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Investigation of the Annual and Periodic Distribution of Malaria Cases in Mogadishu, Somalia: A Retrospective Analysis of Four-Year Data

Mogadişu Somali'de Malarya Olgularının Yıllık ve Dönemsel Dağılımının İncelenmesi: Dört Yıllık Verilerin Retrospektif Analizi

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Abstract

Malaria transmitted mainly by mosquito vectors (female *Anopheles* species) is one of the leading causes of death from infectious diseases in the world. In the fight against malaria, some countries have achieved elimination success with national health policies. However, the efforts of national organizations in the fight against malaria are insufficient in low-income countries and the support of international organizations is needed as a part of the global malaria struggle. Somalia is an easternmost African country, located in the neighboring region which has the highest rates of malaria cases and malaria-related deaths globally, is at risk of malaria epidemics due to its fragile infrastructure. This study was carried out in a tertiary care hospital in Mogadishu and involved the retrospective analysis of 54,748 test results of malaria from 2015 to 2019. The rate of malaria cases tended to decrease over the years, being determined as 4.95%, 0.39%, 0.15%, 0.13%, and 0.1% for 2015, 2016, 2017, 2018, and 2019, respectively. A total of 152 malaria cases were detected, and the incidence of infection was found to be significantly higher in men (0.41%; 107/26,231) than in women (0.16%; 45/28.517) ($p<0.0001$). Standard microscopy and the CareStart Malaria Pf/PAN (HRP2/pLDH) Ag Combo RDT test, which meets the criteria set for rapid diagnostic tests recommended by the World Health Organization, were used for the diagnosis of malaria infections, and the parasite species was identified as *Plasmodium falciparum* in 36.4% (55/151) of the cases. We also observed that malaria cases occurred every month of the year, but there was a remarkable increase in the number of cases for the period between October

2015 and January 2016. In conclusion, our study data show that malaria cases tend to decrease in Somalia as a result of the effective struggles of the national and international health institutions. However, the support of international organizations remains important in achieving the goal of malaria elimination and sustainable struggle with the undesirable effects of the disease in Somalia.

Keywords: Malaria, Rapid diagnostic tests, Elimination, *Plasmodium falciparum*.

Özet

Başlıca sivrisinek vektörler (dişi *Anofel* türleri) aracılığı ile bulaşan sıtma günümüzde dünya genelinde ölüme en sık neden olan enfeksiyon hastalıklarından biridir. Sıtma ile mücadelede bazı ülkeler ulusal sağlık politikaları ile eliminasyon başarısına ulaşmıştır. Bununla beraber kısıtlı kaynaklara sahip ülkelerde ulusal kuruluşların çabaları sıtma ile mücadelede yetersiz kalmakta ve küresel mücadelenin bir parçası olarak uluslararası kuruluşların desteğine ihtiyaç duyulmaktadır. Afrika'nın en doğusundaki bir ülke olan Somali sıtma olgularının ve sıtma ilişkli ölümlerin dünya genelinde en yüksek oranlarda görüldüğü bölgenin komşuluğunda yer alan ve kırılgan altyapısı nedeni ile sıtma salgını için risk altında olan bir ülke konumundadır. Mogadişu'da yerleşik bir üçüncü basamak hastanesinde yürütülen bu çalışmada 2015-2019 yıllarına ait 54.748 sıtma testinin sonucu retrospektif olarak incelendi ve 2015, 2016, 2017, 2018 ve 2019 yılları için sırasıyla %4.95, %0.39, %0.15, %0.13 ve %0.1 olmak üzere vaka sayılarının yıllar içerisinde azalma eğiliminde olduğu saptandı. Toplam 152 sıtma olgusunun saptandığı çalışmada erkeklerde enfeksiyon görülme sıklığı kadınlara göre anlamlı derecede daha yüksek bulundu, erkeklerde %0.41 (107/26,231) ve kadınlarda %0.16 (45/28,517) ($p<0.0001$). Sıtma enfeksiyonu tanısının standart mikroskopi ve Dünya Sağlık Örgütü'nün hızlı tanı testleri için koyduğu kriterleri karşılayan CareStart Malaria Pf/PAN (HRP2/pLDH) Ag Combo RDT testi ile gerçekleştirildiği çalışmada olguların %36.4'ünde (55/151) parazit türü *Plasmodium falciparum* olarak tanımlandı. Sıtma olgularının yılın her ayında ortaya çıktığını gözlemlediğimiz çalışmada, 2015 Ekim ve 2016 Ocak ayları arasını kapsayan bir dönemde olgu sayılarında dikkat çeken bir yükselme belirlendi. Sonuç olarak çalışma verilerimiz sıtma olgularının ulusal ve uluslararası sağlık kuruluşların etkin mücadeleleri ile Somali'de azalma eğilimde olduğunu göstermektedir. Bununla beraber, Somali'de sıtma eliminasyonu hedefine ulaşmada ve hastalığın olumsuz etkileri ile sürdürülebilir mücadelede uluslararası kuruluşların sağladığı desteğin kritik öneme sahip olduğu görülmektedir.

Anahtar Kelimeler: Sıtma, Hızlı tanı testi, Eliminasyon, *Plasmodium falciparum*.

Introduction

Malaria is a vector-borne tropical infectious disease that develops with female anopheles mosquitoes carrying the *Plasmodium* genus parasite inoculate parasitic sporozoites to humans during their feeding [1,2]. Of more than 120 *Plasmodium* species that infect mammals, birds, and reptiles, only six (*Plasmodium falciparum*, *P. knowlesi*, *P. malariae*, *P. ovale wallickeri*, *P. ovale curtisi*, and *P. vivax*) are known to infect humans regularly [3,4]. Certain common features are shared in malaria infections (Figure 1), but epidemiological features, disease pathogenesis and diagnosis, and treatment strategies may differ for infections caused by each species. *P. falciparum* and *P. vivax* pose the greatest threat, causing more than 95% of all human malaria infections, but all species can result in serious illness and death [5].

According to the World Health Organization (WHO) data, more than 80% of the global population lives in areas at risk of at least one major vector-borne disease and more than 700,000 people die each year from vector-borne diseases, including Chagas disease, malaria, dengue fever, schistosomiasis, leishmaniasis, yellow fever, and Japanese encephalitis [6]. Although the distribution area of mosquito vectors for malaria covers a very wide geography, according to the WHO 2019 estimated data, 215 million of the 229 million cases worldwide (94%) are located in the WHO-Africa Region [7]. In 2019, Nigeria (27% of all cases), Democratic Republic of Congo (12%), Uganda (5%), Mozambique (4%), and Niger (3%) had the highest number of cases, and these five African countries accounted for approximately 51% of all global cases [7]. Malaria is also observed as an

important public health problem in other parts of the world. Other countries where malaria cases are most commonly seen include India (20 million cases per year) in the Asia-Pacific region, Papua New Guinea in the West-Pacific region, and Brazil, Colombia, and Venezuela in South America [7].

Globally, the number of malaria cases decreased from 238 million in 2000 to an estimated 229 million in 2019 in 87 countries where malaria is endemic [7]. As a result of comprehensive control strategies, malaria-related deaths decreased steadily worldwide between 2000 and 2019, from 736,000 in 2000 to 409,000 in 2019 [7]. As another reflection of these data, malaria-related deaths in Africa are estimated to have decreased by 44% from 2000 to 2019 [2]. However, malaria continues to be one of the leading infectious diseases, maintaining its importance in the world, and elimination is aimed with global control strategies [8]. The essential path to malaria elimination strategies can be summarized under four topics: (i) vector control (global surveillance, insecticides, environmental improvement), (ii) preventive measures for disease transmission (chemoprophylaxis, vaccine strategies, personal protective measures), (iii) diagnosis of all patients, including suspected cases (performing rapid diagnostic tests in addition to quality-assured microscopy), and (iv) disease treatment strategies [2,7]. While antimalarial treatment applications, are of critical importance in reducing disease progression and mortality rates, they are at risk of being interrupted due to drug resistance problems; and therefore in the fight against malaria, new tools of biotechnology, such as new drugs that can kill gametocytes are being tested and developed to prevent transmission and preventive vaccine studies and research on genetically modified mosquitoes are being conducted [6,9,10].

This study aimed to investigate the incidence of malaria infections and the change in the number of cases over the years in Somalia, where literature data on malaria epidemiology and risk factors are limited.

Material and Method

The study was conducted after obtaining approval from the institutional ethics committee (Ethics Committee of Mogadishu Somalia-Turkey Recep Tayyip Erdoğan Training and Research Hospital, date: 05.12.2019, decision no: 180, number: MSTH/2721). The study group consisted of patients who presented to our hospital for malaria screening or those who voluntarily participated in malaria screening, The personal information of all participants was protected with confidentiality, and the study was conducted in accordance with the principles of the Declaration of Helsinki.

Study group and design

In the study, all malaria tests reported for all suspected cases were retrospectively evaluated over the period between June 2015 and November 2019 in the Medical Microbiology Laboratory of Mogadishu Somalia-Turkey Recep Tayyip Erdoğan Training and Research Hospital. The same test results of the same person within a week were accepted as a single test (including people with both microscopy and rapid test results in a week). Individuals that were found to be positive based on at least one of the parasite screening methods (quality-assured microscopy or a rapid diagnostic test) and have clinical findings consistent with malaria were evaluated as malaria cases.

Microscopic evaluation

A thin smear and a thick film were prepared from the blood samples of the patients with suspected malaria and stained with 5% Giemsa. After the smears were dried in dry hot air for 30 minutes, the slides were examined by an infectious diseases or medical microbiology specialist using immersion oil under a x100 lens.

Malaria rapid diagnostic test

The CareStart Malaria Pf/PAN (HRP2/pLDH) Ag Combo RDT (AccessBio, Somerset, New Jersey, USA) test, which was included in the in vitro diagnosis list of WHO on May 28, 2015, was used as a malaria rapid screening test. This test is designed for the diagnosis of *P. falciparum*, *P. vivax*, *P. malariae*, and *P. ovale* infections, which are four common malaria species. The CareStart™ Malaria HRP2/pLDH (Pf/PAN) Combo RDT assay contains a membrane strip coated with two monoclonal antibodies in two separate lines. One of the monoclonal antibodies is a common (PAN specific) antibody specific for the pLDH of four

Plasmodium species (*P. falciparum*, *P. vivax*, *P. malariae*, and *P. ovale*). The other line contains a monoclonal antibody specific to the HRP2 of *P. falciparum*. Antibodies specific to the pLDH of PAN and HRP2 of *P. falciparum* are distributed on the conjugate pad as absorbed onto gold particles. In the WHO evaluation, the *P. falciparum* panel detection score of the test was determined as 90% at 200 parasites/µl, and the *P. vivax* panel detection score was 94.3% at 200 interference/µl [11,12]. The CareStart™ malaria HRP2/pLDH (Pf/pan) combo malaria diagnostic kit has been reported to provide results comparable to the gold standard method, microscopy [13].

Statistical analysis

At the end of the study, frequency, mean and standard deviation values were calculated, and comparisons were performed using the chi-square and/or Fisher's exact probability test. A p value of <0.05 was considered statistically significant (at the 95% confidence interval). All analyses were undertaken using SPSS v. 22.0 (IBM SPSS Statistics Version 22.0., IBM Corp., Armonk, New York, USA).

Results

A total of 54,748 test results were evaluated in the present study. The mean age of the participants was 34.9±22.9 years, with a range of 1 to 97 years. The positivity rate in the entire study group was found to be 0.28% (152/54,748). The rate of positive test results was 0.16% (45/28,517) in females and 0.41% (107/26,231) in males.

Table 1. Distribution of malaria-positive patients according to years, age groups, and gender.

age groups →		1-10		11-20		21-30		31-40		41-50		51-60		61-70		≥71		Total	
years		F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	n (F:M)	%
2015	positive	8	3	1	8	3	10	5	3	3	5	1	3	4	1	1	1	60	4.95
	negative	89	126	60	71	92	104	79	59	88	67	71	72	65	41	32	35	1,151	
2016	positive	2	5	1	5	0	4	2	7	1	0	1	4	0	2	0	1	35	0.39
	negative	768	1,072	506	452	682	672	636	517	649	452	574	470	455	375	380	385	9,045	
2017	positive	2	0	1	3	0	5	0	1	0	2	2	1	2	2	1	0	22	0.15
	negative	1,252	1,791	808	691	1,400	1,378	1,061	913	1,049	726	791	613	724	574	550	590	14,911	
2018	positive	1	2	2	1	0	2	1	4	0	1	0	2	0	4	0	2	22	0.13
	negative	1,126	1,505	855	714	1,779	1,755	1,360	1,047	1,305	800	1,041	731	781	665	596	546	16,606	
2019	positive	0	4	0	2	0	6	0	1	0	0	0	0	0	0	0	0	13	0.1
	negative	1,037	1,444	798	667	1,347	1,313	974	855	910	580	745	487	564	428	393	341	12,883	
Total 54,748	positive	13	14	5	19	3	27	8	16	4	8	4	10	6	9	2	4	152	0.28
	negative	4,272	5,938	3,027	2,595	5,300	5,222	4,110	3,391	4,001	2,625	3,222	2,373	2,589	2,083	1951	1,897	54,596	

Table 2. Distribution of malaria-positive patients according to years and months.

years		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total (n)	%
2015	positive	-	-	-	-	-	2	1	1	0	10	20	26	60	4.95
	negative	-	-	-	-	-	101	90	229	100	168	78	385	1,151	
2016	positive	12	5	1	0	2	5	4	3	0	0	0	3	35	0.39
	negative	436	719	446	547	863	797	1,310	1,582	412	465	636	832	9,045	
2017	positive	1	1	3	1	2	3	3	1	1	2	4	0	22	0.15
	negative	937	1,202	1,014	856	1,079	1,079	1,371	1,305	1,054	1,233	1,668	2,113	14,911	
2018	positive	0	4	4	2	1	2	3	2	1	0	2	1	22	0.13
	negative	1,666	1,695	1,122	1,727	1,283	828	1,632	1,354	1,662	1,551	1,014	1,072	16,606	
2019	positive	3	0	1	2	0	4	1	0	2	0	0	-	13	0.1
	negative	1,269	1,720	1,004	1,146	1,424	1,354	1,525	1,287	1,276	737	141	-	12,883	
Total 54,748	positive	16	10	9	5	5	16	12	7	4	12	26	30	152	0.28
	negative	4,308	5,336	3,586	4,276	4,649	4,159	5,928	5,757	4,504	4,154	3,537	4,402	54,596	

The incidence of malaria infection was significantly higher in males (p<0.0001). In addition, 18.6% (n= 10,210) of the study group were children aged 10 years and younger and the positivity rate (0.26%; 27/10,210) was similar to the general study group in this group (under 10 years old) (Table 1).

While the rapid diagnosis kit provided a positive result for *P. falciparum* in 36.4% of the samples (55/151), the parasite could not be identified at the species level in the remaining samples. November, December, and January were the months when the number of cases were highest; however, it was observed that malaria cases did occur in all periods of the year (Table 2).

Discussion

The estimated number of malaria cases around the world decreased continuously for five years from 2010 to 2015, with the lowest level being observed as 220 million in 2014 [7]. While it is estimated that there were 218 million predicted malaria cases worldwide in 2015, the number of cases tended to rise again after this date [7]. The "Global technical strategy (GTS) for malaria 2016-2030" project, the details of which are defined by WHO for the global fight against malaria, was accepted by the World Health Assembly in May 2015 and updated in May 2021 [14]. GTS aims to reduce the incidence and mortality rate of malaria by 90% until 2030 based on the 2015 data [7,14]. In our study, we retrospectively analyzed the data on malaria cases in the largest hospital complex located in Mogadishu, operated jointly by the governments of Somalia and Turkey over a 4.5-year period starting from 2015 and observed that the number of cases tended to decrease over the years. We consider that the following positive conditions are among the important reasons for this decrease: (i) joint activities of the Somalia Ministry of Health, Turkish Ministry of Health, WHO, and other international organizations, (ii) free access to diagnostic kits (paid by WHO), (iii) reduction of internal conflict risks, (iv) increase in the possibilities of access to antimalarial drugs in pharmacies in Mogadishu, and (v) development of health institutions and health services in the region in terms of parameters such as "number, capacity, and scope of expertise" [15,16]. An infection prevalence of less than 1% in the population is defined as an indication of the possibility of elimination for national malaria strategies, although this may be misleading as it may be related to the failure of ecological conditions to support transmission [17]. Our study data indicate that malaria infections may occur every month of the year in Somalia. In a remarkable way, relatively high malaria cases were reported from October 2015 until the end of January 2016, indicating that periodic risks may cause an increase in the number of cases in the presence of fragile conditions.

The framework of programs to struggle malaria includes the main headings of preventive measures (chemoprophylaxis, vaccine studies), vector-targeted strategies, and diagnosis and treatment of cases (Figure 1). The WHO data clearly reveal the importance of strong funding support for the continuation of malaria elimination programs, including surveillance studies, supply of diagnostic tests and antimalarial drugs, and vaccine studies [7]. The "Foundation for Innovative New Diagnostics" program of WHO supports the creation of evidence for malaria diagnostic policies by producing regular reports on the quality and performance of rapid diagnostic kits [7]. In this context, rapid diagnosis kits, which were approved by WHO in 2015 and used in our study, are distributed to local health institutions free of charge by the WHO Mogadishu office.

In our study, 17.8% (27/152) of the malaria cases were children under the age of 10 years. A malaria vaccine under study for over 30 years has been reported to provide partial protection against malaria among young African children representing the population most affected by the disease [10]. The first-generation version of this vaccine (RTS,S/AS01, trade name Mosquirix) has been developed against *P. falciparum*, the deadliest malaria parasite worldwide and the most common species in Africa [10]. *P. falciparum* continues its influence in many regions of Africa with varying intensity [17]. Phase 3 clinical tests conducted under the leadership of WHO in seven African countries (Burkina Faso, Gabon, Ghana,

Kenya, Malawi, Mozambique, and the United Republic of Tanzania) have demonstrated the vaccine's potential to prevent malaria and save lives [10]. With the rapid diagnosis kit used in the current study, the presence of *P. falciparum* was identified in 36.4% (55/151) of the tests, indicating that this vaccine is also promising for the Somali population. The Malaria Vaccine Implementation Program, planned by WHO, was established to coordinate and support the routine vaccination under the leadership of the national governments of relevant African countries [10].

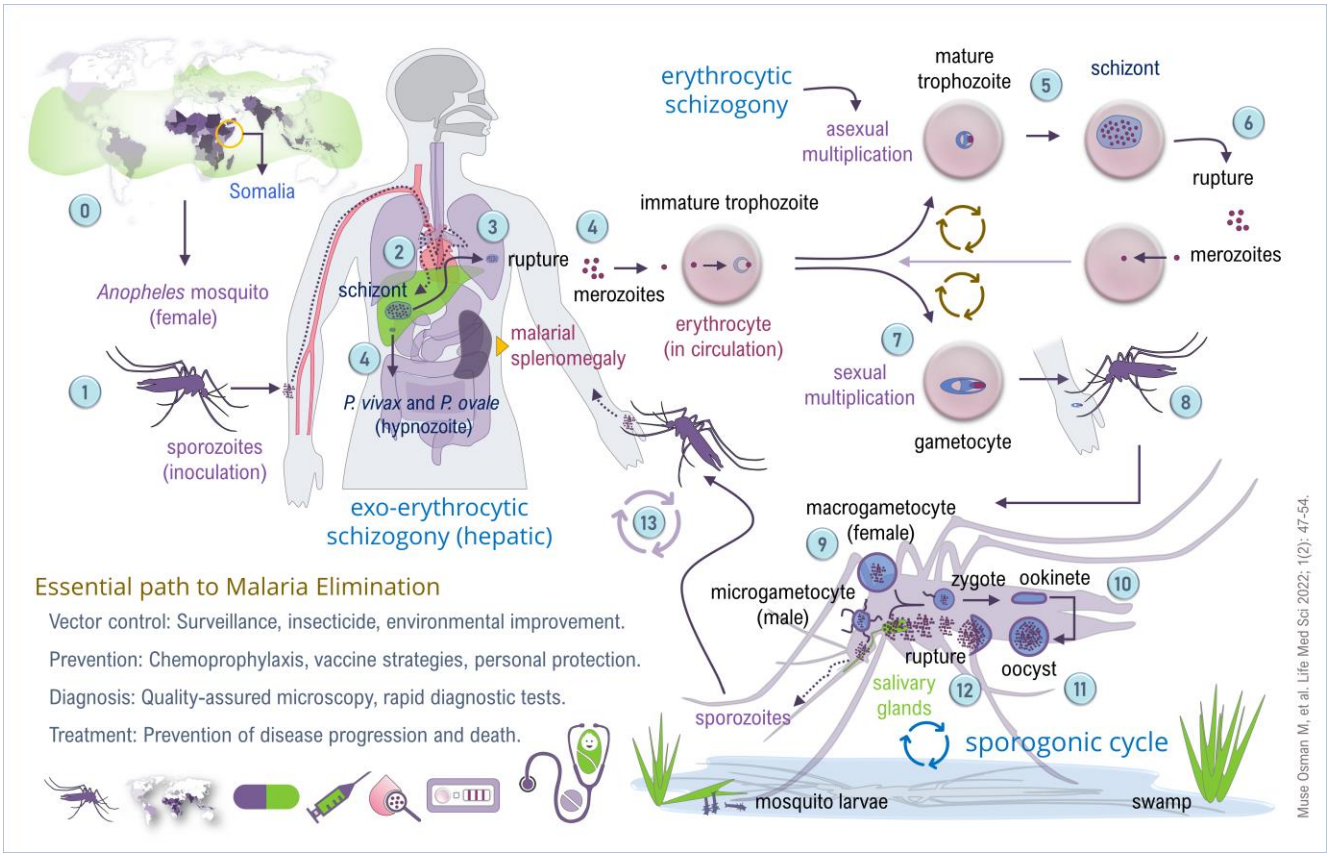

Figure 1. Life cycle of malaria (*illustrated by Fatih Şahiner*). 0. Global spreading of the malaria vector *Anopheles* mosquitoes (green areas) and countries where malaria cases are common. 1. Inoculation of *Plasmodium* spp. parasites in sporozoite form into a person bitten by an infected female anopheles. 2. Sporozoites injected into the bloodstream reach the liver (60 seconds) and reproduce asexually over the next seven to 10 days (exo-erythrocytic schizogony; hepatic schizont). There are no symptoms during this time. 3. Parasites in the form of merozoites exit the liver cells in vesicles and travel from the heart to the capillaries of the lungs. 4. Finally, the vesicles break down and the merozoites are released into the bloodstream (Note: *P. vivax* and *P. ovale* have a dormant stage [hypnozoites] and may persist in the liver and enter the bloodstream weeks or even years later, causing relapses if left untreated) 5. Merozoites attach to and infect erythrocytes and multiply within erythrocytes (asexual multiplication: erythrocytic schizogony; erythrocytic schizont). The ring-stage trophozoites mature into schizonts. The mature schizonts disrupt and release the second-generation merozoites. 6. When cells (erythrocytes) rupture, parasites invade more erythrocytes. *Clinical symptoms, including fever occur simultaneously with the rupture of infected erythrocytes and release of parasite remnants including the malaria pigment (hemozoin) and the putative 'malaria toxin', glycophosphatidylinositol*. 7. In some infected erythrocytes, merozoites enter the sexual erythrocytic stage and transform first into trophozoites, then into schizonts, and then into sexual forms (gametocytes; macrogametocytes-female and microgametocytes-male), which circulate in the bloodstream and are taken up during mosquito bites. 8. Gametocytes are taken up by the mosquito during blood sucking and the mosquito multiplies cycle (sporogonic cycle) of the parasites begins. 9. Gametocytes develop into mature sex cells (gametes) in the mosquito. 10. Microgametes penetrate the macrogametes in the mosquito's stomach and the zygote is formed. Zygotes become motile and elongate (ookinetes) 11. Ookinetes actively penetrate the midgut wall of the mosquito, where they are transformed into oocysts. 12. The oocyst grows, ruptures and releases thousands of active sporozoites. Sporozoites migrate to the salivary glands of mosquitoes. 13. When a mosquito bites another person, the human asexual malaria infection cycle starts again [1,2,7].

In a population at risk for malaria infections, the frequency of being affected by the malarial infection of different gender is determined based on the behaviors and socio-cultural characteristics of women and men in social life, ignoring the immunological, anatomical, and physiological differences associated with male or female gender. In a new study conducted in Ghana in 2021, the data of 203 adult patients were examined, and the rate of women being affected by the infection was found to be approximately twice that of men (126 women and 77 men, respectively; 77 women and 47 men after excluding patients by applying the study criteria of authors) [18]. In the same study, this difference between the men and women was briefly explained by risky behaviors (e.g., the frequency of using insecticide treated nets), which increased the possibility of exposure to mosquito vectors responsible for the transmission of the infectious agent. On the contrary, in our study group (including both adults and children), the incidence of infection in the males was more than two times higher compared to the females [0.41% (107/26,231) vs 0.16% (45/28,517)]. In contrast to Ghana, where the economy is largely dependent on agricultural activities and women actively participate in agricultural activities in wetlands [19], in Somalia, livestock and commercial activities are at the forefront of the economy, and women wear very covered clothes in daily life. It is possible that these behavioral and socio-economic characteristics reduce the risk of infection in women. However, we did not observe any difference between the boys and girls in the group under the age of 10 years [0.24% (14/5,938) vs 0.3% (13/4,272)] (Table 1).

Conclusion

Malaria infection cases in Somalia tend to decrease with the contribution of activities carried out by the National Health authorities and WHO. However, malaria cases being observed any time of the year and the existence of fragile conditions that may cause an epidemic risk reveal the importance of the sustainable fight against malaria and active surveillance in Somalia with the neighboring countries.

Conflict of interest: The authors declare that there is no conflict of interest. The authors alone are responsible for the content and writing of the paper. **Financial disclosure:** There is no financial support for this study. However, the malaria screening kits (rapid diagnosis test) used in our hospital during the study period were provided free of charge by WHO, as is the case with other health institutions in Mogadishu.

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