CHAPTER ONE

Chikungunya Fever

**1.0 Introduction**

Chikungunya fever is an *Aedes* mosquito-borne arboviral disease that pose global public health threat (1). The virus was first discovered in 1952 on the Makonde plateau in Tanzania. The word *Chikungunya* is derived from Makonde verb, *Kungunyala* to mean that which bends up.Chikungunya virus (CHIKV) infection presents as an acute febrile illness with rash and a chronic phase characterized by severe joint and skeletal muscle pains which may persist for Months or even years (2,3). Other clinical manifestations include fever, rash, headache, muscle pain and fatigue which are like dengue symptoms leading to misdiagnosis and underreporting in the absence of specific laboratory diagnostic testing (2). Chikungunya fever induced arthropathy (disease of the joints) has a considerable effect on the quality of life of individuals with chronic disease and results in economic losses especially in developing countries (1).

The virus is transmitted to humans by bites of infected female mosquitoes primarily by *Aedes aegypti* and *Aedes Albopictus* mosquitoes (4). These species also transmit other viruses including dengue, Rift valley fever and Zika with occasional co-infections reported (2). They primarily bite during daylight hours. The virus belongs the genus alphavirus in the *Togaviradae* family (2). It is a single-stranded RNA virus which was first isolated in 1953 in Tanzania (5). In Africa, virus is maintained during inter-epidemic periods in a complex sylvatic cycle-a cyclical transmission between non-human primates and several mosquito species Aedes (Ae) *aegypti*, Ae. *africanus*, Ae. *luteocephalus* and Ae. *furcifer-taylori* with occasional transmission to humans(6–8). In Asia, the virus is maintained in cycles between aedes aegypti or aedes albopictus and humans (8).

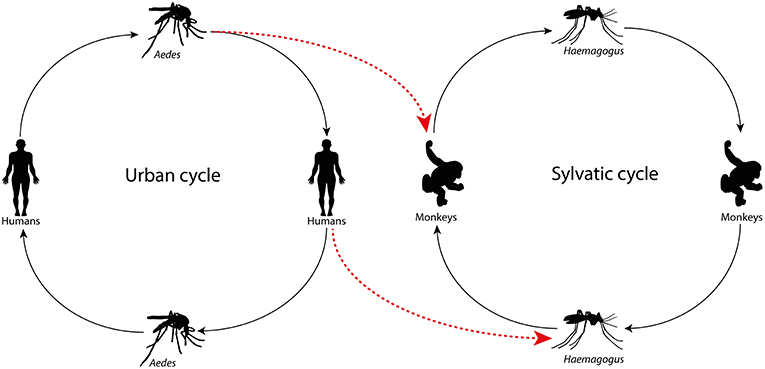


Figure 1: Sylvatic transmission cycle of CHIKV

Between 1960s and 1990s, CHIKV outbreaks were mostly reported in Africa and Asia. Today, CHIKV has been reported in more than 110 countries in East and Central Africa, South and North America, Southeast Asia and Europe because of climate change, increased globalization of commerce and travel which has led to the growth of habitat of Aedes mosquitoes (4,6) .

The virus has spread globally causing a substantial morbidity and socioeconomic burden (1). About 18.7 million Chikungunya virus cases have been reported causing 1.95 million disability adjusted life years (DALYs) between 2011-2020 (1). The CHIKV has been recognized to cause a pandemic, and the virus has been listed by Coalition for Epidemic Preparedness Innovations (CEPI) as a priority pathogen for vaccine development (6). There is no specific antiviral treatment against CHIKV infection, however, several preventive vaccines in different stages of clinical trial development with two vaccines IXCHIQ and VIMKUNYA receiving licensure for use.

**1.2 History of Chikungunya Virus (E. Carey)**

Chikungunya has undergone a remarkable name change because of its similarities between the clinical syndromes caused by dengue and chikungunya virus (7). Between 1961- 1966, E. Carey had been involved in studies of endemic dengue at Vellore, South India and in 1964, a massive wave of CHIKV infection swept through madras state in India. Patients presented certain clinical features that were distinct from the usual classical dengue fever. Carey reviewed previous reports of fever epidemics in India and found a paper dealing with 1923 dengue outbreak in Calcutta. The authors were describing not the epidemic of dengue rather an epidemic similar in all respect associated with CHIKV in 1964 in South India. This discovery let to more intensive research for earlier articles on dengue and acute fever in India and elsewhere. The discovery provided a basis to believe that the term dengue applied in the early 19th century to a clinical syndrome closely related to CHIKV infection and the present-day dengue corresponds to the breakbone fever described by Dr. Rush in 1789.

David Bylon observed an epidemic in Batavia in 1779 and was credited for first published description of dengue. He termed the disease as *knokkle-koorts*, or joint pain and high fever. Around the same time, another outbreak happened in Cairo which was known as *knee trouble* and was characterized by fever which lasted for three days after which the illness increased or diminished according to the disposition of the individual. Fever was accompanied by pains in the joints, knees and inability to move with swelling of the fingers. These two accounts described illness compatible with the CHIKV infection.

The first true description of the clinical features for which the term dengue is presently applied was given by Benjamin Rush who wrote of an epidemic of breakbone fever that was common in Philadelphia from august to September 1780. The outbreak was characterized with severe fever and extremely severe pain in the head, back and limps. The pain in the head sometimes occupied the eyeballs and sometimes affected the neck and arms. Nausea was common and some cases vomiting. Rash appeared on day 3 to day 4. No mention of the joint pain.

In 1824-1825, an enormous epidemic of acute, self-limiting febrile illness was reported in India. James Mellis published Indian epidemic in 1825. He described a classic style epidemic in Calcutta between June to August 1824. The epidemic attacked everyone in Calcutta and later spread to madras and Gujarat on the west. Patients experienced pains in the head and different parts of the body, muscle and joint pains especially in the fingers and toes. Sequalae characterized by continued pain in several joints was also observed. The disease died out in 1825 and reappeared again in India in 1871.

In 1827 – 1828, the disease appeared in west indies. The outbreak was observed on St. Thomas and Santa Cruz which Stedman described as those outbreaks of Indian. The outbreak attacked almost everyone. Sudden onset, joint pains, rash and low mortality. Fever, headache and back pain were observed which abated in 36 hours. Persistence of the pains in the joints for weeks after recovery from acute stage. In St. Thomas, the disease was known as *dandy fever* for English speaking and bouquet for French. Stedman noted that the disease was distinguishable from the one described by Benjamin Rush as breakbone fever. He proposed a name as eruptive articular or rheumatic fever in 1829. His argument was supported by ruan that indeed the west Indian disease was different from that described by Dr. Rush. In the same year, Nicholson published a short letter on an arthritic exanthem which appeared in Antigua in January 1828. His description correlated well with Stedman’s including a more emphasize on the persistent arthritic pains. Still in 1828, the epidemic spread to north America basically in US and Dumaresq described the disease which became very general more than any other previously experienced. He further alluded that this disease could have been brought from Africa with some slave vessels imported into the Havanna. Here, the disease was named as dingee, dengue, danga. The disease was characterized by severe joint pains.

Between 1826 – 1827, cases of breakbone fever like those described by Dr. Rush were seen in Charleston, South Carolina and savannah and in 1828, dengue appeared. Waring contrasted the breakbone fever of 1826 – 1827 in savanna with dengue of 1828 and they lacked resemblance particularly in longer continuance of pain in dengue than in breakbone, more exclusive confinement of pain in joints in dengue than in breakbone, less debility and less tediousness of convalescence. He added that breakbone fever made its invasion by the usual symptoms of fever and pain coming gradually. A remarkable difference was excessive debility in breakbone fever and whereas dengue attacked once, he observed two attacks of breakbone fever in the same individual.

In 1830, Cook described an epidemic in St. Bartholomew from November 1827 - January 1828. The disease was characterized by a sudden onset of pains in the joints of the fingers and toes with fever lasting one to two days and followed with rash. Joint pains remained for several weeks and sometimes months. The epidemic reached Cuba in 1828 and was referred to as dengue which was mainly characterized by persistent pains and fever.

Between 1871-1872, another epidemic fever struck Calcutta. At this time, the term dengue had been adopted by the royal college of physicians and was in use. Patients presented with sudden onset of severe pain of the rheumatic character and more articular than muscular. There was an eruption and convalescence marked by persistence of joint pain. Another dengue eruption appeared around June 1872 characterized by joint pains, 3 to 4-day course, rash and residual arthralgia. In July the same year, editorial mentioned presence of dengue along the western coast of Africa that occurred between July – September 1871. After affecting about 75% of the population, the epidemic abated with onset of cool weather. Nothing but time and patience effected the cure.

In 1881, James Christie published an important epidemiologic account of dengue in Zanzibar in 1870. He described the clinical manifestation of the outbreak characterized by the severity of articular pains like the outbreak that occurred in India. The older inhabitants recognized the disease as one which had been epidemic 48 – 49 years before which was formerly known as *ki-dinga pepo* in Swahili to mean a disease characterized by a sudden cramp-like seizure caused by evil spirits.

From its origin on Zanzibar in 1823, christie believed the disease transferred to India aboard one of the native craft that were constantly passing the African coast and Bombay. He then speculated that a slave vessel had carried infection from east coast of Africa to the west indies where it appeared in 1827. Zanzibar also seemed to be the likely origin of the 1871- 1872 epidemic in India which spread to Aden, Calcutta and Burma.

Between 1901 – 1902, an outbreak occurred in Hong Kong, Burma and India. The epidemic was characterized by pains which affected the small bones of the hand and the feet and persisted for weeks as observed in 1964 chikungunya outbreak in south Indian epidemic. The outbreak that appeared in Madras state presented with severe fever and according to L. Rogers, the disease was typical dengue though fever lasted for longer days and was named *seven-day fever*. The pains were confined to the muscles.

In spite of the early differentiation of the epidemic dengue and the endemic breakbone fever, emphasis was on the distinctiveness of the true dengue towards the end of 19th century and early 20th century, the name dengue was widely used to illness resembling Rush breakbone fever and Rogers seven-day fever. Around this time, the search for the causes of these fevers began.

In 1897 and 1905, epidemics occurred in Australia and was characterized by fever, headache, pain the back and a diphasic fever. It was emphasized that articular pains were not seen in both epidemics. The discomfort observed seemed to be muscular seated in the deep insertions of the muscles. Post febrile rheumatic stage was not observed.

In 1906, Bancroft successfully transmitted the disease to human volunteers via mosquitoes Aedes aegypti and in 1907, Ashburn and Craig working in Philippines reported transmission of dengue fever like one seen in Australia by means of the inoculation of human volunteers with plasma that had passed through a bacteria-tight filter. Both transmission of the disease by Aedes aegypti and ultramicroscopic nature of the agent were confirmed by Cleland and his co-workers in Australia and by siler, Hall and Hitches in Philippines. The former workers that joint pain had frequently been one of the most prominent features of the previous dengue. Ultimately, viruses were established in white laboratory mice and were labelled dengue viruses.

Once experimental transmission of the disease called dengue had been achieved, the clinical features of the experimental infection became the standards by which diagnoses were made. Only little more was heard of Rogers seven – day fever until 1923 when it appeared in India.

In 1923, Knowles and Das Gupta reported dengue to be epidemic in India in Bombay, Lucknow, Delhi and Calcutta. In 1952, an epidemic fever reappeared in east Africa in the southern Tanganyika, an outbreak indistinguishable from Dengue. The name given to this disease by the local people was *chikungunya* means that which bends up and several strains of new chikungunya virus were isolated from the sera of patients in 1953. The virus was found to be serologically quite distinct from the known dengue virus. Clinical description of the chikungunya virus was remarkably like the early accounts of epidemics of dengue. It was proposed based on the record accounts of acute illness characterized by fever, joint pains, rash and persistent arthralgia of all these epidemics were associated with chikungunya or closely related viruses.

**1.3 Chikungunya Virus Phylogeny**

Chikungunya virus is a mosquito-borne virus that belong to the alphavirus genus consisting mainly the arthropod-borne viruses (arboviruses) believed to have a marine origin (9). However, presence of chikungunya virus in sylvatic mosquitoes and nonhuman primates in Uganda and Tanzania suggests that the virus originated in central or east Africa (9). The CHIKV genome consists of a single stranded, positive sense RNA of approximately 11.8kb, encoding nine proteins which are divided into structural and non-structural proteins. The populations of RNA viruses exhibit large genetic variability from two mechanisms, mutation and recombination. Mutation occurs during replication by mis-introduction of nucleotides by error-prone RNA-dependent RNA polymerase that lack proof-reading capacity. Recombination occurs when co-circulating lineages co-infect a host, and mixed segments are incorporated into the descendent viruses. These two processes happen together and encoded in the 3’UTRs of different chikungunya lineages. The resulting variants have highly divergent 3’UTRs that vary in length. Distinctive CHIKV lineages also differ in their 3’UTRs architecture in the copy number and arrangement of the sequence repeats.

Historically, chikungunya virus circulated with four lineages with both enzootic and epidemic transmission cycle (9). The four distinct genotypes which were documented include West African, East-Central-South African (ECSA), Asian and Indian ocean corresponding to their respective geographical origin(3,5,6,8).

Though the origin and history of how CHIKV spreads inside Africa remains elusive (9). The ECSA lineage gave rise to the Indian ocean lineage and has been identified as the origin of recent outbreaks in Brazil and Haiti. The Asian lineage has been constrained in southeast Asia and associated with recent outbreaks Americas. The Indian Ocean lineage circulates in India and surrounding regions with associated outbreaks in south and southeastern Asia as well as Europe.

The ECSA lineage is further divided into two clades; ECSA1 which consist entirely of the ancestral CHIKV sequences and ECSA2 which contains sequences from the Central African Republic, Cameroon, Gabon and the Republic of Congo (6). Further analysis identified 29 temporally and geographically diverse strains from 16 countries in the Western Hemisphere (5). The Asian lineage which caused epidemics in the 1950s split into two clades, the Indian clade which likely went extinct and South-Eastern Asian clade (10). Though the analysis revealed minimal evolution compared to the emergent CHIKV strains. A comprehensive analysis of the historic strains suggests CHIKV originated in Africa and spread episodically to Asia in approximated interval of 50 years (5).

**1.4 Chikungunya Clinical Manifestation**

Chikungunya disease is an acute febrile illness characterized by sudden onset of fever, rash and incapacitating arthralgia (10). The disease may also cause long-lasting joint pain especially in older adults (10). Maternal infection close to delivery may facilitate vertical transmission, leading to severe disease (i.e. encephalopathy and long-term sequelae) (10). The virus is rarely fatal but associated with significant morbidity (11) . symptoms of acute disease include high fever (above 390C), myalgia, arthralgia, arthritis, stiffness, headache and rash, insomnia, and exhaustion post-viremia (12). After acute phase, chronic disease including persistent arthralgia and stiffness is very common. Approximately 50% of detected acute cases have been estimated to develop chronic joint pains that can last for many months (13). Recent studies have revealed other lesser-known clinical manifestations of CHIKV that can contribute to otherwise unrecognized burden including medium to long term neurologic symptoms and inflammatory ophthalmic complications like uveitis (14). Intrapartum CHIKV transmission can cause neonatal encephalitis and poor neurodevelopment outcomes. CHIKV infects multiple human tissues, causing febrile illness characterized by myalgia, polyarthralgia, polyarthritis and rashes (9). The virus had not been associated with life-threatening symptoms before the La Reunion Island outbreak of 2005 that demonstrated a drastic expansion of the disease spectrum. In La Reunion Island and after, higher morbidity has been associated with CHIKV infection including neurological issues such as visual and hearing loss, paralysis, Guillain Barre syndrome and renal complications (9). Approximately 50% of detected acute cases have been estimated to develop chronic joint pain that can last for many months (13). One in 1,000 cases results in death mainly in neonates, infants and the elderly.

**1.5 Risk factors for Chikungunya**

The changing climate**,** urbanization, rapid pace of globalization and remarkable resilience of aedes mosquitoes may drive the spread of habitats for aedes aegypti and aedes albopictus even further (15). There are two major factors driving the global spread of viral vectors Ae. Aegypti and Ae. Albopictus: increasing global temperatures due to climate change and increasing human mobility between endemic and non-endemic countries (16). Mutations that change the protein structure of the virus have been attributed to increased ability to establish infection and outbreaks as the virus becomes better adapted to vector species (9). Due to a mutation in the E1 gene, chikungunya virus infectivity toward Ae. albopictus has increased substantially, resulting in the highly efficient transmission of the species id endemic (16).

1.6 **Chikungunya Epidemiology**

CHIKV is the most prevalent alphavirus transmitted to humans by a bite of infected female Aedes mosquito (6). The virus has caused multiple outbreaks in all continents except Antarctica with most recent in Europe, central and south America (9). With increased global distribution of Aedes mosquitoes and their ability to adapt to urban setting, urban transmission cycle is important (6). During a blood meal on an infected individual, the female mosquito ingests the virus which infects various tissues of the mosquito including the salivary glands. In the subsequent blood meal on humans by the infected mosquito, the virus is deposited in the skin of an uninfected person, infecting the person and perpetuating the viral replication cycle (6).

Since its discovery in 1952, and up until 2000s, a few outbreaks and sporadic cases of chikungunya virus were mainly reported in Sub-Saharan Africa and Southeast Asia (3,6,10,17). Although it occurred in the form of large and brutal epidemic affecting non-immune populations, it was classified as a mildly pathogenic arbovirus and was rated as emerging by the institute of medicine in 1992 (17). The situation changed abruptly when large outbreaks started in Lamu, Kenya in 2004 and 2005-2006 when a strain of CHIKV ravaged southwest Indian ocean islands and adapted rapidly to mosquitoes of Aedes albopictus through mutation in the envelope gene of the virus (10,17). The E1 A226V mutation is associated with increased replication capacity in this worldwide disseminated and invasive vector (10,17). Since 2005, the epidemics in the Indian ocean, India and southeast Asia have accounted for millions of cases locally and resulted in thousands of imported cases in Europe and Americans.

The 2007 Italian outbreak fuelled from a unique patient returning from Northern India during the viraemic phase of infection. Later the virus autochthonous transmission was demonstrated in southern France with two confirmed cases in September 2010 (17). In 2011, the virus spread to New Caledonia and later in the pacific following multiple imported cases (10).

In December 2013, CHIKV cases were reported for the first time in America linked to Asian lineage which caused large outbreaks in St. Martins and other Caribbean islands then spread to Latin America countries (10,17). Four months later, end of March 2014, 9 Caribbean islands were touched with more than 15,000 cases French west indies and first documented cases in south America in French Guyana. One month later, at the end of April 2014, 15 islands of the Caribbean claimed cases and the count reached 35,000 and six fatalities were reported (17). The virus caused sporadic cases in the southern Europe particularly in Italy in 2007 and 2017 and South of France in 2010 and 2014. It has been noted that the potential of worldwide spread of chikungunya is much higher than the risk of dissemination of MERS Coronavirus or Ebola virus, the total number of cases to be expected from introduction of chikungunya in the America, in Europe or in both is undoubtedly incommensurably higher (17).

Therefore, the attention and funding should be considered seriously to build-up and maintain an efficient surveillance system, organize for international coordination and information exchange in a timely manner and develop rapidly countermeasures as advocated by the American committee on arboviruses (17).

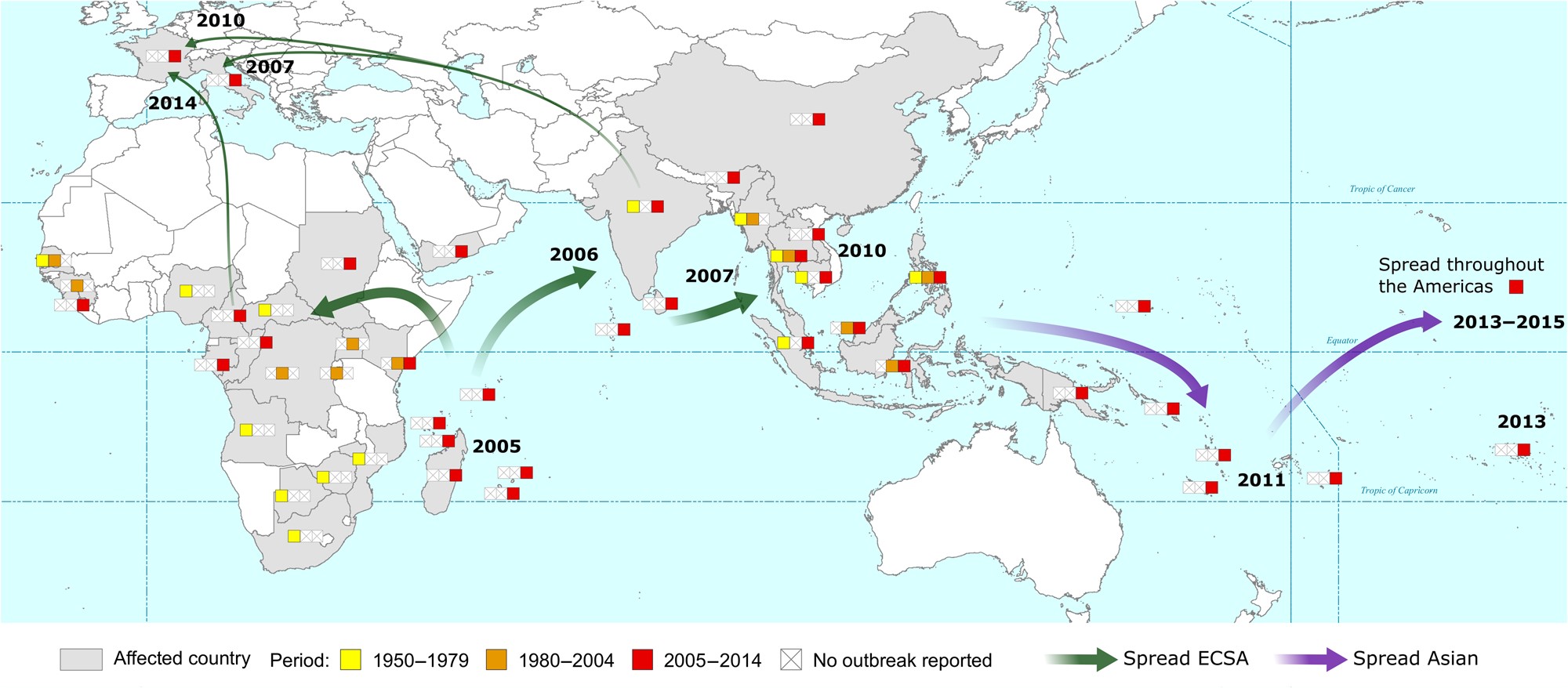


Figure 2: CHIKV geographical distribution. The spread of ECSA and Asian virus genotypes in Africa and Asia between 2005 – 2014

In many places, CHIKV transmission consists of outbreaks followed by periods without circulation (13). Endemic transmissions have also been reported in some parts (13)

**1.7 Chikungunya Epidemics in Africa**

After the first chikungunya outbreak was identified in July 1952 to March 1953 on the Makonde plateau in the Newala and Masisi districts in the southern province of former Tanganyika territory where chikungunya incidence of 23% was reported, several other chikungunya outbreaks have occurred in Africa (10).

The virus spread significantly during the years 1960-1980 causing outbreaks in many African countries: south Africa (1956; 1975 -77)(10), Zimbabwe (1957; 1961 - 62), Democratic republic of Congo (1958; 1960; 2018), Zambia (1959), Senegal (1960; 1997 – 98; 2006; 2015), Uganda (1961-62; 1968) Nigeria (1964; 1969; 1974), Angola (1970-71), Sierra Leone (1978), Central African Republic (1978 – 79) (10). After this period, there was no evidence of circulation of the virus until 1999-2000 when a large outbreak occurred in Kinshasa affecting about 50,000 people. Between 2002 – 2006, few cases of the virus were reported in Equatorial Guinea and in 2004, a large epidemic started on the Kenyan coast spreading to Comoros, La Reunion, Seychelles and Mauritius islands in Indian ocean.

In January 2006, a small outbreak of chikungunya was reported from north-east coastal Madagascar. After 2005, chikungunya transmissions have been reported in several African countries: Cameroon (2006), Gabon (2007-2010), large outbreak were reported in Brazzaville, Republic of Congo (2011, 2018). More recently, large outbreaks were reported in Mandera, Kenya (2016). Kenya experienced another outbreak along the coast in Mombasa (2017-18). In august 2018, a large outbreak was reported in Sudan and Ethiopia (2019).



Figure 3: geographical distribution of chikungunya virus in Africa

**Epidemics of virus disease in southern province of Tanganyika territory in 1952-53**

The epidemic was first recognized in 1952 by Dr. Marion C. Robinson of the Lulindi and Newala Hospitals of the universities mission to central Africa and was notified to the virus research institute by director of medical services, Tanganyika in February 1953 and a team was sent to attempt isolate the virus and carry out general epidemiological studies. On 21 February 1953, collection of serum samples from acute cases and biting insects began. The area covers the three plateau of Rondo, Makonde and mawia along the coast extreme south of Tanganyika. The epidemic was on the Makonde plateau, which slopes gradually to the sea and the plateau surface is 550 – 700m above the sea level. The epidemic was in Newala and Masasi districts, southern Tanganyika territory. The mean annual temperature at Newala was 37.36 in. (949mm.). The rainy season lasts from November – May and the other months are dry seasons. The yearly range of mean daily temperature is 6.70 C – 28.50C. The year 1952 was of unusually high rainfall, total precipitation for the year was 1203 mm.(47.37in.) nearly 27% above the 10-year average.

**South Africa 1956**

Early in April 1956, the attention was drawn to the occurrence of an outbreak of a dengue-like fever in the North-Eastern Transvaal in South Africa (18). The infection was contracted during visits to the bushveld area near Kruger national park in the vicinity of the Oliphant and Letaba river. The patients presented with sudden onset with acute pain in one or more joints. Temperature rose rapidly and often over 1030F on the day after the onset. During febrile period, some patients had a sore throat and headache, suffusion of the conjunctiva (18). The outstanding symptom was joint pain which came on with dramatic suddenness and was often sever. Prolonged convalescence with recurrent joint pains was also observed. It was apparent that these patients were suffering from an unusual illness, and the arrangements were made to identify the cause. Blood samples were collected and screened for various bacterial and viruses. Analysis of the serum sample of one of the patients, the virus differed from the virus of dengue. It was recalled that a dengue like illness had been prevalent in the Newala district of the southern province of Tanganyika in 1952-53 and the disease was called Chikungunya fever. It was considered as a possible virus causing the outbreak in the Eastern Transvaal(18). No documentation of the burden of disease.

**Lamu, Kenya Outbreak (2004)**

An outbreak of CHIKV disease associated with high fever and severe arthralgias was detected in Lamu, Kenya beginning May 2004 peaking in July the same year (11). A sample of 130 specimens were collected and sent to KEMRI for testing and 60 samples tested positive for chikungunya IgM antibodies (11). At least 1,300 cases were documented by the ministry of health meeting a clinical case definition of chikungunya illness (11). A cross-sectional seroprevalence study conducted in Lamu between October 5-9, 2004(9 weeks after the peak of the outbreak). The findings of the study suggested that the outbreak was widespread which affected 75% of the Lamu population. They showed that when those findings were to be extrapolated to the entire population, 13,500 people were affected (11). After this outbreak, other associated outbreaks occurred in Mombasa between November and December 2004. The virus then spread to the Indian ocean islands of Comoros from January to May 2005, reunion island March 2005 to July 2006 and other islands in Indian ocean and in India 2006 (11).

**Indian Ocean Outbreaks (2005-2006)**

An outbreak of CHIKV illness was reported on Comoros Islands in January 2005, which peaked in late March 2005 and lasted until May 2005; 5202 cases reported (19,20). Comoros is an archipelago of four islands and several islets located in the western Indian ocean. The four major islands are Ngazidja (Grande Comore), Mwali (Moheli), Nzwani (Anjouan), and Maore (Manyotte, under French administration). The outbreak predominantly occurred on Ngazidja; the youngest and largest of the islands and is closest to Africa (19). The seroprevalence study was conducted between March 18 - 26, 2005 to define the magnitude of the outbreak and characterize the clinical spectrum of infection with CHIKV (19). Out of 331 serum samples collected, 63% (209/331) samples tested positive for IgM or IgG antibodies to CHIKV. With an attack rate of 63% for CHIKV infection and estimated population of 341,000 people in Ngazidja, extrapolation of the serosurvey data suggest that 214,830 persons were infected during the outbreak (19).

The chikungunya outbreak arrived in La Reunion Island in March 2005 causing two waves (21). The first wave started in March 2005 causing approximately 6,000 cases until December (21). The second wave began in January and by July 2006, it was estimated that more than 266,000 persons were infected from a population of 785,221resulting in a cumulative incidence rate of 34% (21). For the first time deaths were reported during an outbreak. By December 2006, a total of 252 death certificates reported CHIKV infection as a direct or indirect cause of death

1.8 **Chikungunya outbreaks and Seroprevalence studies in Kenya**

The first outbreaks of CHIKV infection were recorded along the coast in Lamu and Mombasa in 2004 (11). The largest documented outbreak of CHIKV occurred in Indian ocean islands and India during 2004 – 2007 (22). The outbreak was associated with high fever and severe protracted arthralgias was detected in Lamu district beginning May and peaking in July 2004, at least1,300 cases were documented by the ministry of Health (11). Out of 130 specimens which were collected and sent to KEMRI for CHIKV antibody testing, 60 were positive of IgM antibodies (23). However, the extent to which CHIKV infection was unknown (23).

A seroprevalence study was conducted between October 5 – 9, 2004 (9 weeks after the peak of the outbreak) to define the magnitude of transmission on Lamu Island (23). Serum samples of 288 were collected from Lamu residents and tested for IgM and IgG antibodies to CHIKV. The participants selected randomly chosen in randomly selected households whether they were ill or not. The results showed that IgM antibodies were detected in 53/288 and IgG antibodies in 206/288 and IgM and/or IgG antibodies were present in 215/288 representing a 75%. If the results were to be extrapolated on the entire population of Lamu island then a total of 13,500(95% CI, 12,458-14328) were affected.

Another outbreak was reported in Mandera east subcounty on 28 May 2016. The outbreak was characterized by febrile illness and joint pains (24). In May 2016, samples were collected for laboratory test for arboviral infections. Out of 10 samples tested for chikungunya virus 7 samples tested positive for CHIKV, and all samples tested negative for dengue, yellow fever and West Nile viruses. As of June 2016, 1,792 cases had been tested positive for chikungunya virus (24). It was estimated that 80% of the population and 50% of the health work force in Mandera town were affected by chikungunya (24).

A human and entomologic investigation of chikungunya outbreak in Mandera was conducted (8). A total of 189 samples were received at the KEMRI viral haemorrhagic fever laboratory 126 samples from Kenya and 63 from Somalia to test the presence IgM antibodies against chikungunya and dengue. Out of 126 samples from Kenya, 113 were samples received from northeastern Kenya and the rest from other towns from Kenya. 100 samples tested positive for CHIKV by either IgM ELISA or RT-PCR. 55 samples tested positive for dengue virus. Out of 55 samples that were positive for dengue IgM antibodies, 16 were positive for chikungunya by RT-PCR. No dengue case was detected by RT-PCR. Sequence analysis of 13 isolates from Mandera associated with the outbreak revealed that the virus is closely related with China isolated in 2010.

A hospital based, cross—sectional survey study was conducted between 2010-2011 to determine the seroprevalence of arboviruses including yellow fever, dengue, chikungunya, and West Nile fever among children aged 1-12 years at two health facilities: Alupe subcounty hospital and KEMRI Alupe clinic. Results showed that a total of 182 out of 656 participants tested positive for any of the arbovirus. Out of these, 29/656 were positive for yellow fever virus, 62/649 were positive for West Nile virus, 36/649 were positive for chikungunya virus, 5/368 were positive for dengue virus1, 59/656 were positive for dengue virus2 and 40/203 were positive for dengue virus3. Neutralizing antibodies were found in 42/54 of the participants, 3/19 to yellow fever, 29/50 to dengue virus2 and 1/55 to West Nile virus. The study confirmed that children under the age of 12 years in Teso South-subcounty are exposed to ongoing arbovirus infections in early life (25). Data were published online

A cross-sectional seroprevalence study was conducted on febrile patients visiting Endebes district hospital, Andersen medical center and Kitale county referral hospital to determine the prevalence of IgA, IgM and IgG antibodies against CHIKV in Mt. Elgon region, Kenya (26). Serum samples were collected and screened using ELISA for IgA + IgG +IgM antibodies. Positive samples were subjected to plaque reduction neutralization assays using a standard PRNT. The results showed that a total of 317 out of 1359 tested positive for CHIKV antibodies. Out of these positive samples, 127 samples tested positive for CHIKV neutralizing antibodies. The findings suggest active circulation of CHIKV in Mt. Elgon though considered as a non-endemic region for the virus (26). Data were published online.

A seroprevalence study was conducted to determine the incidence of dengue virus, chikungunya virus, plasmodium and Leptospira infections among Kenyan children with acute undifferentiated febrile illness using molecular diagnostics. Children less than 18 years of age who presented with acute febrile illness (≤ 5 days duration) and no localizing signs or symptoms were enrolled at 4 study sites in coastal and western Kenya. Serum samples from 385 acute febrile illness children who presented to 1 of 4 clinical sites were tested using microscopy and real time molecular assays for DENV, CHIKV, malaria and leptospiral. Out of 254 patients, 158 tested positive for malaria. Similarly, out of 93 patients, 32 patients tested positive for CHIKV(27) .

**1.9 Seasonality of Chikungunya virus**

The chikungunya outbreaks recurrence in Africa and Asia is often preceded by long periods spanning several years or decades with minimal or no cases (28). Recurrence can be explained by several factors such as absence of neutralizing antibodies in younger age groups after a period of epidemiological silence (28). Additionally, recurrences of CHIK in west Africa have been attributed to enzootic CHIKV periodically causing spillover infections in people who enter or live near forests causing individual cases and small outbreaks. The periodicity of these spillover cases appears to be driven by changes in herd immunity among non-human primate enzootic hosts.

**1.10 The Global burden of Chikungunya**

A systematic review of published literature and surveillance records was conducted to estimate the global burden caused by CHIKV and Zika virus between 2010 – 2019 (14). DALY estimates for each disease were calculated according to standard methods outlined in the Global Burden of Diseases guidelines. It was estimated that chikungunya virus causes 106, 089(794 – 9,058,541) DALYs annually. They also report that 114 countries have experienced autochthonous transmission of chikungunya virus.

In another study to estimate the burden of chikungunya, serocatalytic models were fit to 49 age specific seroprevalence studies collected from 29 countries. Countries were classified as endemic or epidemic. Results showed that endemic locations had a mean annual probability of infection among the susceptible population was 0.024(95% CI 0.018 – 0.035) ranging from 0.0017 – 0.074 in all endemic locations. In epidemic setting, the mean annual probability of infection was 0.016 (95% CI 0.013-0.024) ranging from 0.0004 -0.065 across all locations. The mean percentage of the susceptible population that gets infected during outbreaks was 8.4% (CI 7.2 – 9.1%). Using the estimated FOI, globally there are 35,300,000 infections per year (95% CI 20,900,000 – 56,500,000). Most affected world health organization region is southeast Asia, followed by Africa and the Americas. The burden in the endemic locations was estimated to be 13,800,000 infections largely driven by India and 21,600,000 infections in epidemic countries. It was estimated that these infections lead to 17,700,000 symptomatic cases, 848,000 with chronic sequelae and 3,700 deaths. It was estimated that there were 121,000 disability adjusted life years lost to CHIKV each year (13).

**1.11 The Burden of Chikungunya in Africa**

Kenya, Mozambique, Rwanda and Tanzania have been classified as endemic countries for chikungunya in Africa, majority as epidemic countries with Cape Verde, Eritrea, Gambia, Guinea-Bissau, Cote d’Ivoire, South Sudan, Swaziland and Togo reporting no case but have high levels of vectors and neighboring countries where transmission has been reported (13). The endemic

**1.12. The burden of Chikungunya in Kenya**

Kenya has been classified as an endemic country for chikungunya transmission (13).

**CHAPTER TWO**

Chikungunya Vaccines

2.0 History of Chikungunya vaccines

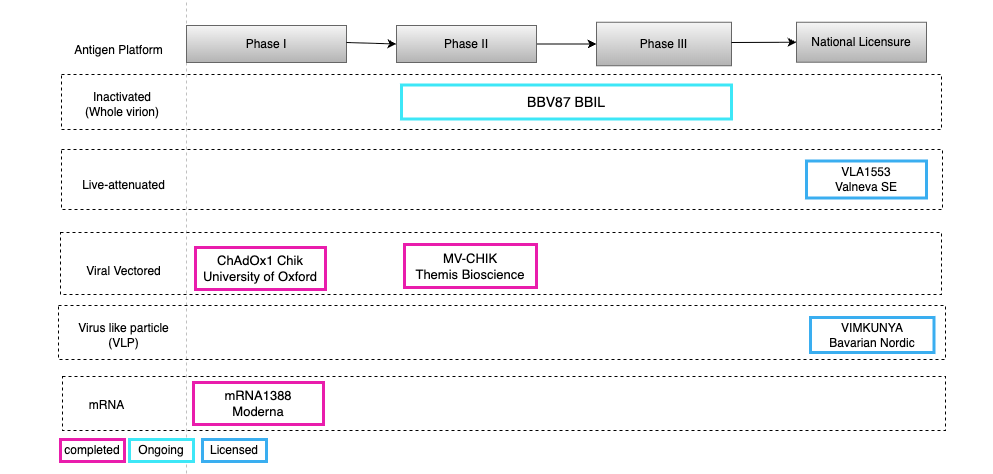
The history of chikungunya vaccine started in 1967 with a formalin inactivated approach at the Walter Reed Army Institute of Research in Washington DC (WRAIR). Pre-clinical phase to assess the immune response elicited in mice and rhesus macaques by a formalin inactivated CHIKV vaccine was done. The African CHIKV strain 168 isolated in southern Tanganyika between 1952 -1953 was used to develop the vaccine. The Chik-embryo (CE), suckling-mouse-brain (SMB) and green monkey kidney cells (GMKC) were used in the experimental vaccines. CE induced poor immunogenicity while good immune responses were induced by SMB and GMKC and the later was selected for further development as SMB poses a risk of inducing encephalitis in humans. Since the vaccine elicited homologous protection in mice, researchers assessed heterologous protection in rhesus macaques using the African CHIKV strain 168, the CHIKV strain E.103 isolated from a pool of 78 Ae. Africanus mosquitoes at Zika forest, Asian strain BAH-306 and the Indian CHIKV strain C-266. 8 Rhesus macaques were vaccinated with three doses of 1 ml each of GMKC – prepared CHIKV strain 168 administered subcutaneously on day 0,7 and 21. Homologous (CHIKV strain 168) and heterologous (CHIKV strain E.103, Asian strain BAH-306, Indian CHIKV strain C-266) challenges were performed 30 days after vaccination. The results yielded complete absence of viremia upon either homologous or heterologous challenge in all vaccinated monkeys. This was the first demonstration of the homologous and heterologous protective efficacy by a formalin-inactivated CHIKV vaccine in pre-clinical models using mice and rhesus macaques (29).

In 1971, formalin-inactivated CHIKV was prepared, and a phase 1 clinical trial was initiated from Thai human samples. Four CHIKV isolates were grown in green monkey kidney culture cells for 10 passages to eliminate any potential hepatitis and other adventitious viruses. A phase 1 clinical trial to assess the vaccine in 16 young volunteers of 21-25 years was done. Two groups of 8 with the first group, received two 0.5ml of the vaccine and the second group 1ml of the vaccine at an interval of 28 days. Local and systemic reactions were assessed for 12 days. Most subjects developed neutralizing antibodies on day 14 after the first vaccination and the second administration prompted the development of significant neutralizing antibody titers with values of log10 neutralization indices of 2.7 for either doses and thus concluded that two 0.5 ml vaccine doses administered 28 days apart are sufficient to elicit good antibody responses.

Production of formalin-inactivated CHIKV vaccine generates costs and safety concerns. This concerns prompted the development of live-attenuated CHIKV vaccine by scientists at the US Army Medical Research Institute of infectious Diseases (USAMRIID) in Maryland in 1986. The vaccine seed virus CHIK 81/clone 25 was obtained from CHIK strain 15561 during the 1962 outbreak of chikungunya disease in Thailand. The original viraemia serum sample was obtained by the US Army Medical component at the SEATO Laboratory in Bangkok, Thailand. The virus was serially passaged 10 times in primary GMK cell culture at WRAIR, and the 11th passage was received from WRAIR and was used as starting material for developing an attenuated clone for production of a live vaccine for human use. CHIK strain 15561 was subjected to 18 plaque-to-plaque in MRC-5 cultures before CHIK 181/clone 25 was selected as a vaccine seed based on homogeneous small plaque size, suckling mouse a virulence, reduced monkey viraemia and genetic stability. Oligonucleotide mapping demonstrated differences between parent and clone. Vaccine elicited neutralizing antibody and protected mice and rhesus monkey against challenge. After the challenge, viraemia’s were absent in vaccinated monkeys. Vaccine was then produced and tested in accordance with governmental regulatory requirements for human use. Following these results, CHIK 181/clone 25 entered phase 1 clinical trial at USAMRIID and the University of Maryland center for vaccine development :reference (30).

**2.1 Chikungunya vaccines Licensure**

After decades with few effective tools to combat CHIKV, substantial investment by CEPI has led to the licensure by FDA and EMA the first two chikungunya vaccines, IXCHIQ(VLA1553) and VIMKUNYA (13,16). Other four chikungunya vaccine candidates are in development.



**2.1 VLA1553/IXCHIQ Vaccine**

IXCHIQ was the first chikungunya vaccine to be licensed by US Food and Drug Administration (FDA) in November 2023, European Medicines Agency (EMA) in May 2024 and Health Canada in June 2024. The vaccine was approved for individuals 18 years of age and older who are at increased risk of exposure to chikungunya virus (31).

**Challenges in Deployment of the Vaccine**

Due to unpredictable nature of chikungunya epidemiology, licensure was obtained through correlates of protection (13). A major hurdle in the optimal deployment of the vaccines is the limited understanding of the underlying burden of CHIKV around the world and how best to deploy the vaccine (13,16). There is also poor understanding of where the virus circulates, hampering the development of vaccine investment cases (13). Gavi alliance which helps lower income countries purchase vaccines has placed CHIKV vaccine on a learning agenda, which means that it does not feel there is sufficient information available to make informed decisions on the likely impact of the vaccine (13). One of the significant obstacles to address the spread of chikungunya is lack of data involving its burden (32). This knowledge gap is driven by frequent clinical misdiagnosis with other pathogens such as dengue and limited access to confirmatory testing (13).

**CHAPTER THREE**

**Chikungunya Modelling Frameworks**

**3.0 Modelling Chikungunya virus**

|  |  |
| --- | --- |
| Author | Model |
| Diego Ruiz et al., 2012 |  |
| Laith et al., 2013 |  |
| Folashade et al.,2016 |  |
| Xiaomei et al., 2019 |  |
| Gilberto et al., 2019 |  |
| Bijal et al., 2020 |  |
| Ruchi et al 2020 |  |
| Haque et al. 2021 |  |
| Mlyashimbi et al., 2022 |  |
| Seema et al 2022 |  |
| Joseph Y et al., 2023 |  |

Table 1: static models

**Serological modeling of Arboviruses**

Antigen-specific antibodies are immunological markers of past infections. Analyzing blood samples from a representative sample of a population can be used to characterize the distribution of these antibodies in a population. Population based serological surveys are key tools in epidemiology to characterize the level of population immunity and reconstruct the past circulation of pathogens (33). For example, if a pathogen is characterized by low level endemic circulation with FOI of 2% per year, we expect that seroprevalence will slowly increase with age. Analyzing how seroprevalence changes with age, serocatalytic models can reconstruct the historical force of infection.

Several serocatalytic models have been developed to estimate force of infection (FOI) from age stratified seroprevalence data. Force of infection is the rate at which susceptible individuals become infected. Antibodies are immune markers of past infection (33). Population immunity and estimates of future infections can be assessed from seroprevalence studies based on the seropositivity status of the population. When age of the participants is available their seropositivity status can be used to reconstruct the history of circulation of a pathogen. Serocatalytic models can be used to test competing modes of circulations (e.g. endemic circulation vs irregular outbreaks) accounting for cross reactivity or characterize transmission following an intervention.

**R packages for Serological analysis**

1. Rsero Package

Rsero is an R package that implements a series of serocatalytic models and estimates the FOI from age-stratified seroprevalence data using Bayesian methods. The package contains features to perform model comparison and visualize model fit. The package can support serosurvey study design in a variety of epidemic situations. It provides a framework to study the dynamics of past pathogen circulation from serological survey data. Rsero is a package that provides a pipeline of methods to store age-stratified serological datasets, analyze and estimate the history of circulation of a pathogen and compare different transmission models. Statistical inference is done Bayesian approaches.

FOI can be estimated from age-stratified serological survey data using the classical theory of serocatalytic models. One year is considered as the time unit. The probability that an individual of age a is seropositive depends on the FOI of the pathogen during their lifetime. A 1-year-old seropositive child will have been exposed to the pathogen during the first year of their life. If λ is the FOI during the first year of life, the probability that the child is seropositive at year 1 is 1 – exp (-λ1). The cumulative FOI of an individual of age n over their lifetime is related to the probability this individual is seropositive at sampling

P = 1 – exp ( (1)

**Mode of Transmission**

Several models are implemented to describe different changes in the temporal dynamics of the FOI in the Rsero package. They include models of constant or piecewise transmission alongside new approaches to model outbreaks.

1. Constant model is a simple model assuming FOI is constant through time. The probability that an individual of age n is seropositive is given by

P = 1 – exp(-nλ) (2)

1. Independent model allows FOI to vary every year. The probability of being seropositive at age n follows equation 1
2. Piecewise-constant model is a model which extends the constant model to consider several periods of constant FOI. The model can be used to describe the impact of an intervention which decreases the annual risk of infection in an endemic setting. For example, if the transmission was constant with annual FOI λA before changing T years ago to an annual FOI λB, then the probability for an individual of age n to be seropositive is

P = 1 – exp(-nλB) if n T

P = 1- exp(-nλB) exp (-(n-T) ( λA - λB)) in n > T (3)

1. The outbreak model describes epidemic spread of a pathogen i.e., assuming there have been k epidemics in the past where k is fixed. The FOI at year *i* is given as a sum of k Gaussians. Each gaussian is centered on Tk, the peak epidemic time of the focal outbreak is given by

λI = k exp (-(I – Tk)2) (4)

Where k  = αk2.

In epidemic k, the FOI is maximal at the peak Tk; 1-exp(-αk) is the attack rate of the outbreak defined by the overall probability of infection over the course of the outbreak

1. The outbreak + constant model is a model where FOI is constant and punctuated by outbreaks. The FOI during year *i* is λI = λC + k exp (-(I – Tk)2) where λC represents the constant part of the FOI.

**Seroreversion**

Not all infections provide lifelong immunity- seroreversion means switching from seropositivity to seronegativity. The seroreversion is captured by a seroreversion rate p. if FOI is a constant λ , then the probability that an individual of age n is seropositive is given by

P =

**Imperfect sensitivity and specificity**

The models can account for uncertainty in the seroprevalence estimates resulting from imperfect sensitivity and specificity of the assays. The *se* and *sp* the sensitivity and specificity of the assays respectively. In a scenario of perfect sensitivity se=sp=1, seroprevalence is the same as the proportion of the population infected by the pathogen. If we denote this proportion the probability for an individual to be observed as seropositive is

**Setting different categories for risk of infection**

For differential susceptibility to a pathogen, it is possible to estimate the FOI for different subjects define by individual characteristics e.g. sex, region etc.

**Parameter inference**

The models are fitted to data using MCMC algorithm implemented in the *rstan* package which provides the interface to stan, a probability programming language for specifying and fitting Bayesian models.

Bayesian inference

Prior distribution

A prior distribution of a parameter is the probability distribution that represents uncertainty about the parameters before observing the data. Multiplying the prior distribution and the likelihood function leads to the posterior distribution of the parameter.

References

1. De Roo AM, Vondeling GT, Boer M, Murray K, Postma MJ. The global health and economic burden of chikungunya from 2011 to 2020: a model-driven analysis on the impact of an emerging vector-borne disease. BMJ Glob Health [Internet]. 2024 Dec 3 [cited 2025 Jul 29];9(12). Available from: https://gh.bmj.com/content/9/12/e016648

2. Nsoesie EO, Kraemer MUG, Golding N, Pigott DM, Brady OJ, Moyes CL, et al. Global distribution and environmental suitability for chikungunya virus, 1952 to 2015. Eurosurveillance [Internet]. 2016 May 19 [cited 2025 Jul 30];21(20):30234. Available from: https://www.eurosurveillance.org/content/10.2807/1560-7917.ES.2016.21.20.30234

3. Zeller H, Van Bortel W, Sudre B. Chikungunya: Its History in Africa and Asia and Its Spread to New Regions in 2013–2014. J Infect Dis [Internet]. 2016 Dec 15 [cited 2025 Aug 1];214(suppl\_5):S436–40. Available from: https://dx.doi.org/10.1093/infdis/jiw391

4. Chikungunya [Internet]. [cited 2025 Jul 30]. Available from: https://www.who.int/health-topics/chikungunya#tab=tab\_1

5. Lanciotti RS, Lambert AJ. Phylogenetic Analysis of Chikungunya Virus Strains Circulating in the Western Hemisphere. Am J Trop Med Hyg [Internet]. 2016 Apr 1 [cited 2025 Jul 30];94(4):800. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC4824221/

6. Bartholomeeusen K, Daniel M, LaBeaud DA, Gasque P, Peeling RW, Stephenson KE, et al. Chikungunya fever. Nature Reviews Disease Primers 2023 9:1 [Internet]. 2023 Apr 6 [cited 2025 Jul 29];9(1):1–21. Available from: https://www.nature.com/articles/s41572-023-00429-2

7. Halstead SB. Reappearance of chikungunya, formerly called Dengue, in the Americas. Emerg Infect Dis. 2015;21(4):557–61.

8. Konongoi SL, Nyunja A, Ofula V, Owaka S, Koka H, Koskei E, et al. Human and entomologic investigations of chikungunya outbreak in Mandera, Northeastern Kenya, 2016. PLoS One [Internet]. 2018 Oct 1 [cited 2025 Aug 7];13(10):e0205058. Available from: https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0205058

9. De Bernardi Schneider A, Ochsenreiter R, Hostager R, Hofacker IL, Janies D, Wolfinger MT. Updated Phylogeny of Chikungunya Virus Suggests Lineage-Specific RNA Architecture. Viruses [Internet]. 2019 Aug 29 [cited 2025 Aug 14];11(9):798. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC6784101/

10. Russo G, Subissi L, Rezza G. Chikungunya fever in Africa: a systematic review. Pathog Glob Health [Internet]. 2020 Apr 2 [cited 2025 Aug 7];114(3):136. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC7241529/

11. Sergon K, Njuguna C, Kalani R, Ofula V, Onyango C, Konongoi LS, et al. Seroprevalence of Chikungunya Virus (CHIKV) Infection on Lamu Island, Kenya, October 2004. Am J Trop Med Hyg [Internet]. 2008 Feb 1 [cited 2025 Aug 8];78(2):333–7. Available from: https://www.ajtmh.org/view/journals/tpmd/78/2/article-p333.xml

12. Flandes X, Hansen CA, Palani S, Abbas K, Bennett C, Caro WP, et al. Vaccine value profile for Chikungunya. Vaccine [Internet]. 2024 Jul 25 [cited 2025 Aug 9];42(19):S9–24. Available from: https://www.sciencedirect.com/science/article/pii/S0264410X23009155?via%3Dihub

13. Ribeiro dos Santos G, Jawed F, Mukandavire C, Deol A, Scarponi D, Mboera LEG, et al. Global burden of chikungunya virus infections and the potential benefit of vaccination campaigns. Nat Med [Internet]. 2025 Jul 1 [cited 2025 Aug 11];31(7):2342–9. Available from: https://www.nature.com/articles/s41591-025-03703-w

14. Puntasecca CJ, King CH, Labeaud AD. Measuring the global burden of chikungunya and Zika viruses: A systematic review. PLoS Negl Trop Dis [Internet]. 2021 Mar 1 [cited 2025 Aug 17];15(3):e0009055. Available from: https://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0009055

15. Chen LH, Fritzer A, Hochreiter R, Dubischar K, Meyer S. From bench to clinic: the development of VLA1553/IXCHIQ, a live-attenuated chikungunya vaccine. 2024 [cited 2025 Aug 11]; Available from: https://doi.org/10.1093/jtm/taae123

16. Maure C, Khazhidinov K, Kang H, Auzenbergs M, Moyersoen P, Abbas K, et al. Chikungunya vaccine development, challenges, and pathway toward public health impact. Vaccine [Internet]. 2024 Dec 2 [cited 2025 Aug 13];42(26):126483. Available from: https://www.sciencedirect.com/science/article/pii/S0264410X24011654

17. Lamballerie de X. Globalization of Chikungunya: 10 years to invade the world. [cited 2025 Aug 8]; Available from: http://www.astmh.org/Content/NavigationMenu/Publications/IntheNews/ACAV\_Chikungunya\_docu

18. The occurrence of a dengue-like fever in the North-Eastern Transvaal. I. Clinical features and isolation of virus - PubMed [Internet]. [cited 2025 Aug 4]. Available from: https://pubmed.ncbi.nlm.nih.gov/13421902/

19. Sergon K, Yahaya AA, Brown J, Bedja SA, Mlindasse M, Agata N, et al. SEROPREVALENCE OF CHIKUNGUNYA VIRUS INFECTION ON GRANDE COMORE ISLAND, UNION OF THE COMOROS, 2005. Centers for Disease Control and Prevention. 2007;

20. A major epidemic of chikungunya virus infection on Reunion Island, France, 2005-2006 - PubMed [Internet]. [cited 2025 Aug 8]. Available from: https://pubmed.ncbi.nlm.nih.gov/17978079/

21. Yaseen HM, Simon F, Deparis X, Marimoutou C. Estimation of Lasting Impact of a Chikungunya Outbreak in Reunion Island. Epidemiology: Open Access [Internet]. 2012 Jan 27 [cited 2025 Aug 7];2(2):1–6. Available from: https://www.omicsonline.org/estimation-of-lasting-impact-of-a-chikungunya-outbreak-in-reunion-island-2161-1165.S2-003.php

22. Njenga MK, Nderitu L, Ledermann JP, Ndirangu A, Logue CH, Kelly CHL, et al. Tracking epidemic Chikungunya virus into the Indian Ocean from East Africa. J Gen Virol [Internet]. 2008 [cited 2025 Aug 5];89(Pt 11):2754. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC3347796/

23. (PDF) Seroprevalence of Chikungunya virus (CHIKV) infection on Lamu Island, Kenya, October 2004 [Internet]. [cited 2025 Aug 5]. Available from: https://www.researchgate.net/publication/5596653\_Seroprevalence\_of\_Chikungunya\_virus\_CHIKV\_infection\_on\_Lamu\_Island\_Kenya\_October\_2004

24. Chikungunya | WHO | Regional Office for Africa [Internet]. [cited 2025 Aug 7]. Available from: https://www.afro.who.int/health-topics/chikungunya

25. Inziani M, Adungo F, Awando J, Kihoro R, Inoue S, Morita K, et al. Seroprevalence of yellow fever, dengue, West Nile and chikungunya viruses in children in Teso South Sub-County, Western Kenya. International Journal of Infectious Diseases [Internet]. 2020 Feb 1 [cited 2025 Aug 5];91:104–10. Available from: https://pubmed.ncbi.nlm.nih.gov/31712089/

26. Kageha S, Ngoi JM, Kubo T, Morita K, Mwau M, Author C. Chikungunya Seroprevalence among Patients Presenting with Febrile illnesses in selected health facilities in Mt. Elgon region, Kenya. medRxiv [Internet]. 2024 Apr 27 [cited 2025 Aug 5];2024.04.26.24306414. Available from: https://www.medrxiv.org/content/10.1101/2024.04.26.24306414v1

27. Waggoner J, Brichard J, Mutuku F, Ndenga B, Heath CJ, Mohamed-Hadley A, et al. Malaria and chikungunya detected using molecular diagnostics among febrile Kenyan children. academic.oup.comJ Waggoner, J Brichard, F Mutuku, B Ndenga, CJ Heath, A Mohamed-Hadley, MK SahooOpen forum infectious diseases, 2017•academic.oup.com [Internet]. 2017 [cited 2025 Aug 6]; Available from: https://academic.oup.com/ofid/article-abstract/4/3/ofx110/3858096

28. de Souza WM, de Lima STS, Simões Mello LM, Candido DS, Buss L, Whittaker C, et al. Spatiotemporal dynamics and recurrence of chikungunya virus in Brazil: an epidemiological study. Lancet Microbe [Internet]. 2023 May 1 [cited 2025 Aug 11];4(5):e319. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC10281060/

29. Reyes-Sandoval A. 51 years in of Chikungunya clinical vaccine development: A historical perspective. Hum Vaccin Immunother [Internet]. 2019 Oct 3 [cited 2025 Aug 18];15(10):2351–8. Available from: https://pubmed.ncbi.nlm.nih.gov/30735447/

30. Levitt NH, Ramsburg HH, Hasty SE, Repik PM, Cole FE, Lupton HW. Development of an attenuated strain of chikungunya virus for use in vaccine production. Vaccine [Internet]. 1986 Sep 1 [cited 2025 Aug 18];4(3):157–62. Available from: https://www.sciencedirect.com/science/article/pii/0264410X86900034?via%3Dihub

31. FDA Approves First Vaccine to Prevent Disease Caused by Chikungunya Virus | FDA [Internet]. [cited 2025 Aug 11]. Available from: https://www.fda.gov/news-events/press-announcements/fda-approves-first-vaccine-prevent-disease-caused-chikungunya-virus

32. Puntasecca CJ, King CH, Labeaud AD. Measuring the global burden of chikungunya and Zika viruses: A systematic review. PLoS Negl Trop Dis [Internet]. 2021 Mar 1 [cited 2025 Aug 13];15(3):e0009055. Available from: https://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0009055

33. Hozé N, Pons-Salort M, Metcalf CJE, White M, Salje H, Cauchemez S. RSero: A user-friendly R package to reconstruct pathogen circulation history from seroprevalence studies. PLoS Comput Biol [Internet]. 2025 Feb 1 [cited 2025 Aug 12];21(2):e1012777. Available from: https://journals.plos.org/ploscompbiol/article?id=10.1371/journal.pcbi.1012777