) Transient ciliogenesis involving Bardet-Biedl syndrome proteins is a fundamental characteristic of adipogenic differentiation. *Proc. Natl. Acad. Sci. U. S. A.* 106, 1820–1825
51. Hilgendorf, K.I. *et al.* (2019) Omega-3 fatty acids activate ciliary FFAR4 to control adipogenesis. *Cell* 179, 1289–1305
52. Imhoff, O. *et al.* (2011) Bardet-Biedl syndrome: a study of the renal and cardiovascular phenotypes in a French cohort. *Clin. J. Am. Soc. Nephrol.* 6, 22–29
53. Moore, S.J. *et al.* (2005) Clinical and genetic epidemiology of Bardet-Biedl syndrome in Newfoundland: a 22-year prospective, population-based, cohort study. *Am. J. Med. Genet. A* 132A, 352–360
54. Aliferis, K. *et al.* (2012) Differentiating Alstrom from Bardet-Biedl syndrome (BBS) using systematic ciliopathy genes sequencing. *Ophthalmic Genet.* 33, 18–22
55. Forsythe, E. and Beales, P.L. (2013) Bardet-Biedl syndrome. *Eur. J. Hum. Genet.* 21, 8–13
56. Diamond, T. *et al.* (2021) Hepatic ciliopathy syndromes. *Clin. Liver Dis. (Hoboken)* 18, 193–197
57. Quatredeniers, M. *et al.* (2022) The renal inflammatory network of nephronophthisis. *Hum. Mol. Genet.* Published online January 19, 2022. https://doi.org/10.1093/hmg/ddac014
58. Song, C.J. *et al.* (2017) Inflammation and fibrosis in polycystic kidney disease. *Results Probl. Cell Differ.* 60, 323–344
59. Ong, A.C. *et al.* (2015) Autosomal dominant polycystic kidney disease: the changing face of clinical management. *Lancet* 385, 1993–2002
60. Norman, J. (2011) Fibrosis and progression of autosomal dominant polycystic kidney disease (ADPKD). *Biochim. Biophys. Acta* 1812, 1327–1336

61. Gomez Perdiguero, E. *et al.* (2015) Tissue-resident macrophages originate from yolk-sac-derived erythro-myeloid progenitors. *Nature* 518, 547–551
62. Liu, F. *et al.* (2020) Distinct fate, dynamics and niches of renal macrophages of bone marrow or embryonic origins. *Nat. Commun.* 11, 2280
63. Karihaloo, A. *et al.* (2011) Macrophages promote cyst growth in polycystic kidney disease. *J. Am. Soc. Nephrol.* 22, 1809–1814
64. Zimmerman, K.A. *et al.* (2019) Tissue-resident macrophages promote renal cystic disease. *J. Am. Soc. Nephrol.* 30, 1841–1856
65. Li, Z. *et al.* (2021) Resident macrophages in cystic kidney disease. *Kidney360* 2, 167–175
66. Revell, D.Z. and Yoder, B.K. (2019) Intravital visualization of the primary cilium, tubule flow, and innate immune cells in the kidney utilizing an abdominal window imaging approach. *Methods Cell Biol.* 154, 67–83
67. Klink, B.U. *et al.* (2020) Structure of the human BBSome core complex. *Elife* 9, e53910
68. Singh, S.K. *et al.* (2020) Structure and activation mechanism of the BBSome membrane protein trafficking complex. *Elife* 9, e53322
69. Hildebrandt, F. *et al.* (2011) Ciliopathies. *N. Engl. J. Med.* 364, 1533–1543
70. Torres, V.E. *et al.* (2007) Autosomal dominant polycystic kidney disease. *Lancet* 369, 1287–1301
71. Focsa, I.O. *et al.* (2021) Clinical and genetic heterogeneity of primary ciliopathies (Review). *Int. J. Mol. Med.* 48, 176
72. Andreotti, F. *et al.* (2017) Mechanisms, pathophysiology, and management of obesity. *N. Engl. J. Med.* 376, 1490–1491