Chikungunya Virus Arthritis: Implications of Acute and Chronic Inflammation Mechanisms on Patient Management

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Abstract

In the past decade, arboviruses – *arthropod-borne* viruses – have been the focus of public health institutions worldwide following a spate of devastating outbreaks. Chikungunya virus (CHIKV), an arbovirus that belongs to the alphavirus genus, is a re-emerging arthritogenic virus that has caused explosive outbreaks since 2006, notably in the Reunion Island, and more recently in the Caribbean, South America, India and South-East Asia. The severity of arthritic disease caused by CHIKV has prompted public health authorities of affected countries to develop specific guidelines to tackle this pathogen. CHIKV disease (CHIKVD) manifests first as an acute stage of severe joint inflammation and febrile illness, which later progresses to a chronic stage, where patients can experience debilitating and persisting articular pain for extended periods. This review aims to provide a broad perspective on current knowledge of CHIKV pathogenesis by identifying key clinical and experimental studies that have contributed to our understanding of CHIKVD to date. In addition, this review explores the practical aspects treatment and management of both acute and chronic CHIKVD patients based on clinical experience during CHIKV outbreaks. Finally, recent findings on potential therapeutic solutions - from antivirals to immunomodulatory drugs - are reviewed to provide both viral immunologists and clinical rheumatologists with a balanced perspective on the nature of a re-emerging arboviral disease of significant public health concern, and an insight into future therapeutic approaches to better address the treatment and management of CHIKVD patients.

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Arthropod-borne viral infections have recently dominated the headlines following a spate of significant outbreaks around the world. Mosquito-borne viruses such as chikungunya virus (CHIK) have sparked the attention of global health organizations and led to robust public health interventions. Outbreaks of CHIKV had been reported in Africa, Asia, and the Indian and Pacific Oceans since the mid-2000s; a small number of cases have also been reported within Europe (1,2). In 2013 the CHIKV epidemic reached the Americas and is now spreading through South America with over one million cases reported (3-5). In the United States, travel-associated cases have been reported in 37 states (1) within mainland US (as of January 2017) with cases of local transmission also reported in Florida (2). Of more pressing concern, India (3) and most South-East Asian countries have been subjected to unabated epidemics that have proven difficult to control. Propitious climate, facilitated by seasonal monsoons and urban areas where mosquito vector control is lacking, along with a reservoir of naïve populations, are major contributing factors to the risk posed by locally-acquired CHIKV infections in developing countries (4). While developed economies that have experienced significant numbers of traveler-imported cases are somewhat equipped to manage CHIKV disease (CHIKVD) burden, less economically developed regions of the world – in particular densely populated urban areas - where epidemics have become most resilient have seen their public health systems overwhelmed by the disease (3,5). CHIKV disease (CHIKVD) symptoms include a range of acute manifestations, among which fever, severe joint and muscle pain, headache and rashes are prominent. However, there is mounting concern over the persistence of long-lasting manifestations associated with debilitating effects of the disease and the deterioration of overall health and quality of life (6-9). Among these, chronic arthralgia/arthritis (joint pain/joint stiffness plus joint swelling, respectively), musculoskeletal injury, and fatigue - and, to a lesser extent, neurocognitive and sensorineural manifestations - have been shown to contribute significantly to the economic burden of the disease (10,11). Although vaccines have reached human trials (12,13) and a range of potential antiviral compounds have undergone preclinical evaluation (14,15), no licensed vaccines or antiviral therapies are available. Both acute and chronic disease manifestations are of significant concern, and there are currently no specific, approved drugs to treat either forms of the disease. Chronic CHIKVD poses important questions with regard to public health policy and interventions: should efforts be directed at managing
the early stages of CHIKVD, to limit the consequences on population productivity and lessen the burden of a “patient rush” on – often under-equipped – hospitals, or should more attention be focused on limiting the development of chronic disease, associated with persistent rheumatic disease and a risk of long-term impact on population health?

In this review, we address the key pathophysiological mechanisms that drive acute and chronic CHIKV arthritis, arguably the most incapacitating phenotype among long-lasting CHIKVD manifestations, based on recent animal experimental disease models and epidemiological studies. We explore the latest findings in therapeutic development aimed at both limiting viral spread and at immune and inflammatory mechanisms, and address the implications of such findings in current and future clinical management of CHIKVD patients.

CHIKVD is recognized as an emerging biphasic disease described as an acute infection followed by persistent symptoms. Hence, experts have defined 3 successive clinical stages, taking into account known difficulties in practical management. In symptomatic, infected individuals, the first stage encompasses an acute incapacitating febrile viremic phase together with residual symptoms. The two last stages are characterized by persistent manifestations: a post-acute stage (from 21 days post-onset of illness (p.o.i.) to the end of the 3rd month) and a chronic stage (beyond 3 months). Schematically, the disease progresses to resolution without sequelae either spontaneously or after treatment, or to a persistence of articular and general symptoms, or to aggravation due to an inflammatory or degenerative process.

**Acute and post-acute CHIKV stage (≤ 3 months post-onset of infection)**

After a silent incubation of four to seven days, the acute stage of CHIKVD can be divided into two distinct phases, the viremic phase (5 to 10 days) and the post-viremic phase (6 to 21 days) (16-18). In the most common manifestations, the former is marked by unusually high-grade fever (>39°C) with
abrupt onset, accompanied by severe incapacitating oligo- or polyarthralgia/arthritis (≤4 or ≥4 joints, respectively, with debilitating and sudden injury), myalgia, headache, backache and cutaneous rash, whereas the latter is characterized by apyrexia (no fever), polyarthralgia/arthritis, and to a lesser extent, myalgia, pruritus, fatigue, and lymphadenopathies and anorexia, though the latter are less common (18). Joint pain is most often symmetric, additive and affects large and smaller articulations of both upper (wrists > phalanges > shoulders > elbows) and lower (ankles > knees > feet > hips) limbs. Atypical locations include vertebral, temporomandibular or sternoclavicular articulations (17). Stiffness and swelling, indicative of synovitis, are observed in ankles, phalanges, wrists and toes, but very exceptionally in larger joints (17). After infection, immunoglobulin (Ig) M antibody isotype appears within days of onset of symptoms and neutralizing anti-CHIKV IgG typically by the second week (19).

Acute CHIKV infection (<21 days post-onset of infection; p.o.i.) is consistently associated with a strong host antiviral type 1 interferon (IFN) response. RNA-Seq analysis of acute CHIKV infection in a mouse model revealed that approximately half of all up-regulated genes following infection are interferon-stimulated genes (ISG) (20,21). IFN-α is detected early in infection, reaching levels of 700 pg/mL in plasma of CHIKV-infected patients, coinciding with onset of disease symptoms (16,22). During the acute stage, patient viremia has been shown to directly correlate with production of cytokines such as IFN-α and interleukin (IL)-6 (20,22). Although this correlation is lacking in other studies, it is broadly accepted that all patients experiencing acute CHIKV infection produce high levels of IFN-α (20). This is reciprocated in animal models of acute CHIKVD (21,23-25). Indeed, CHIKV infection is generally lethal in mice deficient in type I IFN responses, while arthritogenic manifestations are exacerbated in IFN-α/β-deficient mice infected with CHIKV (24-26). Furthermore, downstream effectors of IFN induction such as ISGs play key roles in inhibiting CHIKV infection (21), and a strong IFN response is associated with a milder course of disease in mice infected with alphaviruses (24,25,27). The ability of alphaviruses to develop mechanisms to inhibit host IFN induction and signaling (28) also highlights the importance of this early innate immune response in controlling infection (29).
In addition to a potent type I IFN response, the acute stage of CHIKV infection (< 21 days post-onset of infection, p.o.i.) is also associated with elevated patient plasma levels of multiple soluble factors, including pro-inflammatory cytokines and chemokines (CCL2, MIF, CCL4, CXCL10, IL-6, IL-8 and IL-16), anti-inflammatory cytokines (IL-1RA, IL-10 and IL-13), growth factors (G-CSF, GM-CSF, VEGF and SCGF-β) and other mediators (IFN-γ, IL-4, IL-7 and CXCL9) (20-22,30-33). This results in intense monocyte trafficking to the infected tissues (33), along with strong activation of both CD8+ T and NK cells to assist in clearing the virus (20,34). The post-acute stage of CHIKVD (4th week to 3rd month p.o.i.) is characterized by very polymorphous manifestations prolonging the initial inflammatory symptoms (acute arthritis) by diverse rheumatic disorders, including periarticular involvements, slowly regressive enthesitis, tenosynovitis and bursitis, together with non-rheumatic and systemic symptoms (18). This stage is marked by persistence of IL-6 and GM-CSF secretions and production of IL-17 in the most afflicted patients, or by eotaxin and hepatocyte growth factor in those who have fully recovered (20).

**Chronic CHIKV stage (> 3 months post-onset of infection)**

While 50 to 60% of acute patients recover fully or with mild to moderate sequelae, some evolve to a chronic stage which can last up to several years (18,35). Beyond the post-acute stage, chronic CHIKVD is thus characterized by persistence of arthritic conditions associated with long-term sequelae, such as fatigue and depression, stemming from prior rheumatic conditions – although not a pre-requisite - or in the wake of an authentic arthritogenic alphaviral infection. Chronic CHIKVD is associated with high levels of circulating IL-6 and IL-12 (22,31,36). IL-6 is specifically expressed in the affected joints and could stimulate the release of RANKL while inhibiting the action of its decoy receptor osteoprotegerin (OPG) released by osteoblasts, resulting in osteoclastogenesis and severe bone loss, as has been shown in mice (37-40). Indeed, while bone loss is not a defining feature in chronic CHIKV arthritic patients, recent studies appear to implicate IL-6 as a possible biomarker of chronic CHIKVD (41,42). In addition to soluble host factors responsible for regulating antiviral
responses, CD4+ T cells have been shown to be a major driver of arthritic disease during CHIKV infection in mouse models (43,44), but also in the follow-up of chronic CHIKV arthritis patients (45). Moreover, impaired NK cell function was shown to be associated with chronic arthritis in infected patients (46). Importantly, regulatory T cells (Treg), a subset of CD4+ T cells, may have a role in moderating excessive CHIKV-induced immune responses. Treg compete with naive T cells to interact with antigen-presenting dendritic cells. Treg interacting with dendritic cells were shown to cause mature dendritic cells to downregulate co-stimulatory signaling, curtailing the expansion of CHIKV-specific CD4+ T cells, thus effectively reducing CHIKV-induced joint swelling in mice (47). B cells and neutralizing antibodies, especially IgG3 subclass, are also critical for CHIKV clearance (19,48).

The immaturity of cellular and humoral immunity may, if only partly, explain the rarity of chronic arthralgia in children before the age of three years.

The role of myeloid cells is less clear at the chronic stage of human CHIKV infection, where they may be involved in the clearance of infected cell debris which, when acting as a source of pathogen-associated molecular patterns (PAMPs), may trigger or drive chronic CHIKV (49). Monocytes and macrophages recruited to the sites of inflammation during CHIKV infection dominate the cellular infiltrates (23,33). However, in studies using mice deficient for the chemokine receptor CCR2, the monocyte/macrophage infiltrate was replaced by a severe neutrophil infiltrate, exacerbating CHIKV-induced inflammation and cartilage damage (50). Thus, although recruited CCR2+ monocytes/macrophages contribute to inflammation, they may also be required to prevent excessive pathology or promote resolution of disease following CHIKV infection.

Interestingly, a recent RNA-Seq analysis of CHIKV-infected mouse tissues found good concordance with the expression of genes reported to be up-regulated in CHIKV patients (21). Most genes were associated with inflammation, indicating consistent pro-inflammatory gene expression in both mouse and non-human primate models and CHIKV-infected patients. This suggests that chronic CHIKV may represent an extension of acute disease rather than an activation of, or progression to, new
inflammatory immunopathologies; anti-inflammatory treatments that effectively target acute CHIKV may thus also have utility in treating chronic CHIKV (21). Prominent in the RNA-Seq analysis of CHIKV infection in mice was the serine protease granzyme A (21). Granzyme A was found to have a pro-inflammatory role in CHIKV in mice and was also found in the sera of CHIKV-infected non-human primates and in symptomatic CHIKV-infected patients. The mouse granzyme A inhibitor, Serpinb6b, significantly reduced foot swelling in CHIKV infected mice, presenting granzyme A as a potential therapeutic candidate (21). Granzyme A was also recently shown to be important in a mouse model of rheumatoid arthritis (51) - a further example of the significant overlap between CHIKV arthritis and rheumatoid arthritis (43,45).

**Chronic CHIKV arthritis and implications for therapeutic interventions**

*Risk Factors for Development of Chronic CHIKV arthralgia/arthritis.* Although most patients recover from acute CHIKV infection within days or weeks, instances of persistent post-CHIKV rheumatic illness – chronic and incapacitating joint morbidities - have been reported in a recent systematic review (35), and in up to 40% of patients on average in a recent meta-analysis (52). This often painful, incapacitating chronic CHIKV - of a primarily rheumatic nature, though as noted earlier, prior rheumatic conditions are not a pre-requisite - can persist for years in some patients and is highly detrimental to their health and quality of life (6-10,36,52-63). Although there have been numerous studies focusing on acute CHIKV infection and potential treatments, the mechanisms that cause CHIKV to progress to a chronic stage are still poorly understood.

One hypothesis is that comorbidities such as pre-existing joint disease (e.g., osteoarthritis or rheumatoid arthritis), metabolic syndrome features (e.g., hypertension, obesity or diabetes mellitus) or both, prior to CHIKV infection may exacerbate CHIKV-induced arthritis or increase patient susceptibility to developing chronic CHIKV polyarthritis (54,58,59). In the TELECHIK population-based study, pre-existing rheumatic disorders and metabolic syndrome were both associated with long-lasting rheumatic pain in an unadjusted analysis (10,58), yet a separate study from La Réunion...
found that only 2.8% of the hospital-based patients that suffered from chronic CHIKVD had pre-existing joint disease (59). Given such discrepancies across studies, any link between pre-existing joint disease and increased risk of developing CHIKV-induced arthritic disease remains unproven (35,53,57,62,63). Risk factors for the development of chronic CHIKV arthralgia include age over 45 years (54,58), severe (9,54,58) or long duration acute CHIKVD (9,60), a high viral load (>10⁹/mL) during the viremic phase (36), and an intense CHIKV immune response in the post-viremic phase (58).

**The Case for Viral Persistence.** For most viral arthritides, arthritic disease is associated with the presence or persistence of replicating virus and/or viral debris in joint tissues (64). A substantial body of evidence supports the view that the same underlying mechanism is responsible for alphaviral arthritides including CHIKV arthritis. Poo et al., reported detection of significant levels of CHIKV RNA in feet up to 100 days post-infection (65) in wild-type C57BL/6 mice, which is consistent with studies in non-human primate models (23,66), and to date, a single clinical observation (36). CHIKV proteins and continued virus-specific T cell responses were also detected throughout the chronic stage of disease in C57BL/6 mice (65) and in Rhesus macaques (66). Importantly, the latter model showed that the intensity of the T cell response was age-dependent, as evidenced by diminished T cell infiltrates and a more pronounced persistence of CHIKV in tissues of aged monkeys (66), in concordance with human pathology (54,58). Interestingly, the hierarchy of the T cell response in humans follows a shift from CD4 to CD8, which may be indicative of a role of ageing in CHIKV immunopathology (67). Although persistent CHIKV RNA has been detected in chronic disease (with dsRNA by itself known to be arthritogenic (68)), mouse models and human studies have been unable to isolate infectious virus from tissue or sera (36,65). Human studies have reported high levels of CHIKV-specific IgM up to 180 days in patients with no pre-existing musculoskeletal disease prior to infection (36,53), and even a case of severe destructive arthritis that exhibited CHIKV-specific IgM over 24 months (55). These findings suggest that the persistence of viral antigens could be a contributing factor for development of chronic CHIKV arthritis, but the underlying molecular
mechanisms are still unknown, and a statistical association between persistent CHIKV-specific IgM and chronic arthralgia has not yet been made (53). Immunohistochemical analysis revealed the presence of CHIKV antigen in muscle satellite cells and perivascular macrophages in muscle and synovial tissue in chronic CHIKVD patients (36,69). This suggests that infection of muscle satellite cells - myogenic precursor cells responsible for postnatal muscle growth and repair - could be an underlying cause of persistence of CHIKV in muscle tissues (69,70).

**Clinical management of CHIKV patients: lessons learnt from a decade of outbreaks**

**Current treatment of acute and post-acute CHIKVD patients (≤ 3 months post-onset of infection).**

The management of CHIKV acute patients involves both supportive care and physical measures, and the physician’s role is important. Given the potential for corticosteroids to exacerbate viral arthritides (71), non-steroid anti-inflammatory drugs (NSAID) remain a key disease management approach (72); however, so far there is hitherto no effective antiviral treatment licensed for clearing the virus. In the acute stage of CHIKVD (≤ 3rd week p.o.i.) the therapeutic aims are to first i) alleviate fever and pain and ii) prevent both short-term (i.e., organ dysfunctions of CHIKV or iatrogenic origin), post-acute and chronic complications. In the post-acute stage (≥ 4th week p.o.i.), the therapeutic strategy is based on a patient-centered approach, whose objective remains seemingly to relieve pain and inflammation and to prevent stiffness, loss of muscle tone and loss of physical fitness and autonomy, and to limit consequences of the inflammatory process (18). In this case, the treatment is primarily based on analgesics and NSAIDs, though it must be noted that NSAID treatment depends on the clinical presentation, since no NSAID class has demonstrated a clear benefit or effectiveness in post-CHIKV symptoms. NSAIDs are prescribed at full dose (within critical tolerance), and their effectiveness reassessed (dose, schedule) during the first week or within the first 10 days. If well tolerated, NSAID treatment can be extended up to several weeks before gradual weaning.
Fever and pain alleviation relies on acetaminophen (e.g., paracetamol) as a first-line medication (level 1). The risk of acute hepatitis is increased due to severe pain (that often requires supratherapeutic doses) or underlying comorbidities (alcohol and other liver diseases, malnutrition) or drug interactions (unwanted self-medications). When acetaminophen fails, second-line treatment consists of adding weak opioids (level 2), for instance, combined codeine with acetaminophen (adult regimen contraindicated during breastfeeding), adding codeine to acetaminophen (restricted for children > 12 years) or adding tramadol to acetaminophen (adult formulation or pediatric formulation for children > 3 years) (18). When second-line treatment fails, morphine (level 3) can be used on a case-by-case basis (extended or immediate action), usually in hospital, under strict monitoring and after assessment of risk-benefit ratio due to possible side-effects (respiratory, neurological, digestive and urinary) (18). Corticosteroids are generally strongly discouraged, given the lack of a clear benefit in the course of severe arthritis, as it has been suggested that they may cause a rebound of arthritis or tenosynovitis (18). Notwithstanding their inherent risk, the use of corticosteroids may, in our experience, be warranted when strictly limited to highly inflammatory polyarticular injury associated with tenosynovitis, severe synovitis, or when NSAID treatment has failed. Generally, a 10 mg/day course of prednisone for five days (with immediate de-escalation within 10 days) was found to be sufficient in moderate NSAID-refractory cases. As a generic rule, the total duration of corticosteroid treatment must be less than four weeks, and must be followed by a switch to an NSAID to avoid deleterious clinical rebound and/or drug induced dependence. Of note, other agents like chloroquine (CHQ) have been shown to be ineffective in relieving acute pain (73) and inconclusive in post-acute pain as a sub-optimal co-treatment with paracetamol (74). In the case of disease-modifying anti-rheumatic drugs (DMARDs, which are discussed further in this review), current consensus suggests no formal indication to initiate DMARD therapy before the last phase of the post-acute stage (8 weeks p.o.i.), even with a specific anti-rheumatic agent such as methotrexate (MTX). Other DMARDs, such as hydroxychloroquine (HCHQ) have not been shown to be effective in the treatment of post-acute CHIKVD (75).
Systemic, cardiac, hepatic, renal and metabolic comorbidities should be monitored closely, as some may be exacerbated by CHIKV or increase the severity and duration of CHIKV-specific arthralgia (18). Together with non-specific routine measures used in arthritis when pain persists beyond the 3rd week, therapeutic strategies often focus on pain management. This usually combines level 1 and level 2 analgesics with, in some cases, antineuropathic drugs (nefopam, antiepileptic or antidepressant pain-relievers) if necessary (DN4 score ≥ 4). As outlined above, topical corticosteroids or infiltration should only be given for local involvement including tenosynovitis, bursitis, capsulitis and tunnel syndrome, or synovitis escaping oral treatment.

**Gaps and perspectives in the treatment of acute and post-acute CHIKVD.** The intensity or duration of pain during the acute phase has been correlated with high viral loads, providing clues to hasten viral clearance. Ribavirin and IFN-α have shown synergistic antiviral activity against CHIKV *in vitro*; however, neither of these are recommended for daily use due to the risk of side effects (76). Favipiravir (T-705), a viral RNA polymerase inhibitor licensed in Japan for influenza virus infections, was also found to inhibit CHIKV replication *in vitro* and could become the focus of studies assessing its usefulness in treating CHIKVD (77). Suramin, a licensed drug for sleeping sickness, has also shown *in vitro* antiviral activity through multiple mechanisms and was effective against mutant CHIKV strains resistant to ribavirin or favipiravir (78). Curcumin, a turmeric-derived compound used as a food additive and in herbal medicine, and its derivatives, exhibit similar antiviral properties through inhibition of virus cell binding in *in vitro* studies (79). Lastly, short peptides pimozides and 5-tetradecyloxy-2-furoic acid (TOFA) were found to exhibit high synergistic antiviral activity, using a genome-wide loss-of-function screen, which further reinforces the usefulness of such a broad genome-wide scale approach in CHIKVD studies (80). Despite significant advances in the development of CHIKV antivirals and substantial mechanistic evidence of *in vitro* efficacy, neither of these compounds have yet been evaluated in humans infected with CHIKV.
Current treatment of chronic CHIKV patients (>3 months post-onset of infection).

The therapeutic approach to chronic CHIKV (CIR) requires a rheumatologist and a pain specialist, as for chronic inflammatory rheumatisms (CIR)(18). It focuses on the characterization of the nosology of each patient according to the presence or absence of inflammatory symptoms (i.e., at least one joint with chronic arthritis), the number of joints involved and the level of clinical inflammatory activity (e.g. joint destruction, extra-articular involvement). Practically, two distinct evolving patterns of persistent CHIKV rheumatic disorders are to be deciphered: the large majority of patients who still have pain beyond three months following acute infection present with varied musculoskeletal injury, which is primarily supported by the development of tendinopathies. Herein, patients should experience substantial improvements with prolonged administration of NSAIDs (strictly limiting the use of corticosteroids) in combination with local complementary therapeutics. In contrast, approximately 5% of patients will experience conditions that fulfil the criteria for CIR, carried by potentially destructive arthritis and synovitis. Little is known about the immune (or viral) mechanisms underlying the progressive form of the disease, arthritis development as well as predictors for this outcome.

Morphological assessment, designed to fulfill current criteria for RA and spondylarthritis (SpA), and biological tests are generally performed to confirm the diagnosis and screen for evidence of inflammatory or destructive mechanisms. Incidentally, RA is the most common post-CHIKV CIR before peripheral SpA – alternatively, non destructive arthritis which does not meet the criteria for RA or SpA is referred to as undifferentiated polyarthritis (UP), and may be indicated after alternative causes (connectivitis, lupus-like syndrome) have been ruled out. Once the type of persistent chronic CHIKV condition has been identified, individualized treatment is proposed based on the diagnosis, functional prognosis and the patient’s condition. Therapeutics aim to preserve the functional outcome in order to reduce the psychosocial impact and improve the health-related quality of life. Ideally, it should begin within the first month of the chronic stage (> 8 weeks of post-acute stage).
Disease-Modifying Antirheumatic Drugs (DMARDs). Chronic CHIKV arthritis shares several characteristics with RA, such as the persistent debilitating arthralgia and exacerbated inflammatory response (31,36,43,45,72). Because of these parallels, CHQ, HCHQ and DMARDs such as MTX and sulfasalazine have been the subject of evaluation in some peri-epidemic clinical trials to treat patients with chronic CHIKV arthritis (74,81-84). When compared with HCHQ alone, cohort studies of patients suffering chronic CHIKV arthritic disease have found reductions in joint pain, disability and impaired activity after treatment with DMARDs, either alone or in combination with HCHQ and corticosteroids (83,84). In contrast, a patient cohort study reported that almost all patients showed progression of disease-induced bone erosion and joint space narrowing upon follow-up despite treatment with HCHQ or DMARDs, and a two-year follow up study involving 625 patients found that treatment of chronic CHIKV arthritis with sulfasalazine and MTX was effective (81,85). Given the conflicting outcomes of some cohort studies, it remains unclear whether specific DMARDs are effective in chronic CHIKV arthritis (84).

However, experts have reached a consensus to propose MTX as first-line treatment (18). Thus, though the efficacy of HCHQ and other DMARDs has not been established per se using RCTs, these should be discussed on a case-by-case basis, either as a complement or as an alternative to MTX (18,63,83). DMARDs must be monitored for effectiveness (DAS 28) and tolerance and should be stopped after a durable remission of several months. The specialist may then refer to national guidelines to choose the second-line drug (combination or replacement, biotherapy). However, in most cases, DMARD treatment may be assessed and stopped for patients in remission after a complete response sustained for several months (up to 2 years). Physical therapy includes rest limitation, maintenance of articular amplitude and muscular tone, lymphatic drainage (compression stockings, manual, or using pressotherapy) (18). Its benefit depends on the extent and the intensity of joint affection and the disease impact in terms of autonomy and quality of life. Taken together, the above-mentioned approaches highly benefit from specialized, multidisciplinary case-by-case advice, including participation in clinical trials that follow national/international guidelines when considering second-line treatment for patients with CIR that fail to respond to first-line treatment. In this context, one
option would involve a regimen switch, a combination with alternative DMARDs, or a biotherapy, either alone or in combination with other DMARDs.

**Gaps and perspectives in the treatment of chronic CHIKVD.** Chronic pain has been linked to CHIKV persistence in synovial macrophages as well as to IL-6 and IL-12 secretion. To the best of our knowledge, neither anti-IL-6 (tocilizumab), anti-IL-12/IL-23p40 (ustekinumab), or anti-CD20 (rituximab) monoclonal antibodies have been tested empirically in chronic CHIKVD, whereas anti-TNF-α (etanercept) therapy has been shown to exacerbate tissue damage in a mouse model of alphaviral arthritis (86). While several approaches (biologic, prophylactic, palliative) are being explored and characterized to manage acute and chronic CHIKVD, the development and testing of such therapeutic solutions should be done in close consultation with rheumatologists.

Therapeutic strategies that target MCPs or CCR2, such as Bindarit®, which was shown to ameliorate CHIKV-induced arthritis and bone loss in a mouse model (87,88), might be considered as one of the promising candidates for further evaluation in the treatment of alphaviral arthritides. Of note, treatment of rheumatoid arthritis patients with CCR2 blockers was also unsuccessful (89). Interestingly, Pentosan polysulfate (PPS; Elmiron®), a glycosaminoglycan currently licensed in the United States for the treatment of interstitial cystitis, has undergone promising clinical trials for the treatment of noninfectious arthritis and has been shown to maintain levels of cartilage proteoglycans in experimental animal models of arthritis (90,91). In CHIKV-infected mice, PPS treatment decreased the level of joint swelling and reduced levels of soluble factors CCL2, IL-6, IL-9, and G-CSF during acute inflammation (92). Although the exact mechanism of action of PPS is unclear, treatment was associated with an early increase in anti-inflammatory IL-10, suggesting an indirect mechanism by which inflammation is dampened (92). As part of the Australian Government Department of Health Therapeutic Goods Administration Special Access Scheme, PPS – currently in Phase II clinical trials - has now been successfully used to treat five patients suffering from Ross River virus (RRV)-induced arthralgia who had failed current standard treatment. RRV is an Australian arthritogenic alphavirus,
and the main cause of viral polyarthritis with clinical manifestations (severe, incapacitating joint pain) similar to those seen in CHIKV arthritis. More recently, targeting CD4+ T cells with clinically approved T cell-suppressive drugs, including sphingosine 1-phosphate receptor agonist fingolimod (also known as FTY720) successfully reduced CHIKV-induced disease in mice by blocking lymphoid egress and subsequent migration of CD4+ T cells into the joints (93). Although the US Food and Drug Administration (FDA) has approved fingolimod as a treatment option for multiple sclerosis in 2010, there are currently no clinical trials to assess efficacy in CHIKV arthritis.

**Neutralizing Antibodies and Prophylactic Treatment.** A number of studies have sought to characterize the neutralizing capacity of CHIKV-specific antibodies, the epitopes responsible for neutralization, and the mechanism of viral inhibition (94-101). Robustly neutralizing monoclonal antibodies (mAbs) to CHIKV bind the E2 envelope glycoprotein with conserved epitopes identified on the A and B domain. Antibodies block multiple steps in the viral lifecycle, including entry and egress, by cross-linking adjacent E2 spikes of the E2 glycoprotein or impeding fusion. Importantly, these broadly neutralizing ultrapotent monoclonal antibodies protect mice against infection by multiple alphaviruses including CHIKV (94-96). Neutralizing antibodies have been tested in mouse models as treatment for persistent CHIKV infection. The effect of a neutralizing anti-CHIKV monoclonal antibody on RAG1−/− mice (devoid of B and T cells) with persistent CHIKV infection was to enable effective clearance of infectious virus in quadriceps muscle tissue and sera, although without reducing CHIKV RNA load in joint tissue (102). A separate study by Poo et al., using polyclonal anti-CHIKV anti-serum to treat persistently-infected RAG1−/− and B cell-deficient μMT mice showed that CHIKV viremia became undetectable from 10 days and 30 days post-administration in RAG1−/− and μMT mice, respectively (65). However, viremia recovered to levels similar to those observed prior to administration of the antibody, indicating that effective clearance of CHIKV may require robust and long-lasting B and T cell responses (65). Further, another study showed that a highly conserved amino acid on the E2 glycoprotein promoted CHIKV persistence in joints and impaired neutralization by antibodies targeting the E2 domain. Mutation of this conserved region
allowed viral clearance and enhanced neutralization, providing the structural basis for the mechanism by which CHIKV evades B cell-mediated clearance in chronic joint infection (97). Finally, Rhesus macaques treated after infection with SVIR001, a recombinant human IgG1 monoclonal antibody (mAb) that recognizes the E2 glycoprotein of CHIKV, showed more robust viral clearance and less severe joint inflammation compared to isotype-treated controls (98). In addition, SVIR001 reduced viral burden at the site of infection – as well as at distant sites – but also diminished the number of activated innate immune cells and levels of pro-inflammatory cytokines and chemokines (98). Therefore, more studies are required on the utility of neutralizing antibodies as a therapeutic approach to persistent CHIKV infection, and to confirm the efficacy of these approaches in humans.

In contrast, studies have shown treating mice prophylactically with CHIKV-specific monoclonal antibodies prevents joint inflammation in CHIKV-infected WT mice and can protect against lethal CHIKV infection in AG129, RAG2 and IFNAR1+/− mice (97-102). CHIKV-specific mAbs administered to RAG1−/− mice prior to infection resulted in drastically reduced viremia and CHIKV RNA levels in ankle joints in mice with persistent CHIKV infection (102). Passive immunization with CHIKV IgG to IFNα/βR−/− mice was also able to prevent mortality from a lethal CHIKV infection (101). However, these studies only examined the effect of using mAbs as prophylaxis for acute CHIKV infection. Thus, our understanding of the prophylactic use of mAbs in chronic CHIKV arthritis is still incomplete and more studies are required. Nevertheless, prophylactic treatment could be effective for individuals at increased risk of CHIKV infection in critical settings, such as mother-to-child transmission (103) or hospitalized individuals with life-threatening acute disease (104).

**Combined biotherapies:** A recent study by Miner et al., sought to target both humoral and adaptive arms of the immune response by using abatacept - a drug approved in 2005 by the FDA for treatment of RA - in combination with an anti-CHIKV neutralizing antibody and assessed their ability to decrease acute joint swelling in CHIKV-infected mice (105). Abatacept, a human IgG fusion protein paired with a CTLA-4 extracellular domain motif, prevents antigen presenting cells from delivering
co-stimulatory signals to T cells. In this study, abatacept reduced T cell accumulation in the joints of infected mice and, in combination therapy with an anti-CHIKV neutralizing antibody, abolished signs of inflammatory disease and markedly reduced levels of chemokines, pro-inflammatory cytokines and infiltrating leukocytes (105). Notwithstanding the promising pre-clinical outcomes and innovative approaches, these candidate therapies - along with the other examples cited earlier in this review, summarized in Figure 1 - warrant further evaluation in the treatment of CHIKV-induced joint pathologies in a clinical setting, keeping in mind the potential risk of immunosuppression when targeting host response mechanisms.

**Conclusion**

Chikungunya virus disease, a previously neglected tropical arthritogenic alphaviral infection, has gained global significance in the last decade after a series of devastating outbreaks, exposing the severe public health and economic burdens as well as the significant risk of acquired disabilities incurred by international travelers. The clinical and immunopathological phenotype of this chronic inflammatory rheumatic disease is reminiscent of rheumatoid arthritis, and studies in animal models of alphaviral infections have unraveled previously unknown mechanisms of disease and brought to light novel therapeutic approaches. Based on its newly recognized public health importance and its strong potential for re-emergence, Chikungunya virus arthritis should be the focus of further experimental forays to develop novel therapeutic approaches but should also gain further attention from rheumatologists worldwide.

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References


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Figure 1: Schematic overview of mechanisms of tissue inflammation leading to alphaviral arthritis. Based on experimental models of alphavirus infection, key cytokine-dependent inflammatory pathways have been identified and targeted to explore therapeutic avenues. Principally, monocyte chemotactic proteins (MCP) such as CCL2 and CCL3, which drive monocytic infiltration into joint and muscle tissue in alphaviral inflammation (50,89) (e.g., CHIKV infection) can be targeted using MCP inhibitors such as Bindarit®, while potent pro-inflammatory cytokines and chemokines such as IL-1, CXCL10 and IL-8 are also the focus of current studies. Scavenging soluble MCPs such as CCL2 restricts CCR2-dependent monocyte/macrophage infiltration, which is required for CHIKV arthritis (87,88). Pentosan polysulfate, or Elmiron® has been shown to limit CHIKV-induced arthritis and cartilage damage by significantly reducing tissue expression of matrix metalloproteinases (MMPs) which drive cartilage damage in alphaviral infection (92) and in experimental models of cartilage damage (91). RNA-Seq analysis of CHIKV-infected tissue in mice has shown a prominent role for Granzyme A (GzmA) in driving arthritic inflammation, and the use of mouse GzmA inhibitor Serpinb6b was found to dampen tissue inflammation and swelling in CHIKV-infected mice (21). In addition to targeting innate inflammatory mechanisms, modulating adaptive immunity has also been shown to reduce tissue infiltration, either by inhibiting priming of CD4+ T cells using Abatacept (105) or by blocking egress of CD4+ T cells from the lymph nodes to the joints (93) using FTY720 (Fingolimod). The role of TNF-in alphavirus inflammation, despite its prominent expression in the tissues of infected mice and humans, appears to rely on a more complex mechanism: inhibition of TNF- using Enbrel® in a mouse model of alphaviral arthritis was shown to significantly exacerbate inflammatory disease and cellular infiltration (86), thereby reinforcing the need for a cautious approach when considering biotherapeutic approaches in the treatment of alphaviral disease such as CHIKVD.