

Characteristics of Symptomatic and Asymptomatic Patients with COVID-19 and Seroprevalence of Anti-SARS-CoV-2 Antibodies in Zavidovići, Bosnia and Herzegovina

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ABSTRACT

Human coronaviruses are agent which cause respiratory illnesses and have been described to be continuously emerging. Seroprevalence of IgM/IgG antibodies was determined by lateral flow immunoassay. Study were include information about participants who had COVID-19 symptoms in the period of pandemic (including symptoms and health status). Among 443 serum samples for detection seroprevalence, 186 (42.0%) were seropositive on specific antibodies (IgM/IgG) or participants who had COVID-19 with or without symptoms. Of the seropositive the age group 25-50 years old had the highest percentage (32.8%) followed by 51-64 years (30.6%). Ninety seropositive patients (out of 186; 48.4%) were detected with the specific symptoms. The most frequently symptoms were fever (n=54, out of 90; 60.0%), muscle pain (46.7%), dry cough and loss of smell (36.7%) and others. More than one symptoms were detected in 59 cases (65.6%). The most common comorbidities were diabetes mellitus (n=12, out of 186; 6.5%), than hypertension (nine; 4.8%) and heart diseases (seven; 3.8%). More than half of seropositive were asymptomatic (n=96, out of 186; 51.6%). Note: In period March 2020 - June 2021, among 3323 samples, 804 (24.2%) were positive on SARS-CoV-19 with RT-PCR. The results showed that 394 (out of 804; 49.0%) positive samples were collected from female and 410 (51.0%) from male. The most prevalent of SARS-CoV-19 viruses were detected in > 65 years old (n=267, out of 804; 33.2%). For preventive control of the pandemic spread of coronaviruses, we need additional testing and information about symptomatic and asymptomatic patients in the municipality.

Keywords: Coronavirus, comorbidity, seroprevalence, symptoms.

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I. INTRODUCTION

Based on data, the World Health Organisation (WHO) declared the disease COVID-19 as a pandemic [1]. As the cause of mild respiratory infections, coronaviruses were discovered in the 1960s. [2].

There is a lot of reports about transmission of the coronaviruses from asymptomatic to symptomatic patients and it is very worrying [3].

Asymptomatic patients may represent 30-60% of all infections [4], [5]. Asymptomatic infection often ends without reporting because most of affected individuals cannot be identified by screening methods, such as temperature check [6]. In early phases of disease, without symptoms or mild symptoms, high concentration of virus could be detected [7].

SARS-CoV-2 has an incubation period of 2 to 14 days and approximately 80% of those that are infected will show mild or no symptoms [8].

Some serological and surveys of SARS-CoV-2 with or

without symptoms have been reported. Li et al. were reported prevalence of symptomatic or asymptomatic patient in China in 71% and 29% cases [6]. In Republic of Korea, Kim et al. were reported symptomatic patient with the percentage of 81% and asymptomatic patients in 19% [9]. In one study from Bahrain, it was estimated that 23% and 77% of those with COVID-19 have been detected such as symptomatic and asymptomatic patients [10].

Sood *et al.* were reported seroprevalence of SARS-CoV-2 in Los Angeles in 4.65% of cases [11]. In Milan, Italy Valenti *et al.* were reported about seroprevalence in healthy blood donors with the percentage of 2.7% [12]. Low seroprevalence (6%) have been reported in Kenyan, especially among those who do not come into contact with the intermediate hosts [13]. High seroprevalence was reported in Iran, where the IgG antibodies were detected in 22% and 34% [14], [15]. Study from the Uganda showed a higher seroprevalence of antibody in general population, in 87.53% of cases [16]. Generally, the global SARS-CoV-2 seroprevalence (for 60 countries) was

9.5%, the highest seroprevalence estimates were in Central and Southern Asia (23%), sub-Saharan Africa (19%) and Latin America and Caribbean (18%), while lowest seroprevalence was in the Eastern and South-eastern Asia (2%), Northern Africa and Western Asia (9%) and Europe and North America (7%), respectively [17].

In Bosnia and Herzegovina there is no information about seroprevalence of specific antibody on SARS-CoV-19 and characteristics of patient with or without symptoms.

The aim of the study was to determine the prevalence of anti-SARS-CoV-2 antibodies and characteristics of symptomatic and asymptomatic patients.

II. MATERIALS AND METHODS

A. Sample Collection

In the period of March 2021 to April 2021, 433 consecutive, non-duplicate serum samples were collected from participants who did not vaccinate with inactivated SARS-CoV-2 vaccine in Health Center of Zavidovići. The population covered by this institution is 35.988 in Zavidovići municipality of Zenica-Doboj Canton, Bosnia and Herzegovina. Informed permission was obtained from each person. Informed consent included persons demographic characteristics (such as years, males, females, residence, and profession). Also, study was include information about symptoms (such as fever, feverishness, dry cough, sore throat, shortness of breath, muscle pain, loss of smell and taste, diarrhea) and information of health status (diabetes mellitus-DM, hypertension-HTA, cerebrovascular insult-CVI, heart diseases, diseases of blood vessels, malignancy and others).

B. Methods

About 5 ml blood samples were taken from all patients. Immunochromatographic test was used to detect antibodies by using Tigsun COVID-19 Combo IgM/IgG Rapid test. Detection of SARS-CoV-2 IgG antibodies: When a positive sample is detected, the SARS-CoV-2 IgG antibodies in the sample bound with colloidal gold-labeled SARS-CoV-2 antigens to form an immune complex. When the complex passes the Test Line (T), it binds to the anti-human IgG monoclonal

antibody immobilized in the Test Line (T) of the device to form a new complex. This generates a colored test band that indicates a positive result. When the SARS-CoV-2 antibody level in the specimen is zero or below the target cutoff, there is not a visible colored band in the Test Line (T) of the device. This indicates a negative result (<http://www.tigsun.com/Content/2020/08-14/1712513045.html>).

C. Statistical Analysis

Descriptive analyses were reported as mean and percentage for different variables for symptomatic and asymptomatic patients. The chi-square test was used to get the correlations between the variables. All analyses were performed by SPSS version 15.0 software. A *P* value less than 0.05 was considered significant.

III. RESULTS

Overall, 443 participants were included in the study. Among 443, 186 (42 %) were seropositive patients or patients who had COVID-19 with or without symptoms, while 257 (58.0%) were seronegative on virus COVID-19.

The most seropositive samples were detected between 25-50 and 51-64 years old (33% and 31%; out of 186; cumulative inc. 4.7 and 7.5), followed by > 65 years old (26%; cumulative inc. 13.0), respectively. The seropositive patients were mostly from rural residence (54%; out of 186) (Table I).

Of the 186 seropositive samples (participants), 90 (48.4%; 47 female and 43 male) patients were symptomatic and 96 (51.6%; 57 female and 39 male) were asymptomatic (Table II).

The median age was similar between symptomatic and asymptomatic patients (51.5 years; range 1-81 and 55 years; range 1-81). Age group from 25-50 were more likely to be symptomatic than older patients (33; 54.1%), while older age groups (51-61 and > 65 years) were more asymptomatic (Table II). Among symptomatic and asymptomatic patient, 46 (55.4%) and 37 (44.6%) were active worker (Table II).

Twenty-nine (15.6%) patients had at least one comorbidity. In both groups, Diabetes mellitus was the most common comorbidity (6.5%), and hypertension was the second most common one (4.8%) (Table II).

TABLE I: SEROPREVALENCE OF ANTI-SARS-COV-2 ANIBODIES AMONG ZAVIDOVIĆI MUNICIPALITY POPULATION

Characteristic	No (%) of participants					No of population	Cumulative incidence/ 1000
	Total N°	Pos N°	%	Neg N°	%		
Gender							
Females	256	104	55.9	152	59.1	18102	5.7
Males	187	82	44.1	105	40.9	17886	4.6
Total	443	186	100	257	100	35988	5.2
Age (years)							
0-14	29	15	8.1	14	5.4	6075	2.5
15-24	29	4	2.2	25	9.7	5422	0.7
25-50	147	61	32.8	86	33.5	13115	4.7
51-64	131	57	30.6	74	28.8	7611	7.5
≥65	107	49	26.3	58	22.6	3765	13.0
Total	443	186	100	257	100	35988	5.2
Place of residence							
Urban	192	85	45.7	107	41.6	8174	10.4
Rural	251	101	54.3	150	58.4	27814	3.6
Total	443	186	100	257	100	35988	5.2

TABLE II: CHARACTERISTICS OF SYMPTOMATIC AND ASYMPTOMATIC PATIENTS WHO HAD COVID-19

Characteristic (No of participants)	Overall	Presenting characteristics of symptoms	
		Symptomatic	Asymptomatic
Number (%)	186	90 (48.4)	96 (51.6)
Gender			
Female	104 (55.9)	47 (45.2)	57 (54.8)
Male	82 (44.1)	43 (52.4)	39 (47.6)
Age (years)			
Mean	50.5	50.5	50.6
Median	53.5	51.5	55.0
0-14	15 (8.1)	6 (40.0)	9 (60.0)
15-24	4 (2.2)	3 (75.0)	1 (25.0)
25-50	61 (32.8)	33 (54.1)	28 (45.9)
51-64	57 (30.6)	27 (47.4)	30 (52.6)
≥65	49 (26.3)	21 (42.9)	28 (57.1)
Place of residence			
Urban	85 (45.7)	48 (56.5)	37 (43.5)
Rural	101 (54.3)	42 (41.6)	59 (58.4)
Occupation			
Active worker	83 (44.6)	46 (55.4)	37 (44.6)
Unemployed	2 (1.1)	1 (50.0)	1 (50.0)
Retired	38 (20.4)	18 (47.4)	20 (52.6)
Student	9 (4.8)	4 (44.4)	5 (55.6)
Kids	9 (4.8)	4 (44.4)	5 (55.6)
House person	45 (24.2)	17 (37.8)	28 (62.2)
Chronic diseases			
DM	12 (6.5)	5 (41.7)	7 (58.3)
HTA	9 (4.8)	4 (44.4)	5 (55.6)
CVI	2 (1.1)	0	2 (100.0)
Heart diseases	7 (3.8)	2 (28.6)	5 (71.4)
Diseases of blood vessels	2 (1.1)	0	2 (100.0)
Oncological	5 (2.7)	3 (60.0)	2 (40.0)
More than two	8 (4.3)	3 (37.5)	5 (62.5)

TABLE III: CHARACTERISTICS OF SYMPTOMATIC PATIENTS IN THE STUDY

Characteristic	No (%) of participants	
	Total N° of positive participants	% of total number
Type of symptoms		
Fever	54	60.0
Feverishness	22	24.4
Sore throat	16	17.8
Dry cough	33	36.7
Shortness of breath	20	22.2
Muscle pain	42	46.7
Loss of smell	33	36.7
Loss of taste	30	33.3
Diarrhea	8	8.9
More than two symptoms	59	65.6
Total participants with symptoms	90	100

Ninety seropositive patients (48.4%; out of 186) were detected with the specific symptoms. The most frequently symptoms were fever (60%; out of 90), muscle pain (46.7%), dry cough and loss of smell (36.7%) and others (Table III).

NOTE: From the beginning of pandemic March 2020 to June 2021, a real-time reverse transcriptase-polymerase chain reaction (RT-PCR) test for SARS-CoV detection was performed using a nasopharyngeal swab in Zavidovići municipality in total of 3323 cases. Among 3323 patients, 804 (24%, cumulative inc. 22.3) were positive on COVID-19. A total of positive samples, 51% were males (cumulative inc. 22.9). The most positive samples were detected in > 65 years

old (33%; cumulative inc. 70.9) followed by 25-50 and 51-64 years old (31% in both age groups; cumulative inc. 18.8 and 32.5), respectively (Table IV).

TABLE IV: RESULTS OF SARS-COV-2 PCR IN THE PERIOD MART 2020 - JUNE 2021

Characteristic (No of participants)	No (%) of patients				No of population	Cumulative incidence/1000
	Positive	Negative	Intermediate	No data		
Gender						
Females (1402)	394 (49.0)	983 (39.9)	7 (29.2)	18 (60.0)	18102	21.8
Males (1921)	410 (51.0)	1482 (60.1)	17 (70.8)	12 (40.0)	17886	22.9
Total (3323)	804 (24.2)	2465 (74.2)	24 (0.7)	30 (0.9)	35988	22.3
Age						
0-14 (100)	9 (1.1)	91 (3.7)	0	2 (6.7)	6075	1.5
15-24 (223)	34 (4.2)	189 (7.7)	1 (4.2)	1 (3.3)	5422	6.3
25-50 (1254)	247 (30.7)	1007 (40.9)	6 (25.0)	8 (26.7)	13115	18.8
51-64 (894)	247 (30.7)	647 (26.2)	7 (29.2)	5 (16.7)	7611	32.5
≥65 (798)	267 (33.2)	531 (21.5)	10 (41.7)	14 (46.7)	3765	70.9
Total (3323)	804 (24.2)	2465 (74.2)	24 (0.7)	30 (0.9)	35988	22.3

IV. DISCUSSION

Human coronaviruses are known to have a wide distribution and endemic to most countries in the world, but usually limited information is available on their presence and circulation, especially from small countries, such as from Bosnia and Herzegovina. This study included 186 patients who had diseases with or without symptoms. Ninety (48.4%) patients had a symptom, and it is higher than in reports from China (29.4%), Bahrain (23.4%) and South Korea (38.0%) [6], [10], [18]. Number of asymptomatic patients was higher (51.6%) than number of symptomatic patients, and it is similar with the reports from China [6] and Republic of Korea [18]. In Iceland, Italy and Indian country number of asymptomatic patients were detected in \approx 44%, which is lower than in our study [19]-[21].

The prevalence of comorbidity was higher in the asymptomatic groups than in symptomatic, such as Diabetes mellitus (58.3% and 41.7%) followed by Hypertension (55.6% and 44.4%), and it is similar with the report from China [22].

Our findings also indicate differences between symptomatic and asymptomatic patients in age groups, with the prevalence of asymptomatic patients slightly increased with age for people older than 50 years. Possible reason for this difference could be a high-risk population in the present study and a greater involvement in community activities [17].

This report indicates that patients with COVID-19 can transmit the disease regardless of their symptomatic status, and if they not identified in a timely manner, they could become moving sources of infection and lead to massive transmission of disease [23]. They must be identified and quarantined to eliminate the transmission of SARS-CoV-2.

Prevalence of antibodies in our study was 42%, respectively, which is higher than in report from Iran, 33% and 22% [14], [15].

Prevalence of antibodies in other studies was 1% in Germany [24], 0.7% in Texas [25], 9% in Austria [26], 11% in Switzerland [27] and 2.7% in Milan, Italy [12]. Some study, for example from Italy showed estimated period-prevalence of COVID-19 varies from 0.35% to 13.3% [28], and meta-analysis study from Rostami et al. showed results varied from 1.5% in South America to 5.3% in Northern Europe [29]. In 2021, two years from the beginning of pandemic, it is expected and reasons for this a little higher seroprevalence of anti-SARS-CoV-2.

Seroprevalence of antibody was higher in females than in males (56% and 44%, respectively), and it is similar with the report from Spain [30], Iran [15], and Uganda [16]. This result was contradictory with the reports from Iran (43% in males and 37% in females) [14] and from South Korea (11% in male and 4% in female) [31]. Seroprevalence of antibody were significantly more prevalent in the age group of 25-50 years and it is similar with the reports from Iran and Switzerland [14], [27]. The highest seroprevalence in other study was detected in the following age groups: 0-5 years in Uganda [16] and more than 60 years in South Korean population [31]. Exposure made age groups 0-5 years and more than 60 years more susceptible to contracting human coronaviruses resulting in the high seropositivity [16].

The highest seroprevalence were detected in active workers (44%), followed by house person, and retired, and it is contradictory with the report from Uganda (42% in pre-school children and 37% in students). The possible cause of this report in our study is the higher risk of exposure and transfer of the coronaviruses between workers compared to other participants.

Fever, followed by muscle pain and dry cough, were the most prevalent of symptoms (60%, 47% and 37%) associated with the seroprevalence of antibodies, and it is similar with the report from Iran [14].

Fever and anosmia were also common findings in report from California [11]. In Germany, fever, dry cough and anosmia were also the most common symptoms correlated with a positive test for anti-SARS-CoV-2 antibodies [32]. Disease of COVID-19 have more specific symptoms such as fever, dry cough and others, than other diseases with similar symptoms.

V. CONCLUSION

In generally, after two years from the beginning of pandemic, it is still small number of seroprevalence in this study, and one of the reasons could be: 1. inadequate time from exposure to form IgG response to COVID-19; 2. a significant number of first responders successfully kill the virus with respiratory tract IgA defences, so the virus never enters in the blood; 3. IgG antibodies once produced, do not persist for very long; 4. and small number of participants included in this study.

In conclusion, human coronaviruses are important emerging pathogens and currently the world is facing a devastating pandemic caused by SARS-2, there is therefore need for continuous viral surveillance. For preventive control of the pandemic spread of coronaviruses, we need additional

testing and information about symptomatic and asymptomatic patients in the municipality.

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CONFLICT OF INTEREST

Authors declare that they do not have any conflict of interest.

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