ORIGINAL ARTICLE

UDC: 616-036.22:[616-022:578.833
DOI: https://doi.org/10.2298/VSP241127027A

Seroprevalence of arboviruses in members of United Nations peacekeeping missions in the Central African Republic

Seroprevalencija arbovirusnih infekcija kod pripadnika mirovnih misija Ujedinjenih nacija u Centralnoafričkoj Republici

Sonja Atanasievska Kujović*, Elizabeta Ristanović*, Nikoleta Djordjevski*, Maja Živić†, Milena Krstić†‡, Nenad Ljubenović†, Jovan Mladenović†, Sofija Trifunović‡, Sonja Marjanović‡

Military Medical Academy, *Institute of Microbiology, †Institute of Epidemiology, Belgrade, Serbia; ‡University of Defence, Faculty of Medicine of the Military Medical Academy, Belgrade, Serbia

Abstract

Background/Aim. Members of the Serbian Armed Forces participating in the United Nations (UN) Multidimensional Integrated Stabilization Mission in the Central African Republic (CAR) – MINUSCA, are exposed to the bites of the *Aedes* genus mosquitoes, which are transmitters of some (or certain) arboviruses. The aim of the study was to determine the exposure, risk factors, and prevention and protection measures against infection with arboviruses in members of peacekeeping missions (PKM). **Methods.** The study included 136 members of UN PKM in CAR in two rotations during 2022/2023, lasting 8 and 6 months. Each respondent had to answer the questions of the designed epidemiological questionnaire for the returnees from PKM. The subjects' blood sera were tested for specific antibodies against Dengue, Zika, and Chikungunya viruses using an enzyme-linked immunosorbent assay (ELISA) test. **Results.** The number of seropositive individuals for arboviruses was 48 (35.3%). Most members, 23 (16.9%), were seropositive for the Dengue virus, slightly fewer for the Chikungunya virus, 14 (10.3%), and the fewest were seropositive for the Zika virus, 11 (8.1%). Our research shows that impregnating uniforms with repellent plays a significant role in preventing mosquito bites and the Zika virus infection. The number of days spent in the mission and the factors of gender and age did not show an influence on the occurrence of arbovirus infections. **Conclusion.** The members of PKM in the CAR have come into contact with Dengue, Zika, and Chikungunya viruses. This indicates that it is necessary to work on the constant improvement of prevention measures and protection of their health, including further serological tests, but also tests for the presence of various infectious microorganisms in this endemic area.

Key words:
arbovirus infections; central african republic; culicidae; hospitals, military; insect repellents; preventive health services; risk factors; surveys and questionnaires; united nations.

Apstrakt

Uvod/Cilj. Pripadnici Vojske Srbije koji učestvuju u Multinacionalnoj operaciji Ujedinjenih nacija (UN) u Centralnoafričkoj Republici (CAR) – MINUSCA, izloženi su tokom boravka u misiji ubodima komaraca roda *Aedes* koji su prenosioci nekih (ili pojedinih) arbovirusa. Cilj rada bio je da se utvrdi izloženost, faktori rizika i mere prevencije i zaštite od infekcije arbovirusima kod pripadnika mirovnih misija. **Metode.** U studiju je uključeno 136 pripadnika mirovnih operacija UN u CAR iz dve rotacije tokom 2022/2023 godine u trajanju od 8 i 6 meseci. Svaki ispitanik morao je da odgovori na pitanja koja su data u dizajniranom epidemiološkom upitniku za povratnike iz mirovnih misija. Krvni serumi ispitanika testirani su *enzyme-linked immunosorbent assay* (ELISA) testom na prisustvo specifičnih antitela na viruse *Dengue*, *Zika* i *Chikungunya*. **Rezultati.** Utvrđeno je 48 (35,3%) seropozitivnih ispitanika na arboviruse. Najviše seropozitivnih pripadnika, 23 (16,9%), bilo je na *Dengue* virus, nešto manje na *Chikungunya* virus, 14 (10,3%), dok je najmanje bilo pozitivno na *Zika* virus, 11 (8,1%). Naše istraživanje pokazuje da impregnacija uniforme repelentom ima značajnu ulogu u prevenciji uboda komaraca i sprečavanju infekcije *Zika* virusom. Broj dana provedenih u misiji, kao i faktor pola i godine starosti nisu pokazali uticaj na pojavu infekcija arbovirusom. **Zaključak.** Pripadnici mirovnih misija u CAR došli su u

Correspondence to: Sonja Atanasievska Kujović, Military Medical Academy, Institute of Microbiology, Crnotravska 17, 11 000 Belgrade, Serbia. E-mail: sonja.atanasievska@gmail.com

kontakt sa virusima *Dengue, Zika* i *Chikungunya*. To ukazuje da je neophodno raditi na stalnom unapređenju mera prevencije i zaštite njihovog zdravlja, uključujući serološka ispitivanja, ali i ispitivanja prisustva različitih infektivnih mikroorganizama tog endemskog područja.

Ključne reči:
infekcija, arbovirus; centralna afrička republika; komarci; bolnice, vojne; repelenti; preventivno-medicinska zaštita; faktori rizika; ankete i upitnici; mirovne snage ujedinjenih naroda.

Introduction

Dengue, Zika, and Chikungunya viruses are arboviruses transmitted by mosquitoes of the genus *Aedes*. These viruses are mainly found in tropical and subtropical regions, but in recent years, an expansion of their geographical range towards temperate zones has been observed. This happens because of the spread of vectors to new areas due to climate change and global warming, so the epidemics caused by these viruses occur on almost all continents [1, 2].

Dengue fever is the most common arbovirus infection, occurring in over 100 tropical and subtropical countries, with approximately 400 million cases reported annually [3, 4]. The infection is caused by one of the four serotypes of the Dengue virus. Symptoms can range from mild to severe (hemorrhagic fever and shock syndrome) and can be potentially lethal. This virus occurs seasonally, with a peak during and after the rainy season when the mosquito population increases [3–5].

The Zika virus was named after the Zika area in Uganda, where it was first isolated in 1947. A person can become infected with the Zika virus through a mosquito bite, but also through sexual contact. The symptoms are mild and last two to seven days. However, infection with this virus during pregnancy can cause microcephaly and lead to congenital brain damage in the fetus. It has also been linked to Guillain-Barre syndrome, neuropathy, and myelitis in children and adults [6, 7].

The Chikungunya virus was first isolated in Tanzania in 1952. In addition to increased fever, the most common symptom of this infection is pain in the joints, which can occur repeatedly throughout a person's life and have a progressive degenerative course in the form of chronic arthritis. Muscle pain, headaches, nausea, and skin rash are also present. In addition to Africa and Asia, imported cases have been registered in Europe and parts of the American continent [8–10].

Infections with Dengue, Zika, and Chikungunya viruses are often asymptomatic or have mild clinical symptoms, such as fever, headache, muscle and joint pain, nausea, and skin rash. Since these diseases have similar symptoms, they can resemble each other, and if there is no laboratory confirmation, patients can often be misdiagnosed. As patients with a mild clinical infection do not report to the doctor, a large number of cases go undetected [2, 11, 12].

The treatment of these diseases is symptomatic. There is specific prophylaxis for Dengue fever only, and that in the form of the quadrivalent vaccine Dengvaxia®, while the vaccines for Zika and Chikungunya are still at the clinical trial stage [13, 14].

Considering the increase in the incidence of arbovirus infections, the question of effective prevention of these diseases arises. Prophylaxis and disease control aim to prevent or reduce the transmission of viruses through vector control and prevention of human-vector contact. This could be achieved by implementing personal protective measures, general hygiene measures, specific prophylaxis (where available), as well as by performing continuous educational work among the population in endemic areas [3, 15].

Members of the Ministry of Defence and the Serbian Armed Forces of the Republic of Serbia have been engaged in a United Nations (UN) Multidimensional Integrated Stabilization Mission in the Central African Republic (CAR) – MINUSCA since September 2014. This country and region are endemic for arboviruses. The climate of the CAR is predominantly tropical, with a rainy season from June to September in the northern regions of the country and from May to October in the south. The maximum annual rainfall is around 1,800 mm in the upper part of the Ubangi region. Across the country, annual average temperatures range from 23 °C in the south to 26 °C in the north [16]. There are not many papers in the literature examining the presence of arboviruses in peacekeeping mission (PKM) participants. Various studies from the literature show the results of different geographical, climatic, and epidemiological areas for members of PKM who come from various countries and have different genetic predispositions [2, 3, 11].

The aim of our study was to examine the extent to which members of PKM are exposed to the risk of infection with arboviruses, which risk factors increase the possibility of infection, as well as the effectiveness of the prevention and protection measures implemented to reduce the risk of infection.

Methods

A total of 136 participants from two rotations of the UN PKM in the CAR during 2022/2023 took part in the study. The first rotation lasted 8 months (66 participants), while the second lasted 6 months (70 participants). As part of the preparation for PKM, the participants were informed about the risks of contracting diseases occurring on the territory of the CAR and introduced to the measures for its prophylaxis and prevention. The participants were vaccinated with the prescribed and recommended vaccine (yellow fever) for entry into the CAR territory.

After returning from PKM, the health status of the participants was monitored for 21 days. On the tenth day upon returning, a blood sample was taken from each individual for further microbiological testing. On this occasion, they completed the specifically designed questionnaire at the Institute of Epidemiology, Military Medical Academy (MMA), Belgrade, Serbia, intended for individual risk assessment. From these questionnaires, the following data were collected: whether and how many mosquito bites they had, whether they used repellents for personal protection and room protection, nets on doors and windows, and whether their uniforms were impregnated with repellent. The research has been approved by the Ethics Committee of the Faculty of Medicine of the MMA (No. 1/11/2024).

The serological detection of Dengue, Zika, and Chikungunya viruses was carried out at the Institute of Microbiology (Department of Microbe Genetics and Immunology) of the MMA. An enzyme-linked immunosorbent assay (ELISA) test (EUROIMMUN, Lübeck, Germany) was used for the detection of specific immunoglobulin (Ig) M and IgG antibodies against Dengue, Zika, and Chikungunya viruses [positive results IgM > 1.1 relative units (RU)/mL, IgG > 22 RU/mL].

Descriptive and analytical statistical methods were used for statistical data processing. Quantitative characteristics are presented as mean ± standard deviation, while absolute numbers with percentages were used for categorical characteristics. The Chi-square test was used to determine the statistically significant difference in the distribution of categorical characteristics, and the Student's *t*-test was used to determine the difference in quantitative characteristics. The program package SPSS version 26.0.0.0 (IBM Corporation, USA) was used for statistical data processing.

Results

Out of the total number of respondents, 60 were women and 76 were men, and the average age of the members of both rotations was 47.2 years. The presence of specific IgM and IgG antibodies against Dengue, Zika, and Chikungunya viruses is shown in Table 1. Out of the total of 136 returnees, antibodies were found in 48 (35.3%) members. The number of seropositive members for the Dengue virus was 23 (16.9%), for the Zika virus, it was 11 (8.1%), and for the Chikungunya virus, it was 14 (10.3%) members.

The presence of IgM, IgG, or both classes of antibodies against two viruses simultaneously in the same person was also observed. The number of these individuals is shown in Table 2. In two persons seropositive for Dengue and Zika, one was positive for IgM antibodies to both viruses, while the other was IgM positive for Dengue and IgG positive for the Zika virus. In the case of co-infection with Zika and Chikungunya viruses, one person had positive IgM antibodies. Among the four samples seropositive for Dengue and Chikungunya, two had positive IgG antibodies, one IgM, while one was positive for both classes of antibodies. Likewise, two individuals in the second rotation were seropositive for all three infections in IgM antibodies.

Out of the 23 members seropositive for the Dengue virus in both rotations, 8 (34.8%) were female, while 15 (65.2%) were male. No statistically significant difference was found ($p = 0.323$). As for age, the average age of seropositive individuals with the Dengue virus was 45.43 ± 7.95 years, while the average age of seronegative individuals was 47.54 ± 10.09 years, with no statistical significance ($p = 0.347$) (Table 3).

Table 1

Presence of specific IgM and IgG antibodies against Dengue, Zika, and Chikungunya viruses in members of the peacekeeping mission

Rotation	Number of tested	Dengue positive			Zika positive			Chikungunya positive		
		IgM	IgG	Total	IgM	IgG	Total	IgM	IgG	Total
First	66 (48.5)	2 (3.0)	4 (6.0)	6 (9.0)	7 (10.6)	0 (0)	7 (10.6)	3 (4.5)	0 (0)	3 (4.5)
Second	70 (51.5)	12 (17.1)	5 (7.1)	17* (24.2)	3 (4.3)	1 (1.4)	4 (5.7)	7 (10.0)	4 (5.7)	11 (15.7)
Total	136 (100)	14 (10.3)	9 (6.6)	23* (16.9)	10 (7.3)	1 (0.7)	11 (8.1)	10 (7.3)	4 (2.9)	14 (10.3)

Ig – immunoglobulin.
All values are given as numbers (percentages).
Note: * one person had both IgM and IgG antibodies present.

Table 2

Presence of simultaneous seropositivity for Dengue, Zika, and Chikungunya viruses in members of the peacekeeping missions

Rotation	Seropositive			
	Dengue and Zika	Dengue and Chikungunya	Zika and Chikungunya	Dengue, Zika, and Chikungunya
First	2 (3.0)	0 (0)	0 (0)	0 (0)
Second	0 (0)	4 (5.7)	1 (1.4)	2 (3.0)
Total	2 (1.5)	4 (2.9)	1 (0.7)	2 (1.5)

All values are given as numbers (percentages).

Out of the 11 seropositive members for the Zika virus observed in both rotations, 6 (54.5%) were female, while 5 (45.5%) were male, with no statistical significance ($p = 0.467$). In terms of age, the average age of those seropositive for the Zika virus was 49.64 ± 11.90 years, while the average age of those seronegative was 46.97 ± 9.59 years, with no statistical significance found ($p = 0.387$) (Table 4).

Out of the 14 members seropositive for the Chikungunya virus in both rotations, 5 (35.7%) were female, while 9 (64.3%) were male, with no statistically significant difference ($p = 0.504$). As for age, the average age of those seropositive for the Chikungunya virus was 46.29 ± 8.93 years, while the average age of those seronegative was 47.29 ± 9.89 years. In this case, no statistical significance was found ($p = 0.717$) (Table 5).

Table 3

Statistical association of risk factors and preventive measures with Dengue virus seropositivity

Parameters	Dengue virus yes (n = 23)	Dengue virus no (n = 113)	OR (95% CI)	*p*
Gender, n (%)				
female	8 (34.8)	52 (46.0)	ref.	0.323
male	15 (65.2)	61 (54.0)	1.60 (0.63–4.07)	
Age (years), mean ± SD	45.43 ± 7.95	47.54 ± 10.09	/	0.347
Number of days in the mission, mean ± SD	207.74 ± 29.55	221.76 ± 31.73	/	0.053
Symptoms, n (%)	9 (39.1)	16 (14.2)	3.90 (1.45–10.49)	0.005
Mosquito bite, n (%)	20 (87.0)	80 (70.8)	2.75 (0.77–9.89)	0.109
Repellents for personal protection, n (%)	22 (95.7)	100 (88.5)	2.86 (0.36–23.02)	0.303
Room protection, n (%)	22 (95.7)	105 (92.9)	1.68 (0.20–14.09)	0.631
Impregnation of uniforms, n (%)	8 (34.8)	50 (44.2)	0.67 (0.26–1.71)	0.403

SD – standard deviation; n – number; OR – odds ratio; CI – confidence interval; ref. – reference.

Table 4

Statistical association of risk factors and preventive measures with Zika virus seropositivity

Parameters	Zika virus yes (n = 11)	Zika virus no (n = 125)	OR (95% CI)	*p*
Gender, n (%)				
female	6 (54.5)	54 (43.2)	ref.	0.467
male	5 (45.5)	71 (56.8)	0.63 (0.18–2.19)	
Age (years), mean ± SD	49.64 ± 11.90	46.97 ± 9.59	/	0.387
Number of days in the mission, mean ± SD	231.0 ± 26.51	218.34 ± 32.02	/	0.206
Symptoms, n (%)	4 (36.4)	21 (16.8)	2.83 (0.76–10.54)	0.108
Mosquito bite, n (%)	11 (100)	89 (71.2)	/	/
Repellents for personal protection, n (%)	9 (81.8)	113 (90.4)	1.82 (0.37–8.84)	0.452
Room protection, n (%)	10 (90.9)	117 (93.6)	0.68 (0.08–6.03)	0.731
Impregnation of uniforms, n (%)	5 (45.5)	53 (42.4)	0.06 (0.01–0.23)	< 0.001

For abbreviations, see Table 3.

Table 5

Statistical association of risk factors and preventive measures with Chikungunya virus seropositivity

Parameters	Chikungunya virus yes (n = 14)	Chikungunya virus no (n = 122)	OR (95% CI)	*p*
Gender, n (%)				
female	5 (35.7)	55 (45.1)	ref.	0.504
male	9 (64.3)	67 (54.9)	1.48 (0.47–4.67)	
Age (years), mean ± SD	46.29 ± 8.93	47.29 ± 9.89	/	0.717
Number of days in the mission, mean ± SD	206.21 ± 26.87	220.89 ± 31.97	/	0.101
Symptoms, n (%)	2 (14.3)	23 (18.9)	0.72 (0.15–3.43)	0.676
Mosquito bite, n (%)	11 (78.6)	89 (73.0)	1.36 (0.36–5.18)	0.652
Repellents for personal protection, n (%)	13 (92.9)	109 (89.3)	1.55 (0.19–12.84)	0.682
Room protection, n (%)	14 (100)	113 (92.6)	/	/
Impregnation of uniforms, n (%)	7 (50.0)	51 (41.8)	1.39 (0.46–4.21)	0.557

For abbreviations, see Table 3.

The frequency of seropositive respondents to arboviruses with the use of preventive measures is shown in Tables 3–5. Preventive measures include the use of repellents for personal protection, the protection of rooms from mosquitoes (use of repellents for rooms and protective nets on windows and beds), and the impregnation of uniforms.

Out of the 23 returnees seropositive for the Dengue virus, 22 (95.7%) reported daily use of repellents for personal use (OR: 2.86; 95% CI: 0.36–23.02; $p = 0.303$). Protection of rooms with repellent and use of protective nets on windows and beds was performed by 22 (95.7%) out of 23 seropositive returnees (OR: 1.68; 95% CI: 0.20–14.09; $p = 0.631$). Eight (34.8%) seropositive returnees impregnated their uniform with repellent (OR: 0.67; 95% CI: 0.26–1.7; $p = 0.403$) (Table 3).

Out of the 11 Zika seropositive returnees, 9 (81.8%) reported daily use of repellent for personal use (OR: 1.82; 95% CI: 0.37–8.84; $p = 0.452$). Protection of rooms with repellent and use of protective nets on windows and beds was performed by 10 (90.9%) out of 11 seropositive returnees (OR: 0.68; 95% CI: 0.08–6.03; $p = 0.731$). Five (45.5%) seropositive returnees impregnated their uniforms with repellent (OR: 0.06; 95% CI: 0.01–0.23; $p < 0.001$). Out of the 125 seronegative members, 53 (42.4%) impregnated their uniforms. In this case, the statistical significance of the association between the use of the uniform impregnation measure to prevent the Zika virus infection was found (Table 4).

Out of the 14 seropositive Chikungunya returnees, 13 (92.9%) reported using repellents daily for personal use (OR: 1.55; 95% CI: 0.19–12.84; $p = 0.682$). All 14 (100%) seropositive returnees used repellents and protective nets on windows and beds. Seven (50.0%) seropositive returnees impregnated their uniforms with repellent (OR: 1.39; 95% CI: 0.46–4.21; $p = 0.557$) (Table 5).

The frequency of Dengue, Zika, and Chikungunya seropositivity with the number of days spent in PKM is shown in Tables 3–5. Returnees from PKM seropositive for the Dengue virus spent 207.74 ± 29.55 days in the mission ($p = 0.053$), indicating a result at the limit of statistical significance. Seronegative returnees spent 221.76 ± 31.73 days in the mission (Table 3).

Seropositive PKM returnees for the Zika virus spent 231.00 ± 26.51 days in the mission ($p = 0.206$), which is not statistically significant. Seronegative returnees spent 218.34 ± 32.02 days in the mission (Table 4).

The Chikungunya virus seropositive PKM returnees spent 206.21 ± 26.87 days in the mission ($p = 0.101$). Again, we did not reach statistical significance in this case. Seronegative returnees spent 220.89 ± 31.97 days in the mission (Table 5).

Based on the questionnaire, Table 6 shows the most common symptoms in the arbovirus seropositive individuals. Out of the total of 23 members seropositive for the Dengue virus, 9 (39.1%) reported symptoms during their stay in the mission. The same symptoms were reported by 16 (14.2%) of the 113 seronegative members of PKM (OR: 3.90; 95% CI: 1.45–10.49; $p = 0.005$) (Table 3).

Out of the total of 11 members seropositive for the Zika virus, 4 (36.4%) reported symptoms during their stay in the mission. The same symptoms were reported by 21 (16.8%) out of the 125 seronegative PKM members (OR: 2.83; 95% CI: 0.76–10.54; $p = 0.108$) (Table 4).

Out of the total of 14 members seropositive for the Chikungunya virus, 2 (14.3%) reported symptoms during their stay in the mission. The same symptoms were reported by 23 (18.9%) of 122 seronegative PKM members (OR: 0.72; 95% CI: 0.15–3.43; $p = 0.676$) (Table 5).

Discussion

Viral infections transmitted by arboviruses are increasingly common diseases worldwide. Vegetation, temperature, and precipitation are some of the factors that influence the arthropod vector and its distribution. In recent years, social, demographic, and climatic changes, population migration, and urbanization have strongly influenced the spread of infections with these viruses, which are of increasing global concern [3, 11].

A large number of infected people on the African continent go unrecognized because of the insufficiently developed health system. More precisely, there is a lack of laboratories and a shortage of health personnel to diagnose a disease. Infected people are often discovered as imported cases through subsequent tests when they return from an endemic area [17]. This is also largely the case with returnees from PKM. Establishing the correct diagnosis is essential for preventing complications, severe clinical manifestations, and chronic forms of the disease.

In our research, 48 (35.3%) returnees were seropositive for arboviruses, i.e., one in three respondents had contact with mosquitoes infected with Dengue, Zika, or Chikungunya viruses. When answering the epidemiological questionnaire questions, the members themselves often stated that there were many mosquitoes during their stay in the CAR. Most members were seropositive for the Dengue

Table 6

The presence of the most common symptoms in seropositive members of the peacekeeping missions (two rotations)

Rotation	Symptoms						Total
	fever	headache	weakness	muscle and bone pain	skin rash	red and itchy eyes	
First	2 (4.17)	1 (2.08)	3 (6.25)	2 (4.17)	1 (2.08)	0 (0)	9 (18.75)
Second	5 (10.41)	5 (10.41)	3 (6.25)	3 (6.25)	0 (0)	2 (4.17)	18 (37.50)
Total	7 (14.58)	6 (12.50)	6 (12.50)	5 (10.42)	1 (2.08)	2 (4.17)	27 (56.25)

All values are given as numbers (percentages).

virus, 23 (16.9%), slightly fewer for the Chikungunya virus, 14 (10.3%), and the fewest members were seropositive for the Zika virus, 11 (8.1%). Our results show that the number of antibodies identified for Dengue and Chikungunya viruses increased approximately 3-fold and 3.7-fold, respectively, in the second rotation compared to the first rotation, while it decreased approximately 2-fold for the Zika virus. In addition, for all three arboviruses, more subjects were found to have positive IgM antibodies (34 subjects) compared to IgG antibodies detected (15 subjects), which may indicate that there is a failure in the use of preventive measures in PKM members towards the end of their stay in the mission.

The Dengue virus infections have occurred more frequently in soldiers and military personnel around the world. The United States (US) military is frequently involved in a variety of PKM. Therefore, the problem of the growing risk of Dengue fever among these members has been recognized, which leads to a significant increase in medical costs and potential impact on combat readiness, jeopardizing and compromising the potential success of these operations. A study conducted in 2017 investigated the presence of antibodies in members of PKM deployed in an endemic area for the Dengue virus. Overall, one thousand samples were analyzed, and neutralizing antibodies against all four serotypes were detected in 7.6% of the subjects. The study showed that better epidemiologic surveillance and rapid determination of the correct infection diagnosis are necessary for preventing infections [18].

In a study conducted on Mongolian PKM members deployed in South Sudan between 2012 and 2013, 632 members were tested for possible seroconversion of the Chikungunya virus. Although there are areas on the South Sudan borders where the Chikungunya virus is endemic, with a low prevalence rate (< 2%), no sample showed seroconversion for this virus [19].

In another study conducted in Central America in 2017, different results were obtained. A cohort of 124 Dutch soldiers was deployed to Belize, Curacao, and Saint Martin for an average of eight weeks each. The soldiers were tested for signs and symptoms of the Chikungunya infection at least fourteen days after their return. The 19 members were tested, and virus-specific IgG antibodies were detected in one member who was on Saint Martin, where an epidemic of this virus was reported in 2013 and 2014 [20, 21].

Few papers in the literature investigate the presence of the Zika virus in PKM participants. One of the few published studies involved 1,420 members of the US military who were in areas where this virus is endemic. ELISA and polymerase chain reaction (PCR) tests were performed on all members. Eleven (0.8%) participants were positive for the Zika virus by PCR test, while 26 (1.8%) were positive by serologic analysis [22].

Dengue, Zika, and Chikungunya viruses are usually detected by laboratory tests – ELISA and PCR. The cross-reactivity between Dengue and Zika viruses, which belong to the same *Flaviviridae* family, can be problematic in serological tests. There is also cross-reactivity between Chikungunya and other alphaviruses. A definitive diagnosis is made using PCR tests, but only in acute infections due to the short duration of viremia [11, 23].

In the first rotation, two members were seropositive for Dengue and Zika viruses. In the second rotation, there was no such combination. Still, four people were seropositive for Dengue and Chikungunya viruses, one for Zika and Chikungunya viruses, and two for all three viruses simultaneously. A similar phenomenon was observed in a retrospective study conducted by a hospital in the Indian district of Kolhapur. Kolhapur district is an endemic area for Dengue, where recurrent epidemics occur annually. The aim of the study was to determine the seropositivity rate for Dengue and Chikungunya infections. The study lasted from January 2021 to August 2022, during which time 3,285 samples were tested for IgM antibodies against Dengue, and 1,823 samples were tested for IgM antibodies against Chikungunya. The antibody positivity for Dengue was 29.4%, and 18.4% for Chikungunya. No significant increase in Dengue seropositivity was observed (29% in 2021, up to 30% in 2022), but 17 samples in 2021 and 12 samples in 2022 were positive for both Dengue and Chikungunya [24].

If we look at the distribution of seropositivity by gender and age, our results show no significant correlation. Studies around the world linking gender and age to seropositivity are conducted with a large number of respondents in all age groups from 6 to 65 years [2, 3, 11]. As our study refers to a military population with a small age range and a similar representation of both sexes, obtaining such data was impossible. Similar to our results, a cross-sectional study of 1,003 respondents in northeastern Tanzania did not identify gender or age as statistically significant predictors of seropositivity. In this study, the influence of risk factors on the incidence of Dengue and Chikungunya virus infections was investigated based on the seropositivity of the subjects. The results showed that environmental factors such as living in a house with uncovered containers had a higher probability of Chikungunya IgM positivity (OR 2.89; 95% CI: 1.76–4.76). People who keep ungulates at home or live near lush vegetation are also at a higher risk of being infected. Similar to our results, the analysis by gender and age in this study showed no statistically significant impact on infection risk [25].

The symptoms and clinical manifestations caused by Dengue, Zika, and Chikungunya viruses can be similar. Some symptoms are not specific to these infections alone, so a number of other infectious diseases can also be suspected. The possibility of co-infections and potential serologic cross-reactions should also not be ruled out. Our results show that out of the 48 seropositive members, 27 (56.25%) reported some of the symptoms characteristic of Dengue, Zika, and Chikungunya virus infections. Had the PKM members been tested when the symptoms appeared, a laboratory analysis would have provided a definitive diagnosis. Unfortunately, these analyses were not carried out during the PKM, so making an accurate diagnosis is impossible. Some of the symptoms are likely due to other infections, most commonly malaria, coronavirus disease 2019, or West Nile virus.

During the outbreak of Chikungunya infections in 2006 and 2007 on the island of La Reunion (in the Indian

Ocean, French territory), it was shown that the risk of infection for French military police officers was equivalent to that of the local population. Out of 770,000 inhabitants, 35.0% were infected in the first six months. The French military police formed a cohort of 662 respondents. They were young or middle-aged men, with an average age of 40. Based on the questionnaires they completed, 23.9% reported symptoms related to the infection. Objectively, 19.3% had IgM or IgG antibodies or both simultaneously, which is almost twice as high as our results (10.3%) [21, 26]. Among the seropositives in the aforementioned study, 3.2% of asymptomatic cases of infection were recorded, whereas in our study, the number of asymptomatic infections was significantly higher and amounted to 14.3%.

If we look at the length of stay of PKM members in correlation with seropositivity, there is no statistically significant difference for any arbovirus. Both rotations spent some time in the CAR in both the rainy and dry seasons, so this risk factor was equal for both rotations, considering that the peak of the mosquito population is in the rainy season. The total number of seropositive members in the first rotation was 16 (24.2%), while in the second rotation, the number of seropositive members was twice as high, 32 (45.7%). The first rotation lasted eight months, i.e., two months longer than the second one. Therefore, an opposite result would have been expected due to the duration of exposure. This leads to the conclusion that other risk factors have a much greater influence than the time spent in the risk area.

In contrast to our results, studies of Dengue seroprevalence in US soldiers serving in Puerto Rico correlate to some extent with duration of exposure and seropositivity. The study used sera from 500 US soldiers serving in Puerto Rico from January through July 2015, collected from the military blood bank. The study was stratified by age and place of birth/residence in endemic areas or non-endemic areas prior to residing in Puerto Rico (87.2% of respondents were born or lived in endemic areas). The study showed that respondents who were not born/lived in endemic areas before arriving in Puerto Rico had a very low risk of being exposed to the Dengue virus [adjusted odds ratio (aOR) = 0.28, $p = 0.001$]. Among them, the risk of exposure to the virus was observed to increase with each year of military service in Puerto Rico (aOR = 1.58, $p = 0.06$). The correlation with age was not significant. Among respondents who were born/lived in endemic areas before coming to Puerto Rico, it was observed that age (aOR = 1.04, $p = 0.02$) and not the duration of military service was associated with the occurrence of Dengue seropositivity. This indicates previous lifetime exposure to the virus [27].

Preventive measures to prevent contact of mission members with the vector include personal protection measures using repellents for personal use, protection of rooms from mosquitoes (use of repellents for the rooms, use of protective nets on windows and beds), and wearing a uniform impregnated with a 0.1% permethrin solution [11]. Further measures to reduce the number of mosquitoes in the area of the camp where the mission members are staying are implemented by the Prevention Department team. These are general hygiene controls, drinking water controls, kitchen controls, proper waste disposal controls, disinfection, disinsection, and pest control measures, both in the camp and the hospital. The presence of animals in the warehouse could also be a risk factor. It has been proven that the presence of hoofed animals (goats, sheep, cows) and lush vegetation near houses is associated with the proliferation of mosquitoes. A very important risk factor is water retention, like rainwater on plants and artificial vegetation, where mosquitoes lay their eggs. Containers with stagnant water, like decorative containers with water, are also common places where mosquito larvae can be found. Places with improper disposal of waste and food are abundant with mosquitoes [28].

The statistical analysis of risk factors and preventive measures in relation to arbovirus seropositivity showed no significant associations in any category, except for the use of uniform repellent impregnation, which was significantly associated with a reduced risk of Zika virus infection. A cross-sectional study conducted in northeastern Tanzania yielded similar results. Of the risk factors, they examined the influence of the use of nets around the bed ($p = 0.63$), the use of nets on the windows ($p = 0.18$), and the number of people sleeping in the room ($p = 0.80$ for two people, $p = 0.13$ for three people, $p = 0.80$ for four or more people). No statistical significance was demonstrated in their study either [25].

Another study tracked the association between the seroprevalence of IgG antibodies to Dengue, Chikungunya, and West Nile viruses and their association with risk factors in Madagascar from 2011 to 2013. The study showed that the risk factors for the occurrence of seropositivity for Dengue and Chikungunya are living near lush vegetation and forests, and daily work in the fields. A protective effect for Chikungunya was exerted by a program that included spraying houses against mosquitoes in the last twelve months ($p < 0.001$) [28].

Considering the potential exposure of our members of PKM to arboviruses in endemic and high epidemiologic risk areas, and based on the results of other investigations, it is necessary in the coming period to strengthen epidemiologic surveillance measures and expand the scope of research to other infectious agents for a longer period of time. In addition to the aforementioned personal and general protection measures, in order to achieve an even better prevention effect, the following must be done: paying attention to the education of the defence personnel about risk factors and the method and importance of protection; strengthening epidemiological surveillance in the warehouses and hospitals; removing animals, dense vegetation, ornamental shrubs, and artificial plants from the warehouses; keeping water containers and flower pots outside of the warehouses and hospitals. In the future, one could think about introducing a vaccine against the Dengue virus. As Dengue and Zika viruses have become increasingly widespread in our geographical area in recent years, a seroepidemiological study should be carried out on their presence in our country and the risks to the military population.

Conclusion

The members of the peacekeeping missions in the Central African Republic have come into contact with Dengue, Zika, and Chikungunya viruses. This shows a need for improving prevention and protection of their health, including serologic testing for the presence of various infectious microorganisms in this endemic area. Our research shows that impregnating uniforms with a repellent plays an important role in preventing mosquito bites and infection with the Zika virus.

Acknowledgement

The investigations are part of the research project MFVMA1/20-24 funded by the University of Defence, the Republic of Serbia.

R E F E R E N C E S

1. *Maia LJ, Oliveira CH, Silva AB, Souza PAA, Müller NFD, Cardoso JDC,* et al. Arbovirus surveillance in mosquitoes: Historical methods, emerging technologies, and challenges ahead. Exp Biol Med 2023; 248(22): 2072−82.
2. *Santos LLM, de Aquino EC, Fernandes SM, Ternes YMF, Feres VCR.* Dengue, chikungunya, and Zika virus infections in Latin America and the Caribbean: a systematic review. Rev Panam Salud Publica 2023; 47: e34.
3. *Cortes N, Lira A, Prates-Syed W, Dinis Silva J, Vuitika L, Cabral-Miranda W,* et al. Integrated control strategies for dengue, Zika, and Chikungunya virus infections. Front Immunol 2023; 14: 1281667.
4. *Kok BH, Lim HT, Lim CP, Lai NS, Leow CY, Leow CH.* Dengue virus infection - a review of pathogenesis, vaccines, diagnosis and therapy. Virus Res 2023; 324: 199018.
5. *Tayal A, Kabra SK, Lodha R.* Management of Dengue: An Updated Review. Indian J Pediatr 2023; 90(2): 168–77.
6. *Bhandari V, Taksande AB, Sapkale B.* Disease Transmission and Diagnosis of Zika Virus. Cureus 2023; 15(11): e49263.
7. *Giraldo MI, Gonzalez-Orozco M, Rajsbaum R.* Pathogenesis of Zika virus infection. Annu Rev Pathol 2023; 18(1): 181–203.
8. *Bartholomeeusen K, Daniel M, LaBeaud DA, Gasque P, Peeling RW, Stephenson KE,* et al. Chikungunya fever. Nat Rev Dis Primers 2023; 9(1): 17. Erratum in: Nat Rev Dis Primers 2023; 9(1): 26.
9. *De Lima Cavalcanti TYV, Pereira MR, de Paula SO, Franca RFO.* A Review on Chikungunya Virus Epidemiology, Pathogenesis and Current Vaccine Development. Viruses 2022; 14(5): 969.
10. *Cai L, Hu X, Liu S, Wang L, Lu H, Tu H,* et al. The research progress of Chikungunya fever. Front Public Health 2023; 10: 1095549.
11. *Soni S, Gill VJS, Anusheel, Singh J, Chhabra J, Gill GJS,* et al. Dengue, Chikungunya, and Zika: The Causes and Threats of Emerging and Re-emerging Arboviral Diseases. Cureus 2023; 15(7): e41717.
12. *Beckham JD, Tyler KL.* Arbovirus Infections. Continuum (Minneap Minn) 2015; 21(6 Neuroinfectious Disease): 1599–611.
13. *Huang Z, Zhang Y, Li H, Zhu J, Song W, Chen K,* et al. Vaccine development for mosquito-borne viral diseases. Front Immunol 2023; 14: 1161149.
14. *Evans-Gilbert T.* Vertically transmitted chikungunya, Zika and dengue virus infections: The pathogenesis from mother to fetus and the implications of co-infections and vaccine development. Int J Pediatr Adolesc Med 2020; 7(3): 107–11.
15. *Achee NL, Grieco JP, Vatandoost H, Seixas G, Pinto J, Ching-Ng L,* et al. Alternative strategies for mosquito-borne arbovirus control. PLoS Negl Trop Dis 2019; 13(1): e0006822. Erratum in: PLoS Negl Trop Dis 2019; 13(3): e0007275.
16. *Tsalefac M, Hiol FH, Mahe G, Laraque A, Sonwa D, Scholte P,* et al. Climate of Central Africa: past, present and future. In: *De Wasseige C, Tadoum M, Eba'a Atyi R, Doumenge C,* editors. The Forests of the Congo Basin - Forests and climate change. Belgium: Weyrich; 2015. p. 37–52.
17. *Massengo NRB, Tinto B, Simonin Y.* One Health Approach to Arbovirus Control in Africa: Interests, Challenges, and Difficulties. Microorganisms 2023; 11(6): 1496.
18. *Hesse EM, Martinez LJ, Jarman RG, Lyons AG, Eckels KH, De La Barrera RA,* et al. Dengue Virus Exposures Among Deployed U.S. Military Personnel. Am J Trop Med Hyg 2017; 96(5): 1222–6.
19. *Adam A, Seidahmed OM, Weber C, Schnierle B, Schmidt-Chanasit J, Reiche S,* et al. Low Seroprevalence Indicates Vulnerability of Eastern and Central Sudan to Infection with Chikungunya Virus. Vector Borne Zoonotic Dis 2016; 16(4): 290–1.
20. *Gilbert DN, Moore WL Jr, Hedberg CL, Sanford JP.* Potential medical problems in personnel returning from Vietnam. Ann Intern Med 1968; 68(3): 662–78.
21. *Frickmann H, Herchenröder O.* Chikungunya Virus Infections in Military Deployments in Tropical Settings-A Narrative Minireview. Viruses 2019; 11(6): 550.
22. *Sugiharto VA, Widjaja S, Hartman LJ, Williams M, Myers TE, Simons MP.* Zika virus surveillance in active-duty U.S. military and dependents through the Naval Infectious Diseases Diagnostic Laboratory. MSMR 2019; 26(7): 18–23.
23. *Varghese J, De Silva I, Millar DS.* Latest Advances in Arbovirus Diagnostics. Microorganisms 2023; 11(5): 1159.
24. *Haldar I, Kulkarni V, Jangale N, Putta S.* Increased chikungunya seropositivity with Dengue co-infection-trend in recent years. J Cardiovasc Dis Res 2023; 14(4): 1–6.
25. *Kajeguka DC, Msonga M, Schioler KL, Meyrowitsch DW, Syrianou P, Tenu F,* et al. Individual and environmental risk factors for dengue and chikungunya seropositivity in North-Eastern Tanzania. Infect Dis Health 2017; 22(2): 65–76.
26. *Pages F, Faulde M, Orlandi-Pradines E, Parola P.* The past and present threat of vector-borne diseases in deployed troops. Clin Microbiol Infect 2010; 16(3): 209–24.
27. *Pollett S, Kuklis CH, Barvir DA, Jarman RG, Romaine RM, Forshey BM*, et al. The seroepidemiology of dengue in a US military population based in Puerto Rico during the early phase of the Zika pandemic. PLoS Negl Trop Dis 2022; 16(1): e0009986.
28. *Broban A, Olive MM, Tantely ML, Dorsemans AC, Rakotomanana F, Ravalohery JP,* et al. Seroprevalence of IgG Antibodies Directed against Dengue, Chikungunya and West Nile Viruses and Associated Risk Factors in Madagascar, 2011 to 2013. Viruses 2023; 15(8): 1707.

Received on November 27, 2024
Revised on January 21, 2025
Accepted on January 29, 2025
Online First May 2025