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MINIREVIEW

Beyond Fever and Pain: Diagnostic Methods for Chikungunya Virus

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ABSTRACT Chikungunya virus (CHIKV) is an alphavirus that is primarily transmitted by *Aedes* species mosquitoes. Though reports of an illness consistent with chikungunya date back over 200 years, CHIKV only gained worldwide attention during a massive pandemic that began in East Africa in 2004. Chikungunya, the clinical illness caused by CHIKV, is characterized by a rapid onset of high fever and debilitating joint pain, though in practice, etiologic confirmation of CHIKV requires the availability and use of specific laboratory diagnostics. Similar to infections caused by other arboviruses, CHIKV infections are most commonly detected with a combination of molecular and serological methods, though cell culture and antigen detection are reported. This review provides an overview of available CHIKV diagnostics and highlights aspects of basic virology and epidemiology that pertain to viral detection. Although the number of chikungunya cases has decreased since 2014, CHIKV has become endemic in countries across the tropics and will continue to cause sporadic outbreaks in naive individuals. Consistent access to accurate diagnostics is needed to detect individual cases and initiate timely responses to new outbreaks.

KEYWORDS Chikungunya virus, alphavirus, molecular diagnostics, serology, viral culture

Chikungunya virus (CHIKV) is one of over 30 known viral species in the genus *Alphavirus* (family *Togaviridae*). The alphaviruses are enveloped, single-stranded, positive-sense RNA viruses with a genome of approximately 11.8 kb that encodes five structural proteins (capsid, E3, E2, 6K, and E1) and four nonstructural proteins (nsP1, nsP2, nsP3, and nsP4) (Fig. 1) (1, 2). CHIKV is primarily transmitted to humans by the *Aedes* species mosquitoes *Aedes aegypti* and *Aedes albopictus*, though rare reports of blood-borne transmission have been documented (3). Outbreaks had occurred in several countries throughout Africa, Southeast Asia, and Polynesia, but the CHIKV pandemic that began in East Africa in 2004 brought this previously obscure arbovirus to prominence and is unrivaled among pandemics caused by the alphaviruses in terms of size and geographic distribution (Fig. 2) (1, 2).

CHIKV causes a higher rate of symptomatic infection than other widespread arboviruses, such as dengue virus (DENV) and Zika virus (ZIKV), though there is evidence that the incidence of symptoms is lineage dependent (4, 5). The reported incubation period for CHIKV ranges from 1 to 12 days, and symptomatic infection typically presents as fever and arthralgia, though less common presentations have been described (1, 2, 6). The diagnosis of a CHIKV infection cannot be confirmed based solely on clinical findings (7, 8). Similar to other arboviruses, confirmation is achieved through the use of molecular and/or serological methods, though CHIKV can be isolated in culture, and antigen-based detection has been reported. This review provides an overview of the available CHIKV diagnostics and highlights aspects of basic virology and epidemiology

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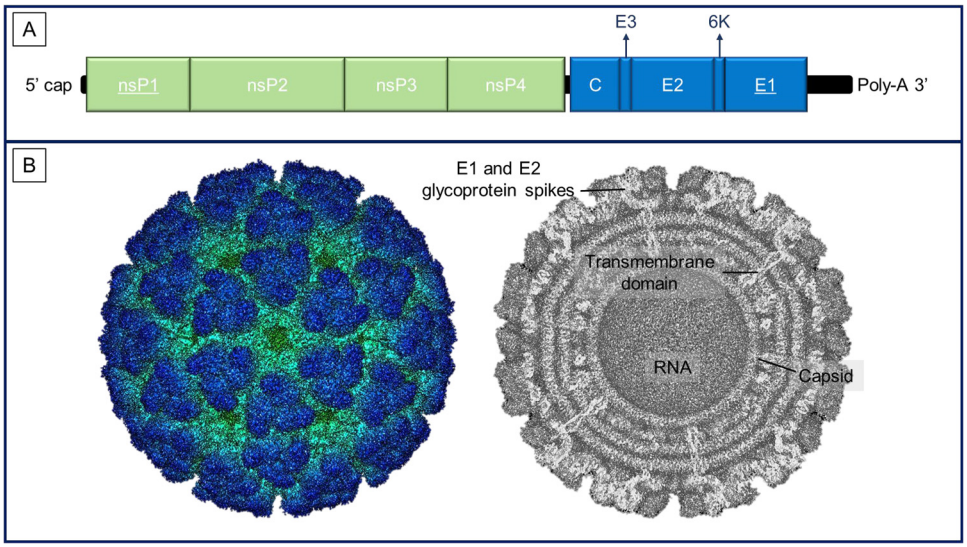

FIG 1 (A) Diagram of the CHIKV genome indicating the relative length of the genes encoding nonstructural (green) and structural (blue) proteins. CHIKV molecular diagnostics have predominantly targeted the nsP1 and E1 genes (underlined), accounting for 10 and 14 of the 32 assays referenced in this review, respectively. (B) The structure of the CHIKV virion determined by electron microscopy is shown, highlighting the E1/E2 glycoprotein spikes on the virion surface, transmembrane domains, and the viral capsid (republished from PDBj.org under the Creative Commons Attribution 4.0 International license [190, 191]).

that pertain to viral detection. As a point of clarification, throughout this review the term chikungunya is used to refer to the clinical illness and CHIKV is used to refer to the virus.

HISTORY AND TRANSMISSION

Descriptions of an illness compatible with chikungunya date back to 1779, including potential outbreaks in the Western Hemisphere (9–11), but CHIKV was not identified

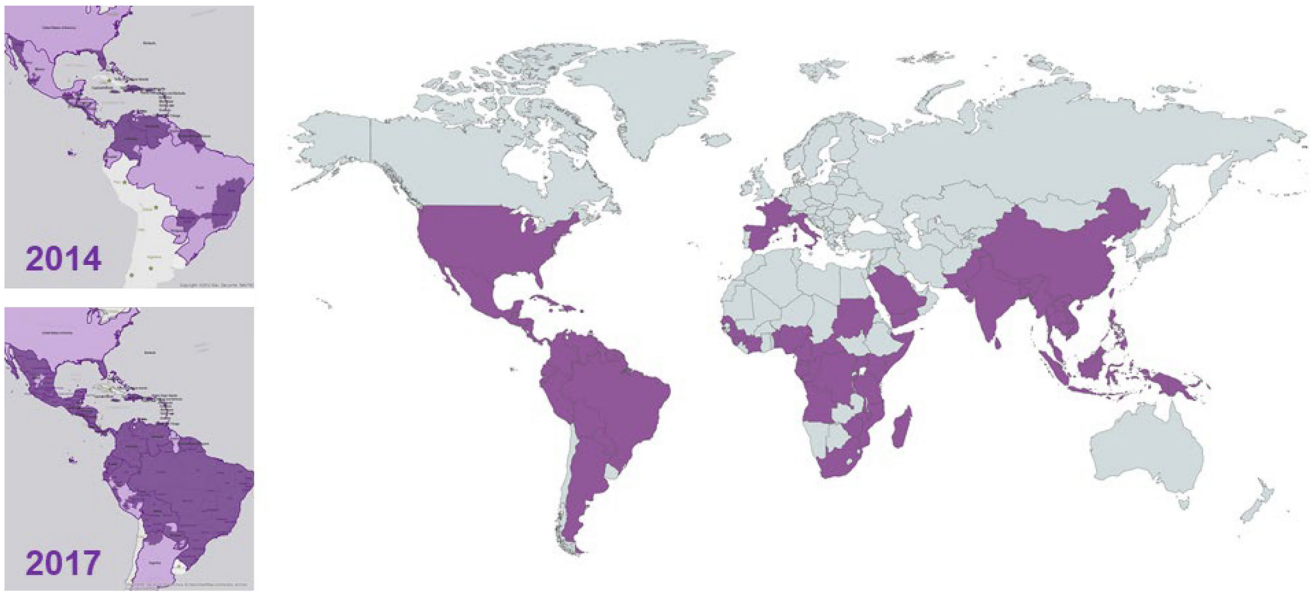

FIG 2 Countries with autochthonous cases of CHIKV (reported through 16 May 2018, dark purple). Inset maps display the geographical spread of CHIKV in the Americas between 2014 and 2017, though overall case numbers decreased ~6-fold during this time period. Regions in dark purple reported autochthonous CHIKV transmission at any time through the year shown. Light purple highlights countries with any CHIKV transmission. Countries in gray had no autochthonous cases; asterisks represent imported cases. Maps were modified from those available at CDC.gov and PAHO.org. Notably, the categorization of Cuba differs between these sources, as autochthonous cases have not been reported to PAHO.

until the 1950s in what is now southern Tanzania (1, 6, 12). Occasional outbreaks also occurred in Asia from the late 1950s through the latter 20th century (1, 12–14). Phylogenetic studies initially identified three CHIKV lineages: the West African, East/Central/South African (ECSA), and Asian lineages(15, 16). However, in 2004, the Indian Ocean lineage (IOL) emerged from an ECSA strain and caused a large outbreak that began in coastal Kenya and spread to the Comoros, La Réunion, and islands of the Indian Ocean (Fig. 2) (12, 13). This new lineage spread across Asia and the South Pacific, with smaller outbreaks occurring in Western Europe following introductions by returned travelers (12, 17–19). In late 2013, the first cases of chikungunya were identified in St. Martin, and CHIKV quickly spread throughout the Caribbean and the Americas (20). This outbreak was almost exclusively caused by Asian lineage strains, with a small number of ECSA lineage infections being reported in Brazil (12, 14, 21). Although the number of cases declined markedly from 2014 to 2017, CHIKV transmission continues in the Americas (PAHO.org), and outbreaks occur in Asia and Africa, with autochthonous cases being documented in Italy as recently as 2017 (22).

During an outbreak, CHIKV is maintained in a human-mosquito-human cycle of transmission by *A. aegypti* and *A. albopictus* mosquitoes, in a pattern similar to that for DENV (1, 23). Attack rates have been relatively low in temperate regions (24), but rates as high as 50 to 75% have been reported during outbreaks in the tropics (25–27). Such high attack rates help explain the occurrence of human coinfections with CHIKV, DENV, and/or ZIKV, which have been reported from the Americas and Asia (7, 28–33), and individuals with these coinfections may present with more severe manifestations (7, 30). A meta-analysis of available publications on CHIKV coinfections revealed that CHIKV-DENV coinfections were most common (44/109 studies), but CHIKV-malaria coinfections were reported in several studies (5/109) (28, 29, 34). During interepidemic periods, CHIKV may be maintained by other mechanisms, leading to persistence in a region and the potential for sporadic outbreaks (35–38).

CHIKV transmission has occurred in regions of endemicity for related alphaviruses, such as O'nyong-nyong virus (ONNV), Mayaro virus (MAYV), Venezuelan equine encephalitis virus (VEEV), and Ross River virus (RRV). Cross protection between alphaviruses has been documented in animal models (39, 40) and humans following natural infection (41) or sequential vaccination (42). In addition to cross protection, there are limited data on the potential for antibody-dependent enhancement (ADE) of alphavirus infections, with *in vitro* studies of RRV (43, 44) and Sindbis virus (45) suggesting ADE of alphaviruses at low antibody titers. In mice immunized against CHIKV, low IgG titers enhanced CHIKV infection (46, 47), but vaccine-elicited antibodies after RRV vaccination did not enhance CHIKV or RRV infection even at low titers (48). ADE from prior alphavirus exposures has not been documented in human infection (44), and the clinical significance of these experimental findings remain unclear.

ACUTE CLINICAL PRESENTATION AND DIFFERENTIAL DIAGNOSIS

Although it has been reported that a high proportion of CHIKV infections are symptomatic (80 to 97%) (1, 27, 49–51), the ratio of symptomatic to inapparent CHIKV infections (S/I ratio) has varied markedly in the literature (5, 27, 52–56). Factors associated with the S/I ratio include the presence of preexisting neutralizing antibodies to CHIKV (52, 55) and patient age (52, 54, 56, 57). Notably, both negative (54, 57) and positive (52) associations between age and symptomatic infection have been reported. Finally, the S/I ratio appears to be lineage dependent. In a recent reevaluation of the literature, it was found that ~50% of infections with Asian lineage CHIKV strains versus ~80% of infections with ECSA lineage strains (predominantly IOL strains) result in symptomatic infections (5). Insufficient data are currently available to estimate the S/I ratio for infections with CHIKV strains of the West African lineage.

The classic triad of symptomatic chikungunya includes an abrupt-onset febrile illness, severe and often debilitating arthralgias, and a rash (Fig. 3) (1, 2, 6). Clinical disease evolves rapidly, and symptomatic patients may present within 2 to 3 days of symptom onset (1, 25, 58–61). Fever can be marked (39 to 40°C) and occurs in the

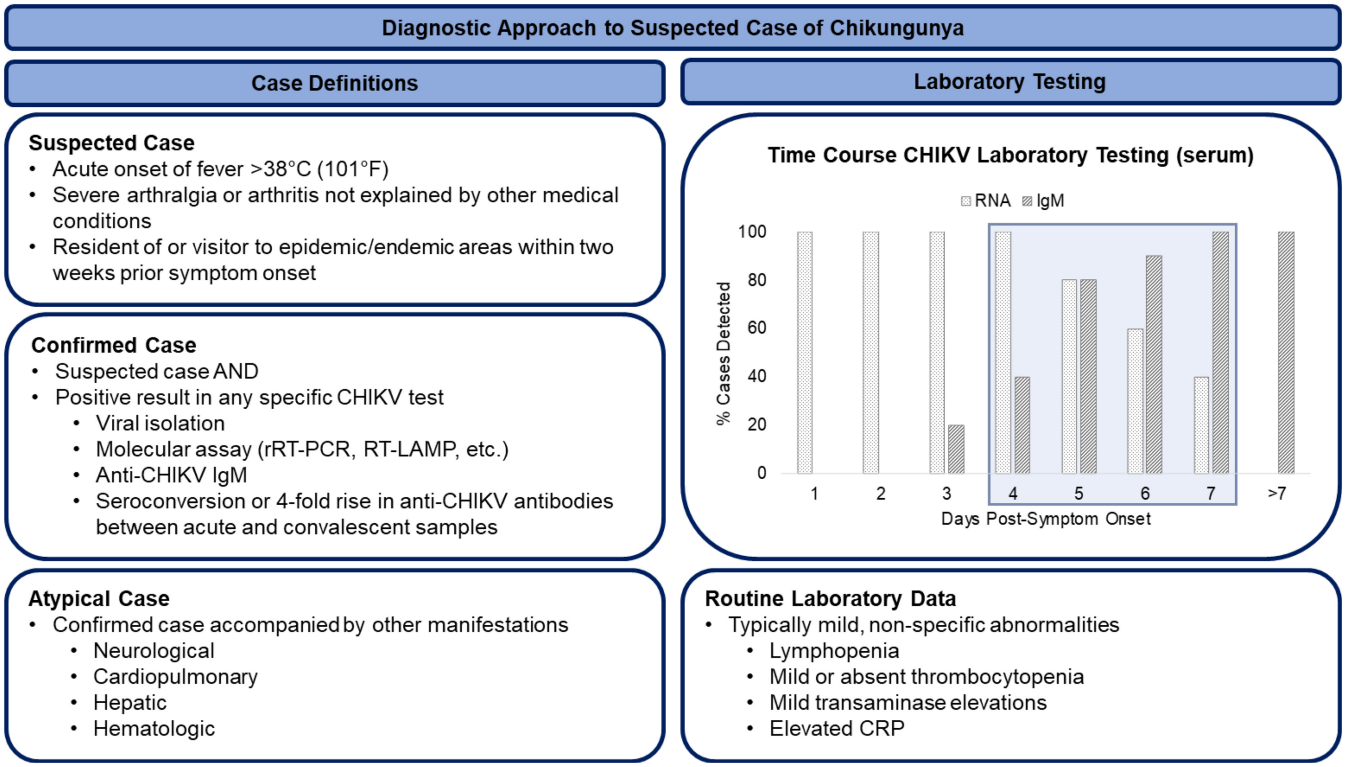

FIG 3 Case definitions and diagnostic approach to suspected chikungunya cases. The proposed time course for CHIKV diagnosis using serum was derived from published reports (103–106). The sensitivity of RNA detection in serum declines between days 4 and 7 as anti-CHIKV IgM becomes detectable. Anti-CHIKV IgG may become detectable at a similar time point (105). RT-LAMP, reverse transcription–loop-mediated isothermal amplification; CRP, C-reactive protein.

majority of cases (80 to 90%), including atypical cases (62). Arthralgia develops around the time of fever onset and is reported at presentation in 85 to 90% of cases (8, 25, 57–60, 63, 64). Joint involvement is bilateral, though not always symmetric, and most commonly involves the knees, ankles, and joints of the upper extremities (metacarpophalangeal, interphalangeal, and metatarsal joints, elbows, and shoulders) (25, 58, 65, 66). Arthritis with joint swelling and tenderness occurs in ~30% of cases (25, 65, 67). A skin rash develops in 40 to 60% of cases (1, 2, 25, 64, 65, 68, 69), though this may not be present at the initial visit (65, 70). Rashes are most often maculopapular and may be pruritic, though bullous and purpuric lesions have been reported (8, 25, 62, 69). Headache and gastrointestinal complaints, such as anorexia, nausea, vomiting, and abdominal pain, are also commonly reported in chikungunya cases (25, 57, 65, 66, 69). Hemorrhagic manifestations, however, are rare (~5% of cases) (25, 59, 67, 71).

Host factors also affect the clinical presentation of CHIKV infections. It has been observed that young children present with arthralgia less frequently than older children and adults (52, 54, 57, 72). Patients older than 65 years of age may present a more complicated clinical picture during acute chikungunya, with more frequent manifestations other than arthralgia, including high rates of neurological complaints (73). Limited data are available from immunocompromised hosts, but chikungunya manifestations and severity do not appear to differ among solid organ transplant recipients (74). Atypical chikungunya cases, defined as cases with predominant manifestations other than fever and arthralgia, have been reported (Fig. 3) and are estimated to occur in ~1% of symptomatic cases (62, 75). The incidence of severe and atypical disease has a bimodal distribution, with peaks occurring among neonates and older adults, who often have comorbid illnesses (57, 62, 64, 72, 75). In a large series of such cases from La Réunion, patients with cardiovascular disease (heart failure, arrythmias, and pericarditis) and neurological disease accounted for >50% of individuals with atypical cases (62). Neurological manifestations have included meningoencephalitis, Guillain-Barré syndrome, optic neuropathy, and retinitis, among others (33, 62, 75).

Clinical diagnosis does not reliably differentiate CHIKV infections from other etiologies on the differential, such as other alphaviruses (MAYV, ONNV), flaviviruses (most commonly DENV and ZIKV), and nonviral pathogens, which include, but are not limited to, *Plasmodium* species, *Leptospira*, rickettsia, and *Salmonella*. In settings of CHIKV cotransmission with DENV and ZIKV, a presumptive diagnosis of chikungunya was correct in only 10 to 40% of cases (7, 8, 31). Clinical prediction algorithms have been evaluated to differentiate CHIKV infections from infections with other etiologies, but these have not been independently validated in separate cohorts (59, 63, 66, 72, 76). Results from routine laboratory tests are generally not specific for chikungunya. Abnormal results that have been observed in cases include lymphopenia ($<$500 to $<$1,000 $\times$ 10^6 lymphocytes/liter) without leukopenia, mild thrombocytopenia ($>$100,000 $\times$ 10^6 platelets/liter), mild transaminase elevations (transaminase levels 2 to 3 times the upper limit of normal), and an elevated C-reactive protein level (Fig. 3) (25, 59, 60, 66, 71). Moderate to severe thrombocytopenia favors the diagnosis of dengue rather than chikungunya, particularly if hemorrhagic signs are present (59, 60, 66, 76). However, these laboratory findings occur in other diseases on the differential and do not provide accurate diagnostic information.

CHIKV DIAGNOSTICS

Biosafety. CHIKV is a risk group 3 pathogen and requires biosafety level 3 (BSL-3) precautions, based on a number of cases associated with laboratory exposures (77, 78), and it is a category C priority pathogen according to the National Institute of Allergy and Infectious Diseases (NIAID) (79). CHIKV infections among laboratory personnel from fieldwork, work with infected mosquitoes, and isolation of live virus from field materials have been reported (80, 81). There have been no case reports of laboratory transmission from blood products; however, a nurse reported acquiring CHIKV by drawing the blood of an infected patient (80). As such, continued caution is recommended when handling infected blood products or live virus in the laboratory due to the potential for transmission. These factors have limited the number of facilities that can safely work with live virus and impacted the testing that is currently available.

Because of the geographic spread of CHIKV, diagnostic approaches that can be practiced in available, low-biosafety-level facilities are needed. As whole-virus preparations are needed for many applications, such as the plaque reduction neutralization test (PRNT), varied strategies of viral inactivation have been studied. It has been reported that standard heat inactivation protocols with a 30-min incubation at 56°C are insufficient for CHIKV, which may require incubations over 2 h to be fully inactivated (82). Complete inactivation of CHIKV was achieved by 1,5-iodonaphthyl azide (INA) treatment. However, a reduction in the capacity to bind to anti-CHIKV antibodies was observed (83). Recent work showed that treatment with UV at 0.09 J/cm^2 was sufficient to inactivate an Asian strain of CHIKV. The resulting inactivated virus was replication deficient, but the procedure did not affect the integrity of the virus, and structural epitopes were unaltered (84).

Viral culture. Despite their use for decades as the "gold standard" for viral detection in clinical virology, cell culture and viral isolation are no longer commonly used for routine diagnostic purposes (85). The procedure requires time for viral growth and subsequent identification as well as specialized equipment and skilled laboratory staff for performance (86). However, the benefit of cell culture is that it allows for biological amplification of the virus and the isolation of strains from human infections. These isolates permit further characterization of viral species and provide invaluable resources to clarify immune responses and perform fundamental basic virology research with contemporaneous strains (86, 87).

Although molecular methods, detailed below, provide a highly sensitive means of detection, false negatives may occur in specimens with low levels of virus, reverse transcription-PCR (RT-PCR) inhibitors, or strains that harbor mutations in target regions. Inoculation of suspected arbovirus-containing human samples onto cell cultures may allow for replication of the virus to high titers, followed by confirmation using immu-

nofluorescence or RT-PCR (6, 88, 89). Furthermore, the semiunbiased nature of viral culture has allowed detection of coinfections with unexpected viral pathogens while simultaneously documenting that these are replicating viruses in the human host (90, 91). The isolation of CHIKV can be performed from serum collected up to 7 days after illness onset, and the virus has also been isolated from human cerebrospinal fluid (CSF) (92) and pools of adult female *A. aegypti* mosquitoes (93).

Both mosquito and mammalian cell culture systems have been used to isolate CHIKV and study viral replication and pathogenesis (79, 94). It has been shown that CHIKV establishes a persistent noncytopathic infection in *A. albopictus* C6/36 mosquito cells, while it causes strong cytopathic effects (CPE) and apoptosis in mammalian cells (94–96). CHIKV culture in C6/36 cells permits viral titers that are 100-fold higher than those in Vero cells, though C6/36 cells may not be readily available in clinical diagnostic laboratories (95). In addition to Vero cells (93, 97), CHIKV has been isolated on other mammalian cell lines, including LLC-MK2 (90), BHK-21 (98, 99), and 293T (100) cells. Also, a combination of successive passages in different cell types has been described (101). Notably, C6/36 cell lines are significantly more permissive for more recent CHIKV isolates, suggesting that the increased infectivity and the recent epidemic may derive from evolution of the CHIKV genome beyond simply the E1-A226V substitution (101).

Molecular testing. Molecular testing remains central to the confirmation of chikungunya (Fig. 3). The viral load in acute CHIKV infections can exceed 11.0 $\log_{10}$ copies/ml of serum, particularly in neonates (7, 70, 102), and the sensitivity of RNA detection remains high through the first 4 to 5 days of illness in most series (103–107). A variety of molecular assays for CHIKV have been published or are commercially available (see Table S1 in the supplemental material). Reported assays include conventional RT-PCR (69, 108–112), real-time RT-PCR (rRT-PCR) (102, 105, 113–120), isothermal methods (110, 121–124), and multiplex assays (125–138). No molecular gold standard by which to evaluate reported assays in practice exists, and the decision to implement a particular test depends on the relative advantages and disadvantages of the method along with the capabilities in a given laboratory.

Consistent with molecular diagnostics in general, real-time methods for CHIKV have proven more sensitive than conventional RT-PCR (112, 115, 116, 121), though this has not been shown in all studies (111). Comparisons between real-time methods have not demonstrated clear differences in assay performance (105, 118, 124, 132, 136), and CHIKV detection in published multiplex assays does not appear to be decreased relative to that in monoplex tests (125, 126, 133, 135, 137, 138). Multiplex assays facilitate testing for a set of pathogens in all patients, and the utility of this approach has been demonstrated in regions with transmission of multiple arboviruses and/or malaria (7, 34). Evaluations of assay exclusivity have been variable. At a minimum, CHIKV molecular tests should be evaluated against DENV, ZIKV, and a panel of alphaviruses that includes MAYV and ONNV, which is the alphavirus most closely related to CHIKV and which may cross-react in molecular assays (124, 136).

Few independent comparisons of available molecular assays have been reported (106), and in a large external quality assessment (EQA) of molecular testing for CHIKV, laboratory performance was not associated with the use of a particular assay or commercial reagents (139). Notably, in this assessment, 50% of laboratories (30/60) failed to meet the acceptable performance standard ($\leq$1 false-negative result and no false-positive results in a set of 12 samples) (139). These findings highlight the complexity of molecular testing in general and the ongoing need for assay harmonization across laboratories. An earlier study demonstrated that the distribution of a preformulated CHIKV rRT-PCR significantly improved sensitivity at participating sites (140). A molecular reference reagent has also been developed for use in the FDA approval process (141, 142), though no molecular test has received FDA approval for CHIKV detection, and use of this reagent outside of the development process has not been reported.

Serum and plasma are the most common specimen types used for CHIKV RNA detection, and all reported methods require nucleic acid extraction for optimal performance. One rRT-PCR using cell culture supernatants without extraction was evaluated, but it remains unclear if this protocol can be applied to clinical samples (114). Given the high viral loads observed in acute CHIKV infections, there has not been the impetus to evaluate alternative specimen types as has been done for ZIKV infections, where average viral loads are significantly lower (7). CHIKV RNA has been amplified from a large number of specimen types other than serum/plasma (143, 144), though most published data describe the testing of CSF, in which both CHIKV RNA and antigen have been detected (33, 143, 145, 146). The viral load in CSF appears to be lower than that in serum, but the duration of CHIKV RNA detection in CSF may be prolonged (143, 145). Finally, both dried blood spots and dried serum spots have been evaluated as specimen types for CHIKV RNA detection by rRT-PCR (147, 148). Both specimens demonstrated ~93% sensitivity compared to serum, indicating that these represent a reasonable specimen for use in resource-limited areas or to improve patient acceptance of sample collection, such as with small children.

Serological testing. Serological testing provides diagnostic information and valuable insight into the immune responses to CHIKV infection. CHIKV-specific IgM and IgG antibodies develop in response to infection, and several methods for the detection of these isotypes during the acute and convalescent phases of infection have been reported. Commercial and in-house enzyme-linked immunosorbent assays (ELISAs) to detect antibodies against whole viral antigen have been the preferred methods. Commercial ELISAs from several companies (including IBL, EuroImmun, InBios, and Abcam) have demonstrated acceptable performance (149), though these may cross-react with other alphaviruses, such as ONNV and MAYV (150). EQA studies of CHIKV serology have also been performed. These generally demonstrate poor sensitivity for anti-CHIKV IgM detection, though they demonstrated better performance for IgG assays (139, 151). In the most recent EQA, commercial IFAs offered the best sensitivity overall, and in-house ELISAs were more sensitive than commercial ELISAs (139).

IgM capture ELISAs may provide improved performance, and two separate groups have described in-house assays whose results demonstrated high concordance with PRNT results from the CDC (152, 153). PRNTs are highly accurate but labor-intensive and require BSL-3 facilities. Although rarely performed in clinical laboratories, PRNT remains in use for diagnosis when available due to its high specificity (103). Attempts have been made to develop versions of the PRNT using noninfectious virus replicon particles or chimeric viruses that do not require enhanced biosafety procedures (154, 155). Commercial, rapid diagnostic tests for anti-CHIKV IgM and IgG have been developed (e.g., SD Bioline and OnSite Chik) but have very low sensitivity and specificity (150, 156–158).

An important consideration for the use of serological testing is the extended duration of antibody detection following acute infection. CHIKV-specific IgM levels tend to peak at between 4 and 20 days after symptom onset, but these may not wane for 11 to 14 months (103) and CHIKV-specific IgM has been detected as late as 18 months postinfection by direct ELISA (143). Therefore, anti-CHIKV IgM detection in the acute phase of infection can provide only a presumptive diagnosis. CHIKV-specific IgG remains detectable long after infection and may indicate lifetime protection. IgG generally rises after IgM, though IgG antibodies to CHIKV E2 epitopes have been detected as early as 6 days after the onset of symptoms (159). In addition to serum and plasma, anti-CHIKV antibodies have been detected in CSF from patients with neurological manifestations (sensitivity, 80%; specificity, 87%) (145).

Other methodologies have recently been developed to simplify and/or improve upon traditional ELISAs. Serum spotted onto filter paper demonstrated 98.2% concordance with frozen samples when evaluated for serological testing on St. Martin (148). The use of multiple antigenic E2 peptides (160, 161), the preparation of E1 or E2 proteins (162–164), or the use of recombinant monoclonal E2 antibodies (165) has shown high specificity compared to whole-virus detection. Seroreactivities to E1 and E2

differ slightly, with high specificities for both by IgM capture ELISA but higher sensitivity for E2 than for E1 (90% compared to 78%) (166). The use of such reagents in capture ELISAs may eliminate the biohazard risk associated with the use of whole-virus preparations (166, 167). Additionally, these assays are more specific and may resolve issues related to cross-reactivity among alphaviruses. ONNV polyclonal antibodies weakly neutralize CHIKV, but the misdiagnosis of CHIKV for ONNV is possible, as between 71 and 86% of monoclonal antibodies to CHIKV also neutralized ONNV (41). Highly specific epitope-blocking ELISAs to the E2 protein do not show significant cross-reactions with other alphaviruses (168) yet identify antibodies that cross-protect between CHIKV lineages (159).

Antigen detection. Antigen capture assays are also in development, though these are used less commonly than antigen-based methods for DENV. An immunochromatographic assay using monoclonal antibodies against the E1 protein was developed to detect CHIKV antigen in serum (169), but this test was found to be sensitive only for the ECSA lineage (89%) and not for the Asian lineage (33%) (170). Another antigen capture ELISA reported 96% concordance with real-time RT-PCR results for acute-phase samples from 200 subjects in India (146), and a test for whole CHIKV antigen in acute-phase samples had an overall agreement of 94% with RT-PCR (171). As these assays continue to develop and improve, antigen-based detection may provide rapid methods for CHIKV diagnostic confirmation and expand testing to laboratories without the capacity for molecular methods.

PROGNOSIS, OUTCOME, AND PREVENTION

Several agents have demonstrated activity against CHIKV *in vitro*, and monoclonal antibodies have shown efficacy in animal models (172–175), but currently, there is no specific antiviral treatment for CHIKV and management remains symptomatic. With the exception of joint pain, the symptoms of acute chikungunya typically resolve over a few days to 1 week (1, 68). Approximately 50% of patients have chronic arthralgia and/or arthritis at 3 to 6 months, and over 25% of patients may still have symptoms at 12 months, which negatively impacts quality of life during recovery (68, 176–180). Although significant heterogeneity regarding the rate of chronic joint pain following chikungunya exists in the literature, older age (>35 to 60 years) and preexisting joint disease have been consistently associated with this outcome (176, 177). Persistent joint symptoms may occur more often in women (177, 179), and one meta-analysis observed a nonsignificant trend toward more frequent chronic manifestations following infections with ECSA lineage strains (50%) than following infections with Asian lineage strains (36%) (178).

During recent outbreaks, mortality from CHIKV infections (attributable and associated) has been 14 to 80/100,000 inhabitants (181–183). Disease severity and excess deaths increase markedly in individuals >70 years old, and in the Dominican Republic, patients over 80 years old experienced a case fatality rate of 4.5% (62, 181, 182, 184). There are data indicating that the CHIKV viral load at presentation is associated with disease severity. Average viral loads are highest among neonates and the elderly, and viral loads are higher in hospitalized cases (7, 65, 66, 102, 184). However, the clinical applicability of these findings remains to be established.

Vector control is the primary mode of CHIKV prevention, though published data support the concept that a CHIKV vaccine could be highly efficacious and provide lasting immunity. Long-term protection against a following infection by CHIKV has been observed in areas of endemicity (185, 186), and a number of candidate vaccines have demonstrated promising results in preclinical studies. A few vaccines have now entered phase I and II clinical trials; however, the design and completion of phase III trials will be a significant challenge in the setting of a low number of cases during interepidemic periods. A complete discussion of CHIKV candidate vaccines is beyond the scope of this minireview, and these have recently been reviewed elsewhere (187–189).

CONCLUSION

Although the number of chikungunya cases has decreased since 2014, CHIKV has become endemic in countries across the tropics and has the capacity to cause sporadic outbreaks in naive individuals. This creates new challenges for CHIKV detection and surveillance as clinical cases become less frequent and may be misdiagnosed if accurate laboratory tests do not remain available. Furthermore, decreased case numbers conceal the need for improved diagnostics and prognostics that could identify individuals at high risk for chronic disease or poor outcomes and target prevention efforts to curb future explosive outbreaks.

SUPPLEMENTAL MATERIAL

Supplemental material for this article may be found at https://doi.org/10.1128/JCM.00350-19.

SUPPLEMENTAL FILE 1, PDF file, 0.1 MB.

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