Review

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Protozoan parasites and type I interferons: a cold case reopened

Daniel P. Beiting

Department of Pathobiology, University of Pennsylvania, Philadelphia, PA, USA

Protozoan parasites, such as *Plasmodium*, *Toxoplasma*, *Cryptosporidium*, trypanosomes, and *Leishmania*, are a major cause of disease in both humans and other animals, highlighting the need to understand the full spectrum of strategies used by the host immune system to sense and respond to parasite infection. Although type II interferon (IFN-γ) has long been recognized as an essential antiparasite immune effector, much less is known about the role of type I interferons (IFN-α and -β) in host defense, particularly *in vivo*. Recent studies are reviewed which collectively highlight that type I IFN can be induced in response to parasite infection and influence the outcome of infection.

IFNs in host defense

IFNs were first discovered in the late 1950s as soluble effector molecules released into the supernatant of virus-infected cell cultures [1]. When transferred to new cells, this supernatant 'interfered' with viral replication, thereby garnering the name interferon. In the decades since their discovery IFNs have been intensively studied for their antiviral activity, resulting in the identification of three classes of molecules that are distinguished by function and receptor usage. Type I IFNs – the original interfering factor in Isaacs and Lindenmann's experiments – consist of many separate molecules, encoded by distinct genes, which are expressed by an abundance of cell types in response to viral infection. The ability of most cells to both produce and respond to type I IFN allows autocrine and paracrine effects in virtually any tissue. By contrast, type II IFN consists only of single member, IFN-γ, which is produced by a limited range of cell types, with T cells and natural killer cells acting as the major source, and which has proven to be crucial for host defense to intracellular pathogens. Much less is known about the recently identified type III IFNs (IFN-λ), but they appear to elicit similar responses to type I IFN [2]; however, the expression of their receptor is more restricted [3,4].

Type I and II IFNs elicit similar but distinct intracellular signaling cascades after binding to their receptors. Type I IFNs activate the receptor-associated kinases, JAK1 (Janus kinase) and TYK2 (tyrosine kinase 2), leading to phosphorylation and subsequent heterodimerization of cytosolic transcription factors, STAT1 (signal transducer and activator of transcription) and STAT2, which then interact with the transcription factor, IRF9 (IFN regulatory factor 9), to form a complex termed the IFN-stimulated gene factor 3 (ISGF3) [5]. ISGF3 translocates to the nucleus where it binds to IFN-stimulated response elements (ISRE) composed of a YAGTTTC(A/T)YTTTYCC motif [6] that is found in the promoters of IFN-stimulated genes (ISGs). By contrast, type II IFN activates JAK1 and JAK2, leading to STAT1 homodimer formation, nuclear translocation, and binding to γ-activated sequences (GAS) containing a $TTCN_{2\text{-}4}GAA$ motif [7].

In the same way as type I IFN has been extensively explored in the context of viral infections, protozoan infections have proven to be an important context in which to understand type II IFN. The potent antiviral activity of type I IFN, taken together with early studies showing weak or no activity of type I IFN on parasites or bacteria *in vitro*, and the finding that type II IFN is essential in immunity to *Toxoplasma gondii* [8,9], *Cryptosporidium parvum* [10], *Leishmania major* [11], and *Trypanosoma cruzi* [12], has led to the dichotomous view that type I IFN is antiviral but is of little importance during parasite infections, where type II IFN responses dominate. Recent data suggest that this dichotomy is overly simplistic. As genome-scale approaches such as transcriptomics are leveraged to interrogate the host–parasite relationship, investigators are capturing a more complete picture of the innate immune pathways activated during parasite infection. Consequently, it is becoming apparent that protozoa, similarly to viruses, are inducers of type I IFN [13], raising questions about how this pathway might contribute to parasite control and disease pathogenesis. The significance of type I IFN signaling is discussed below for specific parasitic diseases.

Plasmodium falciparum

Plasmodium falciparum initiates human infection after transmission by its mosquito vector. Sporozoite-stage parasites injected into the skin during mosquito feeding rapidly enter circulation and travel to the liver, where they infect hepatocytes and initiate an asymptomatic phase of infection. Over the course of a few weeks, individual sporozoites develop into exoerythrocytic forms and undergo rapid cell division and differentiation, eventually giving rise to thousands of merozoites [14]. Once released from the liver, merozoites invade red blood cells, initiating the symptomatic phase of infection characterized by cyclical fevers and, in some cases, life-threatening anemia, seizures, and coma [15].

Corresponding author: Beiting, D.P. (beiting@vet.upenn.edu).
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Although liver-stage *Plasmodium* infection has long been described as clinically silent [14], it is not immunologically silent, and recent studies reveal that type I IFNs play an important role in regulating this stage of infection [16–18]. Portugal and colleagues carried out transcriptomic analysis of livers from infected mice [17], and observed predominantly a type I IFN signature, including upregulation of *Mx2* and *Ifit1* – genes known to be rapidly induced during viral infection and functionally antiviral [19,20]. Through a series of elegant experiments, Liehl *et al*. went on to show that the type I IFN response in the liver is evident within 36 h of infection from mosquitoes. This response required the receptor for type I IFN (IFNAR), suggesting that the signature is amplified by an autocrine feed-forward loop in hepatocytes – that is, liver cells secrete and respond to their own IFN, resulting in increased IFN production. IFNAR-deficient mice, or mice selectively lacking IFNAR on hepatocytes (thereby blocking this autocrine feedback), fail to control parasite replication in the liver, and consequently accumulate a higher parasite burden in the liver and blood [16,18]. Interestingly, unlike type II IFN, which elicits strong cell-autonomous killing of intracellular parasites, including *Plasmodium* liver-stage parasites [21,22], hepatocyte type I IFN did not promote intrinsic control of parasite replication. Rather, autocrine type I IFN signaling in the liver recruits natural killer T cells, which then mediate parasite killing by producing type II IFN [18]. The importance of type I IFN in limiting liver-stage *Plasmodium* could have important clinical implications for treating malaria, particularly because restriction of parasites in the liver is a major goal of malaria vaccines currently in Phase III clinical trials [23,24]. Moreover, type I IFNs have long been used as safe and effective treatment of viral infections in the liver [25]. Although liver infection is transient with *P. falciparum*, other species, such as *P. vivax*, establish a persistent liver infection which serves as a reservoir of parasites resulting in chronic blood infections [26,27]. In light of the high morbidity caused by relapsing *P. vivax*, and the fact that it causes of one of the most prevalent human malaria infections on the planet [28], it will be important to explore whether type I IFN can limit either the frequency or intensity of relapsing disease with *P. vivax*.

P. falciparum blood-stage infection also induces a type I IFN signature in human peripheral blood mononuclear cells, which resolves following chemotherapy [29], and similar type I IFN responses have been observed in *Plasmodium*-infected mice [29–33]. Using a *P. berghei* mouse model of severe malaria, Sharma and colleagues showed that animals deficient in type I IFN signaling survive an otherwise lethal infection [29], implicating type I IFN in disease pathogenesis. These data contrast with the host-protective role described for type I IFN during liver-stage infection [16,18], and suggest that type I IFN can have contrasting roles, perhaps as parasites move through their life cycle, migrate to distinct tissues or cell types, or trigger different inflammatory programs.

Toxoplasma gondii

Toxoplasma is an orally transmitted parasite that initiates infection in the gastrointestinal tract, and subsequently disseminates via blood circulation to all major organs and tissues. In most healthy hosts, a robust immune response is mounted that effectively controls parasite replication, but some parasites evade this response, differentiate to a slowly dividing stage, and establish a latent infection in the central nervous system. Although the genus *Toxoplasma* contains only one species, *T. gondii*, sexual recombination has resulted in strains that vary dramatically in virulence and host immune activation. The population genetic structure of *Toxoplasma* is dominated by three archetypal lineages – termed types I, II, and III – but 'atypical' strains that fall outside of this classification are also found. A transcriptomic survey of the host response to 29 different *T. gondii* strains revealed that a subset of atypical strains induce a type I IFN response in macrophages and fibroblasts [34]. Similarly, another recent study confirmed that canonical strains do not induce type I IFN, but showed that a close relative of *Toxoplasma*, *Neospora caninum*, activates a robust type I IFN response that is sufficient to control a viral challenge *in vitro* [35].

Whether *Toxoplasma* strains or *Neospora* induce type I IFN *in vivo* and what, if any, consequence this might have for parasite replication or immune pathology, remain open questions. Early *in vitro* studies with recombinant type I IFN showed either weak or no effect on parasite replication, in contrast to type II IFN, which mediates potent cell-intrinsic immunity [36–38]. Similarly, infection of IFNAR-deficient mice with a canonical type II strain showed no impact on acute infection [36], but this study did not allow infections to progress to the chronic phase. Interestingly, both *Neospora* and atypical *Toxoplasma* strains are associated with disease in immunocompetent hosts and demonstrate a propensity for vertical transmission from mother to fetus [39–42]. Moreover, studies in mice have shown that impaired type II IFN leads to enhanced vertical transmission in mouse model of transplacental toxoplasmosis [43], suggesting a possible scenario in which atypical strains and/or *Neospora* induce type I IFN, resulting in repression of type II IFN and more severe disease. Finally, given the recent findings that type I IFN has immunomodulatory effects during chronic viral and bacterial infections, it will be important to test whether signaling through this pathway influences latent *Toxoplasma* infection in the brain.

The observation that only a few atypical strains, but no canonical strains of *Toxoplasma*, induce type I IFN *in vitro* suggests that the majority of strains either lack the molecular signature to induce this response or that they have evolved additional mechanisms to suppress type I IFN signaling. Surprisingly, the addition of heat-killed, rather than live, *Toxoplasma* to cultures resulted in robust type I IFN signaling by representative members of each archetypal lineage, suggesting that *Toxoplasma* is capable of triggering type I IFN [35]. To test whether live *Toxoplasma* suppress this response, Beiting *et al.* carried out competition assays between *Toxoplasma* and *Neospora*, both of which infect the same cells, and found that *Neospora* could no longer induce type I IFN in cells previously infected with *Toxoplasma*. Collectively, these results demonstrate that *Toxoplasma* possesses both a type I IFN-inducing factor, as well as mechanism to prevent the innate

response to this factor. Furthermore, Rosowski *et al.* showed that although *Toxoplasma* infected cells stimulated with IFN-β appropriately activate and translocate STAT1 to the nucleus, STAT1 is retained on the DNA, failing to recycle and resulting in a poor induction of STAT1 target genes [44]. Collectively, these results show that *Toxoplasma* has evolved strategies for limiting both the induction of type I IFN and the ability of type I IFN to activate STAT1-dependent transcription. One interpretation of these results is that there may be a selective fitness advantage for *Toxoplasma* strains to limit type I IFN responses, and similar suppressive capacity has been shown for *Toxoplasma* and type II IFN signaling [45–49].

Leishmania *and trypanosomes*

Leishmania spp. are transmitted by the bite of a sand fly and are subsequently taken up by macrophages, where they differentiate to a rapidly dividing form and continue a lytic cycle of replication, ultimately resulting in disease ranging from mild to severe. The most common form of the disease, cutaneous leishmaniasis, is characterized by the development of an ulcerated skin lesion at the site of infection. In a mouse model of cutaneous leishmaniasis, Diefenbach and colleagues showed that type I IFN is a crucial component of the innate immune response to *Leishmania major* infection in mice [50,51]. Soon after infection, macrophages produce type I IFN, which is required for nitric oxide production and control of parasite replication. Consequently, mice lacking IFNAR develop non-healing lesions [51]. Moreover, the administration of recombinant type I IFN is sufficient to rescue a susceptible mouse strain from lethal infection [52]. Interestingly, *Leishmania*, similarly to *Toxoplasma*, appears to have evolved a mechanism to counter this innate response. Parasite infection activates macrophage 4E-BP1 (eukaryotic translation initiation factor 4E binding protein 1), a translational repressor, previously shown to be a strong negative regulator type I IFN production through repression of IRF7 expression, an important regulator of type I IFN [53]. Blocking parasite activation of 4E-BP1 during infection, either through chemical or genetic manipulation, restores host cell translation and type I IFN production, markedly impairing parasite replication *in vitro* and *in* vivo [54]. Little is known about *Leishmania* infection and type I IFN in human infections. Genomic profiling of human *L. braziliensis* skin lesions showed induction of type I IFN signaling in both early- (non-ulcerated) and late-stage lesions [55]. However, the nitric oxide-dependent protective effect of type I IFN described in mice may not pertain to humans, where reactive oxygen species, rather than nitric oxide, appear to be important for limiting parasite growth [56]. Furthermore, there is evidence that type I IFN suppresses reactive oxygen species during *Leishmania* infection [57].

Trypanosoma cruzi and *Trypanosoma brucei*, the causative agents of Chagas disease and African sleeping sickness, respectively, are also introduced by an insect vector and invade through the skin. *T. cruzi* is capable of infecting virtually any nucleated cell and, similarly to *Leishmania*, *in vitro* and *in vivo* infection triggers an early and robust type I IFN response [58–60], the magnitude of which correlates with parasite virulence [58]. Infection of IFNAR-deficient mice with *T. cruzi* or *T. brucei* showed that type I IFN signaling had either a modest or no role in parasite restriction [61–63]. In other contexts, perhaps owing to parasite dose, strain used, or route of infection, type I IFN appears to increase disease susceptibility during *T. cruzi* infection by suppressing type II IFN [61,63,64].

Redundancy and crosstalk between type I IFN and type II IFN

Protozoan parasites are generally recognized as potent inducers of type II IFN. The growing appreciation that they can also induce type I IFN provides an opportunity to address fundamental questions in immunoparasitology: are the roles of these two cytokines redundant during parasite infection? How does 'crosstalk' between type I and type II IFN alter the course of infection? Answers to these questions are beginning to emerge. For example, recent studies have shown that, like liver-stage infection, *Plasmodium* blood-stage infection also induces a rapid type I IFN signature in mice [30–33], and this occurs at a time when type II IFN is also produced in abundance [30]. In a carefully conducted study, Kim and colleagues utilized $Ifnar^{-/-}$, $Ifngr^{-/-}$, or mice lacking both receptors, combined with transcriptomics of whole blood, to dissect the contributions of type I and type II IFN signaling to immunity, and found significant redundancy in their ability to regulate target gene expression and limit infection [30]. This finding may explain why other groups report only a modest impact on parasitemia in mice singly deficient in IFNAR [31,36,65]. Additional studies are needed across multiple parasite infections to understand better the extent of functional redundancy when these two cytokines are coproduced. Interestingly, population genetics studies in humans have shown that type II IFN and numerous members of the type I IFN family show strong purifying selection against deleterious mutations, suggesting nonredundant, essential roles for both cytokines [66,67].

Type I IFN and type II IFN have pleiotropic effects on multiple cell types, raising the possibility of crosstalk occurring during protozoan infections. Under homeostatic conditions, this crosstalk is thought to be beneficial because basal type I IFN production primes cells to respond appropriately to both IFN-α/β and -γ [68,69]. The importance of IFN cross-priming extends to immune responses as well, where type I IFN has been shown to promote innate cell production of type II IFN [70,71]. Indeed, this has been suggested to be the mechanistic basis for administration of recombinant type I IFN leading to enhanced parasite control [37,72]. Paradoxically, type I IFN can also repress type II IFN signaling. During infection with the Gram-positive bacterium, *Listeria monocytogenes*, type I IFN increases the susceptibility of mice by downregulating macrophage expression of the type II IFN receptor [73,74]. Immunomodulation of type II IFN signaling by type I IFN could have important implications for protozoan infections, which, similarly to *Listeria*, require a strong type II IFN response to eradicate infection. Consistent with this notion, Haque *et al.* found that type I IFN increased susceptibility of mice during *Plasmodium* blood-stage infection, and IFNAR-deficient mice are protected from lethal infection. The authors went on to show that type I IFN signaling

suppressed the ability of dendritic cells to prime T cells to produce type II IFN [33,75]. This suppressive effect has also been reported in human infections with *Mycobacterium leprae*, where low type I IFN signaling in the skin is associated with high type II IFN and self-healing lesions, whereas high type I IFN is associated with low type II IFN and disseminated leprosy [76]. Similar findings have even been described in viral infections. Chronic lymphocytic choriomeningitis (LCMV) infection is associated with high levels of type I IFN. Remarkably, when LCMV-infected mice were treated with neutralizing antibody to type I IFN during chronic infection, CD4 T cell function and type II IFN production were enhanced and virus was cleared [77,78].

The factors that determine whether type I IFN will have a host-protective or pathogenic role during parasite infection are not fully understood. Interestingly, Mattner *et al.* have reported that low, but not high, doses of recombinant type I IFN are protective during *Leishmania* infection [50,52], whereas studies in viral infections show that the timing and duration type I IFN production are important [79,80]. Experiments with *Listeria* suggest that the route of infection can also dictate whether type I IFN plays a host-protective role, with type I IFN signaling proving essential for survival when infection is initiated via the oral, but not systemic, routes [81]. Collectively, these studies demonstrate that type I IFN can have immunomodulatory effects on type II IFN signaling, that these effects are observed broadly across different types of infections, and that the timing, duration and/or amplitude of type I IFN signaling may be important determinants in the outcome of disease. Considering that many protozoan parasites are endemic in areas of the world where coinfections with viruses would be commonplace, there is a great need to understand better how the immunomodulatory properties of type I IFN might influence parasite infections.

Molecular basis for type I IFN induction during parasite infection

The observation that evolutionarily divergent parasite species trigger type I IFN raises questions about what conserved parasite-associated molecular patterns (PAMPs) are being recognized, and by which host cell receptors. Sensing of viruses is well studied, occurring via either transmembrane Toll-like receptors (TLR), such as TLR3, TLR7, and TLR9, or by cytosolic molecules such as retinoic acid-inducible gene-1 (RIG-I)-like receptors (RLRs), nucleotide oligomerization domain (NOD)-like receptors (NLRs), and absent in melanoma-2 (AIM2)-like receptors (ALRs) [82]. Recent studies suggest that several of these pathways are engaged by parasites as well (Figure 1), and that the exact pathway involved depends on host cell type. For example, non-phagocytic cells, such as fibroblasts, require the cytosolic sensor, RIG-I, to produce type I IFN in response to *Toxoplasma* infection (Figure 1B) [34]. By contrast, plasmacytoid dendritic cells produce low levels of type I IFN after recognition of *Toxoplasma* profilin by TLR12 [83]. In macrophages, activation of type I IFN signaling by *Toxoplasma* and *Neospora* requires TLR3 and the adapter protein TRIF (Toll-like receptor adaptor molecule) [34,35] (Figure 1C). TLR3 was initially identified as a cellular sensor for viral double stranded RNA [84]. Consistent with this role, transfection of macrophages with purified parasite RNA is sufficient to trigger TLR3-dependent type I IFN production [34,35]. TLR3 localizes to the host cell endosomal compartment, but *Toxoplasma* and *Neospora* reside in a non-fusogenic vacuole in the host cell cytosol, raising questions about how receptor and ligand would have the opportunity to interact. It appears that TLR3 is engaged when parasites are either phagocytosed or invade cells but are killed (Figure 1C), leading to fusion with endosomes and acidification [34,35]. In the case of leishmania, actual virus-derived molecules may be responsible for type I IFN induction. Viruses are known to infect various parasitic protozoa [85], and some strains of *Leishmania* harbor a RNA virus belonging to the *Totiviridae* family [86,87]. Compared to strains without virus, leishmania that harbor viruses are associated with more severe disease and activate inflammatory gene expression, including type I IFN, in a TLR3-dependent manner [88,89] (Figure 1E). Taken together with studies showing that TLR3 is capable of recognizing double stranded RNA from helminth parasites [90], as well as damaged self RNA [91], these data indicate that TLR3 has evolved to broadly recognize RNA from phylogenetically diverse species, including parasites.

Two different PAMP–receptor interactions leading to type I IFN responses have been described for *Plasmodium* infection. Liehl *et al.* report that sensing of *P. berghei* sporozoites in the liver requires recognition of parasite RNA by the cytosolic RIG-I-like receptor, MDA5 (melanoma-differentiation associated protein 5), and its adapter, MAVS (mitochondrial antiviral signaling protein) [16] (Figure 1A). By contrast, Miller *et al.* found that MDA5, MAVS, and TLR3 were not necessary to mount a protective type I IFN response to *P. yoelii* in the liver [18], suggesting either that a novel pathway may be involved, or that parasites might be sensed through multiple redundant pathways. The discrepancy in these findings could be explained the use different parasite species. A second PAMP–receptor interaction was identified in studies of blood-stage *P. falciparum*. In an elegant study by Sharma and colleagues [29], the AT-rich motif, ATTTTTAC, which is present over 6000 times in the *P. falciparum* genome, was found to form stem-loop structures that are recognized by the cytosolic DNA sensor, stimulator of IFN genes (STING) and Tank-binding kinase 1 (TBK1). This is proposed to occur when parasite hemozoin – which has been shown to be coated in parasite DNA [92,93] – is released during parasite lysis of red blood cells and is subsequently taken up by phagocytes (Figure 1D). Hemozoin then physically destabilizes the phagolysosome [94] and escapes into the cytosol where parasite DNA and STING/TBK1 interact.

Concluding remarks and future directions

Our understanding of innate immune recognition of protozoan parasites has progressed more slowly than for viral or bacterial pathogens. For decades after their discovery, type I IFNs were mainly seen as the product of innate recognition of viruses, but the past 5–10 years have seen a greater appreciation that type I IFN can be activated by non-viral

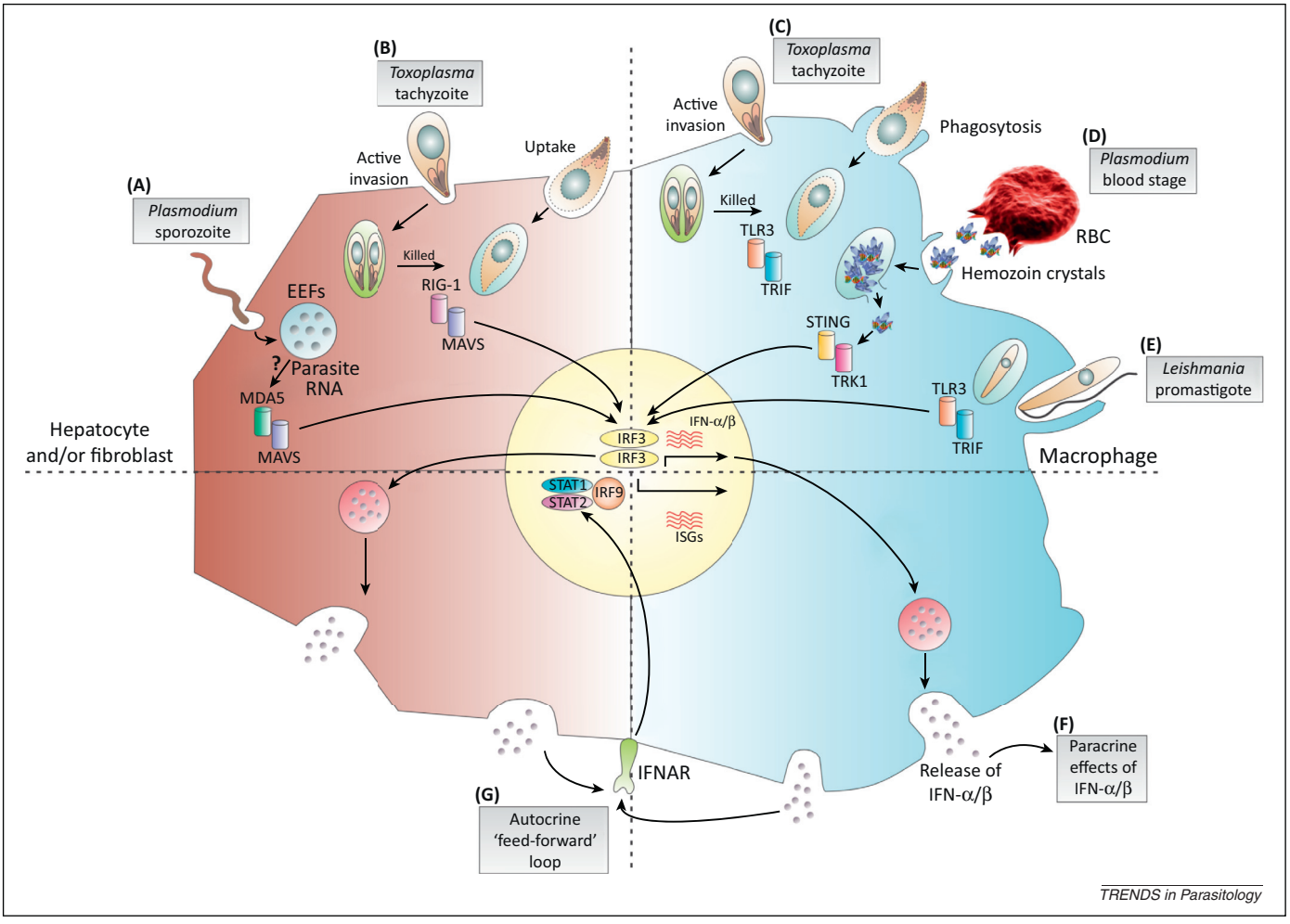

Figure 1. Mechanisms of type I interferon (IFN) signaling during parasite infection. Schematic showing mechanisms by which host cells sense **(A)** liver-stage *Plasmodium*, **(B,C)** *Toxoplasma gondii*, **(D)** hemozoin/DNA complexes from blood-stage *Plasmodium* infection, and **(E)** *Leishmania* strains containing endogenous retroviruses – all culminating in the release of type I IFN. The left side of the schematic represents non-hematopoietic cells such as fibroblasts (infected by *Toxoplasma*) and hepatocytes (infected by *Plasmodium* sporozoites); the right side shows antigen-presenting cells such as macrophages and dendritic cells. The bottom half of the schematic shows release of type I IFN resulting from innate recognition of these parasites. Secreted type I IFN can then act in two ways: **(F)** by exerting paracrine effects on other cells or tissues expressing the IFNAR; and **(G)** by triggering an autocrine feed-forward loop activated within the same cell or tissue, both of which result in transcriptional activation of IFN-stimulated genes (ISGs). Not depicted in this schematic is the *Myd88*-dependent detection of *Toxoplasma* profilin by TLR12 in plasmacytoid dendritic cells [83]. Abbreviation: RBC, red blood cell.

pathogens [95], including protozoan parasites such as *Plasmodium*, *Toxoplasma*, *Leishmania*, and trypanosomes, to name a few. Indeed, the data discussed here collectively show that type I IFN is often induced early after parasite invasion, can amplify its signal through an autocrine feed-forward loop (Figure 1G), and can help to prime an early type II IFN response that is host-protective (Figure 2A). Although numerous gene products induced by type II IFN are known to directly target intracellular parasites for killing, there is little evidence to date that the genes induced by type I IFN are directly parasiticidal. In contexts in which type I IFN protects against parasite infection, the mechanism appears to often be indirect, by recruiting and/or activating other cells that are host-protective. Type I IFN also has potent immunomodulatory effects, and the timing, duration, or intensity the signal may cause a shift from host protection to susceptibility (Figure 2). When this happens, it appears to often involve suppression of type II IFN, which allows increased parasite replication (Figure 2B). Despite this progress, many questions remain (Box 1). For example, a common feature of the protozoan parasites discussed above is the propensity to

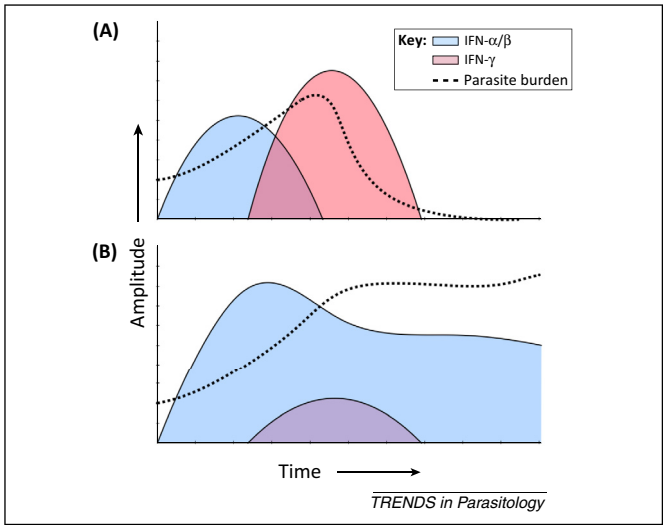

Figure 2. Consequences of type I IFN signaling during parasite infection. Two proposed models depicting possible consequences of type I IFN signaling during parasite infection. **(A)** Early type I IFN (blue curve) enhances a subsequent type II IFN response (red curve), leading to control of parasite replication (broken line). **(B)** A more robust or sustained type I IFN response results in suppression of type II IFN, leading to impaired control of parasite replication.

Box 1. Outstanding questions

- What is the role of type I IFN during chronic parasite infection?
- What are the parasite strategies for immune evasion of type I IFN?
- What is the full spectrum of parasite molecular patterns and host innate immune sensors involved in type I IFN production?
- What factors determine whether type I IFN is host-protective versus pathogenic?
- What role does type I IFN play in hosts harboring virus and parasite coinfections?
- Which, if any, type I IFN-inducible genes are important for cell-intrinsic control of parasites?
- How does type I IFN production change in intensity or duration as parasites move through different tissues or developmental stages?

establish chronic or recurrent infections, and more work is necessary to understand how type I IFN influences this aspect of disease. In addition, although many strategies have been identified by which viruses evade type I IFN responses, little is known about how parasites might accomplish this task, or whether they are under the same evolutionary pressure to do so. Given that type I IFN can suppress type II IFN, it is conceivable that, in some contexts, it may be advantageous for parasites to trigger type I IFN to evade the potent parasiticidal activities of type II IFN.

Finally, it is remarkable that in nearly every setting in which a type I IFN signature was identified during parasite infection, it was revealed using transcriptomics of infected host cells. This highlights that genome-scale, discovery-based approaches in immunoparasitology are an invaluable tool for identifying novel host–pathogen interactions. Moving forward, new resources are needed that enable investigators to mine and query host-response data to compare how type I IFN responses, as well as other host–pathogen interactions, compare across pathogen groups, host species, or cell types, allowing these questions to be addressed in a more systematic way [96,97].

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