Zika Virus Disease

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Faculty

John M. Leonard, MD, Professor of Medicine Emeritus, Vanderbilt University School of Medicine, completed his postgraduate clinical training at the Yale and Vanderbilt University Medical Centers before joining the Vanderbilt faculty in 1974. He is a clinician-educator and for many years served as director of residency training and student educational programs for the Vanderbilt University Department of Medicine. Over a career span of 40 years, Dr. Leonard conducted an active practice of general internal medicine and an inpatient consulting practice of infectious diseases.

Carol Shenold, RN, ICP, graduated from St. Paul's Nursing School, Dallas, Texas, achieving her diploma in nursing. Over the past thirty years she has worked in hospital nursing in various states in the areas of obstetrics, orthopedics, intensive care, surgery and general medicine.

Mrs. Shenold served as the Continuum of Care Manager for Vencor Oklahoma City, coordinating quality review, utilization review, Case Management, Infection Control, and Quality Management. During that time, the hospital achieved Accreditation with Commendation with the Joint Commission, with a score of 100.

Mrs. Shenold was previously the Infection Control Nurse for Deaconess Hospital, a 300-bed acute care facility in Oklahoma City. She is an active member of the Association for Professionals in Infection Control and Epidemiology (APIC). She worked for the Oklahoma Foundation for Medical Quality for six years.

Faculty Disclosure

Contributing faculty, John M. Leonard, has disclosed no relevant financial relationship with any product manufacturer or service provider mentioned.

Contributing faculty, Carol Shenold, RN, ICP, has disclosed no relevant financial relationship with any product manufacturer or service provider mentioned.

Division Planners

Ronald Runciman, MD
Jane C. Norman, RN, MSN, CNE, PhD

Director of Development and Academic Affairs

Sarah Campbell

Division Planners/Director Disclosure

The division planners and director have disclosed no relevant financial relationship with any product manufacturer or service provider mentioned.

Audience

This course is designed for physicians, physician assistants, and nurses in all settings who may identify and act to prevent Zika virus disease.

Accreditations & Approvals

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Course Objective

The purpose of this course is to provide health professionals with a review of the important clinical and public health aspects of Zika virus disease, including the epidemiology of Zika virus migration, modes of transmission, clinical manifestations, approach to diagnosis, and strategies for risk reduction and prevention of infection.

Learning Objectives

Upon completion of this course, you should be able to:

1. Describe the historical background and dynamics of the Zika virus epidemic in the Americas and its potential impact on public health.
2. Discuss and advise patients as to the risks of Zika virus infection in relation to the various routes of transmission.
3. Recognize and manage a patient presenting with characteristic clinical and epidemiologic features of acute Zika virus disease.
4. Discuss the salient features of microcephaly, including the incidence, causative factors, and clinical and pathologic findings unique to congenital Zika virus infection.
5. Select the appropriate laboratory diagnostic tests for Zika virus in relation to a patient's clinical profile and the time elapsed since exposure or onset of symptoms.
6. Devise a management plan for persons with known or suspected infection, including care of the pregnant patient or intimate partner.
7. Using knowledge of Zika virus shedding by infected men and the risk of sexual transmission, counsel patients and couples on the importance and recommended duration of safe sex practice.
8. Using knowledge of vector transmission and the behavior of Aedes aegypti mosquitoes, devise an effective strategy for avoiding bites, limiting exposure, and eliminating mosquito-breeding habitat.

INTRODUCTION

Zika virus (ZIKV) is one in a series of related human arboviral pathogens that has migrated out of Africa and Asia into the Americas during the past two decades [1; 2; 3]. Arboviruses are transmitted by blood-feeding arthropod vectors, principally mosquitoes and ticks. ZIKV and other arboviruses are maintained in cycles of transmission between a competent vector and susceptible vertebrate species in the environs. Like yellow fever, dengue, and chikungunya viruses, the vector for ZIKV is the *Aedes* mosquito and epidemic outbreaks within susceptible population groups are sustained by a mosquito-human-mosquito transmission cycle.

Until recently, ZIKV disease was rarely reported and of little-known consequence; circulation of the virus was largely confined to a mosquito-nonhuman primate-mosquito transmission cycle in forested terrain across a portion of east Africa and the adjacent Asian Pacific [1]. In 2007, the first outbreak of human ZIKV disease appeared on Yap Island in Micronesia. This was followed in 2013 by a large-scale epidemic in French Polynesia. Subsequent outbreaks were reported on other Pacific islands as ZIKV migrated in epidemic fashion across the Pacific and into the Americas. With the aid of a highly competent vector (the *A. aegypti* mosquito) having ready access to large, susceptible population groups, successive outbreaks of ZIKV disease spread rapidly throughout South and Central America and the Caribbean, including Puerto Rico [49].

Primary ZIKV infection is often asymptomatic or causes a relatively mild, self-limited illness. However, infection during pregnancy may lead to transplacental transmission of virus to the fetus, resulting in arrested neurologic development, microcephaly, and related congenital anomalies. There is growing evidence linking ZIKV infection with post-infectious Guillain-Barré syndrome. In February 2016, following the report of a marked increase in newborn infant microcephaly some months following a large ZIKV outbreak in Brazil, the World Health Organization (WHO) declared ZIKV disease a public health emergency of international concern [6]. A short time later, the Centers for Disease Control and Prevention (CDC) elevated its response to level 1, the highest for the agency [7]. Because exposure to ZIKV during pregnancy is linked to microcephaly in newborn infants, pregnant women and those who may become pregnant are advised to avoid travel to regions of ongoing ZIKV transmission, and all travelers are urged to take enhanced precautions in areas where ZIKV is circulating.

Following the 2015–2016 outbreak in this hemisphere, reported cases of ZIKV disease in the United States occurred predominantly in travelers to affected areas and in women having intimate sexual contact with men infected while traveling to regions with ongoing mosquito transmission. Because of ongoing ZIKV circulation in many nearby regions of the hemisphere and an increasing number of ZIKV disease cases among travelers visiting or returning to the United States, ZIKV disease and ZIKV congenital infection were added to the list of nationally notifiable conditions. The CDC Arboviral Disease Branch provides periodic updates of confirmed ZIKV cases reported in the United States and its territories [8].

In 2016, a total of 5,168 cases of ZIKV disease were reported in the United States, of which 4,897 cases were in travelers returning from affected areas, 224 were acquired through presumed local mosquito-borne transmission in Florida (218) and Texas (6), and 47 cases were acquired through other routes, primarily sexual transmission [8]. The number of reported cases from U.S. territories totaled 36,512, of which the great majority was acquired through presumed local mosquito-borne transmission.

Puerto Rico (35,375 cases) was particularly affected, followed by U.S. Virgin Islands (986 cases) and American Samoa (131 cases). An estimated 25% of the population of Puerto Rico may have become infected by the end of 2016 [50]. The number of reported ZIKV disease cases in the United States and its territories began to decline in 2017. In 2018, there were 72 cases reported in the states, all in travelers returning from affected areas; 148 cases were reported in the territories, nearly all acquired through local mosquito-borne transmission [8]. Since 2019, no local mosquito-borne ZIKV transmission has been reported in the United States and no confirmed ZIKV cases reported from U.S. territories.

The potential for a localized or regional outbreak of ZIKV disease in the United States is significant given the level of travel exposure, opportunities for ZIKV migration, and the prevalence of *A. aegypti* mosquitoes along the southern and southeastern rim of the country [9]. Although local mosquito-borne transmission accounted for small clusters of cases in south Florida and Texas during 2016–2017, no such cases were reported in 2018, and the CDC reports no current local transmission of ZIKV in the continental United States [8].

This course is designed to address knowledge gaps, enhance clinical skills, and provide management strategies for healthcare providers who may be called upon to assess or advise international travelers, women who are pregnant or of childbearing age, or persons with febrile rash illness who have recently returned from endemic areas.

HISTORICAL BACKGROUND AND EPIDEMIOLOGY

ZIKV is a single-stranded RNA virus closely related to dengue and belonging to the family of flaviviruses [10]. Like other flaviviruses that infect humans, ZIKV is transmitted by the bite of an infected mosquito.

ZIKV was first identified in 1947, after being isolated from an ill rhesus monkey caged in the Zika Forest of Uganda as part of a sentinel surveillance program for yellow fever [1]. One year later, the virus was isolated from *A. africanus* mosquitoes recovered from the same forest. In subsequent decades, documentation of human ZIKV infection was provided by population-based serologic studies of arbovirus infection in parts of Africa and Asia, combined with occasional case reports of human ZIKV isolation in association with febrile illness. By 1981, ambient human seropositivity for ZIKV had been reported from Nigeria, Uganda, other nearby African countries, and parts of Asia, including India, Malaysia, the Philippines, and Indonesia [4].

In April 2007, on Yap Island, Federated States of Micronesia in the western Pacific, physicians became aware of an outbreak of mild dengue-like illness characterized by rash, conjunctivitis, and arthralgia. The ensuing investigation was the first population-based epidemiologic study of a human ZIKV epidemic [11]. The outbreak lasted four months; 49 confirmed and 57 suspected cases were identified. ZIKV RNA was detected in serum samples obtained from patients during the acute phase of illness. No dengue or other arbovirus RNA was detectable. A household survey and serologic study conducted on a select sample of the population revealed that 414 of 557 participants (74%) were positive for immunoglobulin M (IgM) antibody to ZIKV, indicating recent infection. Among seropositive participants, 19% reported recent illness consistent with ZIKV

infection. Investigators estimated that 5,005 of 6,892 Yap residents (73%) 3 years of age or older had been infected with ZIKV during the outbreak; moreover, approximately 80% of infections had been asymptomatic or too mild to prompt medical attention. The *A. hensilii* mosquito was judged to be the vector, though no virus or viral RNA could be detected in any pools of trapped mosquitoes [11].

In 2013–2014, an outbreak of ZIKV infection was reported from French Polynesia, a territory consisting of 67 islands arranged in five archipelagoes located in the South Pacific. Between October 2013 and February 2014, the regional sentinel surveillance network recorded 8,262 suspected cases of ZIKV disease [12]. Of 746 samples sent for laboratory confirmation, 396 (53%) were confirmed by reverse transcription polymerase chain reaction (RT-PCR). An estimated 28,000 cases of ZIKV-like illness were seen during the course of the epidemic (about 11% of the population of French Polynesia). The *A. aegypti* mosquito was considered the principal vector and the clinical features of acute infection were similar to those reported in the Yap outbreak. The first reports of postinfectious neurologic complications of ZIKV (e.g., Guillain-Barré syndrome) emerged during the French Polynesia outbreak [12]. Subsequent outbreaks in 2014–2015 were reported on other Pacific islands, including New Caledonia, Easter Island, Cook Islands, and Samoa.

In early spring 2015, ZIKV was identified as the cause of an outbreak of febrile rash illness in Bahia State, Brazil, the first indication that the virus had migrated to the Americas. In the months that followed, a progressive, widespread outbreak of ZIKV disease among adults and children occurred in 29 Brazilian states, followed in turn by an unexpected and significant increase in the number of reported infants born with microcephaly [6]. By December 2015, the number of suspected cases of ZIKV disease had reached 56,318. Brazilian authorities estimate that 500,000 to 1,500,000 persons were infected with ZIKV during the first 18 months of the outbreak. In 2016, outbreaks of ZIKV disease and evidence of continuing mosquito-borne transmission were reported from 73 countries and territories, primarily in Latin America and the Caribbean [42].

New and important aspects of ZIKV disease and transmission emerged from investigations during the recent series of regional epidemics. It is now established that ZIKV infection during pregnancy may have serious adverse effects on the developing fetus, resulting in excess fetal loss or congenital ZIKV disease [13; 14]. There is growing realization that ZIKV infection is linked to the observed increases in the incidence of post-infectious Guillain-Barré syndrome reported in several locales following the 2015–2016 outbreak [15]. Finally, sexual transmission of ZIKV by men with recent symptomatic infection has been documented, adding a further element of complexity to strategies for prevention in women who are pregnant or may become pregnant [16].

ZIKA VIRUS TRANSMISSION

Whether cases are epidemic or endemic, ZIKV is primarily vector-borne, transmitted by the bite of an infected *Aedes* mosquito. In addition, other modes of transmission are now known to be important in human ZIKV disease. These include sexual transmission from an infected male to female and male partners; transplacental transmission from mother to fetus during pregnancy, leading to congenital ZIKV disease; and perinatal transmission from a viremic mother to her newborn infant [16; 17]. Because of theoretical concern that blood transfusion and tissue/organ transplantation could also serve as vehicles of transmission, the U.S. Food and Drug Administration has recommended universal screening of donated whole blood and components for ZIKV in the United States and its territories [48].

MOSQUITO-BORNE TRANSMISSION

As noted, the *A. aegypti* mosquito is the principal vector of transmission for most human arbovirus infections, including yellow fever, dengue, chikungunya viruses and ZIKV [2; 3]. *A. aegypti* is one of several species belonging to the *Aedes* genus (*Stegomyia* subgenus) of mosquitoes [10]. *Aedes* species are distributed in various combinations throughout tropical and subtropical regions of the world, having adapted in different ways to prevailing climate and habitat. In remote rainforests of Africa, where ZIKV circulates in a mosquito-nonhuman primate-mosquito transmission cycle, *A. africanus* is the primary vector. In heavily populated and urban areas of Latin America and the Caribbean, the vector of transmission for outbreaks of human ZIKV disease is *A. aegypti* and, to a lesser extent, *A. albopictus*.

There is also variability among *Aedes* species with respect to vector competence (i.e., the intrinsic ability of a vector to transmit a disease agent) and vectorial capacity (i.e., the overall effectiveness of a vector to sustain and propagate a disease outbreak in a given location) [3; 4]. *A. aegypti* and *A. albopictus* have comparable vector competence for ZIKV transmission, but *A. aegypti* exhibits greater vector capacity among human population groups, perhaps because of its behavior and adaptation to an urban environment [18].

In times past, *A. aegypti* thrived on nonhuman hosts, laying its eggs in water collected in tree holes and the axils of forest plant leaves; in recent decades, this mosquito has adapted to an urban habitat and shows a preference for a human host over other mammals [19]. It flourishes in crowded areas with no piped water, inadequate trash disposal, and ineffective domicile barrier protection, such as that afforded by screened doors and windows. A single female deposits its eggs at multiple sites, such as stagnant water found in cemetery vases, pet bowls, abandoned barrels, and automobile tires. Adult mosquitoes of both sexes feed on nectar and fruit, but females require blood protein in order to fully develop their eggs. Thus, only the female mosquito bites humans.

A. aegypti is an aggressive daytime biting mosquito, and feeding is most intense in the hours around dawn and dusk. The bite itself is barely perceptible. Female *A. aegypti* mosquitoes are stealth feeders, approaching victims from behind and biting on ankles and elbows—a "sneak attack" that avoids being noticed [19]. This mosquito does not feed sufficiently with a single bite; it is a "sip feeder" that bites multiple humans in the course of a blood meal, thereby optimizing the vector capacity of a single mosquito carrying the virus. The female prefers shady areas for rest and is adept at hiding in closets and under beds, later to emerge for a nocturnal blood meal.

With the exception of mountainous regions above 3,500 feet, the range of *A. aegypti* and *A. albopictus* includes most of Latin America and the Caribbean and extends into parts of the contiguous United States. While the prevailing range of *A. aegypti* within the United States is limited to south Texas along the Mexican border, south Florida, and coastal areas of the gulf and southern Atlantic states, climate conditions are favorable for periodic expansion into adjacent states [9]. The *A. albopictus* species is acclimated to a milder climate and has a broader range that extends from the eastern seaboard through the Southeast and a portion of the Midwest, and throughout the Southwest. Of public health concern is the following scenario: a local area of ZIKV circulation among *A. aegypti* mosquitoes and humans becomes established in the United States, from which *A. albopictus* emerges as a secondary vector with potential for a more widespread outbreak in other parts of the country.

HUMAN-TO-HUMAN AND SEXUAL TRANSMISSION

Although infected mosquito-borne transmission accounts for most cases of ZIKV disease, human-to-human transmission has been reported through direct exposure to infected body fluids by sexual contact, blood transfusion, laboratory exposure, and both intrauterine and intrapartum transmission [54]. ZIKV RNA has been detected in saliva, urine, semen, vaginal secretions, and other body fluids following acute infection [20].

Epidemiologic investigation and published case reports have demonstrated that a man with symptomatic ZIKV infection can transmit the virus to his partner through intimate sexual contact, including vaginal, anal, and likely oral sex [16; 20; 21; 53]. Within the United States and other countries having no ZIKV circulation, cases of well-documented ZIKV disease have been reported in women whose only risk exposure was sexual contact with a symptomatic male partner who had recently traveled from an area with ongoing ZIKV transmission. An illustrative case report is that of a woman, 24 years of age, living in France, who on February 16, 2016, became ill with fever, arthralgia, myalgia, and a pruritic rash [21]. Samples of urine and saliva collected on the third day of illness were positive for ZIKV RNA by RT-PCR, and the serum tested positive for acute phase ZIKV IgM antibody. There is no known ZIKV circulation in that part of Europe, and the patient had no history of travel to an endemic area. She did report that on several occasions in the week prior to onset of symptoms she had sexual contact (vaginal intercourse and oral sex) with a man who had just returned from a two-month stay in Brazil. He also had experienced a febrile rash illness with arthralgia during the four days prior to his departure for France on February 10. Urine and semen samples obtained 16 days after onset of his symptoms tested positive for ZIKV RNA by RT-PCR. ZIKV was isolated by culture from semen samples obtained on day 18 and on day 24 following onset of his illness [21].

The full spectrum of behaviors and circumstances by which ZIKV is transmitted sexually is not yet known, nor is the duration of infectivity certain. In one reported case, ZIKV RNA was detected in semen up to 62 days after onset of symptoms, and replication-competent ZIKV has been isolated from semen at least two weeks after onset of illness [22]. Another study found that ZIKV RNA detection in semen is common following acute infection and may persist for as long as six months, but shedding of infectious ZIKV is less common and limited to a few weeks [51]. In a 2018 cohort study of 295 infected participants in Puerto Rico, including 94 men who provided semen specimens, the 95th percentiles for time until the loss of ZIKV RNA detection were 41 days in serum, 34 days in urine, and 120 days in semen [54]. Less than 5% of participants had detectable ZIKV RNA in saliva or vaginal secretions. ZIKV was isolated 10% of semen specimens in a period ranging from 15 to 38 days after onset of illness. In this longitudinal study, 11% of men had detectable ZIKV RNA at 90 days and 5% at 120 days; however, isolation of infectious virus from semen was only possible at high viral loads and for a maximum period of 38 days.

Most reported cases of sexual transmission involved vaginal penetration or anal sex with men before, during, or shortly after a symptomatic illness consistent with ZIKV disease [22]. One case of suspected female-to-male sexual transmission of ZIKV has been reported [53]. Sexual transmission has also been reported from a man with subclinical infection, and the CDC considers that the incidence of virus shedding in semen and its persistence after infection are likely similar for symptomatic and asymptomatic men infected with ZIKV [55].

The consistent and correct use of latex condoms is known to reduce substantially the risk of acquiring sexually transmitted infections, including those caused by viruses. The CDC recommends that men infected with ZIKV use condoms or abstain from sex for at least three months after symptom onset (if symptomatic) or last possible exposure (if asymptomatic) to minimize the risk of sexual transmission [52]. If the partner is pregnant, then condom use or abstinence is recommended for the duration of the pregnancy.

VERTICAL TRANSMISSION

As noted, acute ZIKV infection during pregnancy may result in transplacental transmission of infection to the developing fetus. Evidence for congenital ZIKV infection includes demonstration of ZIKV in the placenta and products of conception following spontaneous abortion, identification of ZIKV RNA in amniotic fluid by RT-PCR, and virologic and serologic studies of infants born with microcephaly. The true incidence and natural history of this phenomenon, including the importance of such factors as gestational age, level and duration of viremia, and the role of immune enhancement by pre-existing heterologous anti-flavivirus antibodies, is currently unknown [23]. Studies of fetal outcomes in pregnancy show that maternal-fetal transmission of ZIKV occurs in all trimesters of pregnancy, whether infection in the mother is symptomatic or asymptomatic [57]. In a cohort of 291 pregnant women with laboratory confirmed ZIKV infection at any stage of pregnancy, maternal-fetal transmission was documented in 26% (76/291) of fetuses/newborns. Of the 76 ZIKV positive fetuses/newborns, 45% had normal findings, 41% moderate-to-severe complications compatible with congenital Zika syndrome, and 14% sustained fetal loss [58].

Two cases of intrapartum transmission of ZIKV from a newly infected, viremic mother to her newborn infant have been reported [4]. One infant was considered to be asymptomatic; the other child developed a rash and transient thrombocytopenia. Although ZIKV has been identified in breast milk, there have been no reports of transmission through breastfeeding.

ZIKA VIRUS DISEASE

CLINICAL MANIFESTATIONS

Acute Illness in Adults and Children

In epidemic settings, the majority of primary ZIKV infections are either asymptomatic or characterized by mild febrile illness with rash, conjunctivitis, myalgia, and arthralgia lasting three to six days. The incubation period for ZIKV is not well defined; it is considered to be similar to that of other mosquito-borne flaviviruses—usually less than one week and in the range of 3 to 10 days.

The first detailed description of the illness caused by acute ZIKV infection is a self-reported case study in 1964 of a young (28 years of age) European research worker at the East Africa Virus Research Institute in Uganda [24]:

The illness began with a slight frontal headache in the evening, followed the next morning by an aching sensation in the back and thighs and the appearance of a maculopapular rash covering the face, neck, trunk, and upper arms. Throughout day 2, the rash, which was non-itching, spread gradually to involve all four extremities, including the palms of the hands and the soles of the feet. Toward midday the patient was febrile (99.4° F) and experiencing malaise accompanied by pain in the back and a frontal headache. By the evening of day 2, the temperature had returned to normal, the rash was beginning to fade from the back and neck, and the patient felt better apart from slight headache. On day 3, the patient felt no ill effects and the temperature remained normal. The rash persisted on the trunk and extremities, faded slowly throughout days 3 and 4, and disappeared completely on day 5. No other signs or symptoms were noted during the illness.

This worker had visited Zika Forest 21 days before onset of illness and had been bitten by mosquitoes during a period when ZIKV was being isolated from mosquitoes collected in the forest. ZIKV was isolated from the patient's blood by mouse inoculation studies, and a rise in anti-ZIKV antibody was demonstrated. The clinical features of the infection—a

mild, self-limited febrile illness with malaise, myalgias, headache, and exanthema—was likened to that seen with other arthropod-borne viruses such as West Nile and chikungunya [24].

A more complete description of the natural history of acute ZIKV infection is provided by clinical observations collected during the investigation of the four month-long epidemic on Yap Island in 2007 [11]. Based on results of a seroepidemiologic survey, only about 18% of 5,005 islanders estimated to have been infected developed symptoms attributable to ZIKV. In the course of this investigation, information regarding symptoms and signs was obtained from 31 of 49 confirmed cases (63%) of ZIKV disease. The most common clinical characteristics were macular or papular rash (90%), fever (65%), arthritis or arthralgia (65%), non-purulent conjunctivitis (55%), myalgia (48%), and headache (45%) [11]. Other symptoms included retro-orbital pain, edema, and vomiting. The median duration of rash was 6 days (range: 2 to 14) and arthralgia was 3.5 days (range: 1 to 14). There were no hospitalizations or deaths attributed to ZIKV illness in the course of this outbreak.

A similar pattern of illness was observed in a cohort study from Rio de Janeiro, wherein pregnant women who had experienced fever and rash illness within the previous five days were enrolled in a surveillance study designed to assess the cause of illness and to monitor subsequent maternal health and fetal development [25]. During the period from September 2015 through February 2016, 72 of 88 women enrolled tested positive for acute ZIKV infection by RT-PCR on blood, urine, or both. All women had rash, as this was an inclusion criterion; the prevailing pattern was a descending macular or maculopapular exanthema accompanied by pruritus in 94% of patients. Arthralgia was reported in 65% of ZIKV-positive women, conjunctival injection was seen in 58%, and lymphadenopathy (generalized or regional) was present in 41%. Fever was documented in only one-third of patients and, when present, was low-grade and of short duration. Nausea and vomiting were reported in 21%, and respiratory findings were evident in 7% [25].

From these observations emerges a distinctive, though nonspecific, clinical ZIKV syndrome: an acute onset descending maculopapular rash (often with pruritus), conjunctival injection, arthralgia, myalgia, and low-grade fever. Lymphadenopathy and respiratory symptoms are uncommon. ZIKV disease should be considered in patients with any combination of these symptoms who have traveled to areas with ongoing transmission in the two weeks preceding onset of illness. Rare manifestations of acute ZIKV infection, based on isolated case reports, include meningoencephalitis, myelitis, thrombocytopenic purpura, and ocular complications [4; 26; 27; 59].

Because dengue and chikungunya viruses have the same vector of transmission and share similar geographic distributions and clinical profiles with ZIKV, clinicians should consider diagnostic evaluation and management for these possibilities when encountering patients with suspected ZIKV disease. The differential diagnosis of ZIKV disease also includes malaria, rubella, measles, parvovirus, adenovirus, enterovirus, leptospirosis, rickettsiosis, and group A streptococcal infections [5].

Neurologic Complications of ZIKV Disease

ZIKV is a neurotropic virus, which accounts for its association with post-infectious Guillain-Barré syndrome and with infant microcephaly and related neurologic abnormalities that follow intrauterine infection.

Guillain-Barré Syndrome

Guillain-Barré syndrome is an acute, progressive, motor axonal neuropathy characterized by flaccid paralysis. It is considered to be immune-mediated, often triggered by infection. Most patients recover after many weeks or months, often requiring prolonged hospitalization, respiratory ventilation support, and management of other complications that are costly and burdensome to any health system.

An unexpected increase in the number of patients presenting with Guillain-Barré syndrome has been observed in countries experiencing large outbreaks and ongoing transmission of ZIKV. During the 2013–2014 ZIKV outbreak in French Polynesia, in the space of four months' time, 38 cases of Guillain-Barré syndrome were diagnosed among an estimated 28,000 persons who sought medical care for suspected ZIKV disease [12]. Based on annual reported cases of Guillain-Barré syndrome for the prior four years, the number expected in a four-month period was three or less. Among the ZIKV-associated cases, 73% were male and the mean age was 46 years; 15 patients were admitted to the intensive care unit, and 9 required mechanical ventilation; there were no reported deaths. Clusters of ZIKV-associated Guillain-Barré syndrome have been reported in eight countries: French Polynesia, Brazil, El Salvador, French territory of Martinique, Colombia, Suriname, the Bolivarian Republic of Venezuela, and Honduras [4].

In addition to postinfectious Guillain-Barré syndrome, other neurologic complications have been reported following ZIKV infection. These include meningoencephalitis, myelitis, acute transient polyneuritis, and chronic inflammatory demyelinating polyneuropathy [59].

Microcephaly and Congenital Zika Syndrome

Microcephaly is a rare pediatric disorder often associated with other congenital anomalies and developmental complications. Microcephaly may be classified as prenatal (congenital) or postnatal (developing sometime after birth); only the former has been linked with ZIKV infection. The growth of the fetal brain in utero approaches maximum volume after 21 weeks' gestation, a process that influences the size of the newborn infant skull. Microcephaly is the clinical (or radiographic) finding of a head size unexpectedly small for a given stage of development; it is usually defined as an occipitofrontal head circumference below the third percentile for gestational age and sex [4]. Injury that causes sequence disruption of normal fetal brain growth can lead to microcephaly. The common causative factors are genetic perturbations, maternal illness (e.g., infection), and exposure to teratogenic substances. The incidence of congenital microcephaly is estimated to be 2 to 10 cases per 10,000 live births [4].

The subsequent clinical course of children born with microcephaly is difficult to predict; when intrauterine infection is the cause, the severity and prognosis are determined in large part by gestational age of onset and the presence of additional neurologic deficits. Common childhood sequelae include developmental delay, hearing loss, vision defects, impaired intellectual ability, and seizures.

In November 2015, after alarming reports of an epidemic of microcephaly in northern Brazil, later attributed to maternal-fetal ZIKV infection, the Brazilian Ministry of Health set up a surveillance system for cases of microcephaly and other malformations possibly linked with ZIKV. From November 2015 to June 2016, 7,830 suspected cases were reported, of which 40% were judged confirmed or probable cases of congenital ZIKV syndrome based on clinical and epidemiologic investigation [28]. For this six-month period, the number of newborns with microcephaly associated with confirmed ZIKV infection represents a 40-fold increase in monthly reports of microcephaly in Brazil prior to 2015. In the course of this case-by-case investigation, clinicians discovered that 1 in 5 children with definite or probable congenital ZIKV infection presented with head circumference in the normal range. Thus, the sensitivity of microcephaly alone to detect cases of congenital ZIKV was 83%, increasing to 87% when history of maternal rash was included [28].

Knowledge about the risks of adverse fetal outcomes following maternal ZIKV infection has gradually emerged from studies conducted during the outbreaks of ZIKV infection in 2014–2016. In the Rio de Janeiro cohort study previously noted, a cohort of pregnant women with definite ZIKV infection, contracted between week 6 and week 36 of gestation, were agreeable to fetal monitoring with serial ultrasonography [28]. Subsequent fetal abnor-

malities were identified in 12 of 42 (29%) patients, including growth restriction, cerebral calcifications, microcephaly, and other brain malformations. There were two fetal deaths after 30 weeks' gestation (4.8%) [25]. As of January 2018, 3,720 cases of congenital birth defects associated with maternal ZIKV infection in the Americas had been reported to the Pan American Health Organization [60].

In comparison with other known causes of congenital infection (e.g., toxoplasmosis, rubella, cytomegalovirus, herpes simplex virus, syphilis), the microcephaly associated with ZIKV infection has a unique phenotype consistent with fetal brain disruption [4; 29]. The severity of fetal brain injury and the characteristic clinical and pathologic features associated with congenital Zika syndrome are illustrated by the following case report. An expectant mother, 25 years of age, developed a febrile rash illness during the 13th week of gestation while living and working in northern Brazil. Ultrasonography performed at 14 and 20 weeks' gestation indicated normal fetal growth and anatomy. At 29 weeks, the patient reported reduced fetal movements and follow-up imaging showed early signs of fetal anomalies. Ultrasonography at 32 weeks' gestation confirmed intrauterine growth retardation, microcephaly, and numerous calcifications in various parts of the brain. Because of severe brain disease and a poor prognosis, the pregnancy was terminated by request at 32 weeks' gestation. Autopsy examination revealed microcephaly, widely open Sylvian fissures, small cerebellum and brain stem, almost complete agyria, and internal hydrocephalus of the lateral ventricles. Numerous calcifications of variable size were found in the cortex and subcortical white matter of frontal, parietal, and occipital lobes. Microscopic examination of the brain showed extensive neuronal destructive change with multifocal filamentous neuronal calcifications, diffuse astrogliosis, and degeneration of the long descending tracts within the brain stem and spinal cord. Tissue samples were positive for ZIKV by RT-PCR assay and the complete genome of ZIKV was recovered from the fetal brain [30].

The distinctive characteristics of microcephaly associated with congenital ZIKV infection, evident on neuroimaging and by pathologic examination, include extensive intracranial calcifications, severe cortical atrophy and malformation, hypodensity of the white matter, cerebellar hypoplasia, and ventriculomegaly. ZIKV infection during pregnancy has also been linked to other adverse outcomes, including excess miscarriage and stillbirths, ocular defects, hearing loss, and impaired growth in infants.

In April 2016, after careful consideration of available data, CDC epidemiologists concluded that a causal relationship exists between prenatal ZIKV infection and infant microcephaly and other neurologic abnormalities [14; 29]. Supporting evidence includes the timing of infection during prenatal development is consistent with the defects observed; the observed phenotype is specific, involving microcephaly and associated brain anomalies in infants with confirmed congenital ZIKV infection; ZIKV has been identified in brain tissue of affected fetuses and infants.

The U.S. Zika Pregnancy and Infant Registry (USZPIR) was established to better define the incidence of birth defects among infants born to mothers with laboratory-confirmed ZIKV infection during pregnancy. Among 6,799 live-born infants in USZPIR born between December 1, 2015 and March 31, 2018, 4.6% had one or more Zika-associated birth defect; in a subgroup of pregnancies with positive RT-PCR for ZIKV infection, the percentage was 6.1% [61]. The reported defects were microcephaly, intracranial calcifications, ventriculomegaly, corpus callosum and cortical gyrus pattern abnormalities, chorioretinal atrophy or scarring, and optic nerve abnormalities. One-third of infants had more than one defect reported. ZIKV-associated birth defects were reported with exposures throughout pregnancy but were more prevalent among infants born to mothers exposed early in pregnancy. Approximately two-thirds of pregnant women in this cohort reported asymptomatic infections. These findings identify brain and ocular defects that might prompt

suspicion of prenatal ZIKV infection, providing early evidence for the reemergence of ZIKV disease in geographic regions without ongoing comprehensive ZIKV surveillance [61].

The absence of identifiable birth defects does not exclude the risk of later ZIKV-associated abnormalities such as hearing loss, visual impairment, and developmental delay. In a cohort study of 70 Columbian infants with in utero ZIKV exposure but without signs of congenital Zika syndrome at birth, cranial ultrasonography and neurodevelopmental assessments were conducted at one or two time points at 4 to 8 months and/or 9 to 18 months of age. Nonspecific neuroimaging findings of lenticulostriate vasculopathy, subependymal cysts, and choroid cysts were present in one-third of study subjects, considered a marker of subtle brain injury potentially associated with impaired neuromotor development [62]. Functional assessment revealed an evolving decline from test norms in some subjects, with differences in social cognition and mobility domains that grew larger over time [62]. The decline in the social cognition domain over time was considered to be an indication of impaired neurocognitive development and the need for longer-term neurodevelopmental evaluation in all infants exposed to ZIKV in utero.

DIAGNOSIS

The possibility of ZIKV disease should be considered in the patient with a compatible clinical syndrome (e.g., febrile rash illness with arthralgia and conjunctivitis) and epidemiologic risk factors such as residence or travel to an area of active ZIKV transmission within the previous two weeks or sexual contact with a person known or suspected of recent ZIKV infection. Laboratory confirmation relies on molecular detection of the viral genome (via RT-PCR) in blood or body fluids and serologic assay for acute-phase ZIKV-specific IgM antibody.

The U.S. Food and Drug Administration has issued emergency use authorization for multiple diagnostic tools for ZIKV, including the Triplex Real-Time RT-PCR assay and the Zika MAC-ELISA for anti-ZIKV IgM [31]. These have been distributed to qualified laboratories that are certified to perform high-complexity tests in the United States. Clinicians should contact local and state health departments to facilitate diagnostic testing. Information on the types and selection of diagnostic tests for ZIKV is available from the CDC [31].

MOLECULAR DIAGNOSTIC TESTING

In the first week after onset of symptoms, ZIKV disease can often be diagnosed by performing RT-PCR on serum. However, RT-PCR for detection of viral nucleic acid in blood is dependent on timing of the sample in relation to the onset, duration, and level of viremia. The viremia that follows ZIKV infection begins a few days before onset of symptoms and lasts about five to seven days. By the time a patient presents with symptoms, the window of opportunity may be short or the degree of viremia below detectable levels. During acute infection, ZIKV is shed in the urine and detectable viral RNA persists well into the second week. For this reason, the CDC recommends that serum and urine samples be submitted for RT-PCR in patients presenting up to 14 days after onset of illness [31]. A positive RT-PCR result on any sample confirms ZIKV infection, and no additional testing is indicated. A negative RT-PCR result does not rule out the diagnosis; a serum sample should then be tested for anti-ZIKV IgM antibody.

SEROLOGIC TESTING

Serologic testing is indicated in patients who present more than one week after onset of symptoms. ZIKV-specific IgM antibodies develop toward the end of the first week of illness and remain detectable for 12 weeks; neutralizing antibodies (IgG) become detectable in the second week and persist for many years [32]. Therefore, if serum and urine are negative by RT-PCR, serum IgM antibody

testing for ZIKV should be performed. However, serodiagnosis may be complicated by cross-reactivity producing false-positive results in persons exposed to other flaviviruses. When the patient resides or has traveled to areas endemic for other flaviviruses, then anti-dengue virus and anti-chikungunya virus IgM antibody testing should be requested as well.

The ZIKV IgM enzyme-linked immunosorbent assay (ELISA) is used for the qualitative detection of anti-ZIKV IgM antibodies in serum and cerebrospinal fluid; however, positive test results can be difficult to interpret because of cross-reactivity with related flaviviruses, which precludes identification of the specific infecting virus [32]. This is especially problematic in persons residing in endemic areas or previously vaccinated against flaviviruses. When the IgM ELISA test result is presumed positive, equivocal, or inconclusive, the serum sample must be forwarded to the CDC (or a CDC-designated laboratory) for confirmation by plaque-reduction neutralization testing (PRNT). PRNT measures virus-specific neutralizing antibodies and is able to discriminate between cross-reacting antibodies in primary flavivirus infections [32].

CONGENITAL ZIKV DISEASE

Laboratory testing for congenital ZIKV infection is recommended for infants born to mothers with laboratory evidence of ZIKV infection during pregnancy and those who have abnormal clinical findings suggestive of congenital Zika syndrome [64]. If intrauterine ZIKV infection is suspected during the course of pregnancy and amniocentesis is performed, fluid should be tested by RT-PCR. When an infant is born with microcephaly or intracranial calcifications to a mother with potential ZIKV infection during pregnancy, the infant should be evaluated for congenital ZIKV infection. Interim guidelines for the evaluation and testing of infants with possible congenital ZIKV infection have been published by the CDC, and updated guidance is available at the CDC website [33; 64]. Because ZIKV serologic testing of blood obtained from the umbilical cord can be misleading, the current recommendation is to obtain a sample of blood, and cerebrospinal fluid if available, directly from the infant within two days of birth for RT-PCR and anti-ZIKV IgM testing. Frozen and fixed placenta tissue samples should be submitted for RT-PCR and ZIKV-specific immunohistochemical analysis at specialized reference laboratories [33]. Healthcare providers should consult with local or state health department and review CDC guidance [64]. Preapproval is required prior to submission of any tissue specimens to CDC laboratories. For infants with any positive or inconclusive test findings for ZIKV infection, healthcare providers should report the case to state, territorial, or local health department and assess the infant for long-term sequelae.

CASE DEFINITION AND NOTIFICATION

As noted, ZIKV disease and congenital ZIKV infection have been added to the list of nationally notifiable diseases. Healthcare providers are encouraged to report probable and confirmed ZIKV disease cases to the local or state health department in order to facilitate diagnosis and mitigate the risk for local transmission in areas where Aedes species mosquitoes are active. The WHO has developed an interim case definition for acute ZIKV disease [45]. A suspected case is defined by the presence of rash and/or fever combined with at least one of the following: arthralgia, arthritis, or nonpurulent conjunctivitis. A probable case is a person with this syndrome and a positive IgM antibody test against ZIKV plus an epidemiologic link (i.e., contact with a confirmed case or residence/travel to an endemic area within the previous two weeks). A confirmed case requires laboratory confirmation of recent ZIKV infection by either molecular diagnostic testing (e.g., presence of ZIKV RNA in serum or other body fluids) or serologic test results specific for ZIKV and exclusive of other flaviviruses (e.g., positive anti-ZIKV IgM ELISA confirmed by PRNT).

The CDC is working with the Council of State and Territorial Epidemiologists (CSTE) to improve surveillance and promote effective prevention and control of ZIKV transmission within the United States. The CSTE has developed an interim position statement intended to standardize case definitions and establish criteria for the classification of ZIKV infection [34]. When evaluating a patient with suspected, probable, or newly confirmed ZIKV infection, healthcare providers should consult this source or contact the local or state health department.

As noted, the USZPIR was established to gather information about the timing, absolute risk, and spectrum of outcomes associated with ZIKV infection during pregnancy. It is accessible at https://www.cdc.gov/pregnancy/zika/research/registry.html. The data collected through this registry will be used to update recommendations for clinical care, to plan for services for pregnant women and families affected by ZIKV, and to improve prevention of ZIKV infection during pregnancy [43]. The CDC also offers consultation services as part of the registry, which may be accessed by calling (770) 488-7100 or emailing ZIKAMCH@cdc.gov.

CLINICAL MANAGEMENT ISSUES

GENERAL TREATMENT

There is no specific antiviral therapy for ZIKV infection; treatment is supportive and directed toward relief of symptoms. When the diagnosis is uncertain and dengue, or co-infection with dengue, is a possibility, the patient should be managed expectantly for each. In consideration of dengue, aspirin and nonsteroidal anti-inflammatory drugs should be avoided and the patient should be monitored for signs of progression to hemorrhagic fever or shock [32]. Patient education and secondary prevention are important, especially in regard to sexual transmission and risk reduction in pregnancy. All pregnant women with molecular or serologic evidence of recent ZIKV infection should be evaluated and managed (monitored) for adverse pregnancy outcomes.

CARE OF THE PREGNANT PATIENT

In 2016, the CDC published interim guidance for healthcare providers caring for pregnant women with possible ZIKV exposure and those with confirmed or suspected ZIKV infection [35]. As of November 2019, dengue virus is causing large outbreaks in many areas of the world, with low to no ZIKV transmission being reported globally. In response, the CDC updated its ZIKV and dengue testing guidance for evaluating persons living in or traveling to areas with active dengue transmission and risk of ZIKV infection [63]. The CDC's updated Zika (and dengue virus) testing guidance for providers, including management of pregnant individuals, is available at https://www.cdc.gov/zika/hc-providers/testing-guidance.html. As noted, the CDC provides a 24-hour consultation service for health officials and healthcare providers managing pregnant women as a component of the USZPIR.

At present, the general recommendations for women residing in the United States and its territories are [35; 63]:

- Routine ZIKV testing is not recommended for asymptomatic pregnant women living in or with recent travel to the United States or its territories.
- Pregnant women are advised not to travel to areas with active ZIKV transmission.
- Pregnant women who must travel to one of these areas should strictly follow steps to avoid mosquito bites and prevent sexual transmission during the trip.
- Pregnant women with a sex partner who has traveled to or lives in an area with active ZIKV transmission should use condoms or other barrier methods to prevent infection or abstain from sex for the duration of the pregnancy.

Evaluation of the Symptomatic Pregnant Patient

Pregnant women who report symptoms or signs consistent with acute ZIKV disease, and who had recent travel to areas with active dengue transmission and risk of Zika, should have serum and urine specimens collected as soon as possible for diagnostic testing up to 12 weeks after symptom onset [63]. The following tests should be performed at the same time: dengue and ZIKV RT-PCR on the serum specimen, and ZIKV RT-PCR on the urine specimen. IgM serologic testing should be done for dengue only. Reports of prolonged detection of ZIKV RNA in symptomatic pregnant women support longer time frames for performance of molecular diagnostic testing [35]. ZIKV IgM testing is not recommended because Zika IgM antibodies can persist for months to years following infection and there is notable cross-reactivity between dengue IgM and Zika IgM antibodies in serologic tests [63]. A positive RT-PCR confirms the diagnosis of recent maternal ZIKV infection. If either the dengue RT-PCR or dengue IgM antibody test is positive, this provides adequate evidence of a dengue infection and no further testing is indicated [63].

For symptomatic pregnant women who have had sex with someone who lives in or recently traveled to an area with risk for Zika, serum and urine specimens should be collected as soon as possible up to 12 weeks after onset of symptoms. Only the Zika RT-PCR test should be performed; if positive (confirmed by repeat testing on newly extracted RNA from the same specimen) the diagnosis of maternal ZIKV infection is established [63].

Evaluation of the Asymptomatic Pregnant Patient

ZIKV testing should be considered for asymptomatic pregnant women who have traveled to areas with risk of Zika outside the United States and its territories, and those who have had potential sexual exposure (i.e., sexual contact without barrier/condom method with a person who lives in or has traveled to an area with risk for Zika). Although not routinely recommended, testing may be considered on a case-by-case basis using a shared physician-patient decision-making model, taking into account patient preferences and values, clinical judgment, degree of exposure, and a balanced assessment of risks and expected benefits [56]. If testing is conducted, then serum and urine specimens should be collected for dengue/ZIKV RT-PCR and dengue IgM serologic testing using the 2- to 12-week time frame from last exposure [63]. Asymptomatic pregnant women residing in areas with active ZIKV transmission (and thus ongoing possible ZIKV exposure) should be offered surveillance serum and urine RT-PCR testing for ZIKV infection three times during pregnancy as part of routine obstetric care [56].

Management of the Pregnant Patient with ZIKV Infection

Updated interim guidance provided by the CDC also includes recommendations for prenatal and postnatal management of the pregnant patient with confirmed or possible ZIKV, and for those carrying a fetus with prenatal ultrasound findings consistent with congenital Zika virus syndrome (***Table 1***) [35; 63].

CLINICAL MANAGEMENT OF A PREGNANT WOMAN WITH SUSPECTED ZIKA VIRUS INFECTION		
Interpretation of Laboratory Results	**Prenatal Management**	**Postnatal Management**
Recent Zika virus infection Recent flavivirus infection, specific virus cannot be identified	Consider serial ultrasounds every three to four weeks to assess fetal anatomy and growth.[a] Decisions regarding amniocentesis should be individualized for each clinical circumstance.[b]	*Live births*: Cord blood and infant serum should be tested for Zika virus by rRT-PCR, and for Zika IgM and dengue virus IgM antibodies. If CSF is obtained for other reasons, it can also be tested. Zika virus rRT-PCR and IHC staining of umbilical cord and placenta are recommended. *Fetal losses*: Zika virus rRT-PCR and IHC staining of fetal tissues are recommended.
Presumptive recent Zika virus or flavivirus infection[c]	Consider serial ultrasounds every three to four weeks to assess fetal anatomy and growth.[a] Amniocentesis might be considered; decisions should be individualized for each clinical circumstance.[b]	*Live births*: Cord blood and infant serum should be tested for Zika virus by rRT-PCR, and for Zika virus IgM and dengue virus IgM antibodies. If CSF is obtained for other reasons, it can also be tested. Zika virus rRT-PCR and IHC staining of umbilical cord and placenta should be considered. *Fetal losses*: Zika virus rRT-PCR and IHC staining of fetal tissues should be considered.
Recent dengue virus infection	Clinical management in accordance with existing guidelines.	
No evidence of Zika virus or dengue virus infection	Prenatal ultrasound to evaluate for fetal abnormalities consistent with congenital Zika virus syndrome.[a] *Fetal abnormalities present*: Repeat Zika virus rRT-PCR and IgM test; base clinical management on corresponding laboratory results. *Fetal abnormalities absent*: Base obstetric care on the ongoing risk for Zika virus exposure risk to the pregnant woman.	

CSF = cerebrospinal fluid, IgM = immunoglobulin M, IHC = immunohistochemical, PRNT = plaque reduction neutralization test, rRT-PCR = real-time reverse transcription–polymerase chain reaction.

[a]Fetal abnormalities consistent with congenital Zika virus syndrome include microcephaly, intracranial calcifications, and brain and eye abnormalities.

[b]Healthcare providers should discuss risks and benefits of amniocentesis with their patients. It is not known how sensitive or specific rRT-PCR testing of amniotic fluid is for congenital Zika virus infection, whether a positive result is predictive of a subsequent fetal abnormality, and if it is predictive, what proportion of infants born after infection will have abnormalities.

[c]rRT-PCR or PRNT should be performed for positive or equivocal IgM results as indicated. PRNT results that indicate recent flavivirus infection should be interpreted in the context of the currently circulating flaviviruses. Because of the overlap of symptoms and areas where other viral illnesses are endemic, evaluate for possible dengue or chikungunya virus infection.

Source: [35] *Table 1*

PREVENTION

TRAVEL TO ENDEMIC AREAS

International travelers visiting regions with ongoing mosquito-borne transmission of ZIKV, chikungunya virus, and dengue should plan carefully and exercise caution. Updated CDC information and guidance is available at https://wwwnc.cdc.gov/travel/page/zika-information.

The CDC guidelines for preventing mosquito bites recommend limiting mosquito exposure and avoiding bites by taking the following steps [36]:

- Cover exposed skin by wearing long-sleeved shirts and long pants.
- Use insect repellents that are registered with the Environmental Protection Agency (EPA) and contain one of the following active ingredients: diethyltoluamide (DEET), picaridin, IR3535, oil of lemon eucalyptus, para-menthane-diol, or 2-undecanone. Always use as directed. Pregnant and breastfeeding women can use all EPA-registered insect repellents, including DEET, according to the product label. Most repellents, including DEET, can be used on children older than 2 months of age. To apply, adults should spray insect repellent onto hands and then apply to a child's face. If it might be difficult to find recommended repellent at your destination, pack enough to last the entire trip.
- Use permethrin-treated clothing and gear (e.g., boots, pants, socks, tents). These items may be purchased pretreated or treated when necessary. (Please note that permethrin is not effective in Puerto Rico.)
- Stay and sleep in screened-in and air-conditioned rooms whenever possible.
- Sleep under a mosquito bed net if air-conditioned or screened rooms are not available or if sleeping outdoors.
- Mosquito netting can be used to cover infants younger than 2 months of age in carriers, strollers, or cribs to protect them from mosquito bites.

PREVENTION OF SEXUAL TRANSMISSION

The CDC recommends that men who have traveled to or reside in an area with active ZIKV transmission and their pregnant partners should consistently and correctly use condoms during sex (i.e., vaginal intercourse, anal intercourse, and oral sex) or abstain from sex for the duration of the pregnancy [22; 52; 55]. The purpose is to avoid even a minimal risk of sexual transmission, given the potential for adverse fetal effects when ZIKV is contracted during pregnancy. Pregnant women should discuss their male sex partner's history of travel to areas with active ZIKV transmission and any history of illness consistent with ZIKV disease with their healthcare provider.

Men with known (or presumed) ZIKV disease and their non-pregnant sex partners who want to reduce the risk for sexual transmission of ZIKV are advised to use condoms consistently and correctly during sex or to abstain from sex [55]. The recommended duration of consistent condom use or abstinence depends on whether the man had confirmed infection or clinical illness consistent with ZIKV disease and whether he is residing in an area with ongoing transmission. In weighing the level of risk and a couple's concern about sexual transmission of ZIKV, several factors should be considered [22]. The risk for acquiring mosquito-borne ZIKV infection depends on the duration and extent of exposure to infected mosquitoes and the steps taken to prevent mosquito bites. Viral transmission is of particular concern

during pregnancy; therefore, a couple's resolve and strategy for prevention of unintended pregnancy should be taken into account, including use of the most effective contraceptive methods. The CDC interim guidance now recommends [52; 55]:

- Men with known infection or possible ZIKV exposure who are planning to conceive with their partner should wait at least three months after symptom onset or last possible exposure before engaging in unprotected sex.
- For couples who are not trying to conceive, men should consider using condoms or abstaining from sex for at least three months after symptom onset or last exposure to minimize their risk for sexual transmission of ZIKV.
- Men with ZIKV disease or possible ZIKV exposure whose partner is pregnant should consistently use condoms during sex or abstain from sex for the duration of the pregnancy.
- If a couple has a female partner and only she travels to an area with risk of ZIKV disease, the couple should consider using condoms or not having sex for at least two months after the female partner returns (even if she is asymptomatic) or from the start of the female partner's symptoms or date of diagnosis.

The longer precautionary period for men is because ZIKV persists longer in semen than in other body fluids, including vaginal secretions, urine, and blood.

ZIKV VACCINE

Because of the burden imposed by complications of ZIKV infection, especially in women of childbearing age, development of a ZIKV vaccine seems a compelling strategy for prevention. Vaccines have been developed against other arboviruses, including dengue and West Nile viruses, and a DNA vaccine has been shown to protect against ZIKV in a mouse model studied in Brazil [46]. However, virologists anticipate it would likely take several years to bring a ZIKV vaccine to implementation, in part because the immunology of flavivirus infection poses potential barriers to a safe, predictable strategy, particularly in endemic areas where exposure to multiple flavivirus infections is common. Just as cross-reactivity of antibodies elicited by related flaviviruses confounds diagnosis, so may antibody cross-reactivity impact vaccine efficacy and safety [47]. Among the issues to be considered is the phenomenon of immune enhancement, whereby immunologic memory acquired from earlier flavivirus infection (or vaccination) may, in response to a new flavivirus infection, lead to an IgG antibody/Fc receptor/lymphocyte-mediated augmentation of infection with prolonged viremia, possibly increasing severity and risk for complications. As an example, one consideration could be whether a ZIKV immunization campaign in an area endemic for dengue virus infection places the population at greater risk for more serious illness from dengue virus infection (i.e., hemorrhagic fever and shock). Although a ZIKV vaccine could hold greater promise for population groups outside areas endemic for other flaviviruses, its implementation (after years of development) would need to take into account the durable effect of mosquito control programs and the lingering scope of ZIKV circulation.

VECTOR CONTROL

Strategies employed for prevention of the transmission of ZIKV and other mosquito-borne flaviviruses are directed toward elimination or control of mosquito vectors and interruption of human-mosquito contact. At present, *A. aegypti* mosquito is the principal target of these efforts. Adult *A. aegypti* females feed on humans, rarely travel more than 100 yards from where they hatched, and live for about 10 days. They proliferate in standing rainwater or any of the many nearby stagnant sources associated with urban habitat, such as blocked gutters, bird baths, flowerpots, abandoned food and beverage containers, and construction sites. Control activities are aimed at eliminating immature and adult mosquito habitats in and around homes, out buildings, workplaces, schools, and other venues where people gather.

The CDC recommends an integrated mosquito management plan that employs a combination of methods based on mosquito biology, life cycle, and behavior [37]. An important feature of this approach is that all stakeholders become involved: health department and mosquito control specialists, government agencies, and community citizens and neighbors. The five key elements of this approach are [37]:

- Conduct mosquito surveillance: Monitor places where eggs are laid and young mosquitoes are found; track populations and the virus they are carrying; and determine which insecticide will be effective.
- Remove places where mosquitoes lay eggs: Target public places, parks, roadside dumps, yards, and neighborhoods, including weekly removal of common sources of standing water.
- Control young mosquitoes (larvae and pupae): Dump or remove standing water; use EPA-registered larvicides to treat water-holding structures and containers.
- Control adult mosquitoes: When surveillance shows that adult mosquito populations are increasing or spreading virus, professionals may apply EPA adulticides to reduce the number of mosquitoes in the area, delivered by backpack sprayers, trucks, or airplanes.
- Monitor control programs: Conduct additional studies to assess the effectiveness of mosquito control efforts.

NOVEL APPROACHES TO VECTOR CONTROL

Two novel approaches that have shown considerable promise are the genetic control of *A. aegypti* mosquitoes and the development of mosquitoes that are resistant to arbovirus infection. The first field-trialed genetic control strategy is known as the Release of Insects carrying Dominant Lethal (RIDL) genes and involves the mass rearing of *A. aegypti* that have been genetically modified to express a repressible lethal gene [38]. During the rearing period in insectaries, mosquitoes are provided with a dietary supplement not present in nature (tetracycline), and this supplement represses the lethal gene activation [39].

Only male mosquitoes are released, and these compete with wild males to mate with wild females. Offspring do not survive to the adult stage because they do not receive the dietary additive in the wild. Lines of RIDL males have been shown to have minimal fitness costs (i.e., they are competitive with wild males) and a field release in Bahia, Brazil, reportedly achieved a 95% reduction in local mosquito populations [41].

An alternative approach is to infect the vector with endosymbiotic bacteria, which interrupt arbovirus replication within mosquitos. The Eliminate Dengue project has demonstrated that *Wolbachia* bacteria from *Drosophila* fruit flies can prevent dengue virus transmission in *A. aegypti* mosquitoes without significant fitness costs [40]. *Wolbachia* has also been shown to inhibit the replication of additional arboviruses, such as chikungunya virus and yellow fever virus, strongly suggesting potential inhibitory effects against ZIKV [41].

Whereas RIDL is a self-limiting approach (i.e., the genetic modification is not perpetuated in wild mosquito populations), *Wolbachia*-based control strategies rely on this endosymbiont successfully invading wild mosquito populations through a reproductive phenotype known as cytoplasmic incompatibility. This results in the generation of nonviable offspring when an uninfected female mates with a *Wolbachia*-infected male. By contrast, *Wolbachia*-infected females produce viable progeny when they mate with either infected or uninfected males, a reproductive advantage over uninfected females. Release of *Wolbachia*-infected *A. aegypti* mosquitoes produced a successful invasion of wild mosquito populations in Australia, and releases are ongoing in dengue virus-endemic countries such as Indonesia, Vietnam, and Brazil [41].

What effect either RIDL or *Wolbachia* will have on arboviral transmission and epidemiology remains to be determined. Mathematical models of dengue virus transmission based on the dynamics of viral infection in humans and mosquitoes predict that one strain of *Wolbachia* (wMel) would reduce the basic reproduction number of dengue virus transmission by 70% [44]. Models of dengue virus transmission control with RIDL also predict efficacy in reducing disease burden. These projections suggest that such strategies could have a direct impact on transmission of ZIKV and other arboviruses in countries such as Brazil, where *A. aegypti* is the principal vector [41].

An important benefit of these environmentally friendly, species-specific approaches is the reduced dependence they pose for insecticides—an increasingly important feature of future disease vector control. Suppression of the mosquito population, or rendering it arbovirus-resistant, holds great potential in the simultaneous control of Zika, dengue, chikungunya, and yellow fever viruses. One hundred fifty countries presently have *A. aegypti* and are vulnerable to future outbreaks. The costs of implementing these novel technologies in Brazil and across the tropics should be considered in the context of the multifaceted benefits they pose in controlling several emerging infectious diseases.

CONSIDERATIONS FOR NON-ENGLISH-PROFICIENT PATIENTS

For patients who are not proficient in English, it is important to provide information regarding prevention strategies, testing recommendations, and signs and symptoms of ZIKV disease in their native language, if possible. The CDC provides patient education posters and handouts in a variety of languages, including Spanish, Portuguese, French, and Vietnamese [45]. Copies of these materials may be accessed at https://www.cdc.gov/zika/fs-posters. When there is an obvious disconnect in the communication process between the practitioner and patient due to the patient's lack of proficiency in the English language, an interpreter is required. Interpreters can be a valuable resource to help bridge the communication and cultural gap between patients and practitioners. Interpreters are more than passive agents who translate and transmit information back and forth from party to party. When they are enlisted and treated as part of the interdisciplinary clinical team, they serve as cultural brokers who ultimately enhance the clinical encounter. In any case in which information regarding treatment options and medication/treatment measures are being provided, the use of an interpreter should be considered.

SUMMARY

In 2013–2016, mosquito-borne ZIKV disease outbreaks swept through the Americas, leaving in their wake thousands of cases of microcephaly and higher rates of post-infectious Guillain-Barré syndrome. Sexual transmission of ZIKV has been identified; maternofetal transmission during pregnancy is now known to cause congenital infection and adverse fetal outcomes. The CDC has determined that evidence supports a causal relationship between prenatal ZIKV infection and microcephaly and other congenital neurologic anomalies. Health agencies have established disease surveillance networks and provided guidance for healthcare providers, travelers, and expectant couples on the best means for avoiding mosquito contact and preventing sexual transmission. More information is needed on the absolute risk and spectrum of fetal outcomes following maternal ZIKV infection during pregnancy, the clinical course and prognosis of infants born with congenital ZIKV syndrome, and the most effective means for vector control and prevention of ZIKV infection.

Implicit Bias in Health Care

The role of implicit biases on healthcare outcomes has become a concern, as there is some evidence that implicit biases contribute to health disparities, professionals' attitudes toward and interactions with patients, quality of care, diagnoses, and treatment decisions. This may produce differences in help-seeking, diagnoses, and ultimately treatments and interventions. Implicit biases may also unwittingly produce professional behaviors, attitudes, and interactions that reduce patients' trust and comfort with their provider, leading to earlier termination of visits and/or reduced adherence and follow-up. Disadvantaged groups are marginalized in the healthcare system and vulnerable on multiple levels; health professionals' implicit biases can further exacerbate these existing disadvantages.

Interventions or strategies designed to reduce implicit bias may be categorized as change-based or control-based. Change-based interventions focus on reducing or changing cognitive associations underlying implicit biases. These interventions might include challenging stereotypes. Conversely, control-based interventions involve reducing the effects of the implicit bias on the individual's behaviors. These strategies include increasing awareness of biased thoughts and responses. The two types of interventions are not mutually exclusive and may be used synergistically.

Works Cited

1. Hayes EB. Zika virus outside Africa. *Emerg Infect Dis*. 2009;15(9):1347-1350.
2. Fauci AS, Morens DM. Zika virus in the Americas: yet another arbovirus threat. *N Engl J Med*. 2016;374(7):601-604.
3. Gubler DJ. The global emergence/resurgence of arboviral diseases as public health problems. *Arch Med Res*. 2002;33(4):330-342.
4. Petersen LR, Jamieson DJ, Powers AM, Honein MA. Zika virus. *N Engl J Med*. 2016;374(16):1552-1563.
5. Hennessey M, Fischer M, Staples JE. Zika virus spreads to new areas: region of the Americas, May 2015–January 2016. *MMWR*. 2016;65(3):55-58.
6. World Health Organization. Zika Virus Disease Outbreak 2015–2016. Available at https://www.who.int/emergencies/situations/zika-virus-outbreak. Last accessed July 1, 2022.
7. Schuler-Faccini L, Ribeiro EM, Feitosa IM, et al. Possible association between Zika virus infection and microcephaly—Brazil, 2015. *MMWR*. 2016;65(3):59-62.
8. Centers for Disease Control and Prevention. Zika Virus. Available at https://www.cdc.gov/zika/reporting/index.html. Last accessed July 1, 2022.
9. Monaghan AJ, Morin CW, Steinhoff DF, et al. On the seasonal occurrence and abundance of the Zika virus vector mosquito *Aedes aegypti* in the contiguous United States. *PLoS Curr*. 2016;1.
10. Kuno G, Chang GJ, Tsuchiva KR, Karabatsos N, Cropp CB. Phylogeny of the genus Flavivirus. *J Virol*. 1998;72(1):73-83.
11. Duffy MR, Chen TH, Hancock WT, et al. Zika virus outbreak on Yap Island, Federated States of Micronesia. *N Engl J Med*. 2009;360(24):2536-2543.
12. European Centre for Disease Prevention and Control. Rapid Risk Assessment: Zika Virus Infection Outbreak, French Polynesia. Available at https://ecdc.europa.eu/sites/portal/files/media/en/publications/Publications/Zika-virus-French-Polynesia-rapid-risk-assessment.pdf. Last accessed July 1, 2022.
13. Mlakar J, Korva M, Tul N, et al. Zika virus associated with microcephaly. *N Engl J Med*. 2016;374(10):951-958.
14. Centers for Disease Control and Prevention. CDC Concludes Zika Causes Microcephaly and Other Birth Defects. Available at https://www.cdc.gov/media/releases/2016/s0413-zika-microcephaly.html. Last accessed July 1, 2022.
15. Broutet N, Krauer F, Riesen M, et al. Zika virus as a cause of neurologic disorders. *N Engl J Med*. 2016;374(16):1506-1509.
16. Hills SL, Russell K, Hennessey M, et al. Transmission of Zika virus through sexual contact with travelers to areas of ongoing transmission—continental United States, 2016. *MMWR*. 2016;65(8):215-216.
17. European Centre for Disease Prevention and Control. Rapid Risk Assessment: Zika Virus Epidemic in the Americas: Potential Association with Microcephaly and Guillain-Barré Syndrome. Available at https://ecdc.europa.eu/sites/portal/files/media/en/publications/Publications/zika-virus-americas-association-with-microcephaly-rapid-risk-assessment.pdf. Last accessed July 1, 2022.
18. Chouin-Carneiro T, Vega-Rua A, Vazeille M, et al. Differential susceptibilities of *Aedes aegypti* and *Aedes albopictus* from the Americas to Zika virus. *PLoS Negl Trop Dis*. 2016;10(3):e0004543.
19. Paz S, Semenza JC. El Niño and climate change-contributing factors in the dispersal of Zika virus in the Americas? *Lancet*. 2016;387(10020):745.
20. Oster AM, Brooks JT, Stryker JE, et al. Interim guidance for prevention of sexual transmission of Zika virus—United States, 2016. *MMWR*. 2016;65(5):120-121.
21. D'Ortenzio E, Matheron S, de Lamballerie X. Evidence of sexual transmission of Zika virus. *N Engl J Med*. 2016;374(22):2195-2198.
22. Oster AM, Russell K, Stryker JE, et al. Update: interim guidance for prevention of sexual transmission of Zika virus—United States, 2016. *MMWR*. 2016;65(12):323-325.
23. Russell PK. The Zika pandemic: a perfect storm? *PLoS Negl Trop Dis*. 2016;10(3):e0004589.
24. Simpson DI. Zika virus infection in man. *Trans R Soc Trop Med Hyg*. 1964;58(4):335-338.
25. Brasil P, Pereira JP Jr, Raja Gabaglia C, et al. Zika virus infection in pregnant women in Rio de Janeiro: preliminary report. *Obstet Gynecol Surv*. 2016;71(6):331-333.
26. Carteaux G, Maquart M, Bedet A, et al. Zika virus associated with meningoencephalitis. *N Engl J Med*. 2016;374(16):1595-1596.
27. Karimi O, Goorhuis A, Schinkel J, et al. Thrombocytopenia and subcutaneous bleedings in a patient with Zika virus infection. *Lancet*. 2016;387(10022):939-940.
28. França GVA, Schuler-Faccini L, Oliveira WK, et al. Congenital Zika virus syndrome in Brazil: a case series of the first 1501 livebirths with complete investigation. *Lancet*. 2016;388(10047):891-897.
29. Rasmussen SA, Jamieson DJ, Honein MA, Petersen LR. Zika virus and birth defects: reviewing the evidence for causality. *N Engl J Med*. 2016;374(20):1981-1987.
30. Mlakar J, Korva M, Tul N, et al. Zika virus associated with microcephaly. *N Engl J Med*. 2016;374(10):951-958.
31. Centers for Disease Control and Prevention. Testing for Zika Virus Infection. Available at https://www.cdc.gov/zika/laboratories/types-of-tests.html. Last accessed July 1, 2022.

32. Rabe IB, Staples JE, Villanueva J, et al. Interim guidance for interpretation of Zika virus antibody test results. *MMWR*. 2016;65(21):543-546.
33. Staples JE, Dziuban EJ, Fischer M, et al. Interim guidelines for the evaluation and testing of infants with possible congenital Zika virus infection—United States, 2016. *MMWR*. 2016;65(3):63-67.
34. Centers for Disease Control and Prevention. Zika Virus Disease and Zika Virus Infection 2016 Case Definition, Approved June 2016. Available at https://ndc.services.cdc.gov/case-definitions/zika-virus-disease-and-zika-virus-infection-2016-06-01. Last accessed July 1, 2022.
35. Oduyebo T, Igbinosa I, Petersen EE, et al. Update: interim guidance for health care providers caring for pregnant women with possible Zika virus exposure—United States, July 2016. *MMWR*. 2016;65(29):739-744.
36. Centers for Disease Control and Prevention. Zika Virus: Prevent Mosquito Bites. Available at https://www.cdc.gov/zika/prevention/prevent-mosquito-bites.html. Last accessed July 1, 2022.
37. Centers for Disease Control and Prevention. What Mosquito Control Programs Do. Available at https://www.cdc.gov/mosquitoes/mosquito-control/community/what-mosquito-control-programs-do.html. Last accessed July 1, 2022.
38. Centers for Disease Control and Prevention. Chikungunya Virus: Transmission. Available at https://www.cdc.gov/chikungunya/transmission/index.html. Last accessed July 1, 2022.
39. Brito C. Zika virus: a new chapter in the history of medicine. *Acta Med Port*. 2015;28(6):679-680.
40. Centers for Disease Control and Prevention. Chikungunya Virus: Clinical Evaluation and Disease. Available at https://www.cdc.gov/chikungunya/hc/clinicalevaluation.html. Last accessed July 1, 2022.
41. Yakob L, Walker T. Zika virus outbreak in the Americas: the need for novel mosquito methods. *Lancet Glob Health*. 2016;4(3):e148-e149.
42. World Health Organization. Zika Situation Report. Available at https://apps.who.int/iris/bitstream/handle/10665/204348/zikasitrep_5Feb2016_eng.pdf. Last accessed July 1, 2022.
43. Centers for Disease Control and Prevention. U.S. Zika Pregnancy and Infant Registry. Available at https://www.cdc.gov/pregnancy/zika/research/registry.html. Last accessed July 10, 2019.
44. Ferguson NM, Kien DT, Clapham H, et al. Modeling the impact on virus transmission of Wolbachia-mediated blocking of dengue virus infection of *Aedes aegypti*. *Sci Transl Med*. 2015;7(279):279ra37.
45. World Health Organization. Zika Virus Disease: Case Definition. Available at https://www3.paho.org/hq/index.php?option=com_content&view=article&id=11117:zika-resources-case-definitions&Itemid=41532&lang=en. Last accessed July 1, 2022.
46. Larocca RA, Abbbink P, Peron JP, et al. Vaccine protection against Zika virus from Brazil. *Nature*. 2016;536(7617):474-478.
47. Marins KAO, Dye JM, Bavan S. Considerations for the development of Zika virus vaccines. *Vaccine*. 2016;34:3711-3712.
48. U.S. Food and Drug Administration. FDA Advises Testing for Zika Virus in All Donated Blood and Blood Components in the US. Available at https://www.fda.gov/news-events/press-announcements/fda-advises-testing-zika-virus-all-donated-blood-and-blood-components-us. Last accessed July 1, 2022.
49. Centers for Disease Control and Prevention. Zika Travel Information. Available at https://wwwnc.cdc.gov/travel/page/zika-information. Last accessed July 1, 2022.
50. Frieden TR, Schuchat A, Petersen LR. Zika virus 6 months later. *JAMA*. 2016;316(14):1443-1444.
51. Mead PS, Duggal NK, Hook SA, et al. Zika virus shedding in semen of symptomatic infected men. *N Engl J Med*. 2018;378:1377-1385.
52. Polen KD, Gilboa SM, Hills S, et al. Update: interim guidance for preconception counseling and prevention of sexual transmission of Zika virus for men with possible Zika virus exposure—United States, August 2018. *MMWR*. 2018;67:868-871.
53. Davidson A, Slavinski S, Komoto K, Rakeman J, Weiss D. Suspected female-to-male sexual transmission of Zika virus—New York City, 2016. *MMWR*. 2016;65:716-717.
54. Paz-Baily G, Rosenberg ES, Doyle K, et al. Persistence of Zika virus in body fluids: final report. *NEJM*. 2018;379:1234-1243.
55. Centers for Disease Control and Prevention. Clinical Guidance for Healthcare Providers for Prevention of Sexual Transmission of Zika Virus. Available at https://www.cdc.gov/zika/hc-providers/clinical-guidance/sexualtransmission.html. Last accessed July 1, 2022.
56. Oduyebo T, Polen KD, Walke HT, et al. Update: interim guidance for health care providers caring for pregnant women with possible Zika virus exposure—United States (including U.S. territories), July 2017. *MMWR*. 2017;66:781-793.
57. Musso D, Ko AI, Baud D. Zika virus infection: after the pandemic. *N Engl J Med*. 2019;381:1444-1457.
58. Pomar L, Vouga M, Lambert V, et al. Maternal-fetal transmission and adverse perinatal outcomes in pregnant women infected with Zika virus: prospective cohort study in French Guiana. *BMJ*. 2018;363:k4431.
59. Leonhard SE, Lant S, Jacobs BC, et al. Zika virus infection in the returning traveler: what every neurologist should know. *Practical Neurology*. 2018;18:271-277.
60. Pan American Health Organization. Zika Cumulative Cases: January 2018. Available at https://www3.paho.org/hq/index.php?option=com_content&view=article&id=12390:zika-cumulative-cases&Itemid=42090&lang=en. Last accessed July 1, 2022.

61. Roth NM, Reynolds MR, Lewis EL, et al. Zika-associated birth defects reported in pregnancies with laboratory evidence of confirmed or possible Zika virus infection—U.S. Zika Pregnancy and Infant Registry, December 1, 2015–March 31, 2018. January 2022. *MMWR*. 2022;71:73-89.

62. Honein MA, Woodworth KR, Gregory CJ. Neurodevelopmental abnormalities associated with in utero Zika virus infection in infants and children: the unfolding story. *JAMA Pediatr* 2020;174:237-238.

63. Centers for Disease Control and Prevention. Testing for Zika, NEW Zika and Dengue Testing Guidance (Updated November 2019). Available at https://www.cdc.gov/zika/hc-providers/testing-guidance.html. Last accessed July 1, 2022.

64. Centers for Disease Control and Prevention. Collecting and Submitting Specimens at Time of Birth for Zika Virus Testing. Available at https://www.cdc.gov/zika/hc-providers/test-specimens-at-time-of-birth.html. Last accessed July 1, 2022.