Trends in
Endocrinology & Metabolism

CellPress
OPEN ACCESS

Review

The evolutionary tug-of-war of macrophage metabolism during bacterial infection

Luís Almeida,[1,*] Ayesha Dhillon-LaBrooy,[1] and Tim Sparwasser[1,*]

The function and phenotype of macrophages are intimately linked with pathogen detection. On sensing pathogen-derived signals and molecules, macrophages undergo a carefully orchestrated process of polarization to acquire pathogen-clearing properties. This phenotypic change must be adequately supported by metabolic reprogramming that is now known to support the acquisition of effector function, but also generates secondary metabolites with direct microbicidal activity. At the same time, bacteria themselves have adapted to both manipulate and take advantage of macrophage-specific metabolic adaptations. Here, we summarize the current knowledge on macrophage metabolism during infection, with a particular focus on understanding the 'arms race' between host immune cells and bacteria during immune responses.

Highlights

Macrophage-intrinsic pathogen recognition processes and cellular metabolism are intimately linked.

M1 and M2 macrophages exhibit distinct metabolic profiles, but cannot be categorized as 'purely glycolytic' nor 'purely dependent on mitochondrial respiration'.

Metabolic adaptations in macrophages depend on the stimulation and are generally tailored to promote microbicidal immune responses.

Macrophage metabolism undergoes changes to promote the production and accumulation of intermediate metabolites that inhibit or interfere with bacterial metabolism.

Certain bacteria can inhibit, modulate, or hijack macrophage metabolism, conferring on them a fitness advantage and allowing them to delay or bypass host immunity.

Immunometabolic dichotomy between infection and homeostasis

The function and phenotype of immune cells vary drastically between homeostasis and infection. While some cells, such as tissue-resident macrophages, exert myriad extra-immunological roles in their respective niches [1–3], others (e.g., naïve T cells and B cells) persist in a quiescent, nonproliferative, and energy-conserving state until activated by adequate signals [4–7]. In the event of an insult like a viral or bacterial infection, innate immune cells such as neutrophils and macrophages serve as the first line of defense. To do this, they must rapidly adopt a pathogen-clearing phenotype in place of their homeostatic functions. In turn, adaptive immune cells, such as lymphocytes, must vigorously proliferate and secrete effector cytokines or immunoglobulins on recognition of their cognate antigens [5,8]. Such contrasting states require the dynamic and carefully regulated engagement and disengagement of various metabolic enzymes and pathways. If one or more key metabolic events are perturbed, the cells may find themselves unable to perform a certain process important for their function. In that aspect, metabolism can be seen as the clockwork of immune cells. Given the fundamental role of the immune system in mediating the outcome of infection and, in turn, the vital role of metabolism in orchestrating immune cell function, it is important to understand which, if any, metabolic disturbances that occur during infection are beneficial or detrimental to the outcome of the disease.

Engagement of pathogen- or danger-sensing receptors in immune cells, such as Toll-like receptors (TLRs) in macrophages or the T cell receptor (TCR) on T cells, is a defining event in the mediation of a metabolic switch that occurs on activation. Distinct intracellular kinases and transcription factors act rapidly to promote the synthesis of glycolytic genes, as well as that of genes involved in lipid and protein synthesis and glutamine anaplerosis, as reviewed elsewhere [5,9]. Importantly, this metabolic switch is fundamental for the development of an immune response and clearance of the triggering pathogen(s). Until recently, the importance of most metabolic pathways in macrophages has been studied in isolation, often with limited consideration of the interplay between the metabolism of two entities at odds with one another: the bacteria and the macrophage. In this review, we briefly

[1]Institute of Medical Microbiology and Hygiene, University Medical Center of Johannes Gutenberg University, Mainz 55131, Germany

*Correspondence:
lalmeida@uni-mainz.de (L. Almeida) and
sparwasser@uni-mainz.de
(T. Sparwasser).

cover the metabolic pathways involved in bacterial-resistance effector innate immune responses performed by macrophages. Then, we discuss how certain bacteria have adapted to modulate, bypass, or hijack macrophage metabolism as a way to evade immune clearance. Unless otherwise specified, the data discussed generally refer to mouse macrophages.

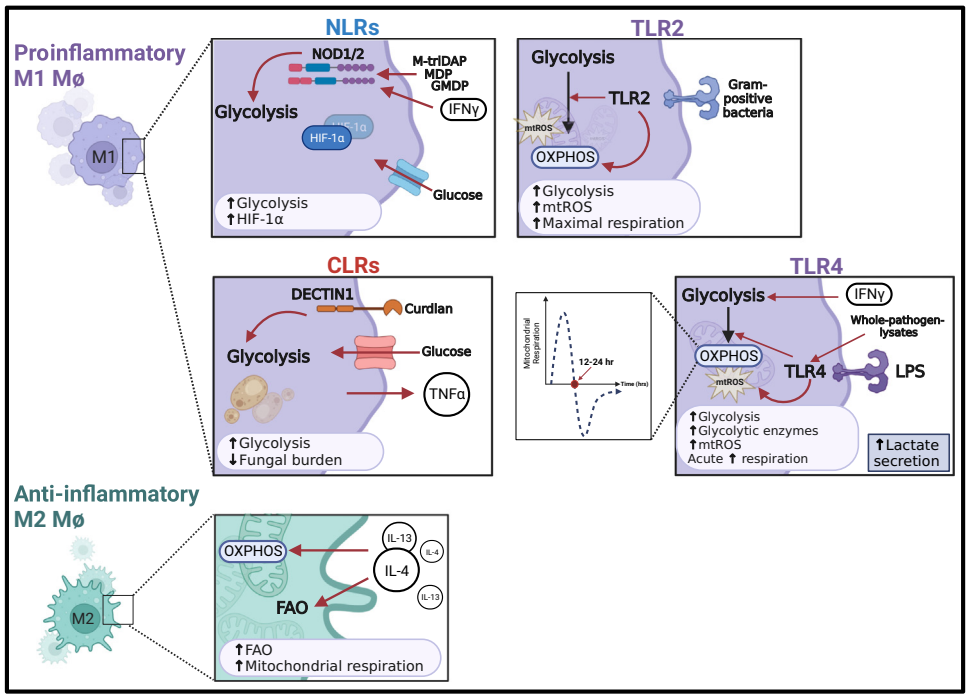
Pathogen-sensing mechanisms trigger metabolic adaptations in macrophages

Macrophages can be broadly divided into two types according to their origin: tissue-resident macrophages and blood-borne monocyte-derived macrophages [3,10,11]. The former are considered to be long lived and arise during development, being replenished throughout life chiefly by self-renewal, with limited import from bone-marrow-derived progenitors [10]. We do not cover here the metabolism of tissue-resident macrophages, focusing instead on blood-circulating monocyte-derived macrophages. However, the metabolic requirements and aspects of tissue-resident macrophages have recently been extensively reviewed elsewhere [12].

Macrophages can respond to pathogen- and damage-associated molecular patterns (PAMPs and DAMPs) early on infection and/or tissue damage [13]. They achieve this through the expression of pattern recognition receptors such as C-type lectin receptors, NOD-like receptors, TLRs, and RIG-I-like receptors, among others [13]. These and others allow the macrophage to generally sense the type of pathogen (bacterial, viral, or fungal) and secrete cytokines and chemokines that assist in orchestrating a specialized response towards that class of pathogen [14]. Monocyte-derived macrophages are generally categorized as M1 or M2 based on whether they display an inflammatory or an anti-inflammatory profile, respectively (Figure 1). It is becoming increasingly apparent, particularly with the advent of single-cell sequencing technologies [15,16], that the M1 versus M2 classification does not provide a complete overview of all of the different macrophage phenotypes [17]. Nevertheless, in this review we consider the metabolism of proinflammatory 'M1' macrophages, which are fundamental in antibacterial responses [18,19]. It is now clear that pattern recognition receptor activation in monocytes is directly linked to metabolic rewiring: Lachmandas *et al.* demonstrated that the majority of pathogen-derived stimuli do activate (aerobic) glycolysis in human monocytes [20]. However, it is also clear that different stimuli lead to different metabolic phenotypes, and there is not a be-all–end-all metabolic state of activated macrophages [12,20,21].

Glycolysis as a hallmark of M1 polarization

Macrophages polarized in the presence of lipopolysaccharide (LPS) and interferon-gamma (IFN-γ) acquire a proinflammatory M1 phenotype (Figure 1), exhibiting higher expression of the glucose transporter Glut1 compared with unstimulated or M2-polarized macrophages [21]. The expression of three other glucose transporters (Glut2, Glut3, and Glut4) is undetectable or only barely detected in all subtypes of macrophages, suggesting both that M1 macrophages exhibit a higher degree of glucose uptake and that Glut1 is the main glucose transporter in this cell type [21]. Activation of monocytes with various TLR ligands or whole-pathogen lysates boosts glucose consumption, lactate production, and/or upregulation of the expression of glycolytic enzyme genes (Figure 2A) [20,22]. Stimulation of human monocyte-derived macrophages with the NOD1 agonist DAP-containing muramyl tripeptide (M-triDAP) (a peptidoglycan degradation product) and the NOD2 agonists muramyl dipeptide and glucosaminyl muramyl dipeptide or the T cell-derived type 1 cytokine IFN-γ equally upregulates glycolysis very quickly *in vitro*, suggesting that an increase in glucose consumption and aerobic glycolysis is a general consequence of pathogen sensing by macrophages [20,22,23]. These data match older observations showing that *ex vivo* exposure of macrophages to endotoxin leads to Glut1 upregulation and a two- to threefold increase in glucose uptake [24]. The upregulation of Glut1 and other glycolysis-associated genes is believed to be mediated by hypoxia-inducible factor (HIF)1α. In addition to regulating metabolism under hypoxia, the HIF



Trends in Endocrinology & Metabolism

Figure 1. Classical differentiation model of monocyte-derived macrophages. On exposure to type 1 stimuli such as interferon-gamma (IFNγ), C-type lectin receptor (CLR), NOD-like receptor (NLR), Toll-like receptor (TLR), or RIG-I-like receptor agonists, monocyte-derived macrophages acquire a proinflammatory 'M1' phenotype characterized by specific metabolic profiles. Type 2 cytokines such as interleukin (IL)-4 and IL-13 drive the acquisition of an anti-inflammatory M2 phenotype with lower levels of glycolysis, with ATP synthesis maintained by mitochondrial respiration and fatty acid oxidation (FAO). Figure created with BioRender.com. Abbreviations: GMP, glucosaminyl muramyl dipeptide; HIF-1α, hypoxia-inducible factor 1α; LPS, lipopolysaccharide; MDP, muramyl dipeptide; M-triDAP, DAP-containing muramyl tripeptide; mtROS, mitochondrial reactive oxygen species; OXPHOS, oxidative phosphorylation; TNFα, tumor necrosis factor alpha.

pathway regulates the expression of genes implicated in central metabolic pathways [25]. HIF1α is upregulated in peritoneal exudate macrophages isolated and stimulated with proinflammatory stimuli such as LPS, LPS and IFN-γ, and curdlan (a dectin-1 ligand) (Figure 2A) [26]. Consistent with this, exposure to the same stimuli or *in vitro* challenge with *Listeria monocytogenes* or *Candida albicans* increases glucose utilization in mouse (or human) macrophages [26]. *In vivo*, mice with a macrophage-specific HIF1α deletion ($Hif1a^{flox/flox} \times Lysm^{Cre}$) fail to mount proinflammatory responses towards intravenous challenge with *L. monocytogenes* or *C. albicans* and exhibit lower serum tumor necrosis factor alpha (TNFα) concentrations, as well as higher pathogen burden compared with wild-type (WT) controls. Inhibition of glycolysis with 2-deoxy-D-glucose (2-DG) (a competitive inhibitor of glycolysis) *in vivo* worsened the *C. albicans* pathogenic burden; *in vitro* it reduced the production of TNFα in macrophages exposed to LPS and curdlan, showing that macrophages rely on glucose to acquire a proinflammatory M1 phenotype. By contrast, interleukin (IL)-4 (a prototypical M2 polarization cytokine) promoted only a marginal increase in both HIF1α and glycolytic flux, suggesting that glycolysis is preferentially engaged during M1 macrophage polarization [26].

Glycolysis fuels bactericidal processes in macrophages

Macrophages kill engulfed pathogens through the fusion of phagosomes with endosomes and, ultimately, lysosomes. The entire process of phagosome maturation results in an acidic organelle rich in antimicrobial peptides and enzymes to promote the killing of the internalized pathogens

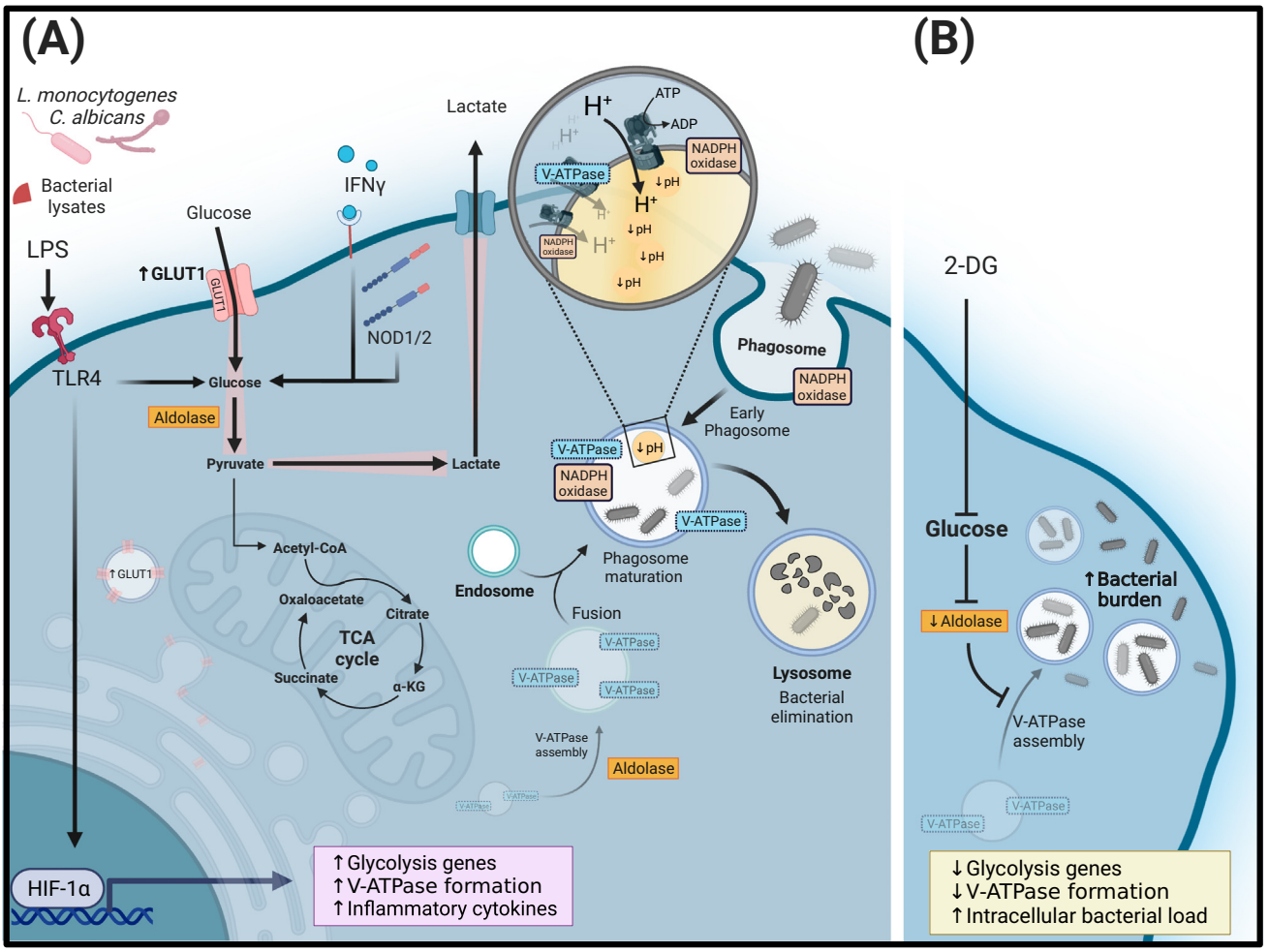


Figure 2. Glycolysis is induced by pathogens and needed for macrophage effector function. (A) Toll-like receptor (TLR) ligands or whole-pathogen lysates promote glucose consumption and lactate production in macrophages. It is believed that this occurs due to upregulation of the expression of glycolytic enzymes by hypoxia-inducible factor (HIF)1α. Glycolysis promotes the acidification of phagosomes by providing ATP used by proton-pumping V-ATPases, promoting bacterial killing. (B) Inhibition of glycolysis impedes the acidification of the phagosome and leads to higher bacterial burden in the cells. Figure created with BioRender.com. Abbreviations: 2-DG, 2-deoxy-D-glucose; α-KG, alpha-ketoglutarate; *C. albicans, Candida albicans; GLUT1, glucose transporter 1; IFNγ, interferon-gamma; L. monocytogenes, Listeria monocytogenes; LPS, lipopolysaccharide; TCA, tricarboxylic acid.*

inside the formed vacuoles, which are enriched in microbicidal proton-pumping V-ATPases, NADPH oxidase, and other enzymes (Figure 2A,B) [27,28]. One way through which glycolysis is required in macrophages is by contributing to the acidification of phagosomes, a prerequisite for the killing of engulfed pathogens. V-ATPases hydrolyze ATP and couple this process to the pumping of protons to the inside of the phagosomes, lowering the pH and contributing both to direct pathogen killing and the activation of microbicidal enzymes in the phagosome [29–31]. When glycolysis is inhibited with 2-DG, bone marrow-derived macrophages (BMDMs) exhibit higher numbers of intracellular live bacteria (Figure 2B) [32]. This effect was most pronounced with *Salmonella typhimurium* but could also be observed with *L. monocytogenes* and *Staphylococcus aureus*. Blocking glycolysis reduces the assembly of a vacuolar-type ATPase complex necessary for the acidification of phagosomes. Assembly of the complex and acidification of the phagosome was prevented by siRNA-mediated ablation of aldolase A in BMDMs. Aldolase A

was shown to interact with the V-ATPase subunit V1B2, and this occurred even in the presence of a catalytically dead aldolase A, demonstrating that the catalytic activity of this enzyme *per se* is not required for the assembly of the vacuolar-type ATPase complex. It is possible that inhibition of glycolysis prevents the normal upregulation of the expression of glycolytic enzymes such as aldolase A, henceforth reducing the amount of aldolase A available for V-ATPase complex assembly [32]. Taking these findings together, it seems clear that macrophages upregulate and require active glycolysis to effectively kill engulfed pathogens.

Mitochondria: using the (ancient) bacteria to destroy the bacteria

Given the scope of this review, it is not possible to provide a complete discussion of the nuanced role of mitochondria in macrophages. For a more detailed discussion, we suggest recently published literature on the topic that offers a more focused overview [33–35]. It has been hinted and suggested that M1 macrophages, in addition to being characterized by high glycolytic rates, further downregulate cellular respiration, while M2 macrophages are mainly respiratory and less glycolytic, relying instead on mitochondrial energy generation pathways such as fatty acid oxidation [34,36,37]. While this concept is broadly accurate for M1 macrophages, particularly because M1-polarizing signals interrupt the tricarboxylic acid (TCA) cycle at key steps (as discussed in this section), recent data challenge this black-and-white paradigm, with the role and importance of mitochondria in macrophages remaining elusive. Divakaruni and others have demonstrated that – unlike what had been previously surmised [37] – mouse and human macrophages do not require fatty acid oxidation to acquire an M2 phenotype, showcasing the complexity of macrophage mitochondrial metabolism [38,39]. Further, lower rates of maximal respiration occur in $CD14^+$ human monocytes exclusively in response to a TLR4 agonist (LPS) [20]. When these cells are exposed to the TLR2 agonist Pam_3CysSK_4 or bacterial lysates, both glycolysis and maximal respiration increase compared with non-activated monocytes (Figure 1). However, in the study, basal cellular respiration was similar in LPS-treated and untreated macrophages. When the two results are taken together, it seems that LPS does not decrease respiration in macrophages but rather, that it prevents its upregulation in response to further challenges (Figure 1).

Data supporting the notion that mitochondrial respiration in macrophages may not be as high as otherwise possible in proinflammatory macrophages include the fact that the TCA cycle is interrupted at two steps in M1 macrophages [36]. One of these 'breaks' is established by the upregulation of the expression of the *Acod1* gene [originally termed immune response gene 1 (Irg1)] in response to LPS [40,41] (Figure 3). *Acod1* encodes aconitate decarboxylase 1 (Acod1) and is one of the most differentially expressed genes in M1 macrophages compared with non-activated macrophages. This enzyme catalyzes the conversion of the TCA cycle metabolite *cis*-aconitate into itaconate leading to the production and accumulation of itaconate in LPS-stimulated macrophages. In turn, itaconate has been described to have antimicrobial properties by promoting macrophage lysosomal biogenesis and bacteriolysis, as well as directly inhibiting bacterial growth [40,42]. It is believed to exert its antibacterial effect by inhibiting isocitrate lyase (the first enzyme of the glyoxylate cycle) in bacteria, which catalyzes the conversion of isocitrate into glyoxylate and succinate [43]. While itaconate production is important in the control of several bacterial infections, such as those caused by *Mycobacterium tuberculosis*, *Coxiella burnetii*, and *S. typhimurium* [42,44–46], other bacteria such as *Yersinia pestis* and *Pseudomonas aeruginosa* are resistant and thrive in the presence of itaconate [47,48], as discussed in the next section. The upregulation of Acod1 and the consequent production of itaconate may be somewhat selective to LPS stimulation since stimulation with M-triDAP leads to only marginal amounts of Acod1 protein and approximately tenfold-lower mRNA levels compared with LPS stimulation [22].

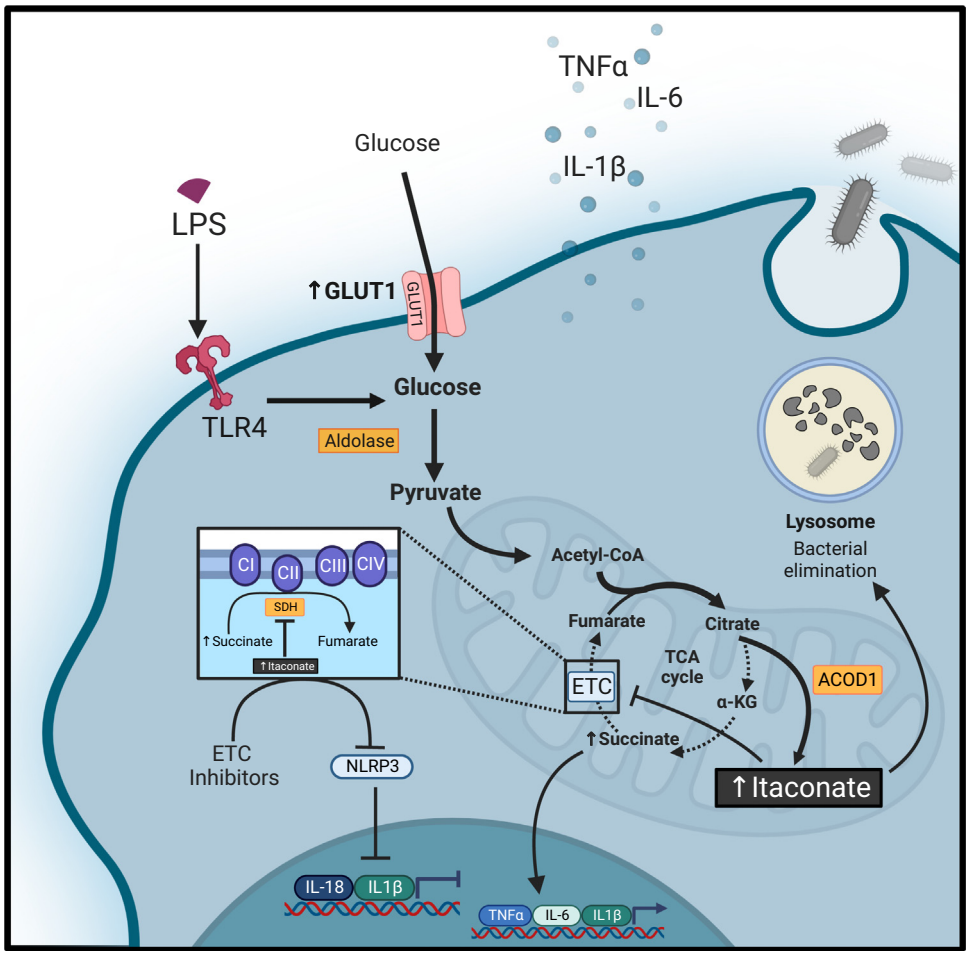


Figure 3. The tricarboxylic acid (TCA) cycle in lipopolysaccharide (LPS)-activated macrophages is interrupted to synthesize itaconate. Itaconate synthesis, as a result of Acod1 activity on LPS activation, contributes to the diversion of carbon flux away from the TCA cycle. In addition, itaconate inhibits succinate dehydrogenase (SDH), leading to succinate accumulation. Notwithstanding, inhibitors of the electron transport chain (ETC) impair the production of proinflammatory cytokines. Figure created with BioRender.com. Abbreviations: α-KG, alpha-ketoglutarate; IL, interleukin; TLR, Toll-like receptor; TNFα, tumor necrosis factor alpha.

Interestingly, despite similar glucose consumption and extracellular acidification rates between LPS-treated and M-triDAP-treated macrophages, the extracellular concentration of lactate was lower in the latter, suggesting that lactate is re-imported and further metabolized to a greater extent by these cells, but the functional and biological relevance of lactate metabolism in response to different stimuli is unknown. Given that lactate in the tumor microenvironment has been reported to promote M2 rather than M1 polarization [49,50], one can speculate that different rates of extracellular lactate import and metabolism may contribute to extracellular lactate clearance and enforcement of M1 polarization and/or the maintenance of an environment permissive for a proinflammatory profile for other immune cells that are also inhibited by lactate [51]. The second break in the TCA cycle that occurs in M1 macrophages is at the level of succinate. Succinate accumulates due to apparently impaired activity of the enzyme succinate dehydrogenase (SDH), particularly in response to LPS; this may be due to increased nitric oxide and/or itaconate concentrations observed on stimulation (Figure 3) [41,52,53]. How each metabolite

contributes to the inhibition of SDH activity is not entirely understood and is likely to depend on the biological context, but regardless, it is now well established that succinate does accumulate in M1 macrophages, which supports both intracellular and extracellular processes driving the production of proinflammatory cytokines, as covered elsewhere [33,35].

While these data may suggest that mitochondrial respiration is dispensable for macrophage activity, other data argue otherwise. When carefully examining the kinetics of oxygen consumption in murine macrophages, Langston *et al.* demonstrated that LPS triggers a small, acute increase in macrophage mitochondrial respiration compared with baseline, with a lower respiratory rate occurring only 12–24 h on LPS exposure [54]. This increase in respiration is largely driven by glucose oxidation, facilitating the production of acetyl-CoA as a substrate for the acetylation of histones bound at the *Il6* and *Il1b* promoters. The authors demonstrate that this acute increase followed by an acute decrease of respiration is largely mediated by the glycerol phosphate shuttle. At the earlier time points (up to ~3 h post-stimulation), the glycerol phosphate shuttle drives glycerol-3-phosphate-dependent forward electron transport. This activity eventually saturates the system, leading to reverse electron transport and impairment of oxidative phosphorylation occurring 12–24 h post-stimulation. The authors further argue that this 'pre-programmed' decrease in respiration is important to mediate a hand-in-hand decrease in IL-6 and IL-1β production as a negative, tolerogenic mechanism. These findings may help to explain why mitochondrial complex I activity is necessary for IL-1β expression in BMDMs [55]. Initially, it was postulated that this was due to complex I-mediated reactive oxygen species (ROS) production, but in a more recent study with LPS-primed macrophages, mitochondrial complex I, II, III, and V inhibitors have been shown to diminish cellular NLRP3 inflammasome activation in BMDMs, which in turn is important for IL-1β and IL-18 production in response to 'two-hit' stimulation with PAMPs/DAMPs (Figure 3) [56]. Inhibition of complex I with piericidin A or of complex III with myxothiazol (or by genetic deletion of mitochondrial complexIII subunit VII) impairs IL-1β secretion (but not transcription) in BMDMs, an effect that is dependent on NLRP3 inflammasome activity [57]. IL-1β secretion in complex I-inhibited cells can be rescued by ectopic overexpression of the *Saccharomyces cerevisiae* homolog of NADH dehydrogenase (NDI1) and in complex III-inhibited or -knockout BMDMs by the *Ciona intestinalis* alternative oxidase (AOX). NDI1 can transfer and AOX can accept electrons from ubiquinone in the mitochondrial electron transport chain (ETC), effectively complementing the energetic deficiency caused by the loss of complexes I and III, respectively. Importantly, and unlike complexes I and III, neither NDI1 nor AOX contributes to ROS production in complex I- and III-deficient cells. Taken together, these data show that decreased mitochondrial ATP – rather than ROS – production may instead mediate the deficient NLRP3-dependent IL-1β secretion that occurs on the inhibition of complex I or III [55]. Therefore, when previous findings are taken together with those of Langston *et al.*, it appears that mitochondria are critically important for the initial production of proinflammatory cytokines in LPS-activated macrophages [54]. Overall, the ETC in macrophages is more complex than initially proposed. It is also possible that macrophages are only minimally reliant on mitochondrial ATP production, but still rely on functional ETC complexes to regenerate NAD^+ levels to keep the TCA cycle and/or NAD^+-dependent metabolic reactions or transcription regulatory processes, such as histone (de-)acetylation by sirtuin enzymes, active [4,58]. *In situ* studies in different body niches and in response to various challenges will be fundamental to understanding these intricacies.

In addition to the yet-to-be-fully understood role of the ETC in macrophages, mitochondria as organelles are undoubtedly important for macrophage function. Mitochondrial enzymes and TCA cycle metabolites such as α-ketoglutarate and succinate are generally required for histone methylation and acetylation processes, which in turn regulate the expression of anti- and proinflammatory genes in most (if not all) immune cell types, including macrophages [4,59–61].

In wound-healing processes, mitochondrial activity has been reported to differ between 'early-stage' and 'late-stage' macrophages (day 4 and 14 post-injury, respectively) [62]. Early-stage macrophages are characterized by lower mitochondrial mass, activity, and ATP production compared with late-stage-injury macrophages. The authors demonstrate that the transition from proinflammatory to an M2 phenotype depends on the sensing of mitochondrially derived ROS and HIF1α stabilization.

Is metabolism an ancient evolutionary battlefield between host and pathogen?

The immune system and pathogens coevolve as the result of a constant tug-of-war: the former must adequately detect and eliminate pathogens, while the latter must evade detection to survive and replicate. Metabolic adaptations triggered by the recognition of conserved PAMPs are suggestive of a conserved advantage that arose throughout evolution. By quickly adapting its metabolism in a way that promotes pathogen clearance on recognition of that class of pathogen, it is intuitive that the host would have a fitness advantage. For example, Kuang *et al.* have recently demonstrated that macrophages quickly deploy transferrin-containing extracellular vesicles in a model of sepsis [63]. These vesicles capture circulating iron, diminishing the access of bacteria to circulatory iron and contributing to infection control by hampering bacterial iron-dependent metabolic processes [63]. However, as the metabolism of one organism changes, so must the other to ensure survival. In the previous sections, we have provided some examples of how host cell metabolism must be carefully adapted to respond to infection. In this section, we attempt to provide some clues that suggest that host and bacterial metabolism is an ongoing tug-of-war or arms race.

A recent preprint study on the evolution of the *Irg1* sequence revealed in a phylogenetic analysis that the metazoan and fungal *Irg1* homologs were intercalated by prokaryotic sequences, suggesting that they were acquired in two different events and not present from a common ancestor [64]. At the same time, *Irg1* homologs are completely absent in entire species and taxa from the same domains of life. Even among mammals, the Hyracoidea and Xenarthra orders do not appear to have any homolog of the *Acod1/Irg1* gene, strongly suggesting that itaconate production *per se* is not fundamentally required for organismal metabolism [64]. Because itaconate is highly reactive, in addition to blocking the prokaryotic glyoxalate cycle, it can also inhibit crucial metabolic enzymes and processes in the host, such as SDH (as mentioned before) and complex II-mediated oxidative phosphorylation [53,64,65]. Therefore, the stringent and highly regulated expression of itaconate in activated macrophages may be necessary to ensure that itaconate does not exert overt toxicity in other cells and tissues. It is possible that the lowering of mitochondrial respiration in LPS-exposed macrophages is partially an unavoidable consequence and trade-off of itaconate production. Further, these data tease the notion that the *Irg1* sequence may have been acquired independently by certain metazoans and fungi to provide a competitive advantage in the presence of itaconate-susceptible bacteria [64].

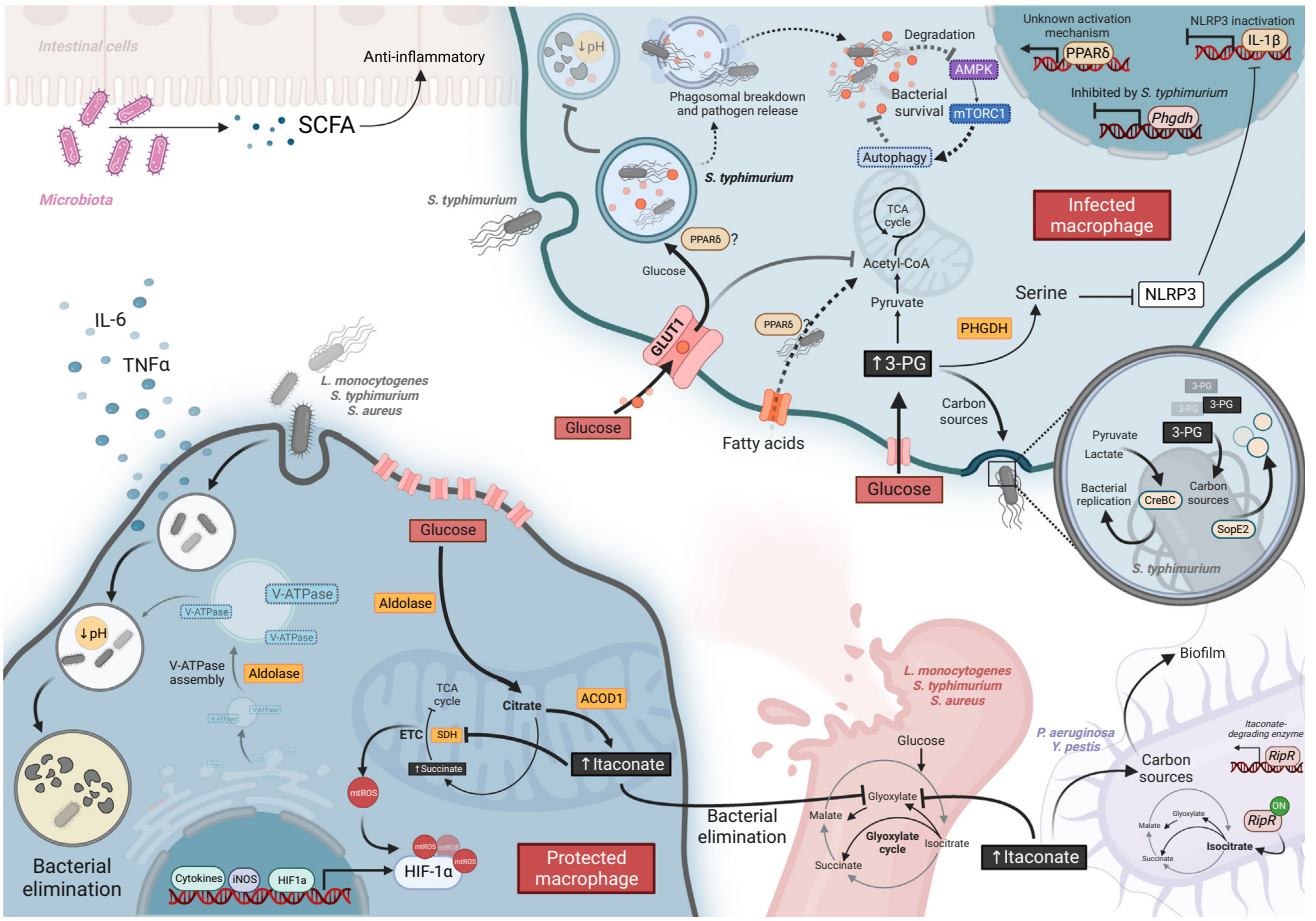
At the same time, different bacteria can degrade macrophage-produced itaconate. *Salmonella* species carry a series of genes encoding itaconate-degrading enzymes, controlled by a protein termed RipR (Table 1 and Figure 4) [44,66]. RipR is activated on binding not to itaconate, but to isocitrate [66]. Because the inhibition of isocitrate lyase by itaconate would lead to accumulation of isocitrate, RipR rescues the glyoxalate cycle flux in bacteria by upregulating itaconate-degrading enzymes on sensing high isocitrate levels. In other bacteria, such as *S. aureus*, normally high glycolytic rates are inhibited by itaconate [67]. As a result, the surviving bacteria isolated from patients with long-term *S. aureus* lung infection appear to have adapted to gluconeogenesis and are selected for extracellular polysaccharide synthesis and consequent biofilm formation (Table 1 and Figure 4), conferring on them the capacity to cause persistent infection

Table 1. Consequences of interfering with macrophage metabolism on antibacterial responses

Bacterium	Change occurring in host		Host factor affected	Consequence and outcome
	Glycolysis	Chemical inhibition of glycolysis (i.e., 2-DG)		
Salmonella typhimurium		↑ Live intracellular bacteria	Aldolase A	Aldolase A is produced during glycolysis and is needed for V-ATPase formation. V-ATPases are needed to reduce the pH of phagosomes leading to bacterial clearance. Inhibition of glycolysis inhibits aldolase A production and thus increases the intracellular bacterial burden[a].
Listeria monocytogenes				
Staphylococcus aureus				
		Bacterially induced inhibition of glycolysis		
S. typhimurium		Induces SopE2 to inhibit host glycolytic enzyme phosphoglycerate dehydrogenase leading to ↓ glycolysis ↓ Glycolysis =↑ 3-phosphoglyceric acid	Phosphoglycerate dehydrogenase and 3-phosphoglyceric acid	↑ 3-Phosphoglyceric acid = ↓ *de novo* serine biosynthesis (↓serine) = ↓ inflammasome and IL-1β activation. Bacteria use 3-phosphoglyceric acid as a carbon source for energy. Promoting bacterial survival.
		Induces Pparδ through an unknown mechanism	Pparδ	Pparδ shifts host cell nutrient utilization from glucose to fatty acids[a]
Mycobacterium tuberculosis		Induces miR-21 in host cells, which inhibits glycolysis.	Phosphofructokinase	Bacteria-induce the expression of miR-21, which inhibits the expression of phosphofructokinase (an enzyme that commits glucose to glycolytic metabolism).
[a]Indicates proposed theories				
	TCA cycle	Host-produced itaconate		
S. typhimurium		↑ Bacterial growth	Itaconate	Macrophage-produced itaconate inhibits the first enzyme in the glyoxylate cycle in bacteria (isocitrate lyase), effectively rendering bacterial growth
L. monocytogenes				
S. aureus				
Coxiella burnetii				
		Bacterial resistance to itaconate		
Salmonella sp.		Degrades itaconate		Itaconate is degraded as a result of bacterial RipR and used by the bacteria as a carbon source for biofilm production and survival
Pseudomonas aeruginosa				Degrades itaconate to use as a carbon source for biofilm production and therefore survival
Yersinia pestis				

[67]. Similar related or unrelated genetic operons, with the purpose of metabolizing itaconate, exist in several other bacteria, including *Y. pestis* and *P. aeruginosa* (Figure 4). Like *Y. pestis* and *P. aeruginosa*, which appear to carry itaconate-degrading genetic operons, *M. tuberculosis* has been reported to metabolize itaconate (Table 1) through its conversion to itaconyl-CoA, followed by hydration to (*S*)-citramalyl-CoA, which is then cleaved by Rv2498c (an enzyme that also participates in lysine catabolism) to form pyruvate and acetyl-CoA [45]. Because both pyruvate and acetyl-CoA are central metabolites that can be used for energy production or shuttled towards biosynthetic pathways, *M. tuberculosis* is able to grow with itaconate as a single carbon source. In a mouse model of aerosol infection, a Rv2498c-deficient strain led to ~90% less bacterial load in the lungs of infected mice at 28, 84, and 112 days post infection, compared with the WT strain [45].

These adaptations allow these (and other) bacteria to not only become resistant to itaconate but to inclusively assimilate it as a carbon source [47,48]. Due to this, itaconate production by macrophages in response to infection with itaconate-metabolizing bacteria may be detrimental to the clearance of pathogens that thrive on itaconate. For example, certain *P. aeruginosa* strains that can metabolize itaconate are less competent at infecting *Acod1*$^{-/-}$ mice, which are unable to



Trends in Endocrinology & Metabolism

Figure 4. Host–bacterial interactions. Top left: Some commensal bacteria at mucosal surfaces produce beneficial anti-inflammatory metabolites that are used and sensed by host cells to mediate homeostasis. Bottom left: summary of macrophage-specific metabolic adaptations that promote pathogen engulfment and killing. Top right: examples of *Salmonella typhimurium*-mediated interference with macrophage metabolism on internalization, conferring protection from the aforementioned bactericidal mechanisms and promoting infection. Bottom right: itaconate inhibits the glyoxalate cycle in bacteria, but certain bacteria such as *Pseudomonas aeruginosa* and *Yersinia pestis* have adapted to metabolize itaconate and can use it as a carbon source rather than be inhibited by it. These data are also summarized in Table 1 in the main text. Figure created with BioRender.com. Abbreviations: 3-PG, 3-phosphoglycerate; AMPK, AMP-activated protein kinase; HIF-1α, hypoxia-inducible factor 1α; IL, interleukin; iNOS, inducible nitric oxide synthase; *L. monocytogenes, Listeria monocytogenes; mTORC1, mammalian target of rapamycin complex 1; mtROS, mitochondrial reactive oxygen species; P. aeruginosa, Pseudomonas aeruginosa; PHGDH, phosphoglycerate dehydrogenase; PPARδ, peroxisome proliferator-activated receptor delta; S. aureus, Staphylococcus aureus; SCFA, short-chain fatty acid; SDH, succinate dehydrogenase; TCA, tricarboxylic acid; TNFα, tumor necrosis factor alpha; Y. pestis, Yersinia pestis.*

produce itaconate [47]. *S. aureus*, despite not metabolizing itaconate, undergoes metabolic changes in the presence of it that promote biofilm production and survival (Figure 4) [67]. Therefore, in *Acod1*$^{-/-}$ mice with pneumonia, *S. aureus* is cleared more rapidly [68]. The importance of itaconate for anti-*S. aureus* responses may, however, be tissue and context dependent, because mice deficient in Acod1 exhibit higher intraocular *S. aureus* bacterial loads in an intraocular infection model [69]. Interestingly, despite the aforementioned capacity of *M. tuberculosis* to process and metabolize itaconate, mice deficient in Acod1 or conditionally lacking it in myeloid cells are more susceptible to *M. tuberculosis* lung infection [70]. Therefore, there appears to be a continuum of adaptation/tolerance to itaconate. Conceivably, the capacity to metabolize itaconate *per se* seems insufficient to determine whether a certain bacterium will thrive and even benefit from the presence of itaconate or whether it may only partially tolerate it and grow in

suboptimal conditions. Further, itaconate itself is now believed to promote beneficial bactericidal-independent effects on host cells, such as boosting phagocytosis, promoting long-term 'trained immunity', and acting as a counterbalancing immunosuppressive metabolite to prevent tissue damage as a result of excessive inflammatory responses [71–73]. From this perspective, understanding whether itaconate production is expected to affect bacterial virulence early during the course of an infection could be exploited to pharmacologically manipulate (or not) host itaconate production to facilitate pathogen clearance.

Given the importance of glycolysis in macrophages and the chief role of macrophages in pathogen clearance, it is equally understandable that some pathogens have evolved to bypass, neutralize, or even take advantage of the increased glycolytic flux in macrophages occurring on pathogen detection. In the cell line RAW 264.7, *S. typhimurium* relies on active macrophage glycolysis and glucose availability for infection [74]. Further, it may even actively trigger the expression of peroxisome proliferator-activated receptor delta (Pparδ) in infected macrophages, although how it does so is unclear (Figure 4) [9,75]. Regardless, because Pparδ acts to shift the intracellular nutrient utilization from glucose to fatty acids [76], it is hypothesized that *S. typhimurium*-induced Pparδ expression enforces a metabolic state that limits glucose utilization by the macrophage, increasing its availability for the bacterium. Accordingly, *S. typhimurium* cannot adequately infect Pparδ-deficient macrophages or mice [75]. It has more recently been proposed that *S. typhimurium* infection also disrupts glycolysis in macrophages as a way to reduce phagosome acidification [32]. In another study, infection of macrophages with live *S. typhimurium* bacteria led to a transient (1 h) activation of AMP-activated protein kinase (AMPK) [77]. Activated AMPK can inhibit mammalian target of rapamycin (mTOR) complex 1 (mTORC1) [78–80], but in *S. typhimurium*-infected cells AMPK and AMPK-interacting partners are targeted for degradation, leading to overt mTORC1 activity and preventing the normal induction of autophagy as a microbicidal process [77]. Pathogen-derived factors interfere with host proteins to dampen the glycolytic flux, not only increasing the amount of glucose available for the pathogen but also preventing microbicidal mechanisms that would otherwise kill the bacteria. As an example, one gene in a genomic region of *S. typhimurium* termed pathogenicity island 1 encodes a protein called SopE2. SopE2 inhibits the expression of host *Phgdh*, the gene encoding phosphoglycerate dehydrogenase, leading to impaired *de novo* serine biosynthesis from the glycolytic intermediate 3-phosphoglycerate (3-PG) (Table 1). As a result of phosphoglycerate dehydrogenase inhibition, the concentrations of 3-PG increase in the cytoplasm of macrophages. Then, 3-PG is freely used up by *S. typhimurium* as a carbon and energy source. Further, without serine synthesis, more glucose is metabolized to pyruvate and/or lactate by the infected macrophage. These two metabolites are sensed by another bacterial protein called CreBC. CreBC activates pathogenicity island 2 genes in response to increasing concentrations of those metabolites, inducing intracellular replication of the pathogen. Because serine deprivation negatively impacts macrophage inflammasome activation and IL-1β production [81,82], the authors postulate that inhibiting the serine biosynthetic branch may additionally provide a survival benefit to the bacteria due to impaired macrophage function. Nonetheless, whether serine supplementation could rescue microbicidal activity in *S. typhimurium*-infected macrophages was not explored. Live *M. tuberculosis* has also been shown to decrease glycolysis and mitochondrial respiration on infection of macrophages. By contrast, the nonvirulent *Mycobacterium bovis* or dead *M. tuberculosis* upregulated aerobic glycolysis in macrophages as expected [83]. *M. tuberculosis* infection leads to persistent expression of the miRNA miR-21, which decreases glycolytic flux by decreasing the expression of the enzyme catalyzing the reaction that commits glucose to glycolytic metabolism [84]. The macrophage-activating cytokine IFN-γ, important in the immune response towards *M. tuberculosis*, inhibits the expression of miR-21 and, consequently, restores glycolysis in macrophages. *M. tuberculosis* infection leads to the presence of lipid bodies in macrophages. These lipid bodies are believed to act as a source of nutrients and a 'safe haven' for intracellular bacteria [85,86]. Interestingly, despite

lipid bodies being important for pathogenicity, and despite the importance of fatty acid synthesis for effector T cell function [87,88], conditional ablation of *de novo* fatty acid synthesis in macrophages (*LysM*^{Cre}*Acaca*$^{flox/flox}$ mice) does not meaningfully affect the capacity of mice to respond to *M. tuberculosis* infection. Hence, the lipid body content can perhaps be sustained from exogenously obtained lipids when intracellular biosynthetic pathways are impaired [88]. These modulations of host metabolism, which appear to occur only with live (and not heat-killed) bacteria, demonstrate that they are not a mere consequence of PAMP or DAMP detection by the macrophage [82]. However, it remains to be fully understood whether such metabolic changes are an active, bacterium-controlled process, a compensatory host response occurring as a consequence of the bacterium outcompeting the host macrophage for nutrient reserves, or a combination of the two. Another host factor that regulates the expression of several genes encoding metabolic enzymes is HIF1α. HIF1α is important in the metabolic reprogramming that occurs in macrophages and for their bactericidal activity, as reviewed elsewhere [25]. *C. burnetii* dampens HIF1α expression in infected macrophages shortly after infection. This effect is dependent on bacterial viability, protein synthesis, and capacity to inject effector proteins into infected macrophages through their type IV secretion system [89]. *C. burnetii* fails to replicate in macrophages under hypoxia, presumably through the induction of HIF1α that diminishes the intracellular content of TCA cycle intermediates in macrophages [90]. This metabolic austerity imposed by HIF1α is suggested to limit the nutrients available for *C. burnetii* replication. Therefore, although no direct molecular mechanism was found between bacterium-derived factors and HIF1α degradation, it is tempting to postulate that *C. burnetii* dampens the intracellular levels of this cytokine to ensure maximal nutrient availability for its own needs [89].

Overall, when one looks at host and bacterial metabolic processes, they appear to be the result of back-and-forth selective pressures, strongly suggesting that metabolism is an ancient and ongoing battlefield between pathogen and host (Figure 4), possibly on the same level or even as part of pathogen recognition versus immune evasion mechanisms. Future studies focusing on the dissection of metabolic pathway conservation and differences between different species may provide evidence that, throughout evolution, species of bacteria have themselves evolved to adapt to the adaptations immune cells acquired to better stave off pathogens. We have summarized some of the important consequences of interfering with host metabolism for the outcome of infection.

Concluding remarks

In recent years, it has become apparent that metabolism is crucial for immune cell function, including macrophages. Initial paradigms stated that proinflammatory M1 macrophages are mainly glycolytic and not reliant on mitochondria, while anti-inflammatory M2 macrophages are the opposite. It is now clear that the metabolism of macrophages is nuanced and cannot be neatly categorized into purely glycolytic and/or respiratory. This is perhaps expected, given that the mammalian body has diverse metabolic niches, and absolute metabolic profiles may not be compatible with all potential niches and pathogens. To add a further layer of complexity, pathogens often rely on their metabolism to thrive, which can be targeted as a way to fight infection but also increases the chance that pathogen metabolism may (co)evolve to bypass or modulate the metabolism of host cells to its advantage. In this sense, metabolic adaptations from both host and microbes may be considered to be conceptually similar to the evolution of different pattern recognition strategies such as TLRs (on the host) and the development of immune evasion mechanisms (on the pathogen); thus, further immunometabolism research may benefit from turning its attention to the metabolism of the pathogens themselves (see Outstanding questions). By understanding the complex and dynamic crosstalk between host and pathogen, it may be possible to develop adjuvant therapies that metabolically benefit the host and/or interfere with bacteria. Finally, it will be equally interesting to understand whether metabolic interplay with the natural microbiota represents a layer of tolerance for the host.

Outstanding questions

Do different combinations of PAMPs and DAMPs selectively modulate the metabolism of macrophages as a way to 'tailor' the response towards the perceived insult?

How fundamental is mitochondrial respiration in macrophages during bacterial infection and to what extent can it replace or be replaced by glycolysis as an ATP source?

Are pathogen-elicited metabolic changes to macrophage metabolism the result of an active process mediated exclusively by bacterium-derived factors, a consequence of metabolic competition between macrophage and bacterium, or a combination of the two?

If certain bacterial species have adapted to macrophage-derived itaconate to use it as a carbon source, would it be beneficial to inhibit the pre-programmed production of itaconate in macrophages as a way to stave off infections with such pathogens?

Can metabolic inhibitors/activators be explored in a context of adjuvant therapies during bacterial infections as a way to promote host cell fitness while impairing bacterial cell survival?

Do pathogenic bacteria 'hijack' tolerance-promoting pathways to delay or bypass immune responses?

It is known that intestinal commensal microbial species produce metabolites, such as spermidine and short-chain fatty acids [91–93], that have anti-inflammatory properties once sensed by various host cells. Likewise, it is possible that certain pathogenic bacteria 'hijack' such metabolite-signaling pathways to delay or bypass immune responses.

Acknowledgments

This work was financially supported by the German Research Foundation (Project Number 318346496, SFB1292/2 TP18) to T.S. This work was also financially supported by the Wilhelm-Sander Foundation (Project ID: 2023.041.1) to L.A. and T.S.

Declaration of interests

No interests are declared.

References

1. Casanova-Acebes, M. *et al.* (2018) Neutrophils instruct homeostatic and pathological states in naive tissues. *J. Exp. Med.* 215, 2778–2795
2. De Filippo, K. and Rankin, S.M. (2020) The secretive life of neutrophils revealed by intravital microscopy. *Front. Cell Dev. Biol.* 8, 603230
3. Nobs, S.P. and Kopf, M. (2021) Tissue-resident macrophages: guardians of organ homeostasis. *Trends Immunol.* 42, 495–507
4. Almeida, L. *et al.* (2021) $CD4^+$ T-cell differentiation and function: unifying glycolysis, fatty acid oxidation, polyamines NAD mitochondria. *J. Allergy Clin. Immunol.* 148, 16–32
5. Almeida, L. *et al.* (2016) Metabolic pathways in T cell activation and lineage differentiation. *Semin. Immunol.* 28, 514–524
6. Caro-Maldonado, A. *et al.* (2014) Metabolic reprogramming is required for antibody production that is suppressed in anergic but exaggerated in chronically BAFF-exposed B cells. *J. Immunol.* 192, 3626–3636
7. Doughty, C.A. *et al.* (2006) Antigen receptor-mediated changes in glucose metabolism in B lymphocytes: role of phosphatidylinositol 3-kinase signaling in the glycolytic control of growth. *Blood* 107, 4458–4465
8. Raza, I.G.A. and Clarke, A.J. (2021) B cell metabolism and autophagy in autoimmunity. *Front. Immunol.* 12, 681105
9. Galli, G. and Saleh, M. (2021) Immunometabolism of macrophages in bacterial infections. *Front. Cell. Infect. Microbiol.* 10, 607650
10. Wu, Y. and Hirschi, K.K. (2021) Tissue-resident macrophage development and function. *Front. Cell Dev. Biol.* 8, 617879
11. Hoeffel, G. and Ginhoux, F. (2018) Fetal monocytes and the origins of tissue-resident macrophages. *Cell. Immunol.* 330, 5–15
12. Wculek, S.K. *et al.* (2022) Metabolism of tissue macrophages in homeostasis and pathology. *Cell. Mol. Immunol.* 19, 384–408
13. Li, D. and Wu, M. (2021) Pattern recognition receptors in health and diseases. *Signal Transduct. Target. Ther.* 6, 291
14. Arango Duque, G. and Descoteaux, A. (2014) Macrophage cytokines: involvement in immunity and infectious diseases. *Front. Immunol.* 5, 491
15. Wen, Y. *et al.* (2021) Macrophage heterogeneity in kidney injury and fibrosis. *Front. Immunol.* 12, 681748
16. Pizzolato, G. *et al.* (2019) Single-cell RNA sequencing unveils the shared and the distinct cytotoxic hallmarks of human TCRVδ1 and TCRVδ2 γδ T lymphocytes. *Proc. Natl. Acad. Sci. U. S. A.* 116, 11906–11915
17. Ruytinx, P. *et al.* (2018) Chemokine-induced macrophage polarization in inflammatory conditions. *Front. Immunol.* 9, 1930
18. Benoit, M. *et al.* (2008) Macrophage polarization in bacterial infections. *J. Immunol.* 181, 3733–3739
19. Thiriot, J.D. *et al.* (2020) Hacking the host: exploitation of macrophage polarization by intracellular bacterial pathogens. *Pathog. Dis.* 78, ftaa009
20. Lachmandas, E. *et al.* (2016) Microbial stimulation of different Toll-like receptor signalling pathways induces diverse metabolic programmes in human monocytes. *Nat. Microbiol.* 2, 16246
21. Freemerman, A.J. *et al.* (2014) Metabolic reprogramming of macrophages. *J. Biol. Chem.* 289, 7884–7896
22. Murugina, N.E. *et al.* (2020) Glycolytic reprogramming of macrophages activated by NOD1 and TLR4 agonists: no association with proinflammatory cytokine production in normoxia. *J. Biol. Chem.* 295, 3099–3114
23. Wang, F. *et al.* (2018) Interferon gamma induces reversible metabolic reprogramming of M1 macrophages to sustain cell viability and pro-inflammatory activity. *EBioMedicine* 30, 303–316
24. Fukuzumi, M. *et al.* (1996) Endotoxin-induced enhancement of glucose influx into murine peritoneal macrophages via GLUT1. *Infect. Immun.* 64, 108–112
25. McGettrick, A.F. and O'Neill, L.A.J. (2020) The role of HIF in immunity and inflammation. *Cell Metab.* 32, 524–536
26. Li, C. *et al.* (2018) HIF1α-dependent glycolysis promotes macrophage functional activities in protecting against bacterial and fungal infection. *Sci. Rep.* 8, 3603
27. Flannagan, R.S. *et al.* (2012) The cell biology of phagocytosis. *Annu. Rev. Pathol.* 7, 61–98
28. Westman, J. and Grinstein, S. (2021) Determinants of phagosomal pH during host–pathogen interactions. *Front. Cell Dev. Biol.* 8, 624958
29. Haas, A. (2007) The phagosome: compartment with a license to kill. *Traffic* 8, 311–330
30. Fairn, G.D. and Grinstein, S. (2012) How nascent phagosomes mature to become phagolysosomes. *Trends Immunol.* 33, 397–405
31. Kissing, S. *et al.* (2015) Vacuolar ATPase in phagosome–lysosome fusion. *J. Biol. Chem.* 290, 14166–14180
32. Gutiérrez, S. *et al.* (2021) *Salmonella* Typhimurium impairs glycolysis-mediated acidification of phagosomes to evade macrophage defense. *PLoS Pathog.* 17, e1009943
33. Wang, Y. *et al.* (2021) Mitochondrial metabolism regulates macrophage biology. *J. Biol. Chem.* 297, 100904
34. Viola, A. *et al.* (2019) The metabolic signature of macrophage responses. *Front. Immunol.* 10, 1462
35. Ryan, D.G. and O'Neill, L.A.J. (2020) Krebs cycle reborn in macrophage immunometabolism. *Annu. Rev. Immunol.* 38, 289–313
36. Mills, E.L. and O'Neill, L.A. (2016) Reprogramming mitochondrial metabolism in macrophages as an anti-inflammatory signal. *Eur. J. Immunol.* 46, 13–21
37. Huang, S.C.-C. *et al.* (2014) Cell-intrinsic lysosomal lipolysis is essential for alternative activation of macrophages. *Nat. Immunol.* 15, 846–855
38. Divakaruni, A.S. *et al.* (2018) Etomoxir inhibits macrophage polarization by disrupting CoA homeostasis. *Cell Metab.* 28, 490–503.e7
39. Namgaladze, D. and Brüne, B. (2014) Fatty acid oxidation is dispensable for human macrophage IL-4-induced polarization. *Biochim. Biophys. Acta* 1841, 1329–1335
40. Michelucci, A. *et al.* (2013) Immune-responsive gene 1 protein links metabolism to immunity by catalyzing itaconic acid production. *Proc. Natl. Acad. Sci. U. S. A.* 110, 7820–7825
41. Jha, A.K. *et al.* (2015) Network integration of parallel metabolic and transcriptional data reveals metabolic modules that regulate macrophage polarization. *Immunity* 42, 419–430
42. Zhang, Z. *et al.* (2022) Itaconate is a lysosomal inducer that promotes antibacterial innate immunity. *Mol. Cell* 82, 2844–2857.e10
43. McFadden, B.A. and Purohit, S. (1977) Itaconate, an isocitrate lyase-directed inhibitor in *Pseudomonas indigofera*. *J. Bacteriol.* 131, 136–144

44. Hersch, S.J. and Navarre, W.W. (2020) The *Salmonella* LysR family regulator RipR activates the SPI-13-encoded itaconate degradation cluster. *Infect. Immun.* 88, e00303–20
45. Wang, H. *et al.* (2019) An essential bifunctional enzyme in *Mycobacterium tuberculosis* for itaconate dissimilation and leucine catabolism. *Proc. Natl. Acad. Sci. U. S. A.* 116, 15907–15913
46. Kohl, L. *et al.* (2023) Macrophages inhibit *Coxiella burnetii* by the ACOD1–itaconate pathway for containment of Q fever. *EMBO Mol. Med.* 15, e15931
47. Riquelme, S.A. *et al.* (2020) *Pseudomonas aeruginosa* utilizes host-derived itaconate to redirect its metabolism to promote biofilm formation. *Cell Metab.* 31, 1091–1106.e6
48. Sasikaran, J. *et al.* (2014) Bacterial itaconate degradation promotes pathogenicity. *Nat. Chem. Biol.* 10, 371–377
49. Chen, P. *et al.* (2017) Gpr132 sensing of lactate mediates tumor–macrophage interplay to promote breast cancer metastasis. *Proc. Natl. Acad. Sci. U. S. A.* 114, 580–585
50. Tao, H. *et al.* (2023) Unveiling the veil of lactate in tumor-associated macrophages: a successful strategy for immunometabolic therapy. *Front. Immunol.* 14, 1208870
51. Lopez Krol, A. *et al.* (2022) Lactate induces metabolic and epigenetic reprogramming of pro-inflammatory Th17 cells. *EMBO Rep.* 23, e54685
52. Cordes, T. *et al.* (2016) Immunoresponsive gene 1 and itaconate inhibit succinate dehydrogenase to modulate intracellular succinate levels. *J. Biol. Chem.* 291, 14274–14284
53. Lampropoulou, V. *et al.* (2016) Itaconate links inhibition of succinate dehydrogenase with macrophage metabolic remodeling and regulation of inflammation. *Cell Metab.* 24, 158–166
54. Langston, P.K. *et al.* (2019) Glycerol phosphate shuttle enzyme GPD2 regulates macrophage inflammatory responses. *Nat. Immunol.* 20, 1186–1195
55. Kelly, B. *et al.* (2015) Metformin inhibits the production of reactive oxygen species from NADH:ubiquinone oxidoreductase to limit induction of interleukin-1β (IL-1β) and boosts interleukin-10 (IL-10) in lipopolysaccharide (LPS)-activated macrophages. *J. Biol. Chem.* 290, 20348–20359
56. Jo, E.-K. *et al.* (2016) Molecular mechanisms regulating NLRP3 inflammasome activation. *Cell. Mol. Immunol.* 13, 148–159
57. Billingham, L.K. *et al.* (2022) Mitochondrial electron transport chain is necessary for NLRP3 inflammasome activation. *Nat. Immunol.* 23, 692–704
58. Imai, S. and Guarente, L. (2014) NAD^+ and sirtuins in aging and disease. *Trends Cell Biol.* 24, 464–471
59. Chen, S. *et al.* (2020) Epigenetic regulation of macrophages: from homeostasis maintenance to host defense. *Cell. Mol. Immunol.* 17, 36–49
60. Niu, Y. *et al.* (2022) Epigenetic modifications in tumor-associated macrophages: a new perspective for an old foe. *Front. Immunol.* 13, 836223
61. Daskalaki, M.G. *et al.* (2018) Histone methylation and acetylation in macrophages as a mechanism for regulation of inflammatory responses. *J. Cell. Physiol.* 233, 6495–6507
62. Willenborg, S. *et al.* (2021) Mitochondrial metabolism coordinates stage-specific repair processes in macrophages during wound healing. *Cell Metab.* 33, 2398–2414.e9
63. Kuang, H. *et al.* (2023) Humoral regulation of iron metabolism by extracellular vesicles drives antibacterial response. *Nat. Metab.* 5, 111–128
64. Szeligowski, R.V. *et al.* (2022) Molecular evolution of IRG1 shapes itaconate production in metazoans and alleviates the "double-edged dilemma" of innate immune defense. *bioRxiv* Published online June 21, 2022. https://doi.org/10.1101/2022.06.17.496652
65. Németh, B. *et al.* (2016) Abolition of mitochondrial substrate-level phosphorylation by itaconic acid produced by LPS-induced Irg1 expression in cells of murine macrophage lineage. *FASEB J.* 30, 286–300
66. Ki, N. *et al.* (2022) Isocitrate binds to the itaconic acid-responsive LysR-type transcriptional regulator RipR in *Salmonella* pathogenesis. *J. Biol. Chem.* 298, 102562
67. Tomlinson, K.L. *et al.* (2021) *Staphylococcus aureus* induces an itaconate-dominated immunometabolic response that drives biofilm formation. *Nat. Commun.* 12, 1399
68. Tomlinson, K.L. *et al.* (2023) *Staphylococcus aureus* stimulates neutrophil itaconate production that suppresses the oxidative burst. *Cell Rep.* 42, 112064
69. Singh, S. *et al.* (2021) Integrative metabolomics and transcriptomics identifies itaconate as an adjunct therapy to treat ocular bacterial infection. *Cell Rep. Med.* 2, 100277
70. Nair, S. *et al.* (2018) *Irg1* expression in myeloid cells prevents immunopathology during *M. tuberculosis* infection. *J. Exp. Med.* 215, 1035–1045
71. McGettrick, A.F. and O'Neill, L.A. (2023) Two for the price of one: itaconate and its derivatives as an anti-infective and anti-inflammatory immunometabolite. *Curr. Opin. Immunol.* 80, 102268
72. Ferreira, A.V. *et al.* (2023) Dimethyl itaconate induces long-term innate immune responses and confers protection against infection. *Cell Rep.* 42, 112658
73. Zhu, X. *et al.* (2021) Itaconic acid exerts anti-inflammatory and antibacterial effects via promoting pentose phosphate pathway to produce ROS. *Sci. Rep.* 11, 18173
74. Bowden, S.D. *et al.* (2009) Glucose and glycolysis are required for the successful infection of macrophages and mice by *Salmonella enterica* serovar Typhimurium. *Infect. Immun.* 77, 3117–3126
75. Eisele, N.A. *et al.* (2013) *Salmonella* require the fatty acid regulator PPARδ for the establishment of a metabolic environment essential for long term persistence. *Cell Host Microbe* 14, 171–182
76. Brunmair, B. *et al.* (2006) Activation of PPAR-δ in isolated rat skeletal muscle switches fuel preference from glucose to fatty acids. *Diabetologia* 49, 2713–2722
77. Ganesan, R. *et al.* (2017) *Salmonella* Typhimurium disrupts Sirt1/AMPK checkpoint control of mTOR to impair autophagy. *PLoS Pathog.* 13, e1006227
78. Huang, J. and Manning, B.D. (2008) The TSC1–TSC2 complex: a molecular switchboard controlling cell growth. *Biochem. J.* 412, 179–190
79. Agarwal, S. *et al.* (2015) AMP-activated protein kinase (AMPK) control of mTORC1 Is p53- and TSC2-independent in pemetrexed-treated carcinoma cells. *J. Biol. Chem.* 290, 27473–27486
80. Gwinn, D.M. *et al.* (2008) AMPK phosphorylation of raptor mediates a metabolic checkpoint. *Mol. Cell* 30, 214–226
81. Chen, S. *et al.* (2020) Serine supports IL-1β production in macrophages through mTOR signaling. *Front. Immunol.* 11, 1866
82. Jiang, L. *et al.* (2021) *Salmonella* Typhimurium reprograms macrophage metabolism via T3SS effector SopE2 to promote intracellular replication and virulence. *Nat. Commun.* 12, 879
83. Cumming, B.M. *et al.* (2018) *Mycobacterium tuberculosis* induces decelerated bioenergetic metabolism in human macrophages. *Elife* 7, e39169
84. Hackett, E.E. *et al.* (2020) *Mycobacterium tuberculosis* limits host glycolysis and IL-1β by restriction of PFK-M via microRNA-21. *Cell Rep.* 30, 124–136.e4
85. Singh, V. *et al.* (2012) *Mycobacterium tuberculosis*-driven targeted recalibration of macrophage lipid homeostasis promotes the foamy phenotype. *Cell Host Microbe* 12, 669–681
86. Gutierrez, M.G. *et al.* (2004) Autophagy is a defense mechanism inhibiting BCG and *Mycobacterium tuberculosis* survival in infected macrophages. *Cell* 119, 753–766
87. Berod, L. *et al.* (2014) *De novo* fatty acid synthesis controls the fate between regulatory T and T helper 17 cells. *Nat. Med.* 20, 1327–1333
88. Stüve, P. *et al.* (2018) *De novo* fatty acid synthesis during mycobacterial infection is a prerequisite for the function of highly proliferative T cells, but not for dendritic cells or macrophages. *Front. Immunol.* 9, 495
89. Hayek, I. *et al.* (2022) *Coxiella burnetii* affects HIF1α accumulation and HIF1α target gene expression. *Front. Cell. Infect. Microbiol.* 12, 867689
90. Hayek, I. *et al.* (2019) Limitation of TCA cycle intermediates represents an oxygen-independent nutritional antibacterial effector mechanism of macrophages. *Cell Rep.* 26, 3502–3510.e6
91. Carriche, G.M. *et al.* (2021) Regulating T-cell differentiation through the polyamine spermidine. *J. Allergy Clin. Immunol.* 147, 335–348.e11
92. Li, M. *et al.* (2018) Pro- and anti-inflammatory effects of short chain fatty acids on immune and endothelial cells. *Eur. J. Pharmacol.* 831, 52–59
93. Vijay, A. *et al.* (2021) The anti-inflammatory effect of bacterial short chain fatty acids is partially mediated by endocannabinoids. *Gut Microbes* 13, 1997559