ARQUIVOS CIENTÍFICOS (IMMES)

Costimulatory molecules and visceral leishmaniasis

Moléculas coestimulatórias e Leishmaniose visceral

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Palavras-chave

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Visceral Leishmaniasis (VL) is an endemic disease caused by the protozoan *Leishmania infantum*, in South America. Most cases of VL have asymptomatic presentation with the outcome related to genetic factors and the host immune response, where a predominance of Th1 response profile and its citokynes provides resistance to the parasite. Costimulatory molecules are directly involved with the outcome of the immune response and several costimulatory pathways have been characterized, some involved with activation, while others with its inhibition. The parasites like Leishmania could manipulate the expression of some costimulatory molecules favoring an immune response profile suitable to ensure its permanence in the host, favoring the development of active forms of the disease. Some experimental models already been used to prove that the manipulation of costimulatory pathways can be used by some pathogens, but in the visceral leishmaniasis remain unclear. It was reviewed the role of costimulatory molecules CD28, CTLA-4, OX-40 and ICOS and how the manipulation of some pathways can be used by the Leishmania to promote its persistence.

Keywords

Moléculas coestimulatórias
Resposta imune
Infecção
Leishmaniose visceral

A Leishmaniose Visceral (LV) é uma doença endêmica causada pelo protozoário Leishmania infantum, na América do Sul. A maioria dos casos de LV tem apresentação assintomática com desfecho relacionado a fatores genéticos e à resposta imune do hospedeiro, onde o predomínio do perfil de resposta Th1 e suas citocinas proporciona resistência ao parasita. Moléculas coestimulatórias estão diretamente envolvidas com o resultado da resposta imune e várias vias coestimulatórias foram caracterizadas, algumas envolvidas com a ativação, enquanto outras com sua inibição. Os parasitas, como a Leishmania, poderiam manipular a expressão de algumas moléculas coestimulatórias favorecendo um perfil de resposta imune adequado para garantir sua permanência no hospedeiro, favorecendo o desenvolvimento de formas ativas da doença. Alguns modelos experimentais já foram utilizados para comprovar que a manipulação das vias coestimulatórias pode ser utilizada por alguns patógenos, mas na leishmaniose visceral permanecem obscuros. Foi revisa o papel das moléculas coestimulatórias CD28, CTLA-4, OX-40 e ICOS e como a manipulação de algumas vias pode ser utilizada pela Leishmania para promover sua persistência.

INTRODUCTION

Visceral leishmaniasis (VL) is an endemic disease in several countries around the world, caused by the protozoan of the genus Leishmania, especially *L. donovani* and *L. infantum* (VAN GRIENSVEN; DIRO, 2019), transmitted by female sand flies during the blood meal (SELVAPANDIYAN et al., 2019). According to data from the World Health Organization (WHO), more than 90% of global cases of VL have been reported in eight countries: Brazil, Eritrea, Ethiopia, India, Kenya, Somalia, South Sudan and Sudan (WHO-WORLD HEALTH ORGANIZATION, 2021).

Most cases of VL have asymptomatic presentation, but the active disease can lead to death in a portion of individuals, even with treatment (RODRIGUES-NETO et al., 2018). Typical manifestations include, weight loss, chronic fever, and hepatosplenomegaly, with pancytopenia on blood examination. The effective immune response against the parasite is associated with macrophage activation and predominance of Th1 cell profile (VAN GRIENSVEN; DIRO, 2019), while susceptibility is associated with the predominance of the Th2 profile and its cytokines (KUMAR; BHATIA; PAI, 2017). The signalling induced by antigenic MHC/peptide interaction with T cell receptor is a prerequisite to T cell activation, but insufficient to initiate T cell responses by itself. Further signalling by costimulatory molecules is crucial to optimal priming, expansion and differentiation of T cells. Thus, during the immune response, the expression of certain costimulatory molecules, and the cytokines induced by them, have a decisive role in the status of the immune response that the individual will present, therefore, it has a central action in the susceptibility or resistance to certain pathogens (BEYERSDORF; KERKAU, 2020; OYEWOLE-SAID et al., 2020; PANNETON et al., 2019).

Conventional treatment for VL is associated with the use of anti-Leishmania drugs, which have variable effectiveness, depending on the individual's immune response and also on

the geographic region in the world where VL was acquired (SELVAPANDIYAN et al., 2019), thus, new therapeutic approaches, such as those that block costimulatory molecules with monoclonal antibodies, for example, are necessary to make treatment more effective and with fewer side effects.

It was reviewed here the most recent knowledge about the role of the costimulatory molecules CD28, CTLA-4, OX-40 and ICOS of T lymphocytes in the immune response. The action on these pathways is a possible form of resistance of parasites, such as Leishmania, to ensure the success in infections.

IMMUNE RESPONSE

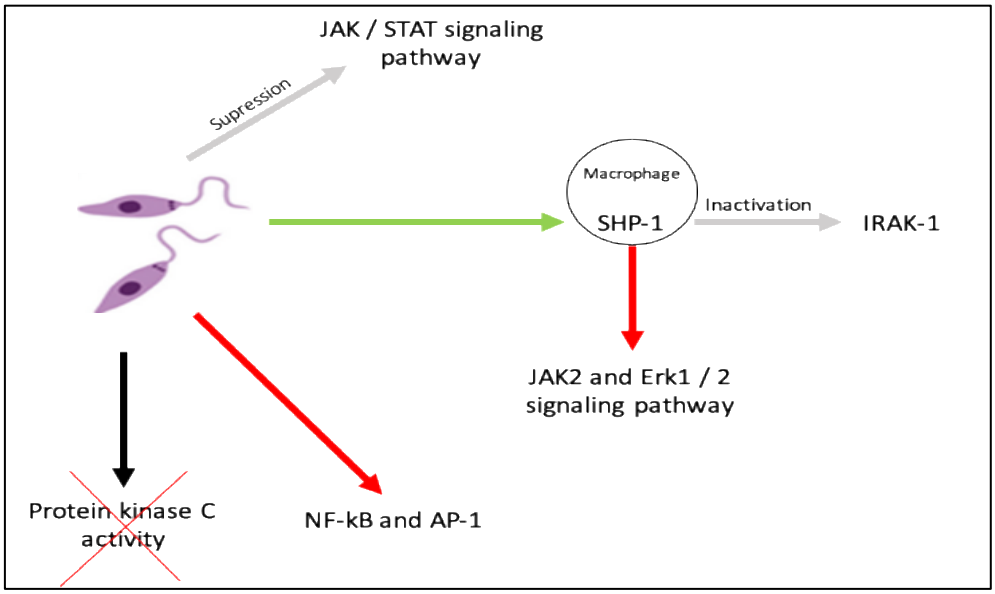
Protective immune response against Leishmania infection is mediated by cellular immunity, Th1 response correlates with resistance, while Th2 is associated with susceptibility to infection (SRIVASTAVA et al., 2012). The immune response to Leishmania can be assessed using experimental animal models that may represent the best way to analyze the infection (MURRAY, 2020; SAINI; RAI, 2020). In these experimental models, the protective immune response observed in the liver includes granuloma formation, which contributes to the parasite elimination. It is well documented that cytokines such as interferon gamma (IFN-γ), interleukin 12 (IL-12) and low levels of tumor necrosis factor (TNF) contribute to parasite control in granulomas (BACELLAR et al., 1996; ENGWERDA et al., 2004). In the spleen, however, active infection is associated with excess of TNF and granuloma formation is clearly decreased or absent. Another major cytokine is interleukin 10 (IL-10) which plays a central role in susceptibility to Leishmania infection in both liver and spleen (MESQUITA et al., 2018; MOULIK et al., 2021).

In a recent past, IL-10 was considered a cytokine strictly related to Th2 profile, showing implication in a large number of pathologies (ANURADHA et al., 2014; HUANG et al., 2014; KUMAR et al., 2013). Nowadays, it is no longer considered an exclusively Th2 cytokine, playing an important role in modulating both Th1 and Th2 immune responses. It has well documented that IL-10 is induced by high levels of TNF-α (ATO et al., 2002), having an proeminent role in controlling TNF-α-induced tissue damage, while promoting parasitic persistence by inhibiting macrophages activation (BELKAID et al., 2001). IL-10 is a regulatory cytokine presumed to be induced as part of a homeostatic network to protect tissues from collateral damage caused by excessive inflammation (KUMAR; NYLÉN, 2012; SUKHBAATAR et al., 2020).

Visceral leishmaniasis (VL) was classically associated with a predominance of Th2 immune response and its preferential expansion. Although VL was associated with a Th2 profile, with elevation of cytokines such as IL-4 and IL-13 (NYLÉN; SACKS, 2007; SUNDAR et al., 1997), many studies suggest that this inclination toward the Th2 profile in human VL is unclear, ie there is no clear dichotomy observed in murine models (KHADEM; UZONNA, 2014). Typically, VL is associated with an increased production of multiple cytokines and chemokines such as IL-10, IL-4, IFN-γ, TNF-α and IL-12 (COSTA et al., 2012; DAYAKAR et al., 2019; SINGH et al., 2012). Although it may colonize other cells such as neutrophils and dendritic cells (OUALHA et al., 2019; LIU; UZONNA, 2012), the main target of Leishmania are the macrophages. Even if these cells have effective mechanisms to destroy intracellular pathogens through toxic metabolites such as nitric oxide and reactive oxygen species, they cannot eliminate the parasite that evades the host immune response by attenuating the proinflammatory signaling pathway (PODINOVSKAIA; DESCOTEAUX, 2015).

Although IL-10-related pathways may explain relevant aspects of parasitic persistence that promotes VL development, other factors include changes in macrophage signaling pathways (SAUNDERS; MCCONVILLE, 2020; SHIO et al., 2012). It has been suggested that Leishmania may activate macrophage protein SHP-1 (Src homology region 2 domain-containing phosphatase-1), which inhibits activation of the signaling pathway of JAK2 (Janus kinase 2) and Erk1 / 2 (extracellular signal-regulated kinases 1 and 2) (Figure 1) (ZHOU et al., 2014).

Figure 1: Changes in macrophage signaling pathways may explain relevant aspects of parasitic persistence that promotes VL development. Leishmania may activate macrophage protein SHP-1, which inhibits activation of the signaling pathway of JAK2 and Erk1 / 2, and SHP-1 also interferes with Toll-like (TLRs) macrophage receptor signaling, inactivating IRAK-1. Moreover, other parasite-mediated changes include blockade of protein kinase C activity, inhibition of NF-kB and transcriptional functions of AP-1 and suppression of JAK / STAT signaling pathway. Green arrow – activation; Red arrows – inhibition; SHP-1 (Src homology region 2 domain-containing phosphatase-1); JAK2 (Janus kinase 2); Erk1 / 2 (extracellular signal-regulated kinases 1 and 2); IRAK-1 (interleukin 1 receptor-associated kinase 1); NF-kB (factor nuclear kappa B); AP-1 (activator protein 1); JAK / STAT (The Janus kinase/signal transducers and activators of transcription).



SHP-1 also interferes with Toll-like (TLRs) macrophage receptor signaling, directly inactivating IRAK-1 (interleukin 1 receptor-associated kinase 1) (ABU-DAYYEH et al., 2008; MARTINY et al., 1999). Other parasite-mediated changes include blockade of protein kinase C activity (DESCOTEAUX A; MATLASHEWSKI G; TURCO SJ, 1992; MCNEELY; TURCO, 1987), inhibition of NF-kB (factor nuclear kappa B) and transcriptional functions of AP-1 (activator protein 1) (CONTRERAS et al., 2010; GHOSH et al., 2002) and suppression of JAK / STAT signaling pathway (The Janus kinase/signal transducers and activators of transcription) (Figure 1) (BLANCHETTE et al., 1999; FORGET; GREGORY; OLIVIER, 2005).

Many of these manipulations are mediated by molecules present on the surface of the parasite, such as LPGs (lipophosphoglycans) and gp63 (PARASHAR; MUKHOPADHYAY, 2017; VIEIRA et al., 2019), resulting in decreased inflammatory cytokines and reactive nitrogen and oxygen intermediates, thus, the parasite can survive and grow inside macrophages.

The clinical evolution of *L. infantum* infection is influenced by the development of the host immune response. Classically, active systemic infection with parasite expansion to the spleen, liver, lymph nodes, bone marrow, and other organs is accompanied by high levels of anti-Leishmania antibodies, decreased interferon-γ (CARVALHO et al., 1985; TRINCHIERI, 1997) and IL-12 production and IL-10 elevation (BACELLAR et al., 1996; SAHA et al., 2007).

The increased production of anti-Leishmania antibodies observed during active disease has been associated with an impaired immune response, since the antibodies produced opsonize the pathogens that, when phagocytized by macrophages, can stimulate the production of IL-10 and TGF-β, in these cells, leading to decreased microbicidal capacity, facilitating the persistence of the parasite (KHADEM; UZONNA, 2014).

During VL, a remarkable immunological feature is the inability of peripheral blood mononuclear cells (PBMCs) to mount a curative immune response (HO et al., 1983; SACKS et al., 1987), which is reflected in the PBMCs failure to effectively proliferate or produce IFN-γ in response to Leishmania antigens (CARVALHO et al., 1985, 1989). This does not appear to be an innate defect since individuals with VL, after treatment, are able to mount an immune response compatible with Leishmania resistance (NYLÉN et al., 2007), so there is a temporary immunosuppression, during acute disease, and parasite-specific. Studies have shown that this inability to produce IFN-γ following specific Leishmania stimulation is not observed during active VL, in studies using whole blood samples (SINGH et al., 2012). But this fact could not be proven in a study with VL patients from Ethiopia that showed at time of diagnosis the IFN -gamma production is

impaired (ADEM et al., 2016). The contradictory findings can be explained by differences in disease severity between countries, genetic variation between parasites or differences in host genetic factors.

After antigenic stimulation, CD4+ and CD8+ T lymphocytes initiate a proliferation program that is directly associated with the acquisition of effector functions and, ultimately, leads to the formation of memory cells (BRUMMELMAN; PILIPOW; LUGLI, 2018). Acquired effector functions are associated with T lymphocytes activation, that promote increased production of transcription factors among them, NF- KB and NFAT (nuclear factor of activated T-cells) (CHAPMAN; BOOTHBY; CHI, 2020). These factors increase the transcription of some genes, and are essential for the profile showed by the cell, that is, it promotes the production of cytokines and other receptors that determine their activation status, such as effector cells, or can determine the expression of molecules related to other functions. Activation-induced changes affect many classes of molecules including, but not restricted to, CD4 and CD8 co-receptor molecules, and these changes are involved with survival, cell cycle control, adhesion, and migration, as well as their effector function (HU; ZOU; SU, 2018). Thus, we can say that T cellular activation is a key immune event that produces broad phenotypic and functional modifications in T lymphocytes which is capable of differentiation into various functional forms like Th-1, Th-2, Th-17, Treg and γδ-T cells etc. These T cells have important role in the inflammatory diseases and also play vital role to prevent our body from hyperimmune activation, auto-immunity, immunodeficiency and the complete execution of infectious agents like Leishmania (JAWED; DUTTA; MAJUMDAR, 2019).

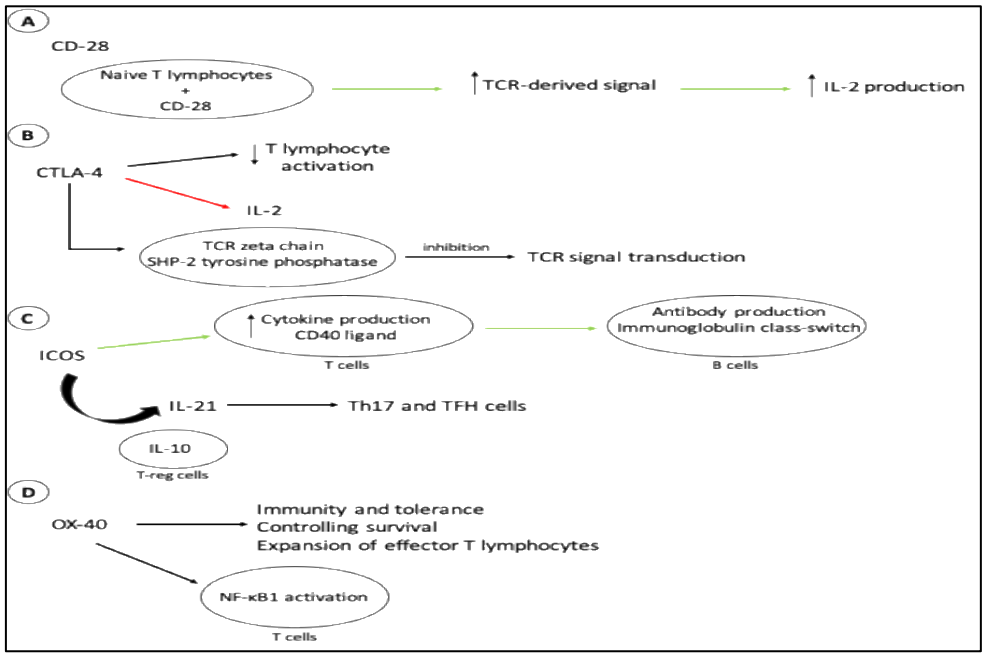
It is well established that for cellular activation to occur two complementary signals are required: the recognition of a specific antigen by the T cell (signal 1), given by the interaction between the T cell receptor (TCR)/ peptide/ main histocompatibility complex (MHC), on the surface of antigen presenting cells (APCs), and a costimulatory signal non antigen-specific (signal 2) provided by interactions between surface molecules of APCs and the T cell (LAFFERTY; ANDRUS; PROWSE, 1980). In fact, early work by Jenkins and Schwartz showed that in the absence of costimulatory signals, T cells become anergic, a state marked by the inability of T lymphocytes to respond to subsequent antigenic stimulation, leading to the model that costimulation has a critical role in controlling the fate of T cell responses: activation or anergy (JENKINS et al., 1990; SCHWARTZ et al., 1989).

Several costimulatory pathways have been characterized, some involved with activation, while others with inhibition of immune response (BEYERSDORF; KERKAU, 2020; OYEWOLE-SAID et al., 2020; WATANABE et al., 2005). Positive signaling pathways are balanced by negative signaling to promote the suitable control of T and B cell activation. Costimulatory molecules such as B7 and CD28, CD40 ligand (CD40L) and CD40, ICOS (inducible co-stimulatory molecule), and ICOS ligand (ICOS-L), and OX40 (CD134) and their ligand, OX40L, are critical for T and B cell activation; Inhibitory molecules such as B7 and CTLA-4 (cytotoxic T lymphocyte antigen-4), and PD-1 (programmed cell death-1) and PD-1 ligand (PD1-L), promote an inhibition of T lymphocyte activation (HUBO et al., 2013; WILLSMORE et al., 2021).

The costimulatory molecules mentioned above belong mainly to three families: the CD28 family, the B7 family and the TNF family (SHARPE, 2009). The CD28 costimulatory molecule is a glycoprotein, a member of the immunoglobulin superfamily, expressed on the T cell membrane as disulfide-linked homodimers (ESENSTEN et al., 2016). It is expressed on the surface of most CD4 T cells and in approximately 50% of CD8 T cells, has like ligands B7-1 (CD80) and B7-2 (CD86) receptors, also members of the immunoglobulin superfamily, and that are expressed in APCs such as dendritic cells, macrophages and activated B cells (SHARPE, 2009). Although B7-1 and B7-2 are co-expressed and share a similar general structure, they differ in their temporal response to stimulus, with induction of B7-2 occurring initially and usually at a much higher level than B7-1, in some APCs (ESENSTEN et al., 2016). B7-1 and B7-2 expression is controlled by cytokines and cell-cell interactions.

CD28 is considered the primary costimulatory molecule, that complements TCR-mediated signaling and promotes T cell activation, and ensures their proliferation and survival. Initial activation of naive T lymphocytes requires CD28 signaling to increase TCR-derived signal and promote IL-2 production for effective expansion of antigen-specific T cells (Figure 2A) (WAKAMATSU; MATHIS; BENOIST, 2013). It has been suggested that CD28 activation is involved in multiple pathways such as PI3K (phosphatidylinositol 3-kinase). In one way, PI3K is required for the activation of other molecules, which in turn regulate many targets along the signaling cascade that are involved in functions as diverse as proteins translation and cellular metabolism (KISHORE et al., 2017). CD28 gene deficient mice or treated with CD28 blocking antibodies exhibit a marked reduction in lymphoproliferative response (GLADOW et al., 2020). In addition, some studies have already demonstrated associations of CD28 gene polymorphisms with the development of some autoimmune and infectious diseases (SORIANO et al., 2002; TEUTSCH et al., 2004).

Figure 2: Costimulatory molecules are involved with activation and inhibition of immune response. A. Initial activation of naive T lymphocytes requires CD28 signaling to stimulate the TCR-derived signal and promote IL-2 production for effective expansion of antigen-specific T cells. **B.** CTLA-4 delivers an inhibitory signal that promotes decreased T lymphocyte activation and inhibits IL-2 production. Its association with TCR zeta chain and with SHP-2 tyrosine phosphatase, in T cells, promote the inhibition of TCR signal transduction. **C.** ICOS promotes increased cytokine production by T lymphocytes and increases CD40 ligand (CD40L) expression, stimulating the production of antibodies by B lymphocytes, and also participates in the immunoglobulin class-switch. ICOS can also regulate IL-21 production, which in turn regulates the expansion of Th17 and TFH cells, and play an important role in the production of IL-10 by Treg cells. **D.** OX-40 plays a central role in immunity and tolerance, controlling survival and expansion of effector T lymphocytes. Its activation strongly contributes to the overall level of NF-κB1 activation in T lymphocytes. Green arrows – activation; Red arrow – inhibition; TCR (T cell receptor); Th17 (T helper 17); TFH (T follicular helper cells); NF-KB1 (Nuclear Factor Kappa B Subunit 1).



CTLA-4, also known as CD152, is expressed on the T lymphocyte membrane. Like CD28 and B7, they are also members of the immunoglobulin superfamily. CTLA-4 binds to the same CD28 receptors, B7-1 and B7-2, but it binds with a much higher affinity to B7 molecules, about 20 times more than CD28 (BRZOSTEK; GASCOIGNE; RYBAKIN, 2016). Although CTLA-4 and CD28 share sequence homology, CTLA-4 acts antagonistically to CD28, and delivers an inhibitory signal that promotes decreased T lymphocyte activation, besides to inhibits IL-2 production and cell cycle progression (Figure 2B) (KRUMMEL; ALLISON, 1996; WALUNAS; BAKKER; BLUESTONE, 1996). CTLA-4 has been suggested to inhibit TCR signal transduction by inhibiting tyrosine phosphorylation of SHP-2 phosphatases (LEE et al., 1998). Studies show that CTLA-4 can act by inhibiting T cell response activation in two different ways: competing with CD28 or sequestering B7 molecules of CD28, and also actively transducing signal through T lymphocytes, via their intracellular domains (SEIDEL; OTSUKA; KABASHIMA, 2018).

The importance of CTLA-4 in the immune response was well documented by experimental models showing that in CTLA-4 knockout mice, an intense lymphoproliferative disorder is observed, which is fatal within 3-4 weeks after birth, supporting the role of this molecule as a negative regulator of T lymphocyte activation, and that it is critical for the control of lymphocyte homeostasis (WATERHOUSE et al., 1995). Furthermore, the role of CTLA-4 in numerous diseases has been demonstrated (GOKHALE et al., 2013; HUANG et al., 2013; WANG et al., 2020). While CD28 is expressed by naive

and activated T cells, CTLA-4 is expressed only in activated cells, at a much lower level.

B7 and CD28 gene families have grown substantially with the exploration of the human genome, and the pathways defined by these new members regulate the activation, inhibition and fine-tuning of T lymphocyte immune responses. Other costimulatory molecules also play a key role in maintaining immune response homeostasis, including ICOS and OX-40. ICOS, a new member of the CD28 family, has great amino acid homology to CD28 and CTLA-4 (HUTLOFF et al., 1999), having ICOS-L (also known as B7-H2, B7RP-1, GL50) as a ligand, a molecule related to B7-1 and B7-2 ligands (LING et al., 2000). Unlike the constitutively expressed CD28 molecule, ICOS is induced on the surface of T lymphocytes only after activation of these cells, being found in small quantities in naive T cells (SOLINAS; GU-TRANTIEN; WILLARD-GALLO, 2020). ICOS ligand is found in APCs, and through its binding to ICOS promotes increased cytokine production by T lymphocytes, as well as increasing CD40 ligand (CD40L) expression, helping in the production of antibodies by B lymphocytes, and also participates in the immunoglobulin class-switch (Figure 2C) (LIU et al., 2021).

ICOS has been suggested to regulate, through a series of signaling cascades, broad characteristics of the T lymphocyte response such as growth, proliferation and survival. Importantly, these primary characteristics could be restricted to specific T cell subpopulations, by the environmental context of the immune response, and their impact on the expression of T lymphocyte lineage-specific transcription factors such as T-bet and GATA-3 (SIMPSON; QUEZADA; ALLISON, 2010). The ICOS / ICOS-L pathway plays a critical role in stimulating effector responses of T lymphocytes and T-dependent B lymphocytes, in addition to regulating T-cell immune tolerance (SOLINAS; GU-TRANTIEN; WILLARD-GALLO, 2020). ICOS has been shown to be an important immune system receptor promoting the precise regulation of effector T lymphocytes, moreover, is important for the generation of the CXCR5 chemokine receptor, in T follicular helper cells (TFH), the only subset of T cells that regulate germinal center reactions and humoral immunity (LE et al., 2018).

Studies using ICOS-deficient mice (ICOS - / -) indicate that this molecule can regulate IL-21 production, which in turn regulates the expansion of Th17 (T helper 17) and TFH cells (PAULOS et al., 2010; SHARPE, 2009). ICOS also plays an important role in the production of IL-10 by Treg cells. It has been observed that ICOS deficiency in humans leads to defective IL-10 and IL-17 production, with impaired affinity maturation and immunoglobulin class switching, in germinal centers, resulting in deep hypogammaglobulinemia (SCHEPP et al., 2017).

Another molecule of great importance in the costimulation process is OX-40, also known as ACT35, CD134 or TNFRSF4. It is a glycoprotein, belonging to the TNFR superfamily (tumor necrosis factor receptor), having like ligand OX-40L (WEBB; HIRSCHFIELD; LANE, 2016). OX-40 is not constitutively expressed in T lymphocytes and is found only after activation of these cells, already its ligand is found in APCs such as macrophages and dendritic cells (MASSARELLI et al., 2019). It has been suggested that the interaction between OX-40 and its ligand plays a central role in immunity and tolerance, controlling survival, expansion of effector T lymphocytes and T cell homeostasis, as well as acting on the generation, reactivation and maintenance of memory T lymphocytes (Figure 2D) (CROFT et al., 2009; HUBO et al., 2013). The interaction of OX-40 with its ligand tightly regulates CD4 and CD8 T cells, and some studies have highlighted its ability to modulate NK and NKT cell function, as well as mediate crosslinking with APCs and various other cell types like mast cells, smooth muscle cells and endothelial cells (BURGESS et al., 2004; IMURA et al., 1996; NAKAE et al., 2006; ZINGONI et al., 2004).

Studies have shown that OX-40 strongly contributes to the overall level of NF-κB1 activation in T lymphocytes (SONG; SO; CROFT, 2008). CD4 T lymphocytes that lose OX-40 do not maintain elevated levels of anti-apoptotic proteins, members of the Bcl-2 family, including Bcl-2, Bcl-xL, and Bfl-1 (ROGERS et al., 2001), a finding that correlates directly with reduced NF-κB1 activity (SONG; SO; CROFT, 2008). Several studies have proven the protective effect of OX-40 / OX-40L blockade, in inflammatory diseases models such as asthma, arteriosclerosis as well as with autoimmune diseases like experimental autoimmune encephalomyelitis, diabetes and colitis (CROFT et al., 2009; KAUR; BRIGHTLING, 2012).

As can be seen, the immune response depends on the proper expression of costimulatory molecules, which can be stimulated or inhibited in certain situations in favor of the host and which can be manipulated by the parasites in their own favor, resulting in active disease in many situations.

DISCUSSION

T cell costimulation plays a decisive role in the final destination of these cells, whether it is activation, with active production of cytokines and effector cells, or with inhibition, modulating the immune response to low levels or even inhibiting the response, leading to an anergy state. The production of cytokines, induced by these costimulatory molecules, can also affect the subtype of T cell response, thus, the induction of different subtypes (Th1, Th2, Th17, ...) may benefit resistance or susceptibility to some diseases. Because of this, costimulatory molecules have been the subject of much research, implying their role in many diseases. Studies

have shown that blocking these molecules can be used as a potential therapeutic target (FANG et al., 2020; WYKES; LEWIN, 2018).

The increase in the levels of CTLA-4 expressed in T lymphocytes, for example, can be used as a strategy of Leishmania to promote anergy of the cellular immune response during active disease, a view that can be reinforced by the findings of Murphy et al. , in an experimental model, which demonstrated that CTLA-4 blockade increases host resistance to infection by *Leishmania donovani*, and Pedicord et al., who demonstrated, in mice, that CTLA-4 blockade increases the number of CD8 memory cells capable to produce IFN-γ and TNF-α (MURPHY et al., 1998; PEDICORD et al., 2011).

It can be suggested that such modulation made by *L. infantum* may favor its survival, since it has been reported that CTLA-4 can induce the TGF-β production (NOUËL et al., 2015), and may thus prevent the production of nitric oxide by macrophages and drive the response to a Th2 profile, since it can also interfere with the IFN-γ prodution, a role that may have special relevance in the development of active VL. Associated with this role, it can also be suggested that the increased expression of CTLA-4 during active VL could, in part, explain the inability of T lymphocytes to mount a suitable cellular response, since it has been suggested that the molecule can modulate the signal threshold necessary for proliferation and cytokine production by T lymphocytes (SMIDA et al., 2013; WAN et al., 2018). Apparently, this response is not completely inhibited, as we can see with data from the production of cytokines in our group that show that after specific stimulus, even during active disease, there is a production of IFN-γ (Unpublished data). The importance of CTLA-4 in the context of visceral leishmaniasis (HAJILOOI et al., 2014) and cutaneous leishmaniasis (AL-GHABBAN et al., 2021) has gained prominence in recent years, having already been found polymorphisms in this gene, associated with these diseases.

The increase in the percentage of OX-40 during active VL may be involved with the development of a Th2 profile, as observed in experimental animal models, favoring the parasite persistence (KAUR; BRIGHTLING, 2012). Other studies also show that signaling by OX-40, preferably, promotes differentiation in the Th2 profile in the absence of IL-12 and independent of IL-4 (ITO et al., 2005; MADDUR et al., 2014). Another well-documented feature of OX-40, is the ability to promote the expansion and survival of effector cells, in this way, could be directly associated with the maintenance of the Th2 response with production of cytokines of this profile (LIN et al., 2018).

It has been suggested that ICOS regulates general features of T lymphocytes such as growth, proliferation and survival (RAO, 2018). Functional studies using knockout mice or blocking antibodies for ICOS have suggested that the costimulatory molecule is important for Th2 immune responses, preferably inducing the production of IL-4 and IL-10 (COYLE et al., 2000; DONG et al., 2001; GONZALO et al., 2001; MCADAM et al., 2000; TESCIUBA et al., 2001; WANG et al., 2013). Several investigation lines have identified PI3K as an important mediator in ICOS signaling. Pharmacological inhibition of PI3K during signaling by ICOS results in reduced production of IL-4, IL-10 and IL-21, demonstrating the importance of PI3K in the secretion of cytokines characteristic of the Th2 and Tfh profile (follicular T helper) (FEITO et al., 2003; GIGOUX et al., 2009).

There are no studies in the literature that have evaluated the ICOS costimulatory molecule in visceral leishmaniasis, but in cutaneous leishmaniasis (LC) Miyahira et al., demonstrated, in an experimental model of *L. major* infection, that the use of blocking antibodies for ICOS or ICOS deficient BALB / c mice have a better prognosis, indicating the role of ICOS in inducing the Th2 profile (MIYAHIRA et al., 2003). In addition, Nouailles et al., has shown, in experimental models of *Mycobacterium tuberculosis* infection, that the absence of ICOS in the late stage of the infection, is responsible for the increase of IFN-γ production by effector CD4 T cells, which favors protection against infection (NOUAILLES et al., 2011).

More broadly, manipulation of costimulatory pathways may provide new targets for immunotherapy, increasing anti-Leishmania immunity. We hope to achieve new ways of combating pathogens in the near future. A good example of manipulating the co-stimulatory pathways, in our favor, was the release of the use of Ipilimumab, a drug based on the blockade of CTLA-4, for the treatment of melanoma (LIPSON; DRAKE, 2011). Thus, based on the literature and preliminary unpublished data, can be infered that approaches that include the molecules, CTLA-4 and ICOS, may have potential success in the control of visceral leishmaniasis, providing additional means of treating the disease. Blocking these molecules can facilitate the parasite elimination by increasing the apoptosis of infected cells and / or the release of reactive intermediates by macrophages.

REFERENCES

ABU-DAYYEH, I. et al. Leishmania-induced IRAK-1 inactivation is mediated by SHP-1 interacting with an evolutionarily conserved KTIM motif. **PLoS neglected tropical diseases**, v. 2, n. 12, p. e305, 2008.

ADEM, E. et al. Successful Treatment of Human Visceral Leishmaniasis Restores Antigen-Specific IFN-γ, but not IL-10 Production. **PLOS Neglected Tropical Diseases**, v. 10, n. 3, p. e0004468, 10 mar. 2016.

AL-GHABBAN, M. J. S. et al. The role of cytotoxic T-Lymphocyte antigen-4+49A/G gene polymorphism in cutaneous leishmaniasis. **Microbial Pathogenesis**, v. 158, p. 105073, set. 2021.

ANURADHA, R. et al. Expansion of parasite-specific CD4+ and CD8+ T cells expressing IL-10 superfamily cytokine members and their regulation in human lymphatic filariasis. **PLoS neglected tropical diseases**, v. 8, n. 4, p. e2762, abr. 2014.

ATO, M. et al. Defective CCR7 expression on dendritic cells contributes to the development of visceral leishmaniasis. **Nature Immunology**, v. 3, n. 12, p. 1185–1191, dez. 2002.

BACELLAR, O. et al. Interleukin-12 restores interferon-gamma production and cytotoxic responses in visceral leishmaniasis. **The Journal of Infectious Diseases**, v. 173, n. 6, p. 1515–1518, jun. 1996.

BELKAID, Y. et al. The role of interleukin (IL)-10 in the persistence of Leishmania major in the skin after healing and the therapeutic potential of anti-IL-10 receptor antibody for sterile cure. **The Journal of Experimental Medicine**, v. 194, n. 10, p. 1497–1506, 19 nov. 2001.

BEYERSDORF, N.; KERKAU, T. [CD28 costimulation and checkpoint inhibition in T cells]. **Der Internist**, v. 61, n. 7, p. 652–659, jul. 2020.

BLANCHETTE, J. et al. Leishmania-induced increases in activation of macrophage SHP-1 tyrosine phosphatase are associated with impaired IFN-gamma-triggered JAK2 activation. **European Journal of Immunology**, v. 29, n. 11, p. 3737–3744, nov. 1999.

BRUMMELMAN, J.; PILIPOW, K.; LUGLI, E. The Single-Cell Phenotypic Identity of Human CD8+ and CD4+ T Cells. **International Review of Cell and Molecular Biology**, v. 341, p. 63–124, 2018.

BRZOSTEK, J.; GASCOIGNE, N. R. J.; RYBAKIN, V. Cell Type-Specific Regulation of Immunological Synapse Dynamics by B7 Ligand Recognition. **Frontiers in Immunology**, v. 7, p. 24, 2016.

BURGESS, J. K. et al. Detection and characterization of OX40 ligand expression in human airway smooth muscle cells: a possible role in asthma? **The Journal of Allergy and Clinical Immunology**, v. 113, n. 4, p. 683–689, abr. 2004.

CARVALHO, E. M. et al. Absence of gamma interferon and interleukin 2 production during active visceral leishmaniasis. **Journal of Clinical Investigation**, v. 76, n. 6, p. 2066–2069, dez. 1985.

CARVALHO, E. M. et al. Antigen-specific immunosuppression in visceral leishmaniasis is cell mediated. **Journal of Clinical Investigation**, v. 83, n. 3, p. 860–864, mar. 1989.

CHAPMAN, N. M.; BOOTHBY, M. R.; CHI, H. Metabolic coordination of T cell quiescence and activation. **Nature Reviews. Immunology**, v. 20, n. 1, p. 55–70, jan. 2020.

CONTRERAS, I. et al. Leishmania-induced inactivation of the macrophage transcription factor AP-1 is mediated by the parasite metalloprotease GP63. **PLoS pathogens**, v. 6, n. 10, p. e1001148, 14 out. 2010.

COSTA, A. S. A. et al. Cytokines and visceral leishmaniasis: a comparison of plasma cytokine profiles between the clinical forms of visceral leishmaniasis. **Memórias do Instituto Oswaldo Cruz**, v. 107, n. 6, p. 735–739, set. 2012.

COYLE, A. J. et al. The CD28-related molecule ICOS is required for effective T cell-dependent immune responses. **Immunity**, v. 13, n. 1, p. 95–105, jul. 2000.

CROFT, M. et al. The significance of OX40 and OX40L to T-cell biology and immune disease. **Immunological Reviews**, v. 229, n. 1, p. 173–191, maio 2009.

DAYAKAR, A. et al. Cytokines: Key Determinants of Resistance or Disease Progression in Visceral Leishmaniasis: Opportunities for Novel Diagnostics and Immunotherapy. **Frontiers in Immunology**, v. 10, 2019.

DESCOTEAUX A; MATLASHEWSKI G; TURCO SJ. Inhibition of macrophage protein kinase C-mediated protein phosphorylation by Leishmania donovani lipophosphoglycan. **Journal of Immunology** (Baltimore, Md. : 1950), v. 149, n. 9, p. 3008–3015, 1 nov. 1992.

DONG, C. et al. ICOS co-stimulatory receptor is essential for T-cell activation and function. **Nature**, v. 409, n. 6816, p. 97–101, 4 jan. 2001.

ENGWERDA, C. R. et al. Distinct roles for lymphotoxin-alpha and tumor necrosis factor in the control of Leishmania donovani infection. **The American Journal of Pathology**, v. 165, n. 6, p. 2123–2133, dez. 2004.

ESENSTEN, J. H. et al. CD28 Costimulation: From Mechanism to Therapy. **Immunity**, v. 44, n. 5, p. 973–988, 17 maio 2016.

FANG, J. et al. Prognostic value of immune checkpoint molecules in breast cancer. **Bioscience Reports**, v. 40, n. 7, p. BSR20201054, 31 jul. 2020.

FEITO, M. J. et al. Mechanisms of H4/ICOS costimulation: effects on proximal TCR signals and MAP kinase pathways. **European Journal of Immunology**, v. 33, n. 1, p. 204–214, 2003.

FORGET, G.; GREGORY, D. J.; OLIVIER, M. Proteasome-mediated degradation of STAT1alpha following infection of macrophages with Leishmania donovani. **The Journal of Biological Chemistry**, v. 280, n. 34, p. 30542–30549, 26 ago. 2005.

GHOSH, S. et al. Leishmania donovani suppresses activated protein 1 and NF-kappaB activation in host macrophages via ceramide generation: involvement of extracellular signal-regulated kinase. **Infection and Immunity**, v. 70, n. 12, p. 6828–6838, dez. 2002.

GIGOUX, M. et al. Inducible costimulator promotes helper T-cell differentiation through phosphoinositide 3-kinase. **Proceedings of the National Academy of Sciences of the United States of America**, v. 106, n. 48, p. 20371–20376, 1 dez. 2009.

GLADOW, N. et al. Treatment of mice with a ligand binding blocking anti-CD28 monoclonal antibody improves healing after myocardial infarction. **PloS One**, v. 15, n. 4, p. e0227734, 2020.

GOKHALE, P. et al. CTLA-4 gene polymorphism at position +49 A>G in exon 1: a risk factor for cervical cancer in Indian women. **Cancer Genetics**, v. 206, n. 5, p. 154–161, maio 2013.

GONZALO, J. A. et al. ICOS is critical for T helper cell-mediated lung mucosal inflammatory responses. **Nature Immunology**, v. 2, n. 7, p. 597–604, jul. 2001.

HAJILOOI, M. et al. The Cytotoxic T Lymphocyte Antigen-4 +49A/G Single Nucleotide Polymorphism Association With Visceral Leishmaniasis. **Jundishapur Journal of Microbiology**, v. 7, n. 10, p. e12143, out. 2014.

HO, M. et al. Immunosuppression in Kenyan visceral leishmaniasis. **Clinical and Experimental Immunology**, v. 51, n. 2, p. 207–214, fev. 1983.

HU, Z.; ZOU, Q.; SU, B. Regulation of T cell immunity by cellular metabolism. **Frontiers of Medicine**, v. 12, n. 4, p. 463–472, ago. 2018.

HUANG, H. et al. Interleukin-10 regulates hepcidin in Plasmodium falciparum malaria. **PloS One**, v. 9, n. 2, p. e88408, 2014.

HUANG, R. et al. Association between cytotoxic T-lymphocyte-associated antigen 4 +49A/G polymorphism and persistent hepatitis B virus infection in the Asian population: evidence from the current studies. **Genetic Testing and Molecular Biomarkers**, v. 17, n. 8, p. 601–606, ago. 2013.

HUBO, M. et al. Costimulatory molecules on immunogenic versus tolerogenic human dendritic cells. **Frontiers in Immunology**, v. 4, p. 82, 2013.

HUTLOFF, A. et al. ICOS is an inducible T-cell co-stimulator structurally and functionally related to CD28. **Nature**, v. 397, n. 6716, p. 263–266, 21 jan. 1999.

IMURA, A. et al. The human OX40/gp34 system directly mediates adhesion of activated T cells to vascular endothelial cells. **The Journal of Experimental Medicine**, v. 183, n. 5, p. 2185–2195, 1 maio 1996.

ITO, T. et al. TSLP-activated dendritic cells induce an inflammatory T helper type 2 cell response through OX40 ligand. **The Journal of Experimental Medicine**, v. 202, n. 9, p. 1213–1223, 7 nov. 2005.

JAWED, J. J.; DUTTA, S.; MAJUMDAR, S. Functional aspects of T cell diversity in visceral leishmaniasis. **Biomedicine & Pharmacotherapy**, v. 117, p. 109098, 1 set. 2019.

JENKINS, M. K. et al. Inhibition of antigen-specific proliferation of type 1 murine T cell clones after stimulation with immobilized anti-CD3 monoclonal antibody. **Journal of Immunology** (Baltimore, Md.: 1950), v. 144, n. 1, p. 16–22, 1 jan. 1990.

KAUR, D.; BRIGHTLING, C. OX40/OX40 ligand interactions in T-cell regulation and asthma. **Chest**, v. 141, n. 2, p. 494–499, fev. 2012.

KHADEM, F.; UZONNA, J. E. Immunity to visceral leishmaniasis: implications for immunotherapy. **Future Microbiology**, v. 9, n. 7, p. 901–915, 2014.

KISHORE, M. et al. Regulatory T Cell Migration Is Dependent on Glucokinase-Mediated Glycolysis. **Immunity**, v. 47, n. 5, p. 875- 889.e10, 21 nov. 2017.

KRUMMEL, M. F.; ALLISON, J. P. CTLA-4 engagement inhibits IL-2 accumulation and cell cycle progression upon activation of resting T cells. **The Journal of Experimental Medicine**, v. 183, n. 6, p. 2533–2540, 1 jun. 1996.

KUMAR, R.; BHATIA, M.; PAI, K. Role of Cytokines in the Pathogenesis of Visceral Leishmaniasis. **Clinical Laboratory**, v. 63, n. 10, p. 1549–1559, 1 out. 2017.

KUMAR, R.; NYLÉN, S. Immunobiology of visceral leishmaniasis. **Frontiers in Immunology**, v. 3, p. 251, 2012.

KUMAR, S. et al. IL-10 production from dendritic cells is associated with DC SIGN in human leprosy. **Immunobiology**, v. 218, n. 12, p. 1488–1496, dez. 2013.

LAFFERTY, K. J.; ANDRUS, L.; PROWSE, S. J. Role of lymphokine and antigen in the control of specific T cell responses. **Immunological Reviews**, v. 51, p. 279–314, 1980.

LE, K.-S. et al. CXCR5 and ICOS expression identifies a CD8 T-cell subset with TFH features in Hodgkin lymphomas. **Blood Advances**, v. 2, n. 15, p. 1889–1900, 7 ago. 2018.

LEE, K. M. et al. Molecular basis of T cell inactivation by CTLA-4. **Science** (New York, N.Y.), v. 282, n. 5397, p. 2263–2266, 18 dez. 1998.

LIN, S.-C. et al. Expression and Regulation of Thymic Stromal Lymphopoietin and Thymic Stromal Lymphopoietin Receptor Heterocomplex in the Innate-Adaptive Immunity of Pediatric Asthma. **International Journal of Molecular Sciences**, v. 19, n. 4, p. E1231, 18 abr. 2018.

LING, V. et al. Cutting edge: identification of GL50, a novel B7-like protein that functionally binds to ICOS receptor. **Journal of Immunology** (Baltimore, Md.: 1950), v. 164, n. 4, p. 1653–1657, 15 fev. 2000.

LIPSON, E. J.; DRAKE, C. G. Ipilimumab: an anti-CTLA-4 antibody for metastatic melanoma. **Clinical Cancer Research: An Official Journal of the American Association for Cancer Research**, v. 17, n. 22, p. 6958–6962, 15 nov. 2011.

LIU, D.; UZONNA, J. E. The early interaction of Leishmania with macrophages and dendritic cells and its influence on the host immune response. **Frontiers in Cellular and Infection Microbiology**, v. 2, 12 jun. 2012.

LIU, Z. et al. Distinct roles of ICOS and CD40L in human T-B cell adhesion and antibody production. **Cellular Immunology**, v. 368, p. 104420, out. 2021.

MADDUR, M. S. et al. Human B cells induce dendritic cell maturation and favour Th2 polarization by inducing OX-40 ligand. **Nature Communications**, v. 5, p. 4092, 9 jun. 2014.

MARTINY, A. et al. Altered tyrosine phosphorylation of ERK1 MAP kinase and other macrophage molecules caused by Leishmania amastigotes. **Molecular and Biochemical Parasitology**, v. 102, n. 1, p. 1–12, 30 jul. 1999.

MASSARELLI, E. et al. High OX-40 expression in the tumor immune infiltrate is a favorable prognostic factor of overall survival in non-small cell lung cancer. **Journal for Immunotherapy of Cancer**, v. 7, p. 351, 16 dez. 2019.

MCADAM, A. J. et al. Mouse inducible costimulatory molecule (ICOS) expression is enhanced by CD28 costimulation and regulates differentiation of CD4+ T cells. **Journal of Immunology** (Baltimore, Md.: 1950), v. 165, n. 9, p. 5035–5040, 1 nov. 2000.

MCNEELY, T. B.; TURCO, S. J. Inhibition of protein kinase C activity by the Leishmania donovani lipophosphoglycan. **Biochemical and Biophysical Research Communications**, v. 148, n. 2, p. 653–657, 29 out. 1987.

MESQUITA, I. et al. The impact of IL-10 dynamic modulation on host immune response against visceral leishmaniasis. **Cytokine**, v. 112, p. 16–20, dez. 2018.

MIYAHIRA, Y. et al. Involvement of ICOS-B7RP-1 costimulatory pathway in the regulation of immune responses to Leishmania major and Nippostrongylus brasiliensis infections. **Immunology Letters**, v. 89, n. 2–3, p. 193–199, 31 out. 2003.

MOULIK, S. et al. Status of IL-4 and IL-10 driven markers in experimental models of Visceral Leishmaniasis. **Parasite Immunology**, v. 43, n. 1, p. e12783, jan. 2021.

MURPHY, M. L. et al. Blockade of CTLA-4 enhances host resistance to the intracellular pathogen, Leishmania donovani. **Journal of Immunology** (Baltimore, Md.: 1950), v. 161, n. 8, p. 4153–4160, 15 out. 1998.

MURRAY, H. W. Targeting IL-27 and/or IL-10 in Experimental Murine Visceral Leishmaniasis. **The American Journal of Tropical Medicine and Hygiene**, v. 103, n. 5, p. 1938–1941, nov. 2020.

NAKAE, S. et al. Mast cells enhance T cell activation: importance of mast cell costimulatory molecules and secreted TNF. **Journal of Immunology** (Baltimore, Md.: 1950), v. 176, n. 4, p. 2238–2248, 15 fev. 2006.

NOUAILLES, G. et al. Impact of inducible co-stimulatory molecule (ICOS) on T-cell responses and protection against Mycobacterium tuberculosis infection. **European Journal of Immunology**, v. 41, n. 4, p. 981–991, 2011.

NOUËL, A. et al. B-Cells induce regulatory T cells through TGF-β/IDO production in A CTLA-4 dependent manner. **Journal of Autoimmunity**, v. 59, p. 53–60, maio 2015.

NYLÉN, S. et al. Splenic accumulation of IL-10 mRNA in T cells distinct from CD4+CD25+ (Foxp3) regulatory T cells in human visceral leishmaniasis. **The Journal of Experimental Medicine**, v. 204, n. 4, p. 805–817, 16 abr. 2007.

NYLÉN, S.; SACKS, D. Interleukin-10 and the pathogenesis of human visceral leishmaniasis. **Trends in Immunology**, v. 28, n. 9, p. 378–384, set. 2007.

OUALHA, R. et al. Infection of Human Neutrophils With Leishmania infantum or Leishmania major Strains Triggers Activation and Differential Cytokines Release. **Frontiers in Cellular and Infection Microbiology**, v. 9, p. 153, 2019.

OYEWOLE-SAID, D. et al. Beyond T-Cells: Functional Characterization of CTLA-4 Expression in Immune and Non-Immune Cell Types. **Frontiers in Immunology**, v. 11, p. 608024, 2020.

PANNETON, V. et al. Inducible T-cell co-stimulator: Signaling mechanisms in T follicular helper cells and beyond. **Immunological Reviews**, v. 291, n. 1, p. 91–103, set. 2019.

PARASHAR, S.; MUKHOPADHYAY, A. GTPase Sar1 regulates the trafficking and secretion of the virulence factor gp63 in Leishmania. **The Journal of Biological Chemistry**, v. 292, n. 29, p. 12111–12125, 21 jul. 2017.

PAULOS, C. M. et al. The inducible costimulator (ICOS) is critical for the development of human T(H)17 cells. **Science Translational Medicine**, v. 2, n. 55, p. 55ra78, 27 out. 2010.

PEDICORD, V. A. et al. Single dose of anti-CTLA-4 enhances CD8+ T-cell memory formation, function, and maintenance. **Proceedings of the National Academy of Sciences of the United States of America**, v. 108, n. 1, p. 266–271, 4 jan. 2011.

PODINOVSKAIA, M.; DESCOTEAUX, A. Leishmania and the macrophage: a multifaceted interaction. **Future Microbiology**, v. 10, n. 1, p. 111–129, 2015.

RAO, D. A. T Cells That Help B Cells in Chronically Inflamed Tissues. **Frontiers in Immunology**, v. 9, p. 1924, 23 ago. 2018.

RODRIGUES-NETO, J. F. et al. CD45RO+ T Cells and T Cell Activation in the Long-Lasting Immunity after Leishmania infantum Infection. **The American Journal of Tropical Medicine and Hygiene**, v. 98, n. 3, p. 875–882, mar. 2018.

ROGERS, P. R. et al. OX40 promotes Bcl-xL and Bcl-2 expression and is essential for long-term survival of CD4 T cells. **Immunity**, v. 15, n. 3, p. 445–455, set. 2001.

SACKS, D. L. et al. An analysis of T cell responsiveness in Indian kala-azar. **Journal of Immunology** (Baltimore, Md.: 1950), v. 138, n. 3, p. 908–913, 1 fev. 1987.

SAHA, S. et al. IL-10- and TGF-beta-mediated susceptibility in kala-azar and post-kala-azar dermal leishmaniasis: the significance of amphotericin B in the control of Leishmania donovani infection in India. **Journal of Immunology** (Baltimore, Md.: 1950), v. 179, n. 8, p. 5592–5603, 15 out. 2007.

SAINI, S.; RAI, A. K. Hamster, a close model for visceral leishmaniasis: Opportunities and challenges. **Parasite Immunology**, v. 42, n. 10, p. e12768, out. 2020.

SAUNDERS, E. C.; MCCONVILLE, M. J. Immunometabolism of Leishmania granulomas. **Immunology and Cell Biology**, v. 98, n. 10, p. 832–844, nov. 2020.

SCHEPP, J. et al. 14 Years after Discovery: Clinical Follow-up on 15 Patients with Inducible Co-Stimulator Deficiency. **Frontiers in Immunology**, v. 8, p. 964, 16 ago. 2017.

SCHWARTZ, R. H. et al. T-cell clonal anergy. **Cold Spring Harbor Symposia on Quantitative Biology**, v. 54 Pt 2, p. 605–610, 1989.

SEIDEL, J. A.; OTSUKA, A.; KABASHIMA, K. Anti-PD-1 and Anti-CTLA-4 Therapies in Cancer: Mechanisms of Action, Efficacy, and Limitations. **Frontiers in Oncology**, v. 8, p. 86, 2018.

SELVAPANDIYAN, A. et al. Innovations for the elimination and control of visceral leishmaniasis. **PLoS Neglected Tropical Diseases**, v. 13, n. 9, 19 set. 2019.

SHARPE, A. H. Mechanisms of Costimulation. **Immunological reviews**, v. 229, n. 1, p. 5–11, maio 2009.

SHIO, M. T. et al. Host cell signalling and leishmania mechanisms of evasion. **Journal of Tropical Medicine**, v. 2012, p. 819512, 2012.

SIMPSON, T. R.; QUEZADA, S. A.; ALLISON, J. P. Regulation of CD4 T cell activation and effector function by inducible costimulator (ICOS). **Current Opinion in Immunology**, v. 22, n. 3, p. 326–332, jun. 2010.

SINGH, O. P. et al. Reassessment of Immune Correlates in Human Visceral Leishmaniasis as Defined by Cytokine Release in Whole Blood. **Clinical and Vaccine Immunology** : CVI, v. 19, n. 6, p. 961–966, jun. 2012.

SMIDA, M. et al. PAG/Cbp suppression reveals a contribution of CTLA-4 to setting the activation threshold in T cells. **Cell communication and signaling**: CCS, v. 11, n. 1, p. 28, 19 abr. 2013.

SOLINAS, C.; GU-TRANTIEN, C.; WILLARD-GALLO, K. The rationale behind targeting the ICOS-ICOS ligand costimulatory pathway in cancer immunotherapy. **ESMO Open**, v. 5, n. 1, p. e000544, 29 jan. 2020.

SONG, J.; SO, T.; CROFT, M. Activation of NF-kappaB1 by OX40 contributes to antigen-driven T cell expansion and survival. **Journal of Immunology** (Baltimore, Md.: 1950), v. 180, n. 11, p. 7240–7248, 1 jun. 2008.

SORIANO, A. et al. Plasma stromal cell-derived factor (SDF)-1 levels, SDF1-3'A genotype, and expression of CXCR4 on T lymphocytes: their impact on resistance to human immunodeficiency virus type 1 infection and its progression. **The Journal of Infectious Diseases**, v. 186, n. 7, p. 922–931, 1 out. 2002.

SRIVASTAVA, A. et al. Identification of TLR inducing Th1-responsive Leishmania donovani amastigote-specific antigens. **Molecular and Cellular Biochemistry**, v. 359, n. 1–2, p. 359–368, jan. 2012.

SUKHBAATAR, O. et al. Activation and IL-10 production of specific CD4+ T cells are regulated by IL-27 during chronic infection with Plasmodium chabaudi. **Parasitology International**, v. 74, p. 101994, 1 fev. 2020.

SUNDAR, S. et al. Circulating T helper 1 (Th1) cell- and Th2 cell-associated cytokines in Indian patients with visceral leishmaniasis. **The American Journal of Tropical Medicine and Hygiene**, v. 56, n. 5, p. 522–525, maio 1997.

TESCIUBA, A. G. et al. Inducible costimulator regulates Th2-mediated inflammation, but not Th2 differentiation, in a model of allergic airway disease. **Journal of Immunology** (Baltimore, Md.: 1950), v. 167, n. 4, p. 1996–2003, 15 ago. 2001.

TEUTSCH, S. M. et al. Association of common T cell activation gene polymorphisms with multiple sclerosis in Australian patients. **Journal of Neuroimmunology**, v. 148, n. 1–2, p. 218–230, mar. 2004.

TRINCHIERI, G. Cytokines acting on or secreted by macrophages during intracellular infection (IL-10, IL-12, IFN-gamma). **Current Opinion in Immunology**, v. 9, n. 1, p. 17–23, fev. 1997.

VAN GRIENSVEN, J.; DIRO, E. Visceral Leishmaniasis: Recent Advances in Diagnostics and Treatment Regimens. **Infectious Disease Clinics of North America, Updates in Tropical Medicine**. v. 33, n. 1, p. 79–99, 1 mar. 2019.

VIEIRA, T. DA S. et al. Intraspecies Polymorphisms in the Lipophosphoglycan of L. braziliensis Differentially Modulate Macrophage Activation via TLR4. **Frontiers in Cellular and Infection Microbiology**, v. 9, p. 240, 2019.

WAKAMATSU, E.; MATHIS, D.; BENOIST, C. Convergent and divergent effects of costimulatory molecules in conventional and regulatory CD4+ T cells. **Proceedings of the National Academy of Sciences of the United States of America**, v. 110, n. 3, p. 1023–1028, 15 jan. 2013.

WALUNAS, T. L.; BAKKER, C. Y.; BLUESTONE, J. A. CTLA-4 ligation blocks CD28-dependent T cell activation. **The Journal of Experimental Medicine**, v. 183, n. 6, p. 2541–2550, 1 jun. 1996.

WAN, R. et al. Screening and antitumor effect of an anti-CTLA-4 nanobody. **Oncology Reports**, v. 39, n. 2, p. 511–518, fev. 2018.

WANG, L. et al. Circulating CTLA-4 levels and CTLA4 polymorphisms associate with disease condition and progression and hepatocellular carcinoma patients' survival in chronic hepatitis B virus infection. **International Immunopharmacology**, v. 82, p. 106377, 9 mar. 2020.

WANG, Y. et al. [Effect of immune response mediated by ICOS signaling pathway on hepatic fibrosis in mice infected with Schistosoma japonicum]. Zhongguo Ji Sheng Chong Xue Yu Ji Sheng Chong Bing Za Zhi = **Chinese Journal of Parasitology & Parasitic Diseases**, v. 31, n. 5, p. 329–336, out. 2013.

WATANABE, M. et al. ICOS-mediated costimulation on Th2 differentiation is achieved by the enhancement of IL-4 receptor-mediated signaling. **Journal of Immunology** (Baltimore, Md.: 1950), v. 174, n. 4, p. 1989–1996, 15 fev. 2005.

WATERHOUSE, P. et al. Lymphoproliferative disorders with early lethality in mice deficient in Ctla-4. **Science** (New York, N.Y.), v. 270, n. 5238, p. 985–988, 10 nov. 1995.

WEBB, G. J.; HIRSCHFIELD, G. M.; LANE, P. J. L. OX40, OX40L and Autoimmunity: a Comprehensive Review. **Clinical Reviews in Allergy & Immunology**, v. 50, n. 3, p. 312–332, jun. 2016.

WHO-WORLD HEALTH ORGANIZATION. Leishmaniasis. Disponível em: <https://www.who.int/data/maternal-newborn-child-adolescent-ageing/advisory-groups/gama/activities-of-gama>. Acesso em: 16 mar. 2021.

WILLSMORE, Z. N. et al. Combined anti-PD-1 and anti-CTLA-4 checkpoint blockade: Treatment of melanoma and immune mechanisms of action. **European Journal of Immunology**, v. 51, n. 3, p. 544–556, mar. 2021.

WYKES, M. N.; LEWIN, S. R. Immune checkpoint blockade in infectious diseases. **Nature Reviews. Immunology**, v. 18, n. 2, p. 91–104, fev. 2018.

ZHOU, X. et al. Leishmania infantum-chagasi activates SHP-1 and reduces NFAT5/TonEBP activity in the mouse kidney inner medulla. **American Journal of Physiology. Renal Physiology**, v. 307, n. 5, p. F516-524, 1 set. 2014.

ZINGONI, A. et al. Cross-talk between activated human NK cells and CD4+ T cells via OX40-OX40 ligand interactions. **Journal of Immunology (**Baltimore, Md.: 1950), v. 173, n. 6, p. 3716–3724, 15 set. 2004.

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