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Seroprevalence of SARS-CoV-2 in Palestine: a cross-sectional seroepidemiological study

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7 3 **Seroprevalence of SARS-CoV-2 in Palestine: a cross-sectional seroepidemiological study**
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For peer review only

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56 27 **Abstract**7
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9 28 Objectives

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12 29 Seroprevalence rates are important indicators to the epidemiology of COVID-19 and the extent of
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14 30 the pandemic given the existence of asymptomatic cases. The purpose of this study is to assess the
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16 31 seroprevalence rate in the Palestinian population residing in the West Bank.
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22 33 Setting

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25 34 The study involved 1355 participants from 11 governorates, including 112 localities in the West
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27 35 Bank, Palestine.
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32 37 Participants

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35 38 Blood samples were collected between 15th June 2020 and 30th June 2020 from 1355 individuals
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37 39 from randomly selected households in the West Bank in addition to 1136 individuals visiting
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39 40 Palestinian medical laboratories between the 1st May 2020 and 9th July 2020 for a *routine checkup*.
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45 42 Primary and secondary outcome measures

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47
48 43 Out of the 2491 blood samples collected, serological tests for 2455 adequate serum samples were
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50 44 done using an Immunoassay for qualitative detection of antibodies against SARS-CoV-2.
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52 45 seroprevalence was estimated as the proportion of individuals who had a positive result in the total
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54 46 SARS-CoV-2 antibodies in the immunoassay
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45 48 Results
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8 49 The random sample of Palestinians living in the West Bank yielded 0% seroprevalence with 95%
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10 50 CI [0,0.0036], while the lab referrals sample yielded an estimated seroprevalence of 0.354% with
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13 51 95% CI [0.0011,0096].
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16 52
1718 53 Conclusions
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20 54 Our results indicate that as of July 2020, seroprevalence in Palestine persist low and is inadequate
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22 55 to provide herd immunity, emphasizing the need to maintain health measures to keep the outbreak
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25 56 under control. Population-based *seroprevalence studies are to be conducted periodically to*
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27 57 *monitor the SARS-CoV-2 seroprevalence in Palestine and inform policy makers about the efficacy*
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29 58 *of their surveillance system.*
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3 **61 Strengths and limitations of this study**
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6 **62 Strengths:**
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9 **63** 1- The random selection of households residents collected between 15th June 2020 and 30th
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11 **64** June 2020. The clusters were selected using probability proportional to size (PPS)
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13 **65** sampling.
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16 **66** 2- The study also included 1136 participants from Medicare laboratory referrals between 1st
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18 **67** May 2020 and 9th July 2020 in 16 branches in the West Bank, which had good
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20 **68** representation of females as well as children
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23 **69** Limitations:
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26 **70** 1- Low representation in females represented in the random sample, the samples from female
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28 **71** were more difficult to obtain due to a cultural inhibition regarding allowing nurses to enter
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30 **72** the households.
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33 **73** 2- The random sample did not include children under the age of 15 to avoid anxiety due to
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35 **74** fear of needles in the study to eliminate personal fears due to blood withdrawal.
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38 **75** 3- Our study only detected Sars-Cov2 antibodies. However, it is important to recognize that
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40 **76** cellular immunity may play a role in providing immunity against SARS-CoV-2
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42 **77** reinfection¹⁸. Further studies aimed at testing cellular immunity are important.
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84 **Introduction**

85 Coronavirus disease 2019, known as COVID-19, is an infectious respiratory disease caused by
86 novel coronavirus SARS-CoV-2 ¹. Since its emergence in Wuhan, China in December 2019 ²,
87 SARS-CoV-2 has spread rapidly around the globe, ultimately being declared by the World Health
88 Organization (WHO) as a global pandemic ^{3,4} and new cases and deaths are being reported daily⁵.
89 Most authorities rely on PCR testing results to estimate number of COVID-19 cases and make up-
90 to-date decisions ⁶. Thus, numbers of patients tested positive for SARS-CoV-2 through PCR
91 testing, symptomatic patients, those admitted to hospitals, or deceased from Covid-19 are updated
92 on a daily basis. However, the data may exclude a fraction of the population with previous mild or
93 asymptomatic COVID-19 that has not been tested by PCR. The proportion of the population who
94 have overcome the infection without being noticed can probably be approximated by testing for
95 antibodies against SARS-CoV-2. Antibodies may confer immunity to repeat infection and a high
96 proportion of immune individuals can attenuate the epidemic. Measures of anti-SARS-CoV-2
97 seroprevalence can also be used to estimate the clinical impact of COVID-19. To this effect,
98 several serological surveys of SARS-CoV-2 have been done worldwide ⁷⁻¹⁶.

99 **Objective:**

100 There is lack of data on the the percentage of undiagnosed Palestinian population with previous
101 mild or asymptomatic COVID-19. Prevalence of COVID-19 among Palestinian remains unknown
102 and many are concerned about this uncertainty. To this end, we conducted a population cross
103 sectional based seroepidemiological study to assess the spread of SARS-CoV-2 throughout the
104 West Bank. The study included 2491 individuals, designed to be representative by cities (1355

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3 105 from randomly selected households and 1136 from laboratory referrals). Elecsys® Anti-SARS-
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5 106 CoV-2 testing was done on 2455 adequate serum samples. Here, we describe the study design and
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8 107 the results of the first wave of the study.
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10 108 **Methods**

11 109 **Study design and participants**

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17 110 The study conducted is a cross-sectional serologic testing study aimed to investigate seropositivity
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19 111 for SARS-CoV-2 in the non-institutionalized Palestinian population residing in the West Bank.

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22 112 The study involved 1355 participants from 11 governorates, including 112 localities
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24 113 (supplementary table 1). 1395 households were selected using 3-stage cluster sampling. The
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26 114 cluster of households or census track is considered to be a geographic location that is comprised
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28 115 of approximately 100 households. The process for conducting cluster sampling was carried as
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30 116 follows : (1) Selecting a cluster of households, (2) Selecting 10 households randomly from each
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32 117 cluster and (3) Selecting a person at random from the selected household. The clusters were
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35 118 selected using probability proportional to size (PPS) sampling (Table 1).
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42 120 To select the number of clusters within each population location: (1) we calculated the sampling
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44 121 interval which equals the total number of households divided by the total number of clusters need
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46 122 to be selected by the sample say for example (m). So the sampling interval $SI = N/m$, where N is
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48 123 the total number of households. (2) selected a random number R_0 between 0 and SI. (3) calculated
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50 124 R_i as $R_0 + i * SI$, a cluster is selected in L_i if R_i belongs to the interval $[C_{i-1}, C_i]$.
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3 125 Field work was carried out between 15th June 2020 and 30th June 2020 by a team of registered
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5 126 nurses, laboratory technicians, nursing students and laboratory technician students from the Arab
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8 127 American University following standardized health protocols (World Health, 2020) .
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11 128 The study also included 1136 participants from Medicare laboratory referrals between 1st May
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13 129 2020 and 9th July 2020 in 16 branches in the West Bank (supplementary table 2) .
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16 130 Participants donated a blood sample for antibodies detection. Blood samples were centrifuged and
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18 131 serum was separated, labelled, stored at -20C at AAUP laboratory until it was used.
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21 132 **Detection of antibodies**

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24 133 Serological tests for 2455 adequate serum samples were done using an Immunoassay for the
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26 134 qualitative detection of antibodies against SARS-CoV-2 (Elecsys[®] Anti-SARS-CoV-2) in human
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28 135 serum by using the Cobas analyzer cobas e 411 (Roche) . The assay uses a recombinant protein
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30 136 representing the nucleocapsid (N) antigen for the determination of antibodies against SARS-CoV-
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32 137 2 with testing time of 18 minutes. We included 6 samples from recovered cases with detected
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34 138 SARS-COV2 antibodies as a positive control.
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38 139 39 40 140 **Statistical Analysis**

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43 141 We estimated seroprevalence as the proportion of individuals who had a positive result in the total
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45 142 SARS-CoV-2 antibodies in the immunoassay.
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48 143 We used Wilson Method With Continuity Correction and Boundary Truncation (WCCBT) to
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50 144 construct 95% CI for the population parameter of seroprevalence ¹⁷ .
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54 145 **Patients and public involvement**

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3 146 The study involved 1355 participants from 11 governorates, including 112 localities and included
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5 147 1136 participants from Medicare laboratory referrals. The development of the research question
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8 148 and outcome measures was based on the public priorities to know the seroprevalence in the
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10 149 Palestinian community. Approval from National ethical committee was obtained
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12 150 (PHRC/HC/737/20). Written informed consent was obtained from the 1355 study participants and
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14 151 approvals were obtained from Medicare laboratories for samples to be tested. Participants were
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16 152 notified of the results of the tests.
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19 153 **Results**

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22 154 Of the 1395 eligible individuals residing in households selected using 3-stage cluster sampling,
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24 155 1355 participants agreed to participate in the study. The proportion of females was lower
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26 156 compared to males (137, 1218 respectively) including 349 in age group (15-24), 314 in age group
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28 157 (25-34), 377 in age group (35-49) and 315 in age group (50+). Out of the 1355 blood samples
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30 158 collected, 1319 serum samples were adequate for testing. None of the tested specimens revealed
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32 159 presence of antibodies against SARS-CoV-2. A 95% CI for the population parameter of
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34 160 seroprevalence was [0,0.0036].
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39 161 Of the 1136 participants from Medicare laboratory referrals in 16 branches. The proportion of
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41 162 males was lower than females (395, 741 respectively) including 71 in age group lower less than
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43 163 15, 173 in age group (15-24), 297 tests in age group (25-34), 290 in age group (35-49) and 305
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45 164 in age group (50+). All serum samples were adequate for testing. Out of the 1136 tested
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47 165 participants, 4 revealed antibodies against Sars-CoV-2 with 95% CI [0.0011,0096].
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52 167 **Discussion**

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3 168 To our knowledge, this is the first SARS-CoV-2 seroprevalence study in Palestine. The findings
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5 169 from this seroprevalence study for SARS-CoV-2 indicate that the estimated seroprevalence of the
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7 170 total SARS-COV2 antibodies persist low. The random sample of Palestinians living in the West
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9 171 Bank yeilded 0% seroprevalence with 95% CI [0,0.0036], while the lab referrals sample yielded
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11 172 an estimated seroprevalence of 0.354% with 95% CI [0.0011,0096]. Seroprevelance in Palestine
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13 173 is very close to that identified in Jordan (0% prevalence)¹⁵, a neighboring country, which was
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15 174 explained to be due to the strict closures implemented by the Jordanian government and the
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17 175 eventual curtailing of infections in Jordan. In comparison to other countries like Spain, Italy. Japan
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19 176 India, Los Angeles, Germany, Switzerland the seroprevalence in Palestine is low⁷⁻¹⁶. It is,
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21 177 however, noteworthy that a comparison with other countries may be problematic due to the timing
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23 178 and the stage of the pandemic which may vary affecting the seroprevalence estimates.
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31 180 A key strength of our study is the random selection of households residents collected between 15th
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33 181 June 2020 and 30th June 2020. The clusters were selected using probability proportional to size
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35 182 (PPS) sampling. This technique ensures getting self-weighting sample which can be used to
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37 183 produce unbiased estimators for the parameters of interest. However, samples from female were
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39 184 more difficult to obtain due to a cultural inhibition regarding allowing nurses to enter the
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41 185 households. Also, random sample did not include children under the age of 15 to avoid anxiety
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43 186 due to fear of needles in the study to eliminate personal fears due to blood withdrawl.
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51 188 As for the lab referals sample, it represents the population of the lab referals between the 1st May
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53 189 2020 and 9th July 2020. The seroprevalence in this sample was 4 positive cases out of the 1136
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55 190 samples tested. Since the seroprevalence within the lab referral population is close to zero , we
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3 191 used Wilson Method With Continuity Correction and Truncation (WCCBT) to construct a 95%
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5 192 confidence interval for the population parameter of seroprevalence [0.0011,0096].
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10 194 Our study only detected Sars-Cov2 antibodies. However, it is important to recognize that cellular
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12 195 immunity may play a role in providing immunity against SARS-CoV-2 reinfection¹⁸. Further
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14 196 studies aimed at testing cellular immunity are important. It is also noteworthy that previous studies
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16 197 have indicated that asymptomatic individuals were reported to have a weaker immune response to
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18 198 SARS-CoV-2 infection and a higher percentage of asymptomatic individuals became seronegative
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20 199 when compared to symptomatic individuals in the early recovering phases. The reduction in
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22 200 neutralizing antibody levels may have implications for immunity strategy and serological surveys
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24 201¹⁹.
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31 203 In conclusion, our study provides estimates of SARS-CoV-2 seroprevalence in Palestine. Our
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33 204 estimate is low indicating that as of July 2020 the population does not have herd immunity. In this
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35 205 situation, health measures have to be taken to keep the outbreak under control. In order to monitor
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37 206 the SARS-CoV-2 seroprevalence in Palestine and inform policy makers about the efficacy of their
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39 207 surveillance system, conducting population-based seroprevalence studies on a regular basis is
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41 208 important.
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48 210 **Acknowledgment**

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51 211 We thank the participants for their cooperation.
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16 217 **Conflict of Interest Disclosures**

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34 223 **Authors contribution**

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40 225 Field work coordination: Mohammad Asia, Imad Abu Khader
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43 226 Data analysis: Faisal Awartani
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46 227 Manuscript writing: Nouar Qutob
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49 228 Manuscript revisions: Nouar Qutob, Faisal Awartani, Zaidoun Salah, Mohammad Asia
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52 229 Methodology and lab work: Nouar Qutob, Khaled Herzallah, Nadeen Balqis, Husam Sallam
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Supplementary Table 1: Governorates and localities included in the households sample

Governorate	Locality
Jenin	Sir
Jenin	Anza
Jenin	Al Araqa
Jenin	Al Mughayyir
Jenin	Al Fandaqumiya
Jenin	Al Judeida
Jenin	Ajja
Jenin	Birqin
Jenin	Meithalun
Jenin	Jaba
Jenin	Silat al Harithiya
Jenin	Arraba
Jenin	Yabad
Jenin	Al Yamun
Jenin	Qabatiya
Jenin	Jenin
Tubas	Ras al Faraa
Tubas	Aqqaba
Tubas	Tammun
Tubas	Tubas
Tulkarm	Kafa
Tulkarm	Kafr Jammal
Tulkarm	Kafr al Labad
Tulkarm	Nur Shams Camp
Tulkarm	Bala
Tulkarm	Tulkarm Camp
Tulkarm	Attil
Tulkarm	Tulkarm
Nablus	Iraq Burin
Nablus	Qusin
Nablus	As Sawiya
Nablus	Majdal Bani Fadil
Nablus	Sabastiya
Nablus	Ein Beit el Ma Camp
Nablus	Beit Dajan
Nablus	Qusra
Nablus	Askar Camp)al Qadeem(
Nablus	Awarta
Nablus	Asira ash Shamaliya
Nablus	Aqraba

Nablus	Beit Furik
Nablus	Balata Camp
Nablus	Nablus
Qalqiliya	Kafr Laqif
Qalqiliya	Jinsafut
Qalqiliya	Sanniriya
Qalqiliya	Azzun
Qalqiliya	Qalqiliya
Salfit	Deir Istiya
Salfit	Haris
Salfit	Biddya
Salfit	Salfit
Ramallah & Al-Bireh	Shabtin
Ramallah & Al-Bireh	Ein Arik
Ramallah & Al-Bireh	Beitin
Ramallah & Al-Bireh	Ein Yabrud
Ramallah & Al-Bireh	Arura
Ramallah & Al-Bireh	Beitillu
Ramallah & Al-Bireh	Deir Abu Mashaal
Ramallah & Al-Bireh	Khirbet Abu Falah
Ramallah & Al-Bireh	Beit Ur at Tahta
Ramallah & Al-Bireh	Shuqba
Ramallah & Al-Bireh	Qibya
Ramallah & Al-Bireh	Al Jalazun Camp
Ramallah & Al-Bireh	Beituniya
Ramallah & Al-Bireh	Ramallah
Ramallah & Al-Bireh	Al Bireh
Jericho & Al-Aghwar	Ein as Sultan Camp
Jericho & Al-Aghwar	Jericho)Ariha(

Jerusalem	Ash Sheikh Sad
Jerusalem	Beit Surik
Jerusalem	As Sawahira ash Sharqiya
Jerusalem	Hizma
Jerusalem	Qalandiya Camp
Jerusalem	Ar Ram & Dahiyat al Bareed
Jerusalem	Anata
Jerusalem	Al Eizariya
Bethlehem	Al Masara
Bethlehem	Wadi Rahhal
Bethlehem	Dar Salah
Bethlehem	Husan
Bethlehem	Hindaza and Bureidaa
Bethlehem	Nahhalin
Bethlehem	Ad Duheisha Camp
Bethlehem	Ad Doha
Bethlehem	Beit Sahur
Bethlehem	Beit Jala
Bethlehem	Beit Fajjar
Bethlehem	Bethlehem (Beit Lahm)
Hebron	An Najada
Hebron	Beit ar Rush al Fauqa
Hebron	Shuyukh al Arrub
Hebron	Al Burj and Al Bira
Hebron	Nuba
Hebron	Al Fawwar Camp
Hebron	Beit Kahil
Hebron	Kharas
Hebron	Beit Awwa
Hebron	Beit Ula
Hebron	Taffuh
Hebron	Beit Ummar
Hebron	Surif
Hebron	Tarqumiya
Hebron	Sair
Hebron	Bani Naim
Hebron	Idhna
Hebron	As Samu
Hebron	Halhul
Hebron	Adahiriya
Hebron	Dura
Hebron	Yatta

Hebron	Hebron (Al Khalil)
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Supplementary Table 2: Governorates and localities included in the lab referrals sample

Governorate	Locality
Jericho	Jericho
Hebron	Dura
Hebron	Yatta
Hebron	Hebron (Al Khalil)
Hebron	Adahiriya
Bethany	Bethany
Bethlehem	Bethlehem
Jenin	Jenin
Ramallah	Ramallah
Ramallah	Silwad
Tubas	Tubas
Tulkarem	Tulkarem
Qalqiliah	Qalqiliah
Nablus	Nablus
Salfit	Bidia
Salfit	Salfit

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported
Objectives	3	State specific objectives, including any prespecified hypotheses
Methods		
Study design	4	Present key elements of study design early in the paper
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection
Participants	6	(a) <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants (b) <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group
Bias	9	Describe any efforts to address potential sources of bias
Study size	10	Explain how the study size was arrived at
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses

Continued on next page

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60**Results**

Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed (b) Give reasons for non-participation at each stage (c) Consider use of a flow diagram
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders (b) Indicate number of participants with missing data for each variable of interest (c) <i>Cohort study</i> —Summarise follow-up time (eg, average and total amount)
Outcome data	15*	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses

Discussion

Key results	18	Summarise key results with reference to study objectives
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence
Generalisability	21	Discuss the generalisability (external validity) of the study results

Other information

Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based
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*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at www.strobe-statement.org.

BMJ Open

Seroprevalence of SARS-CoV-2 in the West Bank region of Palestine: a cross-sectional seroepidemiological study

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Primary Subject Heading:	Epidemiology
Secondary Subject Heading:	Public health
Keywords:	COVID-19, IMMUNOLOGY, EPIDEMIOLOGY

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5 2 **Seroprevalence of SARS-CoV-2** the West Bank region of Palestine **a cross-sectional**
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7 3 **seroepidemiological study**
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56 27 **Abstract**7
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9 28 Objectives

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12 29 Seroprevalence rates are important indicators to the epidemiology of COVID-19 and the extent of
13
14 30 the pandemic given the existence of asymptomatic cases. The purpose of this study is to assess the
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16 31 seroprevalence rate in the Palestinian population residing in the West Bank.
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22 33 Setting

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25 34 The study involved 1355 participants from 11 governorates, including 112 localities in the West
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27 35 Bank, Palestine.
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32 37 Participants

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35 38 Blood samples were collected between 15th June 2020 and 30th June 2020 from 1355 individuals
36
37 39 from randomly selected households in the West Bank in addition to 1136 individuals visiting
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39 40 Palestinian medical laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.
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43 41
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45 42 Primary and secondary outcome measures

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47
48 43 Out of the 2491 blood samples collected, serological tests for 2455 adequate serum samples were
49
50 44 done using an Immunoassay for qualitative detection of antibodies against SARS-CoV-2.
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52 45 seroprevalence was estimated as the proportion of individuals who had a positive result in the total
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54 46 SARS-CoV-2 antibodies in the immunoassay
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3 47
45 48 Results
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8 49 The random sample of Palestinians living in the West Bank yielded 0% seroprevalence with 95%
9
10 50 and an adjusted confidence interval [0 , 0.0043], while the lab referrals sample yielded an
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12
13 51 estimated seroprevalence of 0.354% with 95% and an adjusted confidence interval [0.001325
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15 52 , 0.011566].
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19
20 54 Conclusions
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23 55 Our results indicate that as of July 2020, seroprevalence in Palestine persist low and is inadequate
24
25 56 to provide herd immunity, emphasizing the need to maintain health measures to keep the outbreak
26
27 57 under control. Population-based seroprevalence studies are to be conducted periodically to monitor
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30 58 the SARS-CoV-2 seroprevalence in Palestine and inform policy makers about the efficacy of their
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32 59 surveillance system.
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62 **Strengths and limitations of this study**

63 Strengths:

- 64 1- The random selection of households residents collected between 15th June 2020 and 30th
65 June 2020. The clusters were selected using probability proportional to size (PPS)
66 sampling.
- 67 2- The study also included 1136 participants from Medicare laboratory referrals between 1st
68 May 2020 and 9th July 2020 in 16 branches in the West Bank, which had good
69 representation of females as well as children

70 Limitations:

- 71 1- Low representation in females represented in the random sample, the samples from female
72 were more difficult to obtain due to a cultural inhibition regarding allowing nurses to enter
73 the households.
- 74 2- The random sample did not include children under the age of 15 to avoid anxiety due to
75 fear of needles in the study to eliminate personal fears due to blood withdrawal.
- 76 3- Our study only detected Sars-Cov2 antibodies. However, it is important to recognize that
77 cellular immunity may play a role in providing immunity against SARS-CoV-2
78 reinfection¹. Further studies aimed at testing cellular immunity are important.
- 79 4- It was previously reported that antibodies against SARS-CoV-2 may drop or even
80 disappear in patients with mild Covid-19, which may have led to the inability to detect
81 antibodies in a few previously positive cases

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84 Introduction

85 Coronavirus disease 2019, known as COVID-19, is an infectious respiratory disease caused by
86 novel coronavirus SARS-CoV-2². Since its emergence in Wuhan, China in December 2019³,
87 SARS-CoV-2 has spread rapidly around the globe. A global pandemic was declared by the World
88 Health Organization (WHO)^{4,5} and new cases and deaths are being reported daily⁶.

89 Most authorities rely on PCR testing results to estimate number of COVID-19 cases and make up-
90 to-date decisions⁷. Thus, numbers of patients tested positive for SARS-CoV-2 through PCR
91 testing, symptomatic patients, those admitted to hospitals, or deceased from Covid-19 are updated
92 on a daily basis. However, the data may exclude a fraction of the population with previous mild or
93 asymptomatic COVID-19 that has not been tested by PCR. The proportion of the population who
94 have overcome the infection without being noticed can probably be approximated by testing for
95 antibodies against SARS-CoV-2. Antibodies may confer immunity to repeat infection and a high
96 proportion of immune individuals can attenuate the epidemic. Measures of anti-SARS-CoV-2
97 seroprevalence can also be used to estimate the clinical impact of COVID-19. To this effect,
98 several serological surveys of SARS-CoV-2 have been done worldwide⁸⁻¹⁷.

99 Objective:

100 There is lack of data on the the percentage of undiagnosed Palestinian population with previous
101 mild or asymptomatic COVID-19. Prevalence of COVID-19 infections among Palestinians
102 residing in the West Bank remains unknown and many are concerned about this uncertainty. To
103 this end, we conducted a population cross sectional based seroepidemiological study to assess the
104 spread of SARS-CoV-2 throughout the West Bank. The study included 2491 individuals, designed
105 to be representative by cities (1355 from randomly selected households and 1136 from laboratory

106 referrals). Elecsys® Anti-SARS-CoV-2 testing was done on 2455 adequate serum samples. Here,
 107 we describe the study design and the results of the first wave of the study.

108 **Methods**

109 **Study design and participants**

110 The study conducted is a cross-sectional serologic testing study aimed to investigate seropositivity
 111 for SARS-CoV-2 in the non-institutionalized Palestinian population residing in the West Bank.

112 The study involved 1355 participants from 11 governorates, including 112 localities
 113 (supplementary table 1). We used 3-stage cluster sampling to select 1395 households. The cluster
 114 of households or census track is considered to be a geographic location that is comprised of
 115 approximately 100 households. The process for conducting cluster sampling was carried as
 116 follows : (1) Selecting a cluster of households, (2) Selecting 10 households randomly from each
 117 cluster and (3) Selecting a person at random from the selected household. The clusters were
 118 selected using probability proportional to size (PPS) sampling (Table 1).

120 Table 1: The PPS sampling algorithm

Location	# of households in the location	Cumulative
L1	X1	C1=X1
L2	X2	C2=X1+X2

L3	X3	$C3=X1+X2+X3$
.	.	.
.	.	.
.	.	.
Lk-1	Xk-1	$Ck-1=X1+X2+\dots+Xk-1$
Lk	Xk	$Ck = X1+X2+\dots+Xk$

121

122 To select the number of clusters within each population location: (1) we calculated the sampling
 123 interval which equals the total number of households divided by the total number of clusters need
 124 to be selected by the sample say for example (m). So the sampling interval $SI= N/m$, where N is
 125 the total number of households. (2) selected a random number $R0$ between 0 and SI. (3) calculated
 126 Ri as $R0+i*SI$, a cluster is selected in Li if Ri belongs to the interval $[Ci-1, Ci]$.

127 Field work was carried out between 15th June 2020 and 30th June 2020 by a team of registered
 128 nurses, laboratory technicians, nursing students and laboratory technician students from the Arab
 129 American University following standardized health protocols ¹⁸.

130 The study also included 1136 participants from Medicare[®] medical laboratories network referrals
 131 between 1st May 2020 and 9th July 2020 in 16 branches in the West Bank (supplementary table 2).

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3 132 Participants donated a blood sample for antibodies detection. Blood samples were centrifuged and
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5 133 serum was separated, labelled, stored at -20C at AAUP laboratory until it was used.
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8 134 **Detection of antibodies**

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11 135 Serological tests for 2455 adequate serum samples were done using an Immunoassay for the
12
13 136 qualitative detection of antibodies against SARS-CoV-2 (Elecsys[®] Anti-SARS-CoV-2) in human
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15 137 serum by using the Cobas analyzer cobas e 411 (Roche) . The assay uses a recombinant protein
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17 138 representing the nucleocapsid (N) antigen for the determination of antibodies against SARS-CoV-
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19 139 2 with testing time of 18 minutes. We included 6 samples from recovered cases with detected
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21 140 SARS-COV2 antibodies as a positive control.
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26 27 142 **Statistical Analysis**

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30 143 We estimated seroprevalence as the proportion of individuals who had a positive result in the total
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32 144 SARS-CoV-2 antibodies in the immunoassay.
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36 145 We used Wilson Method With Continuity Correction and Boundary Truncation (WCCBT) to
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38 146 construct 95% CI for the population parameter of seroprevalence ¹⁹ .
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41 147 We calculated the adjusted confidence of interval. Since the specificity of the kit used for the
42
43 148 antibodies test is 100% and 83% respectively ²⁰, we adjusted the confidence interval for
44
45 149 seroprevalence according to the following transformation: Adjusted lower confidence limit=
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47 150 $\max[0, \{a-(1-sp)\}/(sp+se-1)]$ and adjusted upper confidence limit= $\min[1, \{b-(1-sp)\}/(sp+se-1)]$
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50 151 .²¹.
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153 **Patients and public involvement**

154 The study involved 1355 participants from 11 governorates, including 112 localities and included
155 1136 participants from Medicare[®] laboratories referrals. The development of the research
156 question and outcome measures was based on the public priorities to know the seroprevalence in
157 the Palestinian community. Approval from National ethical committee was obtained
158 (PHRC/HC/737/20). Written informed consent was obtained from the 1355 study participants and
159 approvals were obtained from Medicare[®] laboratories for samples to be tested. Participants were
160 notified of the results of the tests.

161 **Results**

162 Of the 1395 eligible individuals residing in households selected using 3-stage cluster sampling,
163 1355 participants agreed to participate in the study. The proportion of females was lower
164 compared to males (137, 1218 respectively) including 349 in age group (15-24), 314 in age group
165 (25-34), 377 in age group (35-49) and 315 in age group (50+). (Figure 1). The majority of the
166 participants did not report having symptoms in the last three months nor prevailing chronic
167 diseases (supplementary table 3 and 4). None of the tested specimens revealed presence of
168 antibodies against SARS-CoV-2. A 95% CI for the population parameter of seroprevalence was
169 [0.0, 0.0036]. A 95% CI for the population parameter of seroprevalence was [0.0, 0.0036], and an
170 adjusted confidence interval of [0, 0.0043].

171 Out of the 1355 blood samples collected, 1319 serum samples were adequate for testing. None of
172 the tested specimens revealed presence of antibodies against SARS-CoV-2. A 95% CI for the
173 population parameter of seroprevalence was [0,0.0036].

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3 174 Of the 1136 participants from Medicare laboratory referrals in 16 branches. The proportion of
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5 175 males was lower than females (395, 741 respectively) including 71 in age group lower less than
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8 176 15, 173 in age group (15-24), 297 tests in age group (25-34), 290 in age group (35-49) and 305
9
10 177 in age group (50+) (Figure 2). Records of symptoms and chronic diseases were unattainable. Out
11
12 178 of the 1136 tested participants, 3 males, ages 38, 58, 59 and 1 female, age 40 revealed antibodies
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14 179 against SARS-CoV-2, with 95% CI [0.0011,0.0096] and an adjusted confidence interval of
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17 180 [0.001325, 0.011566].
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20 181 All serum samples were adequate for testing. Out of the 1136 tested participants, 4 revealed
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22 182 antibodies against Sars-CoV-2 with 95% CI [0.0011,0096].
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28 184 **Discussion**

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32 185 To our knowledge, this is the first SARS-CoV-2 seroprevalence study in Palestine. The findings
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34 186 from this seroprevalence study for SARS-CoV-2 indicate that the estimated seroprevalence of the
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36 187 total SARS-CoV-2 antibodies persist low as of mid of June. The random sample of Palestinians
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38 188 living in the West Bank yielded 0% seroprevalence with 95% and an adjusted confidence interval
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40 189 of [0, 0.0043], while the lab referrals sample yielded an estimated seroprevalence of 0.354% with
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43 190 95% and an adjusted confidence [0.001325, 0.011566].
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45

46 191 The low SARS-CoV-2 antibody prevalence in our population can be explained by the imposed
47
48 192 total lockdown in the first three months when cases were under control and only reached around
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50
51 193 800 cases by May. Seroprevalence in Palestine is very close to that identified in Jordan (0%
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53 194 prevalence)¹⁶, a neighboring country, which was explained to be due to the strict closures
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56 195 implemented by the Jordanian government and the eventual curtailing of infections in Jordan.
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3 196 Also, Seroprevalence in Maine and Montana was 0.4% and 0.5% respectively by end of August
4
5 197 2020²². In comparison to other countries like Spain, Italy, Japan India, Los Angeles, Germany,
6
7 198 Switzerland the seroprevalence in Palestine is low⁸⁻¹⁷. Similarly, the seroprevalence among
9
10 199 Palestinians residing in the West Bank was shown to be lower than that in Israel as reported in the
11
12 200 first conducted serological study of 1,700 tests. The study suggested that 2.5 percent of the Israeli
13
14 201 population, have had the coronavirus²³. It is, however, noteworthy that a comparison with other
15
16 202 countries may be problematic due to the timing and the stage of the pandemic which may vary
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18 203 affecting the seroprevalence estimates.
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26 205 A key strength of our study is the random selection of household's residents collected between
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28 206 15th June 2020 and 30th June 2020. The clusters were selected using probability proportional to
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30 207 size (PPS) sampling. This technique ensures getting self-weighting sample which can be used to
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32 208 produce unbiased estimators for the parameters of interest. However, samples from females were
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34 209 more difficult to obtain due to a cultural inhibition regarding allowing nurses to enter the
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36 210 households. Also, random samples did not include children under the age of 15 to avoid anxiety
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38 211 due to fear of needles in the study to eliminate personal fears due to blood withdrawal.
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43 212 As for the lab referrals sample, it represents the population of the lab referrals between the 1st May
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45 213 2020 and 9th July 2020. The seroprevalence in this sample was 4 positive cases out of the 1136
46
47 214 samples tested. Since the seroprevalence within the lab referral population is close to zero, we
48
49 215 used Wislon Method With Continuity Correction and Truncation (WCCBT) to construct a 95%
50
51 216 confidence interval for the population parameter of seroprevalence [0.0011,0096]. In comparison
52
53 217 to household samples that were collected between 15th-30th of June, laboratory referral samples
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3 218 include a good representation of females and children below the age of 15 (Figure 2). Also, samples
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5 219 were collected starting from May 1st, which may better represent the small peaks of SARS-CoV-
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8 220 2 reported cases in Palestine and thus explain the presence of few positive samples in comparison
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10 221 to null in household samples.

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13 222 Our study only detected SARS-CoV-2 antibodies. It was previously reported that antibodies
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15 223 against SARS-CoV-2 may drop or even disappear in patients with mild Covid-19, which may have
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18 224 led to the inability to detect antibodies in a few previously positive cases^{24,25}. Also, it is important
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20 225 to recognize that cellular immunity may play a role in providing immunity against SARS-CoV-2
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22 226 reinfection¹⁸. Further studies aimed at testing cellular immunity are important. It is also noteworthy
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25 227 that previous studies have indicated that asymptomatic individuals were reported to have a weaker
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27 228 immune response to SARS-CoV-2 infection and a higher percentage of asymptomatic individuals
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29 229 became seronegative when compared to symptomatic individuals in the early recovering phases.
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31 230 The reduction in neutralizing antibody levels may have implications for immunity strategy and
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34 231 serological surveys^{26,27}.

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37 232 In conclusion, our study provides estimates of SARS-CoV-2 seroprevalence in Palestine. Our
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39 233 results reflect the seroprevalence as of mid of June. It is noteworthy that on May 25th, the
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41 234 government of Palestine eased the restrictions following a decline in cases which led to a surge in
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44 235 cases beginning of July, with the epicenter of the epidemic in Hebron accounting for over 70 % of
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46 236 active cases implying a possible increase in seroprevalence after July. In this situation, health
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49 237 measures have to be taken to keep the outbreak under control. In order to monitor the SARS-CoV-
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51 238 2 seroprevalence in Palestine and inform policy makers about the efficacy of their surveillance
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54 239 system, conducting population-based seroprevalence studies on a regular basis is important.

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11
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26 24827
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47 **255 Authors contribution**48
49
50 256 Research design: Nouar Qutob, Zaidoun Salah, Faisal Awartani51
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53 257 Field work coordination: Mohammad Asia, Imad Abu Khader54
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56 258 Data analysis: Faisal Awartani

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3 259 Manuscript writing: Nouar Qutob
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6 260 Manuscript revisions: Nouar Qutob, Faisal Awartani, Zaidoun Salah, Mohammad Asia
7
8

9 261 Methodology and lab work: Nouar Qutob, Khaled Herzallah, Nadeen Balqis, Husam Sallam
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15 263 **Data sharing**

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18 264 Data are available upon reasonable request
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24 266 **Figure Legends**

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26
27 267 Figure 1: (a) Age and (b) gender distribution of the 1355 participants from Palestinian
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29 268 households in the West Bank. Blood samples were collected between 15th June 2020 and 30th
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31 269 June from the participants.
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34 270 Figure 2: (a) Age and (b) Gender distribution of the 1136 participants visiting Medicare
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36 271 laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.
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42 273 **References**

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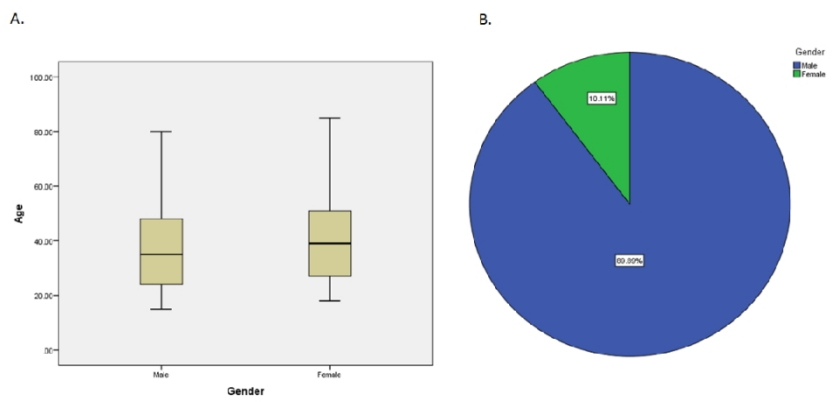


Figure 1
Caption : (a) Age and (b) gender distribution of the 1355 participants from Palestinian households in the West Bank. Blood samples were collected between 15th June 2020 and 30th June from the participants.

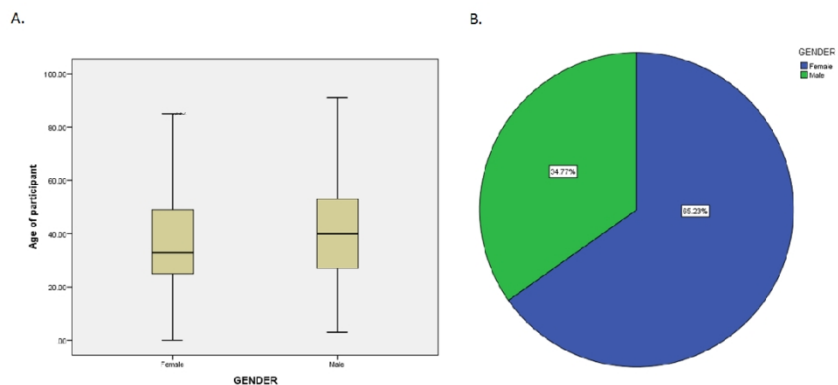


Figure 2

Caption : (a) Age and (b) Gender distribution of the 1136 participants visiting Medicare laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.

Supplementary Table 1: Governorates and localities included in the households sample

Governorate	Locality
Jenin	Sir
Jenin	Anza
Jenin	Al Araqa
Jenin	Al Mughayyir
Jenin	Al Fandaqumiya
Jenin	Al Judeida
Jenin	Ajja
Jenin	Birqin
Jenin	Meithalun
Jenin	Jaba
Jenin	Silat al Harithiya
Jenin	Arraba
Jenin	Yabad
Jenin	Al Yamun
Jenin	Qabatiya
Jenin	Jenin
Tubas	Ras al Faraa
Tubas	Aqqaba
Tubas	Tammun
Tubas	Tubas
Tulkarm	Kafa
Tulkarm	Kafr Jammal
Tulkarm	Kafr al Labad
Tulkarm	Nur Shams Camp
Tulkarm	Bala
Tulkarm	Tulkarm Camp
Tulkarm	Attil
Tulkarm	Tulkarm
Nablus	Iraq Burin
Nablus	Qusin
Nablus	As Sawiya
Nablus	Majdal Bani Fadil
Nablus	Sabastiya
Nablus	Ein Beit el Ma Camp
Nablus	Beit Dajan
Nablus	Qusra
Nablus	Askar Camp)al Qadeem(
Nablus	Awarta
Nablus	Asira ash Shamaliya
Nablus	Aqraba

Nablus	Beit Furik
Nablus	Balata Camp
Nablus	Nablus
Qalqiliya	Kafr Laqif
Qalqiliya	Jinsafut
Qalqiliya	Sanniriya
Qalqiliya	Azzun
Qalqiliya	Qalqiliya
Salfit	Deir Istiya
Salfit	Haris
Salfit	Biddya
Salfit	Salfit
Ramallah & Al-Bireh	Shabtin
Ramallah & Al-Bireh	Ein Arik
Ramallah & Al-Bireh	Beitin
Ramallah & Al-Bireh	Ein Yabrud
Ramallah & Al-Bireh	Arura
Ramallah & Al-Bireh	Beitillu
Ramallah & Al-Bireh	Deir Abu Mashaal
Ramallah & Al-Bireh	Khirbet Abu Falah
Ramallah & Al-Bireh	Beit Ur at Tahta
Ramallah & Al-Bireh	Shuqba
Ramallah & Al-Bireh	Qibya
Ramallah & Al-Bireh	Al Jalazun Camp
Ramallah & Al-Bireh	Beituniya
Ramallah & Al-Bireh	Ramallah
Ramallah & Al-Bireh	Al Bireh
Jericho & Al-Aghwar	Ein as Sultan Camp
Jericho & Al-Aghwar	Jericho)Ariha(
Jerusalem	Ash Sheikh Sad

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6		Sharqiya
7	Jerusalem	Hizma
8	Jerusalem	Qalandiya Camp
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10	Jerusalem	Ar Ram & Dahiyat al
11		Bareed
12	Jerusalem	Anata
13	Jerusalem	Al Eizariya
14	Bethlehem	Al Masara
15	Bethlehem	Wadi Rahhal
16	Bethlehem	Dar Salah
17	Bethlehem	Husan
18	Bethlehem	Hindaza and Bureidaa
19	Bethlehem	Nahhalin
20	Bethlehem	Ad Duheisha Camp
21	Bethlehem	Ad Doha
22	Bethlehem	Beit Sahur
23	Bethlehem	Beit Jala
24	Bethlehem	Beit Fajjar
25	Bethlehem	Bethlehem (Beit Lahm)
26	Hebron	An Najada
27	Hebron	Beit ar Rush al Fauqa
28	Hebron	Shuyukh al Arrub
29	Hebron	Al Burj and Al Bira
30	Hebron	Nuba
31	Hebron	Al Fawwar Camp
32	Hebron	Beit Kahil
33	Hebron	Kharas
34	Hebron	Beit Awwa
35	Hebron	Beit Ula
36	Hebron	Taffuh
37	Hebron	Beit Ummar
38	Hebron	Surif
39	Hebron	Tarqumiya
40	Hebron	Sair
41	Hebron	Bani Naim
42	Hebron	Idhna
43	Hebron	As Samu
44	Hebron	Halhul
45	Hebron	Adahiriya
46	Hebron	Dura
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Hebron	Hebron (Al Khalil)
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For peer review only

Supplementary Table 2: Governorates and localities included in the lab referrals sample

Governorate	Locality
Jericho	Jericho
Hebron	Dura
Hebron	Yatta
Hebron	Hebron (Al Khalil)
Hebron	Adahiriya
Bethany	Bethany
Bethlehem	Bethlehem
Jenin	Jenin
Ramallah	Ramallah
Ramallah	Silwad
Tubas	Tubas
Tulkarem	Tulkarem
Qalqiliah	Qalqiliah
Nablus	Nablus
Salfit	Bidia
Salfit	Salfit

Supplementary Table 3: Prevalence of symptoms reported by participants

Symptom	Yes N=1355	No N=1355	Don't Know N=1355
Fever	3.8%	95.4%	.8%
Sneezing	6.8%	92.4%	.8%
Dry Cough	5.7%	93.5%	.8%
Vomiting	1.1%	98.1%	.8%

Sore and red throat	4.3%	94.9%	.8%
Diarrhea	2.0%	97.2%	.8%
Muscle ache	3.2%	95.9%	.9%
Nasal Congestion	2.8%	96.3%	.8%
Skin rash or change in color of the thumb or toe	1.2%	97.9%	.9%
Loss of sense of taste and smell	.7%	98.5%	.8%
Chest pain	1.7%	97.6%	.8%
Difficulty breathing	1.8%	97.4%	.8%
Loss of ability to speak or move	.6%	98.7%	.8%

Supplementary Table 4: Prevalence of chronic diseases reported by participants

Chronic Disease	Yes N=1355	No N=1355	Don't Know N=1355
Heart disease	5.1%	94.9%	0.0%

Blood pressure	11.9%	88.1%	0.0%
Cancer	.2%	99.8%	.0%
Kidney disease	.7%	99.2%	.1%
Nervous system disease	.7%	99.3%	0.0%
Diabetes	10.1%	89.9%	0.0%
Respiratory disease (Asthma)	4.4%	95.5%	.0%
Hospitalized through the past 6 months	4.2%	95.8%	.0%
On immunosuppressant drug?	1.2%	98.8%	0.0%

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported
Objectives	3	State specific objectives, including any prespecified hypotheses
Methods		
Study design	4	Present key elements of study design early in the paper
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection
Participants	6	(a) <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants (b) <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group
Bias	9	Describe any efforts to address potential sources of bias
Study size	10	Explain how the study size was arrived at
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses

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60**Results**

Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed (b) Give reasons for non-participation at each stage (c) Consider use of a flow diagram
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders (b) Indicate number of participants with missing data for each variable of interest (c) <i>Cohort study</i> —Summarise follow-up time (eg, average and total amount)
Outcome data	15*	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses

Discussion

Key results	18	Summarise key results with reference to study objectives
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence
Generalisability	21	Discuss the generalisability (external validity) of the study results

Other information

Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based
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*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at www.strobe-statement.org.

BMJ Open

Seroprevalence of SARS-CoV-2 the West Bank region of Palestine: a cross-sectional seroepidemiological study

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Keywords:	COVID-19, IMMUNOLOGY, EPIDEMIOLOGY

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56 26 **Abstract**7
8
9 27 Objectives

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12 28 Seroprevalence rates are important indicators to the epidemiology of COVID-19 and the extent of
13
14 29 the pandemic given the existence of asymptomatic cases. The purpose of this study is to assess the
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16 30 seroprevalence rate in the Palestinian population residing in the West Bank.
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22 32 Setting

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25 33 The study involved 1355 participants from 11 governorates, including 112 localities in the West
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27 34 Bank and 1136 individuals visiting Palestinian medical laboratories.
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32 36 Participants

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35 37 Blood samples were collected between 15th June 2020 and 30th June 2020 from 1355 individuals
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37 38 from randomly selected households in the West Bank in addition to 1136 individuals visiting
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39 39 Palestinian medical laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.
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45 41 Primary and secondary outcome measures

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47
48 42 Out of the 2491 blood samples collected, serological tests for 2455 adequate serum samples were
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50 43 done using an Immunoassay for qualitative detection of antibodies against SARS-CoV-2.
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52 44 seroprevalence was estimated as the proportion of individuals who had a positive result in the total
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54 45 SARS-CoV-2 antibodies in the immunoassay
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45 47 Results
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8 48 The random sample of Palestinians living in the West Bank yielded 0% seroprevalence with 95%
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10 49 and an adjusted confidence interval [0, 0.0043], while the lab referrals sample yielded an estimated
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13 50 seroprevalence of 0.354% with 95% and an adjusted confidence interval [0.001325, 0.011566].
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15 51
1617 52 Conclusions
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20 53 Our results indicate that as of mid-june 2020, seroprevalence in Palestine persist low and is
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22 54 inadequate to provide herd immunity, emphasizing the need to maintain health measures to keep
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25 55 the outbreak under control. Population-based seroprevalence studies are to be conducted
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27 56 periodically to monitor the SARS-CoV-2 seroprevalence in Palestine and inform policy makers
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29 57 about the efficacy of their surveillance system.
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60 **Strengths and limitations of this study**

61 Strengths:

- 62 1- The random selection of household's residents collected between 15th June 2020 and 30th
63 June 2020.
- 64 2- Good representation of females and children among the 1136 participants from Medicare
65 laboratory referrals between 1st May 2020 and 9th July 2020.

66 Limitations:

- 67 1- Low representation in females represented in the random sample.
- 68 2- The random sample did not include children under the age of 15.

71 **Introduction**

72 Coronavirus disease 2019, known as COVID-19, is an infectious respiratory disease caused by
73 novel coronavirus SARS-CoV-2 ¹. Since its emergence in Wuhan, China in December 2019 ²,
74 SARS-CoV-2 has spread rapidly around the globe. A global pandemic was declared by the World
75 Health Organization (WHO) ^{3,4} and new cases and deaths are being reported daily ⁵.

76 Most authorities rely on PCR testing results to estimate number of COVID-19 cases and make up-
77 to-date decisions ⁶. Thus, numbers of patients tested positive for SARS-CoV-2 through PCR
78 testing, symptomatic patients, those admitted to hospitals, or deceased from COVID-19 are
79 updated on a daily basis. However, the data may exclude a fraction of the population with previous
80 mild or asymptomatic COVID-19 that has not been tested by PCR. The proportion of the

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3 81 population who have overcome the infection without being noticed can probably be approximated
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5 82 by testing for antibodies against SARS-CoV-2. Antibodies may confer immunity to repeat
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7 83 infection and a high proportion of immune individuals can attenuate the epidemic. Measures of
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9 84 anti-SARS-CoV-2 seroprevalence can also be used to estimate the clinical impact of COVID-19.
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12 85 To this effect, several serological surveys of SARS-CoV-2 have been done worldwide⁷⁻¹⁶.
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15 86 Objective:

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18 87 There is lack of data on the percentage of undiagnosed Palestinian population with previous mild
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20 88 or asymptomatic COVID-19. Prevalence of COVID-19 infections among Palestinians residing in
21
22 89 the West Bank remains unknown and many are concerned about this uncertainty. To this end, we
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24 90 conducted a population cross sectional based seroepidemiological study to assess the spread of
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26 91 SARS-CoV-2 throughout the West Bank. The study included 2491 individuals, designed to be
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28 92 representative by cities (1355 from randomly selected households and 1136 from laboratory
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30 93 referrals). Elecsys® Anti-SARS-CoV-2 testing was done on 2455 adequate serum samples. Here,
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32 94 we describe the study design and the results of the first wave of the study.
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37 95 **Methods**

38 39 40 96 **Study design and participants**

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43 97 The study conducted is a cross-sectional serologic testing study aimed to investigate seropositivity
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45 98 for SARS-CoV-2 in the non-institutionalized Palestinian population residing in the West Bank.

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48 99 The study involved 1355 participants from 11 governorates, including 112 localities
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50 100 (supplementary table 1). We used 3-stage cluster sampling to select 1395 households. The cluster
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52 101 of households or census track is considered to be a geographic location that is comprised of
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54 102 approximately 100 households. The process for conducting cluster sampling was carried as
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3 103 follows: (1) Selecting a cluster of households, (2) Selecting 10 households randomly from each
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5 104 cluster and (3) Selecting a person at random from the selected household. The clusters were
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8 105 selected using probability proportional to size (PPS) sampling (Table 1).
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10

11 106 Table 1: The PPS sampling algorithm
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Location	# of households in the location	Cumulative
L1	X1	C1=X1
L2	X2	C2=X1+X2
L3	X3	C3=X1+X2+X3
.	.	.
.	.	.
.	.	.
Lk-1	Xk-1	Ck-1=X1+X2+...+Xk-1
Lk	Xk	Ck = X1+X2+...+Xk

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42 108 To select the number of clusters within each population location: (1) we calculated the sampling
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44 109 interval which equals the total number of households divided by the total number of clusters need
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46 110 to be selected by the sample say for example (m). So, the sampling interval $SI = N/m$, where N is
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3 111 the total number of households. (2) selected a random number R_0 between 0 and SI . (3) calculated
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5 112 R_i as $R_0 + i * SI$, a cluster is selected in L_i if R_i belongs to the interval $[C_{i-1}, C_i]$.

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8 113 Field work was carried out between 15th June 2020 and 30th June 2020 by a team of registered
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10 114 nurses, laboratory technicians, nursing students and laboratory technician students from the Arab
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12 115 American University following standardized health protocols ¹⁷. The random sample did not
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14 116 include children under the age of 15 and suffered from a low representation in females.

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18 117 The study also included 1136 participants from Medicare[®] medical laboratories network referrals
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20 118 between 1st May 2020 and 9th July 2020 in 16 branches in the West Bank (supplementary table 2).

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24 119 Participants donated a blood sample for antibodies detection. Blood samples were centrifuged and
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26 120 serum was separated, labelled, stored at -20C at AAUP laboratory until it was used.

27 28 29 121 **Detection of antibodies**

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32 122 Serological tests for 2455 adequate serum samples were done using an Immunoassay for the
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34 123 qualitative detection of antibodies against SARS-CoV-2 (Elecsys[®] Anti-SARS-CoV-2) in human
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36 124 serum by using the Cobas analyzer cobas e 411 (Roche). The assay uses a recombinant protein
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38 125 representing the nucleocapsid (N) antigen for the determination of antibodies against SARS-CoV-
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40 126 2 with testing time of 18 minutes. The assay was reported to have a specificity of 100%, while the
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42 127 overall sensitivity was 83.87%, rising to 87.0% at 14 days after onset of symptoms, 87.7% 21 days
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44 128 after, and 100% more than 40 days after ¹⁸. We included 6 samples from recovered cases with
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46 129 detected SARS-CoV-2 antibodies as a positive control.

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53 54 131 **Statistical Analysis**

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3 132 We estimated seroprevalence as the proportion of individuals who had a positive result in the total
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5 133 SARS-CoV-2 antibodies in the immunoassay.

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8 134 We used Wilson Method With Continuity Correction and Boundary Truncation (WCCBT) to
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10 135 construct 95% CI for the population parameter of seroprevalence ¹⁹ .

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13 136 We calculated the adjusted confidence of interval. Since the specificity of the kit used for the
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15 137 antibodies test is 100% and 83% respectively ¹⁸, we adjusted the confidence interval for
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17 138 seroprevalence according to the following transformation: Adjusted lower confidence limit=
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19 139 $\max[0, \{a-(1-sp)\}/(sp+se-1)]$ and adjusted upper confidence limit= $\min[1, \{b-(1-sp)\}/(sp+se-1)]$
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23 140 .²⁰. The Low representation in females represented in the random sample required reweighting by
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25 141 gender. As for the Medicare Labs sample, the obtained estimates from the lab data are considered
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27 142 to be unbiased and did not require reweighting.

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31 32 33 144 **Patients and public involvement**

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36 145 The study involved 1355 participants from 11 governorates, including 112 localities and included
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38 146 1136 participants from Medicare[®] laboratories referrals. The development of the research
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40 147 question and outcome measures was based on the public priorities to know the seroprevalence in
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42 148 the Palestinian community. Approval from National ethical committee was obtained
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44 149 (PHRC/HC/737/20). Written informed consent was obtained from the 1355 study participants and
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46 150 approvals were obtained from Medicare[®] laboratories for samples to be tested. Participants \ were
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48 151 notified of the results of the tests.

49 50 51 52 53 152 **Results**

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3 153 Of the 1395 eligible individuals residing in households selected using 3-stage cluster sampling,
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5 154 1355 participants agreed to participate in the study. The proportion of females was lower
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8 155 compared to males (137, 1218 respectively) including 349 in age group (15-24), 314 in age group
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10 156 (25-34), 377 in age group (35-49) and 315 in age group (50+). (Figure 1). The majority of the
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12 157 participants did not report having symptoms in the last three months nor prevailing chronic
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14 158 diseases (supplementary table 3 and 4). None of the tested specimens revealed presence of
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16 159 antibodies against SARS-CoV-2. A 95% CI for the population parameter of seroprevalence was
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18 [0.0, 0.0036]. A 95% CI for the population parameter of seroprevalence was [0.0, 0.0036], and an
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20 adjusted confidence interval of [0, 0.0043].
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24 162 Of the 1136 participants from Medicare laboratory referrals in 16 branches. The proportion of
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26 163 males was lower than females (395, 741 respectively) including 71 in age group lower less than
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28 164 15, 173 in age group (15-24), 297 tests in age group (25-34), 290 in age group (35-49) and 305 in
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30 165 age group (50+) (Figure 2). Records of symptoms and chronic diseases were unattainable. Out of
31
32 166 the 1136 tested participants, 3 males, ages 38, 58, 59 and 1 female, age 40 revealed antibodies
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34 167 against SARS-CoV-2, with 95% CI [0.0011,0.0096] and an adjusted confidence interval of
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36 [0.001325, 0.011566].
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42 169 All serum samples were adequate for testing. Out of the 1136 tested participants, 4 revealed
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44 170 antibodies against Sars-CoV-2 with 95% CI [0.0011,0096].
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48 49 50 172 **Discussion**

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53 173 To our knowledge, this is the first SARS-CoV-2 seroprevalence study in the Palestine. The
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55 174 findings from this seroprevalence study for SARS-CoV-2 indicate that the estimated
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3 175 seroprevalence of the total SARS-CoV-2 antibodies persist low as of mid of June. The random
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5 176 sample of Palestinians living in the West Bank yielded 0% seroprevalence with 95% and an
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7 177 adjusted confidence interval of [0, 0.0043], while the lab referrals sample yielded an estimated
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9 178 seroprevalence of 0.354% with 95% and an adjusted confidence [0.001325, 0.011566].
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13 179 The low SARS-CoV-2 antibody prevalence in our population can be explained by the imposed total
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15 180 lockdown in the first three months when cases were under control and only reached around 800 cases by
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17 181 May. Our sampling dates ranged from 15th June 2020-30th June 2020 in the random sample and
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19 182 between 1st May 2020 and 9th July 2020 in the lab referrals sample. The GoP extended the state of
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21 183 emergency and imposed total lockdown in the first three months. The samples were collected after
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23 184 this strict lockdown which was accompanied by a drop in disease prevalence. Covid-19 cases were
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25 185 under control. The reported cases by the Palestinian ministry of health at the beginning of the data
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27 186 collection process by mid-June was 689 cases, and the reported cases rose to 2765 cases by the
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29 187 end of June ²¹. However, it takes 10-14 days for antibodies against SARS-CoV-2 to go up,
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31 188 therefore, our results indeed reflect the situation by mid of June. Also, It was previously reported
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33 189 that antibodies against SARS-CoV-2 may drop or even disappear in patients with mild Covid-19
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35 190 ²²⁻²⁴ which may have led to the inability to detect antibodies in a few previously positive cases.
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41 191 Seroprevalence in Palestine is very close to that identified in Jordan (0% prevalence) ¹⁵, a
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43 192 neighboring country, which was explained to be due to the strict closures implemented by the
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45 193 Jordanian government and the eventual curtailing of infections in Jordan. Also, Seroprevalence in
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47 194 Maine and Montana was 0.4% and 0.5% respectively by end of August 2020 ²⁵. In comparison to
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49 195 other countries like Spain, Italy, Japan India, Los Angeles, Germany, Switzerland the
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51 196 seroprevalence in Palestine is low ⁷⁻¹⁶. Similarly, the seroprevalence among Palestinians residing
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53 197 in the West Bank was shown to be lower than that in Israel as reported in the first conducted
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3 198 serological study of 1,700 tests. The study suggested that 2.5 percent of the Israeli population, have
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5 199 had the coronavirus²⁶. It is, however, noteworthy that a comparison with other countries may be
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8 200 problematic due to the timing and the stage of the pandemic which may vary affecting the
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10 201 seroprevalence estimates.

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13 202 A key strength of our study is the random selection of household's residents collected between
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15 203 15th June 2020 and 30th June 2020. The clusters were selected using probability proportional to
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18 204 size (PPS) sampling. This technique ensures getting self-weighting sample which can be used to
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20 205 produce unbiased estimators for the parameters of interest. The sample was supposed to be
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22 206 stratified by gender and age. However, the female response was low and samples from females
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24 207 were more difficult to obtain due to a cultural boundary. In fact, women were less open to
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26 208 welcoming a team of nurses at home for research purposes. Also, the random sample did not
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28 209 include children under the age of 15 to avoid anxiety due to fear of needles in the study to eliminate
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30 210 personal fears due to blood withdrawal. To ensure that the estimated prevalence in the random
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32 211 sample was unbiased, we reweighted the sample by gender. Except for cultural issues resulting in
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34 212 the low response rate from women in the random sample, we do not believe there were any issues
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36 213 that would lead participants to opt out from the study.

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39 214 As for the lab referrals sample, it represents the population of the lab referrals between the 1st May
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41 215 2020 and 9th July 2020. The seroprevalence in this sample was 4 positive cases out of the 1136
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43 216 samples tested. Since the seroprevalence within the lab referral population is close to zero, we
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45 217 used Wilson Method with Continuity Correction and Truncation (WCCBT) to construct a 95%
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47 218 confidence interval for the population parameter of seroprevalence [0.0011,0096]. In comparison
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49 219 to household samples that were collected between 15th-30th of June, laboratory referral samples
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51 220 include a good representation of females and children below the age of 15 (Figure 2). Also, samples

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3 221 were collected starting from May 1st, which may better represent the small peaks of SARS-CoV-
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5 222 2 reported cases in Palestine and thus explain the presence of few positive samples in comparison
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8 223 to null in household samples. The sample was randomly selected by the lab data managers.
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10 224 Although females were not represented among the 1355 adults from Palestinian households in the,
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12 225 they were represented among the 1136 individuals visiting laboratories; out of the 1136, 741, more
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15 226 than 50% of the participants were females (Figure 2). Hence, the obtained estimates from the lab
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17 227 data are considered to be unbiased.

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20 228 Our study only detected SARS-CoV-2 antibodies. It was previously reported that antibodies
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22 229 against SARS-CoV-2 may drop or even disappear in patients with mild Covid-19, which may have
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24
25 230 led to the inability to detect antibodies in a few previously positive cases^{23,22}. Also, it is important
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27 231 to recognize that cellular immunity may play a role in providing immunity against SARS-CoV-2
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29 232 reinfection¹⁸. Further studies aimed at testing cellular immunity are important. It is also noteworthy
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32 233 that previous studies have indicated that asymptomatic individuals were reported to have a weaker
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34 234 immune response to SARS-CoV-2 infection and a higher percentage of asymptomatic individuals
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36 235 became seronegative when compared to symptomatic individuals in the early recovering phases.
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38 236 The reduction in neutralizing antibody levels may have implications for immunity strategy and
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41 237 serological surveys^{24,27}.

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44 238 In conclusion, our study provides estimates of SARS-CoV-2 seroprevalence in Palestine. Our
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46 239 results reflect the seroprevalence as of mid of June. It is noteworthy that on May 25th, the
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49 240 government of Palestine eased the restrictions following a decline in cases which led to a surge in
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51 241 cases beginning of July, with the epicenter of the epidemic in Hebron accounting for over 70 % of
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53 242 active cases implying a possible increase in seroprevalence after July. Reported cases rose to 2765
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56 243 cases by the end of June. By December 2020, the cumulative number of positive cases grew to

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3 244 150,046 cases ²¹ . In this situation, health measures have to be taken to keep the outbreak under
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5 245 control. In order to monitor the SARS-CoV-2 seroprevalence in Palestine and inform policy
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7 246 makers about the efficacy of their surveillance system. With the surge in cases, conducting
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9 247 population-based seroprevalence studies on a regular basis is important.
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18
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21

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25
26 253 Adam Maraw`a, Sharhabeel Nasrallah, Hisham Zahran and Mohammad Barakat
27
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29

30 254 We thank Medicare labs for providing blood samples of individuals visiting their laboratories.
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36 256 **Conflict of Interest Disclosures**

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39 257 No authors reported disclosures.
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48 260 Award/Grant number is not applicable
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51 261
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54 262 **Authors contribution**

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18 268 Methodology and lab work: Nouar Qutob, Khaled Herzallah, Nadeen Balqis, Husam Sallam
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24 270 **Data sharing**

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27 271 Data are available upon reasonable request
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33 273 **Figure Legends**

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36 274 Figure 1: (a) Age and (b) gender distribution of the 1355 participants from Palestinian households
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38 275 in the West Bank. Blood samples were collected between 15th June 2020 and 30th June from the
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40 276 participants.
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43 277 Figure 2: (a) Age and (b) Gender distribution of the 1136 participants visiting Medicare
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45 278 laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.
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52 280 **References**

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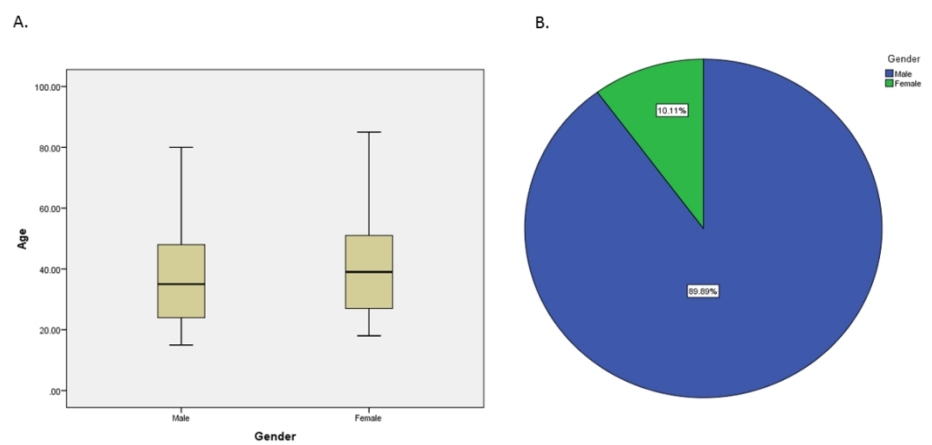


Figure 1: (a) Age and (b) gender distribution of the 1355 participants from Palestinian households in the West Bank. Blood samples were collected between 15th June 2020 and 30th June from the participants.

338x190mm (96 x 96 DPI)

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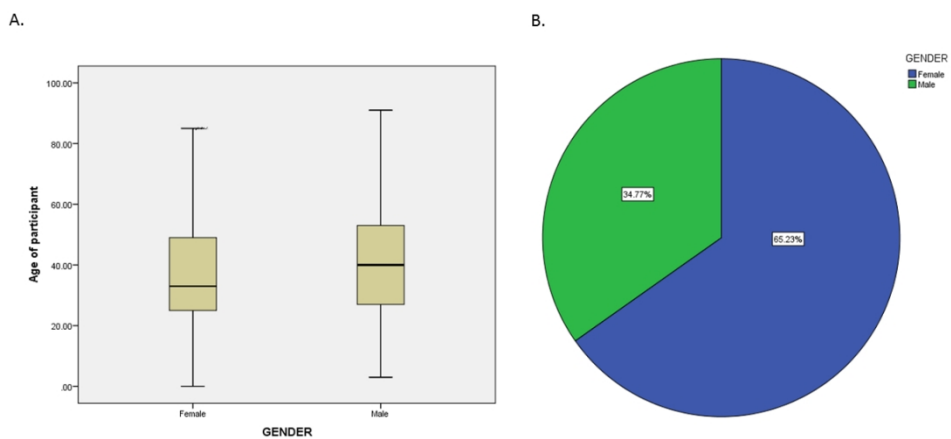


Figure 2: (a) Age and (b) Gender distribution of the 1136 participants visiting Medicare laboratories between the 1st May 2020 and 9th July 2020 for a routine checkup.

338x190mm (96 x 96 DPI)

Supplementary Table 1: Governorates and localities included in the households sample

Governorate	Locality
Jenin	Sir
Jenin	Anza
Jenin	Al Araqa
Jenin	Al Mughayyir
Jenin	Al Fandaqumiya
Jenin	Al Judeida
Jenin	Ajja
Jenin	Birqin
Jenin	Meithalun
Jenin	Jaba
Jenin	Silat al Harithiya
Jenin	Arraba
Jenin	Yabad
Jenin	Al Yamun
Jenin	Qabatiya
Jenin	Jenin
Tubas	Ras al Faraa
Tubas	Aqqaba
Tubas	Tammun
Tubas	Tubas
Tulkarm	Kafa
Tulkarm	Kafr Jammal
Tulkarm	Kafr al Labad
Tulkarm	Nur Shams Camp
Tulkarm	Bala
Tulkarm	Tulkarm Camp
Tulkarm	Attil
Tulkarm	Tulkarm
Nablus	Iraq Burin
Nablus	Qusin
Nablus	As Sawiya
Nablus	Majdal Bani Fadil
Nablus	Sabastiya
Nablus	Ein Beit el Ma Camp
Nablus	Beit Dajan
Nablus	Qusra
Nablus	Askar Camp)al Qadeem(
Nablus	Awarta
Nablus	Asira ash Shamaliya
Nablus	Aqraba

Nablus	Beit Furik
Nablus	Balata Camp
Nablus	Nablus
Qalqiliya	Kafr Laqif
Qalqiliya	Jinsafut
Qalqiliya	Sanniriya
Qalqiliya	Azzun
Qalqiliya	Qalqiliya
Salfit	Deir Istiya
Salfit	Haris
Salfit	Biddya
Salfit	Salfit
Ramallah & Al-Bireh	Shabtin
Ramallah & Al-Bireh	Ein Arik
Ramallah & Al-Bireh	Beitin
Ramallah & Al-Bireh	Ein Yabrud
Ramallah & Al-Bireh	Arura
Ramallah & Al-Bireh	Beitillu
Ramallah & Al-Bireh	Deir Abu Mashaal
Ramallah & Al-Bireh	Khirbet Abu Falah
Ramallah & Al-Bireh	Beit Ur at Tahta
Ramallah & Al-Bireh	Shuqba
Ramallah & Al-Bireh	Qibya
Ramallah & Al-Bireh	Al Jalazun Camp
Ramallah & Al-Bireh	Beituniya
Ramallah & Al-Bireh	Ramallah
Ramallah & Al-Bireh	Al Bireh
Jericho & Al-Aghwar	Ein as Sultan Camp
Jericho & Al-Aghwar	Jericho)Ariha(
Jerusalem	Ash Sheikh Sad

Jerusalem	Beit Surik
Jerusalem	As Sawahira ash Sharqiya
Jerusalem	Hizma
Jerusalem	Qalandiya Camp
Jerusalem	Ar Ram & Dahiyat al Bareed
Jerusalem	Anata
Jerusalem	Al Eizariya
Bethlehem	Al Masara
Bethlehem	Wadi Rahhal
Bethlehem	Dar Salah
Bethlehem	Husan
Bethlehem	Hindaza and Bureidaa
Bethlehem	Nahhalin
Bethlehem	Ad Duheisha Camp
Bethlehem	Ad Doha
Bethlehem	Beit Sahur
Bethlehem	Beit Jala
Bethlehem	Beit Fajjar
Bethlehem	Bethlehem (Beit Lahm)
Hebron	An Najada
Hebron	Beit ar Rush al Fauqa
Hebron	Shuyukh al Arrub
Hebron	Al Burj and Al Bira
Hebron	Nuba
Hebron	Al Fawwar Camp
Hebron	Beit Kahil
Hebron	Kharas
Hebron	Beit Awwa
Hebron	Beit Ula
Hebron	Taffuh
Hebron	Beit Ummar
Hebron	Surif
Hebron	Tarqumiya
Hebron	Sair
Hebron	Bani Naim
Hebron	Idhna
Hebron	As Samu
Hebron	Halhul
Hebron	Adahiriya
Hebron	Dura
Hebron	Yatta

Hebron	Hebron (Al Khalil)
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For peer review only

Supplementary Table 2: Governorates and localities included in the lab referrals sample

Governorate	Locality
Jericho	Jericho
Hebron	Dura
Hebron	Yatta
Hebron	Hebron (Al Khalil)
Hebron	Adahiriya
Bethany	Bethany
Bethlehem	Bethlehem
Jenin	Jenin
Ramallah	Ramallah
Ramallah	Silwad
Tubas	Tubas
Tulkarem	Tulkarem
Qalqiliah	Qalqiliah
Nablus	Nablus
Salfit	Bidia
Salfit	Salfit

Supplementary Table 3: Prevalence of symptoms reported by participants

Symptom	Yes N=1355	No N=1355	Don't Know N=1355
Fever	3.8%	95.4%	.8%
Sneezing	6.8%	92.4%	.8%
Dry Cough	5.7%	93.5%	.8%
Vomiting	1.1%	98.1%	.8%

Sore and red throat	4.3%	94.9%	.8%
Diarrhea	2.0%	97.2%	.8%
Muscle ache	3.2%	95.9%	.9%
Nasal Congestion	2.8%	96.3%	.8%
Skin rash or change in color of the thumb or toe	1.2%	97.9%	.9%
Loss of sense of taste and smell	.7%	98.5%	.8%
Chest pain	1.7%	97.6%	.8%
Difficulty breathing	1.8%	97.4%	.8%
Loss of ability to speak or move	.6%	98.7%	.8%

Supplementary Table 4: Prevalence of chronic diseases reported by participants

Chronic Disease	Yes N=1355	No N=1355	Don't Know N=1355
Heart disease	5.1%	94.9%	0.0%

Blood pressure	11.9%	88.1%	0.0%
Cancer	.2%	99.8%	.0%
Kidney disease	.7%	99.2%	.1%
Nervous system disease	.7%	99.3%	0.0%
Diabetes	10.1%	89.9%	0.0%
Respiratory disease (Asthma)	4.4%	95.5%	.0%
Hospitalized through the past 6 months	4.2%	95.8%	.0%
On immunosuppressant drug?	1.2%	98.8%	0.0%

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation
Title and abstract	1	(a) Indicate the study's design with a commonly used term in the title or the abstract (b) Provide in the abstract an informative and balanced summary of what was done and what was found
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported
Objectives	3	State specific objectives, including any prespecified hypotheses
Methods		
Study design	4	Present key elements of study design early in the paper
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection
Participants	6	(a) <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants (b) <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group
Bias	9	Describe any efforts to address potential sources of bias
Study size	10	Explain how the study size was arrived at
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why
Statistical methods	12	(a) Describe all statistical methods, including those used to control for confounding (b) Describe any methods used to examine subgroups and interactions (c) Explain how missing data were addressed (d) <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of sampling strategy (e) Describe any sensitivity analyses

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60**Results**

Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed (b) Give reasons for non-participation at each stage (c) Consider use of a flow diagram
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders (b) Indicate number of participants with missing data for each variable of interest (c) <i>Cohort study</i> —Summarise follow-up time (eg, average and total amount)
Outcome data	15*	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures
Main results	16	(a) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included (b) Report category boundaries when continuous variables were categorized (c) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses

Discussion

Key results	18	Summarise key results with reference to study objectives
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence
Generalisability	21	Discuss the generalisability (external validity) of the study results

Other information

Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based
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*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at <http://www.plosmedicine.org/>, Annals of Internal Medicine at <http://www.annals.org/>, and Epidemiology at <http://www.epidem.com/>). Information on the STROBE Initiative is available at www.strobe-statement.org.