

Zika Virus Detection with 2013 Serosurvey, Mombasa, Kenya

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Acute Zika virus (ZIKV) infection has not been confirmed in Kenya. In 2018, we used specimens collected in a 2013 dengue serosurvey study in Mombasa to test for ZIKV IgM. We confirmed specific ZIKV IgM positivity in 5 persons. These results suggest recent ZIKV transmission in the coastal region of Kenya.

Zika virus (ZIKV), an arbovirus belonging to the family *Flaviviridae*, was initially isolated in 1947 from the serum of a pyrexial sentinel rhesus macaque in the canopy of the Zika Forest in Uganda (1). In 1970, ZIKV antibodies were reported in humans in Kenya, using immunoassays with limited sensitivity and specificity (2); since then, only past infections have been reported (3,4).

In 2015, the emergence of ZIKV in the Americas (5) raised the possibility that ZIKV infections were also occurring in Kenya but were undetected due to mild or asymptomatic infections and rudimentary birth defect surveillance. In 2016, Cape Verde and, in 2017, Angola reported ZIKV outbreaks associated with microcephaly. Genome sequencing from Angola identified the similarity of the Asian lineage of ZIKV to the American strain (6); this evidence implies that the African lineage may be less likely to cause microcephaly, thus explaining the few reported number of ZIKV-associated microcephaly cases in Africa (7).

To understand prior transmission of ZIKV in Kenya, in 2018, we used specimens collected in 2013 for a dengue serosurvey from Mombasa amidst a dengue outbreak. During May 3–11, 2013, serum specimens were collected from 1,500 consenting household members in 986 randomly selected households (8). Specimens were tested for dengue virus (DENV) by

real-time reverse transcription PCR (rRT-PCR) and DENV IgM ELISA (InBios International, Inc., <https://inbios.com>). Concurrent with this serosurvey, hospital surveillance of suspected dengue patients was established (8).

For our study, we used only the DENV-negative serum specimens and tested them for ZIKV by Centers for Disease Control and Prevention rRT-PCR and ZIKV IgM antibody capture ELISA (MAC ELISA). A ZIKV MAC ELISA specimen with a positive-to-negative ratio ≥ 3.00 was positive and a ratio of 2–2.99 was equivocal. We confirmed all positive specimens with 90% plaque reduction neutralization test (PRNT₉₀) against ZIKV strain MR766 (African lineage) and DENV (ChimeraVax; Sanofi Pasteur, <https://www.sanofi.com>) (9). We defined recent ZIKV infection by a positive result in the ZIKV MAC ELISA and a PRNT₉₀ titer of ZIKV that was 4-fold higher than the titer of DENV. The institutional review board at US Centers for Disease Control and Prevention and Kenya Medical Research Institute Scientific Ethics Review Unit approved the study.

We identified a total of 745 DENV-negative persons from the dengue serosurvey (n = 704) and hospital surveillance (n = 41); median age was 28 (range 0–94) years. None were positive by ZIKV rRT-PCR. Thirty-four (4.6%) were positive by ZIKV MAC ELISA, 7 captured in hospital surveillance; 24 (3.2%) were equivocal and 687 (92.2%) negative. Of the 34 ZIKV MAC ELISA positives, the ZIKV PRNT₉₀ assay confirmed 5 (15.1%, 4 from serosurvey and 1 hospital) as ZIKV, 7 (21.2%) as DENV, 3 (9.1%) as cross-reactive to both viruses, and 18 (54.5%) as negative for ZIKV and DENV; 1 (2.9%) could not be confirmed due to insufficient sample (Table). Of the 5 ZIKV MAC ELISA-positive patients confirmed by ZIKV PRNT₉₀, median age was 57 (range 50–70) years.

We identified 5 persons sampled in 2013 who were positive for ZIKV IgM and confirmed by PRNT₉₀. Of note, the positive participants (median age 57 years) were older than the sample group tested (median age 29 years). The level of transmission of ZIKV in Kenya is unknown, although Kenya has the competent vector (*Aedes aegypti* mosquitoes), and parts of Kenya are ecologically similar to the Zika forest (1). ZIKV has not been detected in Kenya despite recent intensive follow-up of pregnant women in coastal Kenya (10). Our study suggests that ZIKV may circulate in Mombasa and cause asymptomatic disease not captured in hospital surveillance systems. In addition, severely ill ZIKV patients might not have been identified from the original dengue study because it only captured clinical endpoints related to severe dengue.

Table. Test results for 16 participants in a 2013 serosurvey in Kenya whose samples were positive for ZIKV by testing conducted in 2018*

Participant no.	Age, y	CDC ZIKV MAC			DENV PRNT ₉₀ titer	Participant type	Final interpretation of case
		ELISA P/N ratio (IgM)	ZIKV PRNT ₉₀ titer				
2	63	3.56	1:20	<1:20	Serosurvey	ZIKV	
3	50	10.00	>1:320	<1:20	Serosurvey	ZIKV	
5	70	3.17	1:20	<1:20	Serosurvey	ZIKV	
10	NA	3.51	1:20	<1:20	Hospital	ZIKV	
31	51	5.50	>1:320	1:20	Serosurvey	ZIKV	
4	45	3.26	<1:20	1:20	Serosurvey	DENV	
7	36	8.65	1:20	1:320	Serosurvey	DENV	
20	58	13.18	1:20	1:320	Serosurvey	DENV	
25	45	3.70	1:20	>1:320	Serosurvey	DENV	
26	27	5.00	1:80	>1:320	Serosurvey	DENV	
27	42	3.20	1:80	1:320	Serosurvey	DENV	
33	30	10.70	1:20	1:80	Serosurvey	DENV	
1	32	4.49	1:20	1:20	Serosurvey	Cross-reactive	
9	27	3.82	1:20	1:20	Serosurvey	Cross-reactive	
30	58	3.20	>1:320	>1:320	Serosurvey	Cross-reactive	
6	41	16.35	1:20	ND	Serosurvey	Inconclusive	

*CDC, Centers for Disease Control and Prevention; DENV, dengue virus; MAC, IgM antibody capture; NA, not available; ND, not done due to insufficient specimen; PRNT₉₀, plaque reduction neutralization test 90%; ZIKV, Zika virus.

We found 1 participant positive by ZIKV MAC ELISA but negative by ZIKV and DENV PRNT₉₀, suggesting the presence of another co-circulating cross-reactive flavivirus in this region of Kenya (e.g., West Nile virus). This finding merits further investigation to determine all circulating flaviviruses in Mombasa. ZIKV transmission season in Kenya most likely coincides with other arboviruses that share the same vector. Of interest, titers of neutralizing antibodies against ZIKV were low for 3 of the 5 positive participants (1:20 PRNT₉₀ titer), typically observed in acute infections (<10 days after onset of illness). Two participants had high neutralization titers (>1:320), suggesting recent infection (within the previous 90 days).

Our study had some limitations. It was conducted in 2018, so specimens had been archived for 5 years. We also had incomplete demographic and clinical data and could not discount concurrent DENV infections. Finally, we might have underestimated ZIKV positives because of weak neutralization by IgM.

In conclusion, ZIKV may have circulated at low levels in Kenya in 2013. More research is needed to evaluate current ZIKV circulation and characterize other co-circulating flaviviruses. Enhanced surveillance systems, including for microcephaly and other birth defects, could capture ZIKV patients and determine the epidemiology of ZIKV African lineage in this country.

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Mycobacterium bovis **Pulmonary Tuberculosis** **after Ritual Sheep Sacrifice** **in Tunisia**

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A woman in France was diagnosed with pulmonary tuberculosis caused by *Mycobacterium bovis* after a ritual sheep sacrifice in her home country of Tunisia. This investigation sheds light on ritual sacrifice of sheep as a circumstance in which religious tradition and practices can expose millions of Muslims worldwide to this disease.

Mycobacterium bovis is historically responsible for zoonotic, deadly tuberculosis and has seemingly reemerged in countries where it had previously vanished following eradication programs in cattle and the pasteurization of dairy products (1-3). In most cases, *M. bovis* tuberculosis results from consuming unpasteurized milk; extrapulmonary disease

is thus the most frequent clinical manifestation (4-6). Also, *M. bovis* could be an airborne zoonotic pathogen causing pulmonary tuberculosis (7). In western Europe countries, most of *M. bovis* human tuberculosis cases are seen in migrants and are associated with travel to the country of origin (4,5,8,9). *M. bovis* tuberculosis is traced to animal sources, yet reporting of clinical signs and symptoms is often delayed (3). The observation of a woman affected by *M. bovis* tuberculosis who participated in a precisely dated religious practice involving sheep slaughtering provided an opportunity to shed light on these medical aspects.

A 43-year-old unemployed woman born in Tunisia emigrated to France in 2000, married, and had 2 children. The patient had no underlying chronic condition, no medical history, no treatment, no history of smoking, and no toxic habits. She had received the bacillus Calmette-Guérin vaccine during childhood. Her last trip to Tunis and surrounding areas was during July 10-August 28, 2018. The patient denied any contacts with ill persons during her stay. She participated in the Aid-el-Kebir (the Great Festival) Muslim festivities on August 22-23, 2018. After the ritual, her husband slaughtered a veterinary-uncontrolled sheep outside the house by cutting through its neck in an open place and then insufflating air beneath the skin of the dead animal using bellows before butchering the viscera, including lungs, heart, liver, and kidneys, which were put into a container while the digestive tract was put separately into another container. His wife then washed the lungs and the other viscera for ≈2 hours in a confined kitchen, cooked them, and consumed them with her family; she did not experience any injury while butchering the animal.

After her return to France, the patient was apyretic with productive cough, fatigue, and anorexia, which started exactly 22 days after the Aid-el-Kebir festivities ended. The patient attributed symptoms to allergy and did not consult with a healthcare professional. In December 2018, her respiratory tract symptoms persisted; she also developed fatigue, fever, and night sweats and consulted a general practitioner. In January 2019, a computerized tomodensitometry scan confirmed an abscess in the inferior lobe of the patient's left lung with thick walls and an infiltrate in the lower lobe of the right lung. The patient reported fever, cough, expectoration, and anorexia, and lost 5 kg within 1 month (body mass index 15); we found crackles in both the left inferior and right superior lobes. She did not report hemoptysis, and her physical examination was otherwise unremarkable. Blood examination showed an iron deficiency in microcytic anemia, and her leukocyte count was normal.