Minireview

Inflammation and mitochondria in the pathogenesis of chronic Chagas disease cardiomyopathy

João Paulo Silva Nunes[1,2,3], Vinicius Moraes de Paiva Roda[1,2], Pauline Andrieux[4], Jorge Kalil[1,2,3], Christophe Chevillard[4] and Edecio Cunha-Neto[1,2,3]

[1]Laboratory of Immunology, Heart Institute (InCor), Faculdade de Medicina da Universidade de São Paulo, 05403-900 São Paulo, Brazil; [2]Division of Clinical Immunology and Allergy, Faculdade de Medicina da Universidade de São Paulo, 01246-903 São Paulo, Brazil; [3]Institute for Investigation in Immunology (III), Instituto Nacional de Ciência e Tecnologia (INCT), 05403-900 São Paulo, Brazil; [4]Institut National de la Santé Et de la Recherche Médicale (INSERM), Unité Mixte de Recherche (UMR) U1090, Aix Marseille Université, TAGC Theories and Approaches of Genomic Complexity, Institut MarMaRa, 13288 Marseille, France
Corresponding author: Edecio Cunha-Neto. Email: edecunha@usp.br

Impact Statement

This review article highlights the significant impact of mitochondrial dysfunction on Chagas disease (CD) cardiomyopathy. By examining the existing body of research, it provides a comprehensive analysis of the detrimental consequences resulting from impaired mitochondrial function in this cardiac condition. We review new intricate relationship between CD cardiomyopathy, inflammatory response, mitochondrial dysfunction and energy disbalance, elucidating some molecular mechanisms and pathways involved. This review not only enhances our understanding of the disease pathogenesis but also emphasizes the crucial role of mitochondria in cardiac function and its potential as a therapeutic target for mitigating CD cardiomyopathy.

Abstract

Chagas disease (CD), caused by the protozoan parasite *Trypanosoma cruzi*, is a neglected disease affecting around 6 million people. About 30% of CD patients develop chronic Chagas disease cardiomyopathy (CCC), an inflammatory cardiomyopathy that occurs decades after the initial infection, while most infected patients (60%) remain asymptomatic in the so-called indeterminate form (IF). Death results from heart failure or arrhythmia in a subset of CCC patients. Myocardial fibrosis, inflammation, and mitochondrial dysfunction are involved in the arrhythmia substrate and triggering events. Survival in CCC is worse than in other cardiomyopathies, which may be linked to a Th1-T cell rich myocarditis with abundant interferon (IFN)-γ and tumor necrosis factor (TNF)-α, selectively lower levels of mitochondrial energy metabolism enzymes in the heart, and reduced levels of high-energy phosphate, indicating poor adenosine triphosphate (ATP) production. IFN-γ and TNF-α signaling, which are constitutively upregulated in CD patients, negatively affect mitochondrial function in cardiomyocytes, recapitulating findings in CCC heart tissue. Genetic studies such as whole-exome sequencing (WES) in nuclear families with multiple CCC/IF cases has disclosed rare heterozygous pathogenic variants in mitochondrial and inflammatory genes segregating in CCC cases. In this minireview, we summarized studies showing how IFN-γ and TNF-α affect cell energy generation, mitochondrial health, and redox homeostasis in cardiomyocytes, in addition to human CD and mitochondria. We hypothesize that cytokine-induced mitochondrial dysfunction in genetically predisposed patients may be the underlying cause of CCC severity and we believe this mechanism may have a bearing on other inflammatory cardiomyopathies.

Keywords: Mitochondria dysfunction; inflammation, Chagas disease cardiomyopathy, IFN-γ, cell metabolism, antioxidants

***Experimental Biology and Medicine* 2024; 248: 2062–2071. DOI: 10.1177/15353702231220658**

Introduction

Clinical and epidemiological features of Chagas disease

Chagas disease (CD) (*American trypanosomiasis*) caused by the protozoan parasite *Trypanosoma cruzi* is estimated to affect 6 million people in endemic areas of Latin America.[1–3] Due to migration, *T. cruzi*–infected individuals have spread throughout the world, and it is estimated that 400,000 infected persons live in non-endemic countries, mainly in the United States and Europe.[4] One-third of the patients advance into chronic Chagas disease cardiomyopathy (CCC), an inflammatory dilated cardiomyopathy that frequently leads to heart failure and arrhythmia, resulting in fatality. In addition, 10% of individuals develop digestive disorders such as megacolon and megaesophagus, while the remaining patients exhibit an asymptomatic condition throughout their lives referred to as the "indeterminate"

form (IF).[5] CCC is the most prevalent etiology of myocarditis in the world, and the most important cause of non-ischemic cardiomyopathy in endemic regions of Latin America. The annual mortality rate for individuals with chronic Chagasic heart disease varies depending on their risk stratification, with a range of 1–10% in the low-risk group and up to 84% in the high-risk group.[6] The overall mortality rate is estimated at 33%, in which approximately 35–45% are directly linked to the advancement of heart failure. The worsening of heart failure can result in a significant decline in cardiac function, impaired circulation, fluid retention, and the development of life-threatening complications, ultimately contributing to the observed mortality rate.[7] Sudden cardiac death (SCD) accounts for about 50–60% of total mortality, where 80–90% are caused by ventricular tachycardia/ventricular fibrillation (VT/VF) and the remainder by bradyarrhythmia. In general, the rate and complexity of ventricular arrhythmias are higher in CCC, and prognosis is worse than in other cardiomyopathies.[8,9] Non-sustained ventricular tachycardia (NSVT) is recognized as an independent predictor of SCD.[8] In common with other cardiomyopathies, CCC shows myocardial hypertrophy and fibrosis. However, inflammation is particularly important in CCC, as clinical severity in CD is closely associated with presence of myocarditis.[10] The histopathological substrate of CCC is chronic myocarditis and fibrosis, which can occur in electrical conduction tissue or in atrial or ventricular myocardium, leading to conduction system disorders and arrhythmias, or left ventricular dysfunction associated with progressive dilation leading to heart failure. Unfortunately, no vaccine or antiparasitic drug with proven efficacy is available for chronically infected adults, when most patients are diagnosed.[11] The development of effective drugs for CCC is hampered by the limited knowledge of its pathogenesis.

In this review, we conducted a comprehensive search in journal databases for scientific studies examining the effects of the pro-inflammatory cytokines IFN-γ and TNF-α in *in vitro* experiments. Specifically, we focused on studies that investigated the impact of these cytokines on cardiomyocyte cell metabolism and mitochondrial dysfunction, as the cytokine-mediated disruption of redox balance and energy generation has emerged as a prominent topic in CD cardiomyopathy research. Our search encompassed papers published across all time ranges available in public databases (i.e. PubMed, Web of Science, Google Scholar). The following keywords were used in our search: "TNF-α and/or IFN-γ and mitochondrial dysfunction"; "TNF-α and/or IFN-γ and Chagas disease cardiomyopathy"; and "TNF-α and/or IFN-γ in cardiomyocytes" as well as "Chagas disease and mitochondria." We also included any additional papers that were cited in the selected studies. This review can support new studies and may shed light on novel treatment modalities that would be effective in ameliorating the morbimortality of CCC.

Pathogenesis and genetics of CCC

Immune dynamics in acute and chronic *T. cruzi* infection, central role of IFN-γ

The intracellular life cycle of *T. cruzi* is a major target of the antiparasite immune response.[12] While extracellular *T. cruzi* components engage Toll-like receptors (TLR) TLR2 and TLR4,[13] *T. cruzi* RNA and DNA activate endosomal TLR7 and TLR9. TLR engagement promotes the Myd88-mediated activation of NF-κB. The acidification of lysosomes facilitates the escape of *T. cruzi* into the cytoplasm, where it undergoes differentiation into replicative amastigote forms. Amastigote replication in the cytoplasm triggers the activation of inflammasomes, resulting in the release of inflammatory cytokines and activation of NF-κB. This inflammatory stimulus induces expression of pro-inflammatory cytokines including interleukin (IL)-12 in macrophages and dendritic cells, which elicits differentiation of IFN-γ-producing Th1 cells. IFN-γ induces expression of multiple other genes involved in resistance to *T. cruzi*, such as *TNFA* and *NOS2*.[14,15]

Although powerful, the immune response that occurs during acute infection leads to partial parasite control. *T. cruzi* evades complete eradication, leading to the establishment of a chronic persistent infection with low parasitism. *T. cruzi*–infected individuals maintain increased production of inflammatory/Th1 cytokines like IFN-γ and TNF-α as compared to healthy individuals, as a result of persistent stimulus of innate and specific immunity.[16,17] CCC patients show an even more increased number of IFN-γ-producing Th1-T cells accompanied by elevated levels of plasma IFN-γ and TNF-α when compared to IF patients. Conversely, the numbers of IL-10-producing regulatory T cells, as well as Ebi3/IL-27p28, are lower in CCC patients as compared to IF. These observations align with a deficiency in the regulatory arm of the immune response in CCC.[17–20]

The increased proportion of circulating Th1-T cells is reflected in CCC myocardium. IFN-γ is the most upregulated cytokine[21] and the top upstream regulator of the gene expression profile in CCC heart tissue.[21,22] Circulating IFN-γ-producing CCR5 + CXCR3 + Th1-T cells are more abundant in the blood of CCC than IF patients,[23] and the same cells were identified in CCC heart tissue, along with the chemokine ligands of receptors CCR5 and CXCR3, CCL5 and CXCL9, respectively.[24] Together, this suggests that locally produced Th1-T cell-attracting chemokines play a role in the selective accumulation of Th1-T cells in CCC hearts. Thus, we believe the unopposed IFN-γ action in CCC myocardium is linked to the fact that factors that reduce IFN-γ production and/or Th1-T cell differentiation and regulatory T cells are downregulated in CCC patients. This lack of regulation could explain the destructiveness of the inflammatory infiltrate, most likely due to excessive collateral damage by IFN-γ-producing T cells as described in acutely *T. cruzi*–infected mice. IFN-γ is thus considered the culprit of CCC.[25]

Mitochondria in the pathogenesis of CCC

Evidence of mitochondrial dysfunction has been found in hearts of animal models of CD, as well as the myocardium of CCC patients. This is especially relevant for CCC pathogenesis, since mitochondrial dysfunction is a paramount feature of heart failure of diverse etiologies.[26] Nisha Garg and her group pioneered and studied in detail mitochondrial dysfunction in hearts of murine models of acute and

chronic CD and *in vitro* infection models.[27] In addition, two studies reported that IFN-γ transgenic mice exhibit chronic active myocarditis and cardiomyopathy, which is mediated by TNF-α.[28,29] Regarding mitochondrial damage in human CCC, our group described altered expression of mitochondrial genes and 16S mitochondrial rRNA in CCC heart lesions.[21,30] We also found a selective reduction of protein expression of ATP synthase and creatine kinase activity—key mitochondrial energy metabolism enzymes—in CCC heart lesions.[31] Six enzymes belonging to fatty acid beta-oxidation present reduced expression in CCC myocardium, as compared with one or two in non-inflammatory cardiomyopathies.[32] Cevey *et al*.[33] reported that treatment with fenofibrate, a PPARα agonist, was able to induce mitochondrial fatty acid beta-oxidation, restore left ventricular function, reduce myocarditis and fibrosis in a murine model of CCC. A reduction of high-energy phosphates, indicative of reduced ATP production, has been observed *in vivo* in CCC heart tissue.[34] In agreement, mitochondrial DNA content was found to be reduced in CCC heart tissue as compared to healthy or DCM patients.[35] Comparatively, CCC exhibited heightened nitro-oxidative stress in contrast to DCM, further indicating that mitochondrial function is especially disturbed in CCC. Upon performing pathways analysis of proteomic and transcriptomic studies in CCC myocardium, our research team observed an enrichment in the pathways of mitochondrial dysfunction, mitochondrial translation, and quality control. In addition, we observed suppressed pathways associated with the tricarboxylic acid (TCA) cycle, respiratory electron transport chain, lipid oxidation, and transmembrane potential of mitochondria. Notably, these findings were more pronounced in CCC compared to non-inflammatory cardiomyopathies.[21,32]

IFN-γ and TNF-α and energy generation

IFN-γ and TNF-α are essential pro-inflammatory cytokines in the antimicrobial response. Indeed, these cytokines allow a metabolic reprogramming essential to immune cells to activate and fight a pathogen. When innate immune cells such as dendritic cells and macrophages are activated, by a molecular pattern associated with a pathogen/danger (PAMP/DAMP) or metabolism-associated danger signal (MADS) via pattern recognition receptors (PRR), some of these cells become antigen presenting cells (APCs).[36]

The APCs change their metabolism depending on whether they are anti-inflammatory or pro-inflammatory. In a pro-inflammatory role, APCs will release inflammatory cytokines including IL-1, TNF-α, and IL-6, among others. These cytokines contribute to the initiation and amplification of the inflammatory response, which lead to a polarization of Mφ macrophages into M1 macrophages which is the form most adapted to a pro-inflammatory profile, this type of macrophage has a much stronger phagocytic capacity and pathogenic destruction activity.[37–39] M1 macrophages in turn, secrete high levels of pro-inflammatory cytokines such as IL-1β, IL-6, IL-12, and TNF-α,[40,41] which leads to macrophage self-activation but also to natural killer (NK) cell activation and amplification of the Th1 response.[42]

To activate M0 macrophages in M1 with IFN-γ and LPS, cells switch metabolism. M0 macrophages showed a transition of metabolic pathways to produce adenosine triphosphate (ATP), between the oxidative phosphorylation (OXPHOS) pathway and the tricarboxylic acid cycle (TCA) versus energy production by the glycolytic and pentose phosphate pathways (PPPs).[43,44] An alteration of OXPHOS will cause a disruption of TCA, as many positive and negative allosteric regulators control the metabolic flux of this cycle. The disturbed functioning of these two pathways leads to the formation of reactive oxygen species (ROS).[36] Moreover, this alteration could be due to the stimulation of the cells by TNF-α. Indeed, several studies showed that TNF-α leads to production of mitochondrial reactive oxygen species (mtROS) by altering the complex I of the mitochondrial respiratory chain.[45,46] Superoxide, along with nitric oxide, plays a crucial role in the inflammatory and antimicrobial response, as demonstrated in the context of *T. cruzi* infection. This combination generates peroxynitrite (ONOO–), which effectively eradicates intracellular *T. cruzi* amastigotes.[47,48] In addition, it has been shown that stimulation with IFN-γ and TNF-α leads to NO production by induction of nitric oxide synthase 2 (NOS2).[49,50] IFN-γ is also essential for the polarization of T cells into cytotoxic CD8 + T cells, upon activation the Th1-T cells will switch from OXPHOS to glycolysis.[36] Although IFN-γ and TNF-α are essential for the activation and polarization of the immune system toward a pro-inflammatory action, their action in other cell types of the organism—in special, cardiomyocytes—can have a deleterious effect.

Several diseases that are due to chronic inflammation, such as Chronic inflammatory bowel disease,[51] chronic CD,[35] psoriasis,[52] non-infectious uveitis[53] and many others, all have in common the deleterious effect of pro-inflammatory cytokines such as IFN-γ and TNF-α. During chronic inflammation, cells belonging to the inflammatory sites, cardiac, intestinal, epithelial cells are constantly stimulated with inflammatory cytokines. These cytokines will have the effect of switching the cellular metabolism from OXPHOS and TCA to glycolysis.[54] A decrease in ATP is then visible, concomitant with an increase in the generation of mtROS.[31,35,46] NO production was also observed in the case of stimulation with IFN-γ and TNF-α in human cell cardiomyocytes *in vitro* and in a model for Alzheimer's disease by the STAT1/NF-κB/NOS2 pathway.[35,55] IFN-γ and TNF-α also mediated mitochondria depolarization, increased NO, ROS, lipid peroxidation, and decreased creatine kinase activity in other cell types, such as hepatic cells[56] and skeletal muscle cells.[57] The decrease in ATP, the increase in ROS and the switch between OXPHOS and glycolysis may be partly explained by the fact that TNF-α has the ability to reduce the expression of the four subunits of the complex I.[46] Indeed, reduced expression of complex I may cause increase in ROS, a reduction of ATP but also an accumulation of nicotinamide adenine dinucleotide (NADH). NADH inhibits all enzymes in the TCA cycle, so the TCA is possibly altered in this case. In M1 pro-inflammatory macrophages, it has already been said that stopping the TCA causes the modification of the metabolism with citrate which no longer participates in the TCA but joins the

cycle of PPP.[58] Succinate, which activates hypoxia-inducible factor-α (HIF1-α), could be one of the causes of the metabolism change, as HIF1-α promotes glycolysis and PPP.[46] IFN-γ and TNF-α also cause damage to mitochondria via the reduction of mitochondrial membrane potential (ΔΨm) and the decrease in mitochondrial DNA.[35,59]

Stimulation with IFN-γ and TNF-α leads to an increase in glucose and PPP metabolism by impairing the OXPHOS metabolism and the fatty acid pathway. In one study, it was shown that these cytokines decrease the dependency of cardiac cells on the fatty acid pathway and the glutamine oxidation pathway.[35] In addition, a study on patients with CCC, a condition in which IFN-γ and TNF-α are locally present in large amounts, showed a decrease in enzymes of the beta-oxidation pathway in cardiac biopsies.[32] These elements suggest that TNF-α and IFN-γ cause a shift in metabolism between OXPHOS/fatty acid pathway and toward glycolysis/PPP (Figure 1).

Role of IFN-γ and TNF-α in mitochondrial damage

Mitochondrial dysfunction is one of the main pathophysiological mechanisms of chronic disease, like cardiomyopathies and neurodegenerative diseases.[60] Multiple studies demonstrate that inflammatory cytokines such as IFN-γ and TNF-α cause mitochondrial damage in diverse cell types. IFN-γ and TNF-α are abundantly produced in CD in response to the persistent stimulus of *T. cruzi* infection. Multiple studies have shown that these cytokines cause a reduction in oxidative metabolism, in the expression of energy metabolism enzymes, in the activity of the lipid beta-oxidation pathway and mitochondrial ATP production, by an NF-κB dependent pathway.[61] In our research, we have discovered that IFN-γ and TNF-α stimulation of AC16 cardiomyocytes leads to the promotion of nitro-oxidative stress and reduction of mitochondrial copy number, which closely mirror the findings in IFN-γ and TNF-α-rich CCC heart tissue.[35]

OXPHOS is the process by which ATP is generated from the energy released by the transfer of electrons from NADH and flavin adenine dinucleotide (FADH2) to molecular oxygen in the mitochondria. This process is mediated by a series of enzyme supercomplexes, called the electron transport chain (ETC), which are located in the inner mitochondrial membrane.[62]

The energy released by the electron transfer is used to pump protons (H+) across the inner mitochondrial membrane, creating an electrochemical gradient (mitochondrial membrane potential, ΔΨm) which drives ATP synthase for ATP synthesis. TNF-α and IFN-γ also can disrupt cardiomyocyte metabolism, decreasing basal and maximal respiration and ATP production.[63] Recent results showed that IFN-γ caused a dose-dependent reduction in ΔΨm and that TNF-α alone or in combination with IFN-γ decreased mitochondrial, glycolytic, and total ATP in human cardiomyocyte AC16 cells.[35] Furthermore, these two cytokines together change cardiomyocytes energetic metabolism decreasing the dependency of glutamine and fatty acid oxidation and increasing the capacity of glucose oxidation.[35,60]

Mechanistically, TNF-α can induce glucose oxidation by downregulation of peroxisome proliferator-activated receptor-gamma coactivator (PGC-1α) and pyruvate dehydrogenase kinase 4 (PDK4).[64] Through TNF-α stimulation, NF-κB activation is triggered, facilitating its translocation to the nucleus and promoting the interaction between NF-κB p65 subunit and the transcription factor E2F1. The DNA-binding capability of E2F1 is required for the expression of PDK4, which in turn inhibits the pyruvate dehydrogenase complex through phosphorylation.[65]

A significant crosstalk occurs between NF-κB and mitochondrion-protecting proteins. NF-κB signaling downregulates sirtuin-1 (SIRT1) activity through the expression of IFN-γ, ROS and NO.[66] SIRT1, an antioxidant and anti-inflammatory protein, regulates the oxidative respiration and cellular survival and is highly expressed in the heart, acting as an inhibitor of NF-κB inflammatory signals by deacetylating the p65 subunit of the NF-κB complex.[67] While NF-κB stimulates glycolytic energy flux in acute inflammation, SIRT1 inhibits NF-κB and enhances mitochondrial oxidative metabolism through 5′-AMP-activated protein kinase (AMPK), an energy sensor that regulates the metabolism in cardiomyocytes in health and disease.[66] Indeed, treatment of *T. cruzi*–infected mice with SIRT1 and/or AMPK agonists SRT1720, resveratrol and metformin reduced myocardial NF-κB transcriptional activity, inflammation and oxidative stress, resulting in beneficial results for the amelioration of cardiac function.[68,69] Likewise, treatment of AC16 human cardiomyocyte cell line stimulated with IFN-γ/TNF-α with the same agents, NRF2 agonists, or antagonists of NOS2 and NF-κB led to restoration of ΔΨm.[35]

Dysregulated myocardial fatty acid metabolic signals were reported in some *in vitro* research using TNF-α-stimulated cardiomyocytes.[70,71] A study depicted that TNF-α-treated cells have decreased levels of AMPK.[72] The authors showed that activation of AMPK can mediate the transport of fatty acids into mitochondria through phosphorylation and inhibition of acetyl-CoA carboxylase (ACC), an important enzyme that inhibits the fatty acid oxidation, AMPK therefore enhanced the oxidation of fatty acids in HL-1 cardiomyocyte cells.[73] The same group reported that the expression of PPAR-α and PPAR-δ, transcription factors that regulate fatty acid oxidation, is decreased when cells are exposed to TNF-α.[73] TNF-α decreased carnitine O-palmitoyltransferase 1 (CPT-1) expression, which is required for the translocation of long-chain fatty acids into the mitochondrial matrix (a limiting step in beta-oxidation). The unphosphorylated ACC state catalyzes the carboxylation of acetyl-CoA into malonyl-CoA, which decreases fatty acid oxidation by inhibiting CPT-1.[74] Also, the oxidation of fatty acids is deregulated in IFN-γ-treated cardiomyocytes, similar to that observed in TNF-α treatment, where ACC phosphorylation is decreased.[73] Impairment of these enzymes by TNF-α could have, therefore, a great implication for heart dysfunction, as fatty acid oxidation is the primary source of energy for contractile function of cardiomyocytes.

Additional pathways that can be related to energetic dysfunction could be TNF-α induction of class I histone deacetylases that can alter mitochondrial complexes I and II activity[75] and a decreased inositol 1,4,5-trisphosphate (IP3) formation, possibly by decreased activity of

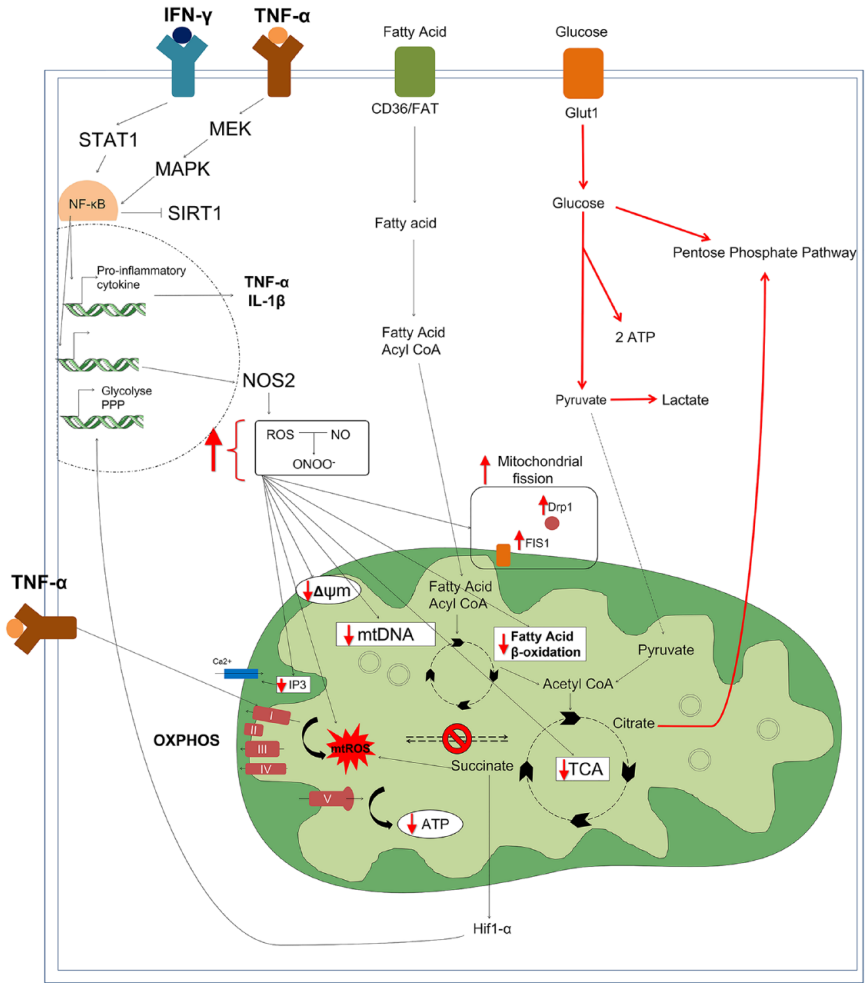

Figure 1. IFN-γ and TNF-α dysregulate energy production in mitochondria. Upon binding to their receptors, IFN-γ and TNF-α trigger the production of pro-inflammatory cytokines, including TNF-α and IL1-β, as well as the generation of nitric oxide (NO) by NOS2. This process occurs through the activation of the STAT1/NF-κB and MEK/MAPK/NF-κB pathways, respectively. Consequently, an increase in NO levels within the cells leads to alterations in mitochondrial functions such as changes in membrane potential, a decrease in mitochondrial DNA (mtDNA), a reduction in ATP production, and an elevation of mitochondrial reactive oxygen species (mtROS). Furthermore, TNF-α-induced reduction in complex I of the respiratory chain enhances mtROS production, further lowering ATP levels and causing an accumulation of NADH. This accumulation disrupts the proper functioning of the tricarboxylic acid (TCA) cycle. Consequently, several compounds, such as succinate, accumulate, which, in turn, activates HIF-1 and promotes glycolysis and the pentose phosphate pathway (PPP). The cytokines can also modulate mitochondrial fusion-fission mechanism by affecting the expression of FIS1 and DRP1 phosphorylation. Stimulation of cells with IFN-γ and TNF-α induces a metabolic shift, leading to alterations in the oxidative phosphorylation (OXPHOS) and TCA pathways. In addition, it results in a decrease in the β-oxidation of fatty acids pathway and an increase in the glucose pathway of the PPP.

glycerol-3-phosphate dehydrogenase enzyme that regulates some lipid synthesis.[76] IP3 can induce $Ca^{2}+$ uptake in mitochondrial matrix and increases ATP production in cardiomyocytes.[77]

The cytokines can have numerous effects on cardiomyocytes, deteriorating their functions and homeostasis (Table 1). TNF-α mediates changes in mitochondrial function leading to dysfunction by several mechanisms. For instance, Shen and co-workers have shown that TNF-α increases the phosphorylation of dynamin 1 like (DRP1) and its translocation to mitochondria in H9c2 cardiomyocytes, likely through increased RhoA GTPase expression.[78] In addition to increased p-Drp1, IFN-γ and TNF-α were shown to induce mitochondria fragmentation by upregulating the expression of the mitochondrial fission protein fission 1 (FIS1).[79] Both DRP1 and FIS1 play key role in mitochondrial fission and their modulation by IFN-γ and TNF-α is reported to change the cell metabolism, to increase the ΔΨm and ROS production and also to dampen basal and maximal respiration and ATP synthesis.[73,79]

Clinical similarities between genetic mitochondriopathies and CD: a whole-exome sequencing study

We have recently used whole-exome sequencing (WES) to identify pathogenic variants in nuclear families with multiple CCC/IF cases, disclosing rare heterozygous pathogenic variants in mitochondrial and inflammatory genes linked to CCC cases. In each family, the identified variants were shared only by family members with CCC, which were absent from IF siblings; such mutations occurred in mitochondrial genes in five out of the six studied families.[80] One of the variants identified is a complete loss of function mutation in the dihydroorotate dehydrogenase (*DHODH*) gene, which supplies electrons to complex III of the ETC. Significantly, incubation

Table 1. Summary of IFN-γ and TNF-α effect in *in vitro* cellular assays.

Stimulation	Effects	Model	Reference
IFN-γ	↓ Basal respiration ↓ Maximal respiration ↓ ATP ↓ Spare respiratory capacity ↓ pACC ↓ FAO	Primary cardiomyocytes from neonatal mouse hearts	Ni *et al.*[63]
IFN-γ	↓ ATP	Primary cardiomyocytes from neonatal rat hearts	Wang *et al.*[61]
IFN-γ	↓ Creatine kinase activity	Human muscle cells	Kalovidouris *et al.*[57]
TNF-α	↓ ATP ↓ Complex I subunits	Sprague-Dawley mitochondria extracted from LV	Mariappan *et al.*[46]
TNF-α	↓ Basal respiration ↓ Maximal respiration ↓ ATP turnover ↓ pAMK ↓ pACC ↓ PGC-1α ↓ CPT-1 ↓ DGAT	HL-1 cardiomyocytes	Lee *et al.*[73]
TNF-α	↓ Basal respiration ↓ Maximal respiration ↓ ATP turnover ↓ total ATP levels ↓ Complex I and II activity	HL-1 cardiomyocytes	Lkhagva *et al.*[75]
TNF-α	↓ Inositol phosphate synthesis ↓ PIP2 synthesis	Neonatal rat cardiomyocytes	Reithmann and Werdan[76]
TNF-α	↑ Glucose oxidation ↓ PGC-1α ↓ PDK4	AC16 cardiomyocytes TNF1.6 transgenic mice hearts	Palomer *et al.*[64]
TNF-α	↑ phospho-DNM1L ↑ Mitochondrial fragmentation	H9c2 cardiomyocytes	Shen *et al.*[78]
IFN-γ and TNF-α	↑ Mitochondrial hyperpolarization ↑ Mitochondrial fragmentation ↓ ATP ↑ phospho-DNM1L ↑ hFIS	H9c2 cardiomyocytes	Buoncervello *et al.*[79]
IFN-γ and TNF-α	↑ Mitochondrial depolarization ↓ Total ATP ↓ Mitochondrial ATP ↓ Glycolytic ATP ↑ Basal respiration ↑ Maximal respiration ↑ Spare respiratory capacity ↓ Fatty acid dependency ↓ Glutamine oxidation dependency	AC16 cardiomyocytes	Nunes *et al.*[35]
IFN-γ and TNF-α	↑ Mitochondrial depolarization ↑ NO ↑ ROS ↑ lipid peroxidation ↑ Apoptosis ↑ STAT1 nitration	Hep3B and primary hepatocytes	Lee *et al.*[56]
IFN-γ and LPS	Metabolic Shift: ↑ Glycolysis ↓ FAO ↓ TCA	Bone marrow–derived macrophages	Liu *et al.*[43]
IFN-γ and LPS	↑ PPP	Bone marrow–derived macrophages	Haschemi *et al.*[44]

IFN-γ: interferon-γ; TNF-α: tumor necrosis factor-α; ATP: adenosine triphosphate; CPT: carnitine O-palmitoyltransferase 1; PDK4: pyruvate dehydrogenase kinase 4; NO: nitric oxide; ROS: reactive oxygen species; TCA: tricarboxylic acid; PPP: pentose phosphate pathway; pACC: phosphorylated acetyl-CoA carboxylase; FAO: fatty acid oxidation; LV: left ventricle; DGAT: diacylglycerol O-acyltransferase; STAT1: signal transducer and activator of transcription 1; LPS: lipopolysaccharide.

with IFN-γ on the human cardiomyocyte cell line treated with an inhibitor of dihydroorotate dehydrogenase brequinar markedly reduced ΔΨm in synergy with IFN-γ and TNF-α, indicating increased mitochondrial dysfunction.[80] Another variant linked to CCC in another family leads to a stopgain variant at exon 8 of RPSUD3, involved in the assembly of the mitochondrial ribosome, creating a truncated version of the RPSUD3 protein, lacking 24% of its C-terminal sequence.

Mitochondriopaties, caused by homozygous pathogenic variants of genes encoded in the nuclear DNA, or in the mitochondrial DNA, are the most common group of monogenic syndromes. They severely damage the energy metabolism and cause mitochondrial dysfunction and functional impairment in tissues with high metabolic demand, such as cardiac and skeletal muscle, nervous tissue, liver, and kidneys. They can present as syndromes affecting individual organs or tissues or combinations of them and each clinical syndrome is associated with a specific gene or gene group bearing the variant; the clinical presentation can vary between intrauterine to the adult age. Significantly, 30–40% of mitochondriopathy patients evolve with cardiomyopathy, heart conduction abnormalities and severe arrhythmia,[81,82] and 15% to gastrointestinal motility disorders, including megaesophagus and megacolon, with denervation of myoenteric nervous plexi, and autonomic nervous system disturbances. The similarity in the proportions of clinical outcomes with those of CD (30% CCC, 10% digestive form motility disorders[83]) is impressive and made us hypothesize that heterozygous pathogenic variants, which may cause a partial reduction in mitochondrial function, may play a role in differential progression for CD in a two-hit phenomenon where cytokine-induced mitochondrial damage is the second hit. Genetic alterations with direct implications to mitochondrial function and immune response can therefore be important components for CD cardiomyopathy and megaesophagus susceptibility and progression.[80,83]

Limitations

This review has compiled most, if not all, relevant scientific data concerning the interplay between genetics, inflammation/cytokines, and mitochondrial dysfunction in the pathogenesis of human CCC, but it did not include reports on animal models of CD. While this review may not directly influence public policies regarding CCC therapy, it significantly improves our understanding of the disease's pathogenesis. Furthermore, it underscores the vital role of mitochondria in cardiac function and highlights their potential as a therapeutic target for mitigating CD cardiomyopathy.

Future directions

This review provides a summary of the significant roles played by genetic factors and IFN-γ and TNF-α in mitochondrial dysfunction in cardiomyocytes, which could explain the worse prognoses for CD cardiomyopathy. The harmful effects of these cytokines together with gene polymorphisms in mitochondria genes could contribute to the progression of cardiac dysfunction. Therefore, targeting antioxidant and mitochondria-sparing pathways that can mitigate cytokine-induced mitochondrial dysfunction holds great potential as a therapeutic approach to alleviate the detrimental effects of IFN-γ and TNF-α. Further research is required to explore the effectiveness and safety of antioxidant- and mitochondria-sparing–based interventions on cytokine-induced mitochondrial dysfunction. Repositioning of clinically approved drugs may pave the way for novel treatments that ameliorate mitochondrial dysfunction and enhance the clinical prognosis and treatment of patients with CD cardiomyopathy, as well as other conditions associated with chronic inflammation and dysregulated inflammatory cytokines.

Conclusions

IFN-γ and TNF-α have negative impacts on mitochondrial function through various mechanisms. These cytokines induce the production of reactive oxygen species (ROS), disrupt the activity of the electron transport chain, disturb mitochondrial membrane potential ($\Delta\Psi m$), and trigger the opening of mitochondrial permeability transition pore. They also cause mitochondrial DNA damage and alter mitochondrial biogenesis, creating a cycle of mitochondrial dysfunction and cardiac injury. The specific implications of IFN-γ and TNF-α in CD cardiomyopathy have not been fully explored, underscoring the unique nature of mitochondrial dysfunction in this context.

Considering the critical role of oxidative stress in the detrimental effects of IFN-γ and TNF-α on mitochondria, therapeutic strategies aimed at enhancing antioxidant defense systems show promise for intervention. Various approaches have been investigated to boost antioxidant pathways, such as administering external antioxidants, modulating internal antioxidant enzymes, activating antioxidant signaling pathways, and utilizing AMPK and sirtuin agonists.

AUTHORS' CONTRIBUTIONS

JPSN, VMdPR, PA, JK, CC, and ECN wrote and revised the manuscript.

DECLARATION OF CONFLICTING INTERESTS

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

FUNDING

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by the Institut National de la Santé et de la Recherche Médicale (INSERM); the Aix-Marseille University (grant no. AMIDEX "Internation-al_2018" MITOMUTCHAGAS); the French Agency for Research (Agence Nationale de la Recherche – ANR) (grant nos "Br-Fr-Chagas," "landscardio"). This work was founded by the Inserm Cross-Cutting Project GOLD. This project has received funding from the Ex-cellence Initiative of Aix-Marseille University – A*Midex a French "Investissements d'Avenir programme" – Institute MarMaRa AMX-19-IET-007. This work was supported by FAPESP (São Paulo State Research Funding Agency Brazil) (grant 2022/00758-0). Brazilian National Research Council (CNPq) provided a Productivity Award for ECN and JK. This work is supported by the CAPES-COFECUB program (Me987/23). CC is supported by the Inserm's PRI/IRP 2022 program (Me987/23). The funders did not play any role in the decision to publish or preparation of the manuscript.

ORCID IDS

João Paulo Silva Nunes https://orcid.org/0000-0003-4355-6669
Edecio Cunha-Neto https://orcid.org/0000-0002-3699-3345

REFERENCES

1. World Health Organization. *Chagas disease (American trypanosomiasis)*. https://www.who.int/health-topics/chagas-disease#tab=tab_1
2. Chagas disease (American trypanosomiasis). https://www.who.int/health-topics/chagas-disease#tab=tab_12021
3. World Health Organization. *Chagas disease (American trypanosomiasis)*. Geneva: WHO, 2021
4. Antinori S, Galimberti L, Bianco R, Grande R, Galli M, Corbellino M. Chagas disease in Europe: a review for the internist in the globalized world. *Eur J Intern Med* 2017;**43**:6–15
5. Cunha-Neto E, Chevillard C. Chagas disease cardiomyopathy: immunopathology and genetics. *Mediators Inflamm* 2014;**2014**:683230
6. Saraiva RM, Mediano MFF, Mendes FS, Sperandio da Silva GM, Veloso HH, Sangenis LHC, da Silva PS, Mazzoli-Rocha F, Sousa AS, Holanda MT, Hasslocher-Moreno AM. Chagas heart disease: an overview of diagnosis, manifestations, treatment, and care. *World J Cardiol* 2021;**13**:654–75
7. Bocchi EA, Bestetti RB, Scanavacca MI, Cunha Neto E, Issa VS. Chronic Chagas heart disease management: from etiology to cardiomyopathy treatment. *J Am Coll Cardiol* 2017;**70**:1510–24
8. Martinelli Filho M, De Siqueira SF, Moreira H, Fagundes A, Pedrosa A, Nishioka SD, Costa R, Scanavacca M, D'Avila A, Sosa E. Probability of occurrence of life-threatening ventricular arrhythmias in Chagas' disease versus non-Chagas' disease. *Pacing Clin Electrophysiol* 2000;**23**:1944–6

9. Pereira FT, Rocha EA, de Paula Martins Monteiro M, Lima Nde A, Rodrigues Sobrinho CR, Pires Neto Rda J. Clinical course after cardioverter-defibrillator implantation: Chagasic versus ischemic patients. *Arq Bras Cardiol* 2016;**107**:99–100
10. Higuchi ML, De Morais CF, Pereira Barreto AC, Lopes EA, Stolf N, Bellotti G, Pileggi F. The role of active myocarditis in the development of heart failure in chronic Chagas' disease: a study based on endomyocardial biopsies. *Clin Cardiol* 1987;**10**:665–70
11. Morillo CA, Marin-Neto JA, Avezum A, Sosa-Estani S, Rassi A Jr, Rosas F, Villena E, Quiroz R, Bonilla R, Britto C, Guhl F, Velazquez E, Bonilla L, Meeks B, Rao-Melacini P, Pogue J, Mattos A, Lazdins J, Rassi A, Connolly SJ, Yusuf S, BENEFIT Investigators. Randomized trial of benznidazole for chronic Chagas' cardiomyopathy. *N Engl J Med* 2015;**373**:1295–306
12. da Matta Guedes PM, Gutierrez FR, Maia FL, Milanezi CM, Silva GK, Pavanelli WR, Silva JS. IL-17 produced during *Trypanosoma cruzi* infection plays a central role in regulating parasite-induced myocarditis. *PLoS Negl Trop Dis* 2010;**4**:e604
13. Rodrigues MM, Oliveira AC, Bellio M. The immune response to *Trypanosoma cruzi*: role of toll-like receptors and perspectives for vaccine development. *J Parasitol Res* 2012;**2012**:507874
14. Cummings KL, Tarleton RL. Inducible nitric oxide synthase is not essential for control of *Trypanosoma cruzi* infection in mice. *Infect Immun* 2004;**72**:4081–9
15. Arantes RM, Marche HH, Bahia MT, Cunha FQ, Rossi MA, Silva JS. Interferon-gamma-induced nitric oxide causes intrinsic intestinal denervation in *Trypanosoma cruzi*-infected mice. *Am J Pathol* 2004;**164**:1361–8
16. Abel LC, Rizzo LV, Ianni B, Albuquerque F, Bacal F, Carrara D, Bocchi EA, Teixeira HC, Mady C, Kalil J, Cunha-Neto E. Chronic Chagas' disease cardiomyopathy patients display an increased IFN-gamma response to *Trypanosoma cruzi* infection. *J Autoimmun* 2001;**17**:99–107
17. Gomes JA, Bahia-Oliveira LM, Rocha MO, Martins-Filho OA, Gazzinelli G, Correa-Oliveira R. Evidence that development of severe cardiomyopathy in human Chagas' disease is due to a Th1-specific immune response. *Infect Immun* 2003;**71**:1185–93
18. Sousa GR, Gomes JA, Fares RC, Damásio MP, Chaves AT, Ferreira KS, Nunes MC, Medeiros NI, Valente VA, Corrêa-Oliveira R, Rocha MO. Plasma cytokine expression is associated with cardiac morbidity in Chagas disease. *PLoS ONE* 2014;**9**:e87082
19. de Araújo FF, Vitelli-Avelar DM, Teixeira-Carvalho A, Antas PR, Assis Silva Gomes J, Sathler-Avelar R, Otávio Costa Rocha M, Elói-Santos SM, Pinho RT, Correa-Oliveira R, Martins-Filho OA. Regulatory T cells phenotype in different clinical forms of Chagas' disease. *PLoS Negl Trop Dis* 2011;**5**:e992
20. Ferreira LR, Frade AF, Baron MA, Navarro IC, Kalil J, Chevillard C, Cunha-Neto E. Interferon-γ and other inflammatory mediators in cardiomyocyte signaling during Chagas disease cardiomyopathy. *World J Cardiol* 2014;**6**:782–90
21. Laugier L, Ferreira LRP, Ferreira FM, Cabantous S, Frade AF, Nunes JP, Ribeiro RA, Brochet P, Teixeira PC, Santos RHB, Bocchi EA, Bacal F, Cândido DDS, Maso VE, Nakaya HI, Kalil J, Cunha-Neto E, Chevillard C. miRNAs may play a major role in the control of gene expression in key pathobiological processes in Chagas disease cardiomyopathy. *PLoS Negl Trop Dis* 2020;**14**:e0008889
22. Cunha-Neto E, Dzau VJ, Allen PD, Stamatiou D, Benvenutti L, Higuchi ML, Koyama NS, Silva JS, Kalil J, Liew CC. Cardiac gene expression profiling provides evidence for cytokinopathy as a molecular mechanism in Chagas' disease cardiomyopathy. *Am J Pathol* 2005;**167**:305–13
23. Gomes JA, Bahia-Oliveira LM, Rocha MO, Busek SC, Teixeira MM, Silva JS, Correa-Oliveira R. Type 1 chemokine receptor expression in Chagas' disease correlates with morbidity in cardiac patients. *Infect Immun* 2005;**73**:7960–6
24. Nogueira LG, Santos RH, Ianni BM, Fiorelli AI, Mairena EC, Benvenuti LA, Frade A, Donadi E, Dias F, Saba B, Wang HT, Fragata A, Sampaio M, Hirata MH, Buck P, Mady C, Bocchi EA, Stolf NA, Kalil J, Cunha-Neto E. Myocardial chemokine expression and intensity of myocarditis in Chagas cardiomyopathy are controlled by polymorphisms in CXCL9 and CXCL10. *PLoS Negl Trop Dis* 2012;**6**:e1867
25. Medina TS, Oliveira GG, Silva MC, David BA, Silva GK, Fonseca DM, Sesti-Costa R, Frade AF, Baron MA, Ianni B, Pereira AC, Chevillard C, Cunha-Neto E, Marin-Neto JA, Silva JS. Ebi3 prevents *Trypanosoma cruzi*-induced myocarditis by dampening IFN-γ-driven inflammation. *Front Immunol* 2017;**8**:1213
26. Brown DA, Perry JB, Allen ME, Sabbah HN, Stauffer BL, Shaikh SR, Cleland JG, Colucci WS, Butler J, Voors AA, Anker SD, Pitt B, Pieske B, Filippatos G, Greene SJ, Gheorghiade M. Expert consensus document: mitochondrial function as a therapeutic target in heart failure. *Nat Rev Cardiol* 2017;**14**:238–50
27. Lopez M, Tanowitz HB, Garg NJ. Pathogenesis of chronic Chagas disease: macrophages, mitochondria, and oxidative stress. *Curr Clin Microbiol Rep* 2018;**5**:45–54
28. Reifenberg K, Lehr HA, Torzewski M, Steige G, Wiese E, Küpper I, Becker C, Ott S, Nusser P, Yamamura K, Rechtsteiner G, Warger T, Pautz A, Kleinert H, Schmidt A, Pieske B, Wenzel P, Münzel T, Löhler J. Interferon-gamma induces chronic active myocarditis and cardiomyopathy in transgenic mice. *Am J Pathol* 2007;**171**:463–72
29. Torzewski M, Wenzel P, Kleinert H, Becker C, El-Masri J, Wiese E, Brandt M, Pautz A, Twardowski L, Schmitt E, Münzel T, Reifenberg K. Chronic inflammatory cardiomyopathy of interferon γ-overexpressing transgenic mice is mediated by tumor necrosis factor-α. *Am J Pathol* 2012;**180**:73–81
30. Benatar AF, García GA, Bua J, Cerliani JP, Postan M, Tasso LM, Scaglione J, Stupirski JC, Toscano MA, Rabinovich GA, Gómez KA. Galectin-1 prevents infection and damage induced by *Trypanosoma cruzi* on cardiac cells. *PLoS Negl Trop Dis* 2015;**9**:e0004148
31. Teixeira PC, Santos RH, Fiorelli AI, Bilate AM, Benvenuti LA, Stolf NA, Kalil J, Cunha-Neto E. Selective decrease of components of the creatine kinase system and ATP synthase complex in chronic Chagas disease cardiomyopathy. *PLoS Negl Trop Dis* 2011;**5**:e1205
32. Teixeira PC, Ducret A, Langen H, Nogoceke E, Santos RHB, Silva Nunes JP, Benvenuti L, Levy D, Bydlowski SP, Bocchi EA, Kuramoto Takara A, Fiorelli AI, Stolf NA, Pomeranzeff P, Chevillard C, Kalil J, Cunha-Neto E. Impairment of multiple mitochondrial energy metabolism pathways in the heart of Chagas disease cardiomyopathy patients. *Front Immunol* 2021;**12**:755782
33. Cevey ÁC, Mirkin GA, Donato M, Rada MJ, Penas FN, Gelpi RJ, Goren NB. Treatment with Fenofibrate plus a low dose of Benznidazole attenuates cardiac dysfunction in experimental Chagas disease. *Int J Parasitol Drugs Drug Resist* 2017;**7**:378–87
34. Leme AM, Salemi VM, Parga JR, Ianni BM, Mady C, Weiss RG, Kalil-Filho R. Evaluation of the metabolism of high energy phosphates in patients with Chagas' disease. *Arq Bras Cardiol* 2010;**95**:264–70
35. Nunes JPS, Andrieux P, Brochet P, Almeida RR, Kitano E, Honda AK, Iwai LK, Andrade-Silva D, Goudenège D, Alcântara Silva KD, Vieira RS, Levy D, Bydlowski SP, Gallardo F, Torres M, Bocchi EA, Mano M, Santos RHB, Bacal F, Pomerantzeff P, Laurindo FRM, Teixeira PC, Nakaya HI, Kalil J, Procaccio V, Chevillard C, Cunha-Neto E. Co-exposure of cardiomyocytes to IFN-γ and TNF-α induces mitochondrial dysfunction and nitro-oxidative stress: implications for the pathogenesis of chronic Chagas disease cardiomyopathy. *Front Immunol* 2021;**12**:755862
36. Sun L, Yang X, Yuan Z, Wang H. Metabolic reprogramming in immune response and tissue inflammation. *Arterioscler Thromb Vasc Biol* 2020;**40**:1990–2001
37. Arango Duque G, Descoteaux A. Macrophage cytokines: involvement in immunity and infectious diseases. *Front Immunol* 2014;**5**:491
38. Atri C, Guerfali FZ, Laouini D. Role of human macrophage polarization in inflammation during infectious diseases. *Int J Mol Sci* 2018;**19**:1801
39. Chávez-Galán L, Olleros ML, Vesin D, Garcia I. Much more than M1 and M2 macrophages, there are also CD169(+) and TCR(+) macrophages. *Front Immunol* 2015;**6**:263
40. Mosser DM, Edwards JP. Exploring the full spectrum of macrophage activation. *Nat Rev Immunol* 2008;**8**:958–69
41. Barrett TJ. Macrophages in atherosclerosis regression. *Arterioscler Thromb Vasc Biol* 2020;**40**:20–33
42. Laskin DL, Sunil VR, Gardner CR, Laskin JD. Macrophages and tissue injury: agents of defense or destruction? *Annu Rev Pharmacol Toxicol* 2011;**51**:267–88

43. Liu L, Lu Y, Martinez J, Bi Y, Lian G, Wang T, Milasta S, Wang J, Yang M, Liu G, Green DR, Wang R. Proinflammatory signal suppresses proliferation and shifts macrophage metabolism from Myc-dependent to HIF1α-dependent. *Proc Natl Acad Sci USA* 2016;**113**:1564–9
44. Haschemi A, Kosma P, Gille L, Evans CR, Burant CF, Starkl P, Knapp B, Haas R, Schmid JA, Jandl C, Amir S, Lubec G, Park J, Esterbauer H, Bilban M, Brizuela L, Pospisilik JA, Otterbein LE, Wagner O. The sedoheptulose kinase CARKL directs macrophage polarization through control of glucose metabolism. *Cell Metab* 2012;**15**:813–26
45. Roca FJ, Whitworth LJ, Prag HA, Murphy MP, Ramakrishnan L. Tumor necrosis factor induces pathogenic mitochondrial ROS in tuberculosis through reverse electron transport. *Science* 2022;**376**:eabh2841
46. Mariappan N, Elks CM, Fink B, Francis J. TNF-induced mitochondrial damage: a link between mitochondrial complex I activity and left ventricular dysfunction. *Free Radic Biol Med* 2009;**46**:462–70
47. Silwal P, Kim JK, Kim YJ, Jo EK. Mitochondrial reactive oxygen species: double-edged weapon in host defense and pathological inflammation during infection. *Front Immunol* 2020;**11**:1649
48. Paiva CN, Medei E, Bozza MT. ROS and *Trypanosoma cruzi*: fuel to infection, poison to the heart. *PLoS Pathog* 2018;**14**:e1006928
49. Salim T, Sershen CL, May EE. Investigating the role of TNF-alpha and IFN-gamma activation on the dynamics of iNOS gene expression in LPS stimulated macrophages. *PLoS ONE* 2016;**11**:e0153289
50. Cortese-Krott MM, Kulakov L, Opländer C, Kolb-Bachofen V, Kröncke KD, Suschek CV. Zinc regulates iNOS-derived nitric oxide formation in endothelial cells. *Redox Biol* 2014;**2**:945–54
51. Resta-Lenert S, Barrett KE. Probiotics and commensals reverse TNF-alpha- and IFN-gamma-induced dysfunction in human intestinal epithelial cells. *Gastroenterology* 2006;**130**:731–46
52. Rendon A, Schakel K. Psoriasis pathogenesis and treatment. *Int J Mol Sci* 2019;**20**:1475
53. Hsu YR, Huang JC, Tao Y, Kaburaki T, Lee CS, Lin TC, Hsu CC, Chiou SH, Hwang DK. Noninfectious uveitis in the Asia-Pacific region. *Eye (London)* 2019;**33**:66–77
54. Rath E, Haller D. Intestinal epithelial cell metabolism at the interface of microbial dysbiosis and tissue injury. *Mucosal Immunol* 2022;**15**:595–604
55. Belkhelfa M, Rafa H, Medjeber O, Arroul-Lammali A, Behairi N, Abada-Bendib M, Makrelouf M, Belarbi S, Masmoudi AN, Tazir M, Touil-Boukoffa C. IFN-gamma and TNF-alpha are involved during Alzheimer disease progression and correlate with nitric oxide production: a study in Algerian patients. *J Interferon Cytokine Res* 2014;**34**:839–47
56. Lee HJ, Oh YK, Rhee M, Lim JY, Hwang JY, Park YS, Kwon Y, Choi KH, Jo I, Park SI, Gao B, Kim WH. The role of STAT1/IRF-1 on synergistic ROS production and loss of mitochondrial transmembrane potential during hepatic cell death induced by LPS/d-GalN. *J Mol Biol* 2007;**369**:967–84
57. Kalovidouris AE, Plotkin Z, Graesser D. Interferon-gamma inhibits proliferation, differentiation, and creatine kinase activity of cultured human muscle cells. II. A possible role in myositis. *J Rheumatol* 1993;**20**:1718–23
58. Cezard A, Monard S, Bréa-Diakite D, Guillon A, Si-Tahar M. [Metabokines reviewed: essential mediators of anti-infectious immunity]. *Med Sci (Paris)* 2021;**37**:342–8
59. Blaser H, Dostert C, Mak TW, Brenner D. TNF and ROS crosstalk in inflammation. *Trends Cell Biol* 2016;**26**:249–61
60. Chevillard C, Nunes JPS, Frade AF, Almeida RR, Pandey RP, Nascimento MS, Kalil J, Cunha-Neto E. Disease tolerance and pathogen resistance genes may underlie. *Front Immunol* 2018;**9**:2791
61. Wang D, McMillin JB, Bick R, Buja LM. Response of the neonatal rat cardiomyocyte in culture to energy depletion: effects of cytokines, nitric oxide, and heat shock proteins. *Lab Invest* 1996;**75**:809–18
62. Andrieux P, Chevillard C, Cunha-Neto E, Nunes JPS. Mitochondria as a cellular hub in infection and inflammation. *Int J Mol Sci* 2021;**22**:11338
63. Ni C, Ma P, Wang R, Lou X, Liu X, Qin Y, Xue R, Blasig I, Erben U, Qin Z. Doxorubicin-induced cardiotoxicity involves IFNγ-mediated metabolic reprogramming in cardiomyocytes. *J Pathol* 2019;**247**:320–32
64. Palomer X, Alvarez-Guardia D, Rodríguez-Calvo R, Coll T, Laguna JC, Davidson MM, Chan TO, Feldman AM, Vázquez-Carrera M. TNF-alpha reduces PGC-1alpha expression through NF-kappaB and p38 MAPK leading to increased glucose oxidation in a human cardiac cell model. *Cardiovasc Res* 2009;**81**:703–12
65. Palomer X, Álvarez-Guardia D, Davidson MM, Chan TO, Feldman AM, Vázquez-Carrera M. The interplay between NF-kappaB and E2F1 coordinately regulates inflammation and metabolism in human cardiac cells. *PLoS ONE* 2011;**6**:e19724
66. Kauppinen A, Suuronen T, Ojala J, Kaarniranta K, Salminen A. Antagonistic crosstalk between NF-κB and SIRT1 in the regulation of inflammation and metabolic disorders. *Cell Signal* 2013;**25**:1939–48
67. Caruso R, Marafini I, Franzè E, Stolfi C, Zorzi F, Monteleone I, Caprioli F, Colantoni A, Sarra M, Sedda S, Biancone L, Sileri P, Sica GS, MacDonald TT, Pallone F, Monteleone G. Defective expression of SIRT1 contributes to sustain inflammatory pathways in the gut. *Mucosal Immunol* 2014;**7**:1467–79
68. Wan X, Wen JJ, Koo SJ, Liang LY, Garg NJ. SIRT1-PGC1α-NFκB pathway of oxidative and inflammatory stress during *Trypanosoma cruzi* infection: benefits of SIRT1-targeted therapy in improving heart function in Chagas disease. *PLoS Pathog* 2016;**12**:e1005954
69. Vilar-Pereira G, Carneiro VC, Mata-Santos H, Vicentino AR, Ramos IP, Giarola NL, Feijó DF, Meyer-Fernandes JR, Paula-Neto HA, Medei E, Bozza MT, Lannes-Vieira J, Paiva CN. Resveratrol reverses functional Chagas heart disease in mice. *PLoS Pathog* 2016;**12**:e1005947
70. Lee TI, Kao YH, Tsai WC, Chung CC, Chen YC, Chen YJ. HDAC inhibition modulates cardiac PPARs and fatty acid metabolism in diabetic cardiomyopathy. *PPAR Res* 2016;**2016**:5938740
71. Lee TI, Kao YH, Chen YC, Tsai WC, Chung CC, Chen YJ. Cardiac metabolism, inflammation, and peroxisome proliferator-activated receptors modulated by 1,25-dihydroxyvitamin D3 in diabetic rats. *Int J Cardiol* 2014;**176**:151–7
72. Li X, Liu J, Lu Q, Ren D, Sun X, Rousselle T, Tan Y, Li J. AMPK: a therapeutic target of heart failure-not only metabolism regulation. *Biosci Rep* 2019;**39**:BSR20181767
73. Lee TI, Kao YH, Baigalmaa L, Lee TW, Lu YY, Chen YC, Chao TF, Chen YJ. Sodium hydrosulphide restores tumour necrosis factor-α-induced mitochondrial dysfunction and metabolic dysregulation in HL-1 cells. *J Cell Mol Med* 2019;**23**:7641–50
74. Zhou W, Tu Y, Simpson PJ, Kuhajda FP. Malonyl-CoA decarboxylase inhibition is selectively cytotoxic to human breast cancer cells. *Oncogene* 2009;**28**:2979–87
75. Lkhagva B, Kao YH, Lee TI, Lee TW, Cheng WL, Chen YJ. Activation of Class I histone deacetylases contributes to mitochondrial dysfunction in cardiomyocytes with altered complex activities. *Epigenetics* 2018;**13**:376–85
76. Reithmann C, Werdan K. Tumor necrosis factor alpha decreases inositol phosphate formation and phosphatidylinositol-bisphosphate (PIP2) synthesis in rat cardiomyocytes. *Naunyn Schmiedebergs Arch Pharmacol* 1994;**349**:175–82
77. Seidlmayer LK, Kuhn J, Berbner A, Arias-Loza PA, Williams T, Kaspar M, Czolbe M, Kwong JQ, Molkentin JD, Heinze KG, Dedkova EN, Ritter O. Inositol 1,4,5-trisphosphate-mediated sarcoplasmic reticulum-mitochondrial crosstalk influences adenosine triphosphate production via mitochondrial Ca2+ uptake through the mitochondrial ryanodine receptor in cardiac myocytes. *Cardiovasc Res* 2016;**112**:491–501
78. Shen YL, Shi YZ, Chen GG, Wang LL, Zheng MZ, Jin HF, Chen YY. TNF-α induces Drp1-mediated mitochondrial fragmentation during inflammatory cardiomyocyte injury. *Int J Mol Med* 2018;**41**:2317–27
79. Buoncervello M, Maccari S, Ascione B, Gambardella L, Marconi M, Spada M, Macchia D, Stati T, Patrizio M, Malorni W, Matarrese P, Marano G, Gabriele L. Inflammatory cytokines associated with cancer growth induce mitochondria and cytoskeleton alterations in cardiomyocytes. *J Cell Physiol* 2019;**234**:20453–68
80. Ouarhache M, Marquet S, Frade AF, Ferreira AM, Ianni B, Almeida RR, Nunes JPS, Ferreira LRP, Rigaud VO, Cândido D, Mady C, Zaniratto RCF, Buck P, Torres M, Gallardo F, Andrieux P, Bydlowsky S,

Levy D, Abel L, Cardoso CS, Santos-Junior OR, Oliveira LC, Oliveira CDL, Nunes MDC, Cobat A, Kalil J, Ribeiro AL, Sabino EC, Cunha-Neto E, Chevillard C. Rare pathogenic variants in mitochondrial and inflammation-associated genes may lead to inflammatory cardiomyopathy in Chagas disease. *J Clin Immunol* 2021;**41**:1048–63

81. El-Hattab AW, Scaglia F. Mitochondrial cardiomyopathies. *Front Cardiovasc Med* 2016;**3**:25
82. Scaglia F, Towbin JA, Craigen WJ, Belmont JW, Smith EO, Neish SR, Ware SM, Hunter JV, Fernbach SD, Vladutiu GD, Wong LJ, Vogel H. Clinical spectrum, morbidity, and mortality in 113 pediatric patients with mitochondrial disease. *Pediatrics* 2004;**114**:925–31
83. Silva KDA, Nunes JPS, Andrieux P, Brochet P, Almeida RR, Kuramoto Takara ACK, Pereira NB, Abel L, Cobat A, Zaniratto RCF, Levy D, Bydlowski SP, Cecconello I, Seguro FCBD Kalil J, Chevillard C, Cunha-Neto E. Chagas disease megaesophagus patients carrying variant *MRPS18B* P260A display nitro-oxidative stress and mitochondrial dysfunction in response to IFN-γ stimulus. *Biomedicines* 2022;**10**:2215