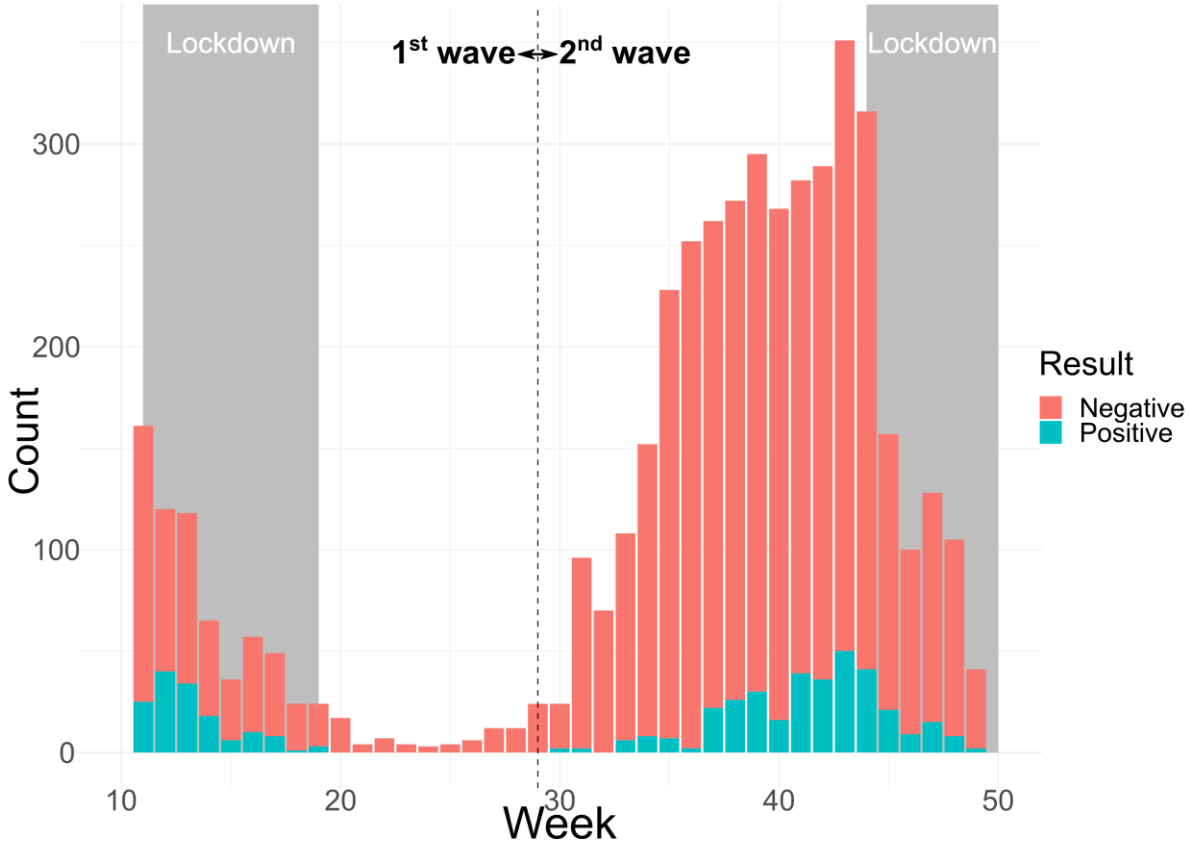
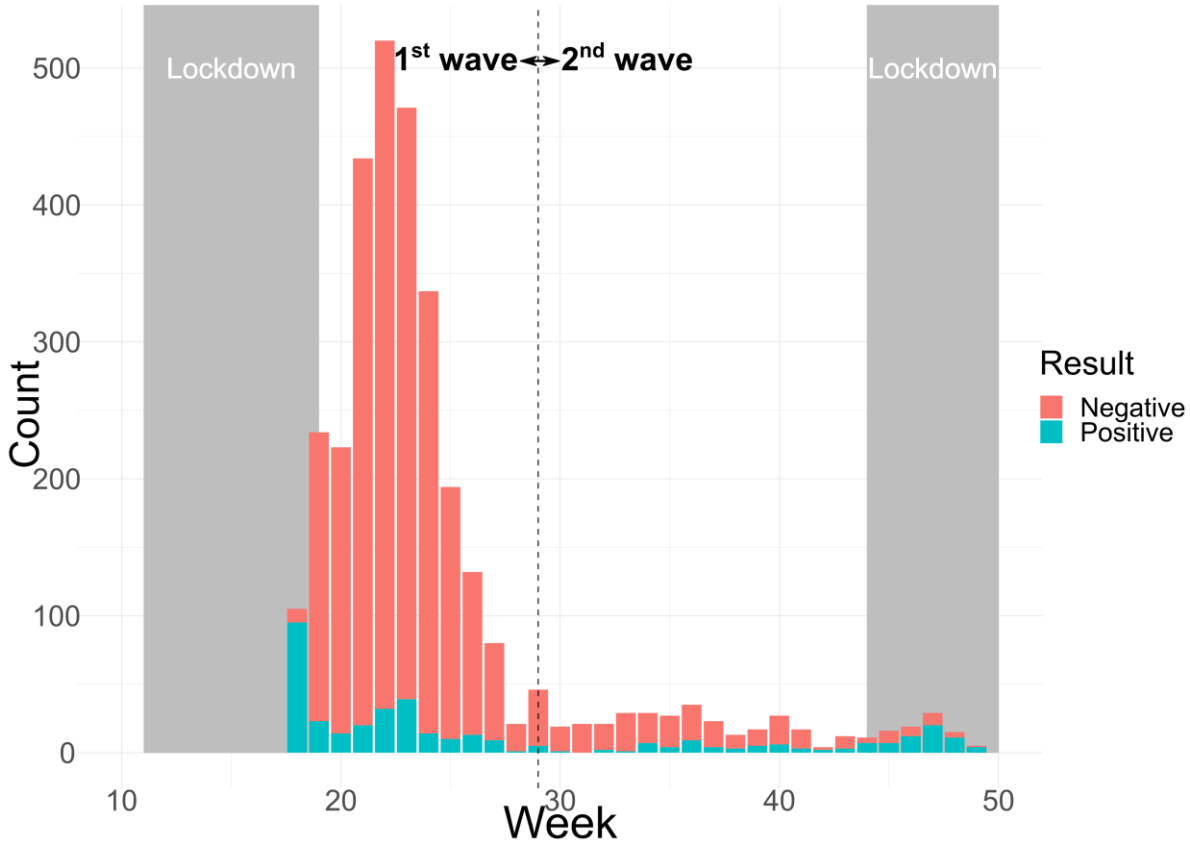
No SARS-CoV-2 reinfection among staff health-care workers: Prospective hospital-wide screening during the first and second waves in Paris

Maxime Wack, Hélène Péré, Nathalie Demory-Guinet, Najiby Kassis-Chikhani, Laurence Janot, Benoit Vedie, Laure Izquierdo, Laurent Bélec, David Veyer

A. Weekly PCR results for health-care workers



B. Weekly serology results for health-care workers



1 **No**

SARS-CoV-2 reinfection among staff health-care workers: Prospective hospital-wide screening during the first and second waves in Paris

Running title: No SARS-CoV-2 reinfection among HCWs

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Summary

Objectives. Risk of reinfection with SARS-CoV-2 among health-care workers (HCWs) is unknown. We assessed the incidence rate of SARS-CoV-2 reinfection in the real-life setting of a longitudinal observational cohort of HCWs from the *Hôpital Européen Georges Pompidou, Assistance Publique-Hôpitaux de Paris*, France, during the first and second waves of COVID-19 epidemic.

Methods. From March to December 2020, HCWs were subjected to molecular and serology testing of SARS-CoV-2. Reinfection was defined as a positive test result during the first wave, either by serology or PCR, followed by a positive PCR during the second wave. Evolution of COVID-19 status of HWCs was assessed by a Sankey diagram.

Results. A total of 7765 tests (4579 PCR and 3186 serology) were carried out and 4168 HCWs had at least one test result during the follow-up period with a positivity rate of 15.9%. No case of reinfection during the second wave could be observed among 102 positive HCWs of the first wave, nor among 175 HCWs found positive by PCR during the second wave who were negative during the first wave.

Conclusions. SARS-CoV-2 reinfection was not observed among HCWs, suggesting a protective immunity against reinfection that lasts at least 8 months post infection.

Keywords: SARS-CoV-2; reinfection; health-care workers; vaccination; epidemiology

Introduction

Health-care workers (HCWs) constitute a vulnerable population at high risk of SARS-CoV-2 infection (1–3). During the first wave of COVID-19 epidemic in France, as of May 13, 2020, more than 4,500 professionals were infected and 4 dead from COVID-19 in the *Assistance Publique-Hôpitaux de Paris* (AP-HP, which represents the largest group of university hospitals in Europe, accounting for about 100 000 employees).

SARS-CoV-2 infection elicits both humoral and cellular, mucosal and systemic specific immunity (4–6). A key question yet to be addressed is whether SARS-CoV-2 infection induces long-lasting protective or sterilizing immunity. Our understanding of the immune correlates of protection for SARS-CoV-2 infection and their durability remains limited and depends mainly on previous knowledge gained from SARS-CoV-1, the most closely related virus known to affect humans (5,7). Furthermore, a number of cases of SARS-CoV-2 reinfection in humans have now been reported a few months after initial infection (8,9), including HCWs (10), challenging the possibility of durable protective immunity. These findings may have implications for the need of continued protective measures and of further vaccination for persons previously infected with SARS-CoV-2 (9).

In France, the SARS-CoV-2 epidemic officially began on January 24, 2020, with the first confirmed case of COVID-19 imported from China. Early April, the number of deaths from coronavirus increased dramatically, with more than 10,000 people dying during that period of time. The first wave of the COVID-19 epidemic was contained due to the first national lockdown that extended from March 17, 2020 until May 10, 2020, the peak of the first wave being reached on the 1st of April 2020. During August 2020, COVID-19 cases began to rise again. On October 28, France entered a second nationwide lockdown with progressive lifting starting from December 15. The peak of the second epidemic wave was reached on the 20th of November 2020, but the epidemic has

since remained at a high plateau level. The duration period between the peaks of the first and second waves was 7.8 months (234 days).

Our institution, the *Assistance Publique-Hôpitaux de Paris*, has largely promoted both molecular and serological testing for SARS-CoV-2 among healthcare staff, from the first wave of the COVID-19 epidemic (1). Molecular testing for SARS-CoV-2 of symptomatic hospital employees with COVID-19-related symptoms started on February 24, 2020. In addition, starting early April 2020, the *Assistance Publique-Hôpitaux de Paris* offered HCWs serological screening for SARS-CoV-2 to assess the prevalence of the infection during the first epidemic peak.

The intensity of COVID-19 epidemic in France in 2020 with a high risk of re-exposure to SARS-CoV-2 infection for HCWs, as well as the availability of our centralized data-capture system of all molecular and serological SARS-CoV-2 tests results, prompted us to assess the risk of SARS-CoV-2 reinfections in a real-life setting in HCWs from our institution.

Materiel and method

Study design and participants recruitment.

The study was designed as an observational cohort with longitudinal analysis, focused on molecular detection of SARS-CoV-2 RNA and immune response to SARSCoV-2 in volunteers HCWs from the *Hôpital Européen Georges Pompidou*, *Assistance Publique-Hôpitaux de Paris*, tested during the first and second epidemic waves for suspected SARS-CoV-2 infection, contact tracing and SARS-CoV-2 infection serological surveillance. Data included HCW ages, SARS-CoV-2 PCR and SARS-CoV-2 serology sampling dates and results. HCWs gender was not available. All molecular and serological results were fully pseudonymized. July 15 2020 was considered as the limit between the first and second epidemic waves.

During the first epidemic wave, the HCW participants were categorized into two groups according to their molecular and serological results: positive if any SARS-CoV-2 PCR test and/or serology were positive, and negative if every SARS-CoV-2 PCR test and serology were negative. During the second epidemic wave, the participants were categorized into two groups according to their molecular results, as positive if any SARS-CoV-2 PCR test was positive, and negative if every SARS-CoV-2 PCR test were negative. In addition, HCW participants included during the first wave but not tested during the second wave, and those included during the second wave but not tested during the first wave were categorized as participants with not available results (NA). Finally, reinfection was defined as a positive result during the first wave, either by serology or PCR, followed by a positive PCR during the second wave. If iterative PCR sampling occurred, reinfection was considered if a negative result was surrounded by 2 positive results. Furthermore, if 2 PCRs were positive with more than 30 days between the sample dates without any negative results in between, results were individually assessed by a virologist to decide if reinfection was confirmed or if remnant RNA from the original infection was still detected when the cycle threshold (C_t) value of the PCR was over 33 (11). Positive serology during the first wave followed by positive PCR during the second wave was also considered a reinfection.

SARS-CoV-2 testing. Molecular detection of SARS-CoV-2 was carried out from flocked nasopharyngeal swab samples using Allplex™ 2019-nCoV Assay (Seegene, Seoul, Korea), a multiplex real-time PCR assay that detects three coronavirus target genes (E gene, RdRP gene and N gene) in a single tube, according to the manufacturer's instructions, as previously described (12). Abbott SARS-CoV-2 IgG assay detecting IgG against SARS-CoV-2 nucleoprotein was used on Architect analyzer (Abbott Architect™ i2000), according to manufacturer's instructions, as previously described (13). Index value threshold for positivity was 1.4. Qualitative results were used for analysis.

Statistical analysis. Categorical variables are presented as number (percentage and its [95% confidence interval]), and numeric variables as median or mean (SD or IQR). The number of test results was computed on a weekly basis. The evolution of the status for COVID-19 of HWCS between the two waves was assessed using a Sankey diagram, as described (14).

Ethics statement. Our observational study was carried out in accordance with the Declaration of Helsinki with no sampling addition to usual procedures. Swab and serum specimens were obtained only for standard diagnostic following medical prescriptions in the service of occupational medicine of our institution, and further eventual care. Under these conditions, the study was exempt from informed consent application, according to the French public health code (*Code de la Santé Publique*, article L 1121-1.1; https://www.legifrance.gouv.fr/). The dataset was completely anonymous and did not contain any identifiable personal health information.

Results

A total of 7765 tests, including PCR and serology, were collected from March 5, 2020 to December 4, 2020 by the occupational medicine service at the *Hôpital Européen Georges Pompidou*, Paris.

As depicted in the Figure 1, the weekly number of PCR tests evolved in accordance to the two waves of the epidemic of cases of SARS-CoV-2 infection in Paris. The number of serology tests per week evolved according to the curve resembling the first wave of the epidemic due to an institutional campaign for SARS-CoV-2 serology testing among HCWs between May and July.

Overall, 4579 PCR and 3186 serology tests were performed with positivity rates of 10.7% and 12.1%, respectively. A total of 4168 HCWs (median age, 34.3 years; IQR, 20.9 years) had at least one result, either PCR or serology, during the entire period with a positivity rate of 15.9%. There were 690 HCWs with at least one PCR result during the first wave *versus* 2340 during the

second wave with a positivity rate of 21.3% and 14.1%, respectively. Conversely, 2737 HCWs had a serology result during the first wave *versus* 417 during the second one, with a positivity rate of 9.8% and 27.8%, respectively (Table 1).

Among the 302 positive HCWs (either by serology or PCR) during the first wave, 102 were tested by PCR at least once during the second wave (154 total PCRs), and all their PCR results were negative (Figure 2). Furthermore, among the 330 PCR-positive HCWs during the second wave, 190 had a previous result from the first wave. Among those 190 that were tested in the first wave, there were 9 positive PCRs and 14 positive serology (15 total HCWs), and 170 negative serology and 45 negative PCRs (175 HCWs). All those 15 positive HCWs in the first wave were only positive in serology in the second wave. Among the 2657 negative HCWs during the first wave, 175 had a positive PCR result (185 tests) and 965 (1859 tests) had a negative result during the second wave. Overall, no case of reinfection during the second wave could be observed among the HCWs population in our hospital. The observed reinfection incidence was thus 0% [95% CI: 0 - 3.55%].

Discussion

We herein assessed the incidence rate of SARS-CoV-2 reinfection in the real-life setting of a longitudinal observational cohort of HCWs from one of the major university hospitals of the *Assistance Publique-Hôpitaux de Paris*, France, during the first and second waves of COVID-19 epidemic in 2020. The longitudinal prevalence rates of SARS-CoV-2 infection among HCWs, mostly symptomatic infections due to the design of the study, mimicked the curves of both waves of the COVID-19 epidemics in France with an overall rate of SARS-CoV-2 infection around 16%, demonstrating possible occupational risk for SARS-CoV-2 infection in HCWs despite barrier protection measures within the community and hospital settings. Among the included SARS-CoV-2 infected HCWs tested for SARS-CoV-2 PCR swab and/or IgG serology in both the first and second

waves of the COVID-19 epidemic in Paris, no case of SARS-CoV-2 reinfection could be evidenced. Thus, the risk of SARS-CoV-2 reinfection among HCWs appears negligible, despite the high level of exposure to the virus during epidemic waves. Taken together, these observations demonstrate that HCWs are at high risk of SARS-CoV-2 infection. SARS-CoV-2 reinfection was infrequent in study HCWs, highly suggestive of a protective immunity against reinfection lasting at least 8 months following the primary infection.(1–3,15)

More than a seventh (15.9%) of HCWs showed at least one positive biological marker for SARS-CoV-2 during the nearly 8 months of study inclusion. The risk of SARS-CoV-2 infection in included HCWs strictly followed the waves of the COVID-19 epidemic in France, during which between 3% and 7% of French people will have been infected by SARS-CoV-2 (16). Thus, our findings suggest that HCWs are likely at supplementary risk for occupational SARS-CoV-2 infection, as previously reported in Paris and other settings (1–3,15).

The possibility of SARS-CoV-2 reinfection has raised important issues about the strength and durability of the immune response to primary infection, which are key factors in predicting the course of the pandemic (8). We herein show SARS-CoV-2 reinfection was infrequent or absent among HCWs infected during the first wave of COVID-19 epidemic, despite their community and occupational re-exposure during the second wave. Our observations are in agreement with the low risk of SARS-CoV-2 reinfection generally reported (8,17), although the rates of reinfection with SARS-CoV-2 can vary widely, depending on the criteria used, and also because it is difficult to make the diagnosis with certainty, as previously pointed (18). By including the stringent criteria of viral genomic data to distinguish reinfection from persistent viral carriage, only 16 documented individual cases of reinfection confirmed by sequencing have been reported in the literature at the end of 2020 (8,19). Recently in Qatar, the risk of SARS-CoV-2 reinfection was estimated to be as low as 1 case of reinfection per 5000 infected individuals (17). However, a 2,800 person study found no symptomatic re-infections over a ~118 days window (20), and a 1,246 person study

observed no symptomatic re-infections over 6 months (21). Overall, SARS-CoV-2 re-infection is considered a possible but rare event (8), in keeping with our observational study based on field results from a well-documented hospital cohort.

SARS-CoV-2 infection consistently elicits neutralizing antibodies targeting the spike protein (in addition to other viral antigens), as well as CD8+ and CD4+ T cell responses (4–6). However, the duration of effective immunity to SARS-CoV-2 remains unknown, and the issue of waning immunity and reversion to a SARS-CoV-2 susceptible state over months to years is raised (22). Our observations of infrequent SARS-CoV-2 reinfection in hospital setting during the first and second waves of COVID-19 epidemic in Paris are strongly suggestive of efficient and protective immunity against reinfection that lasts at least 8 months post primary infection. These observations are in keeping with measurable immune memory in the three major branches of adaptive immunity (CD4+ T cell, CD8+ T cell, and humoral immunity) in ~95% of subjects 5 to 8 months post symptom onset, indicating that durable immunity against secondary COVID-19 disease is a possibility in most individuals (23). Obviously, further follow-up of SARS-CoV-2-infected HCWs over time may allow characterization of potential effects of SARS-CoV-2-specific immunity waning.

The need to vaccine previously SARS-CoV-2-infected individuals remains the matter of debate (9,24). It is possible that a fraction of the SARS-CoV-2-infected population with low immune memory would become susceptible to re-infection relatively soon (23). Current recommendations for COVID-19 vaccination from the World Health Organization (25) and the Centers for Disease Control and Prevention (USA) (26) do not consider the fact of having been infected with SARS-CoV-2. The National Health Service (UK) recommends to temporarily postponing vaccination for 4 weeks after the onset of COVID symptoms (27). In France, the *Haute Autorité de Santé*, Saint-Denis, has recommended that there is no need to systematically vaccinate people who have already developed a symptomatic form of COVID-19 (28). Our observations of HCWs infected by SARS-CoV-2 harboring durable protection against reinfection for at least 8

months indicate that COVID-19 vaccination does not *a priori* constitute a priority in case a previous infection, especially if vaccine doses are limited. As vaccine candidates advance worldwide, serostatus for COVID-19 could be relevant in HCWS before vaccination. If it is determined that a single SARS-CoV-2 exposure induces long-lasting protective immunity, a positive serology test could indicate that an individual does not require vaccination or should not receive priority for vaccination (29). Finally, in case of vaccination in HCWS who have already been infected, the vaccination schedule could also possibly be simplified to a single dose.

Our study has some limitations. Except for HCWs that were recently exposed to positive patients or colleagues (contact tracing), only HCWs volunteering for testing were included and probably most of them had symptomatic infection. Furthermore, the possibility could be that reinfections (if they occur) would have been asymptomatic or well tolerated and that they went unnoticed without a biological diagnosis. Finally, HCWs in our institution were exposed to the SARS-CoV-2 variants circulating in France until December 2020, and our study does not evaluate the possibility of reinfection by other variants, such as the new U.K. variant of SARS-CoV-2, which various modeling exercises have estimated to be up to 70% more transmissible than the previously circulating form of the virus (30).

In conclusion, SARS-CoV-2 reinfection appears to be a rare phenomenon, if it exists, in HCWs despite their high level of exposure to the virus during epidemic peaks. This feature may suggest that effective immunity against SARS-CoV-2 infection develops after primary infection and lasts for at least 8 months, and that it is able to protect against reinfection. Finally, HCWs already infected with SARS-CoV-2 do not constitute a priority population for vaccination, especially as vaccine doses remain rationed.

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Author's contribution. HP, LB, DV designed the research. NDG, KCN, LJ provided clinical care and biological specimens. LB, DV, LI, BV analyzed the samples. MW, DV performed statistical analysis. HP, MW, LB, DV drafted the manuscript. All authors read and approved the final manuscript.

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Table 1. SARS-CoV-2 RNA PCR and SARS-CoV-2-specific IgG serology results carried out among HCWs in *Hôpital* Européen *Georges Pompidou*, Paris, France, during the first and second waves of the COVID-19 epidemic in 2020.

	First wave	**Second wave**	**Total**
HCWs tested by PCR and/or serology [*n*]	2902	2506	4168
Age of tested HCWs [*median (IQR); years*]	37.1 (20.6)	32.1 (19.5)	34.3 (20.9)
SARS-CoV-2 PCR and serology [*n*]	3510	4255	7765
PCR [*n*]	759	3820	4579
[*percent of positive (CI)*]	19.5% (16.7 – 22.5)	8.9% (8.0 – 9.8)	10.7% (9.8 – 11.6)
Serology [*n*]	2751	435	3186
[*percent of positive (CI)*]	9.8% (8.7 – 10.9)	26.7% (22.6 – 31.1)	12.1 % (11.0 – 13.3)
HCWs tested positive by PCR and/or serology [*n*]	302	379	666
[*percent (CI)*]	10.4% (9.3 – 11.6)	15.1% (13.7 – 16.6)	15.9% (14.9 – 17.1))
HCWs tested by both PCR and serology [*n*]	525	251	1565
[*percent of positivity by PCR or serology (CIs)*]	41.5% (37.3 – 45.9)	47% (40.7 – 53.4)	31.3% (29.0 – 33.7)
[*percent of positivity by both PCR and serology (CIs)*]	21.5% (18.1 – 25.3)	26.7% (21.3 – 32.6)	11.6% (10.1 – 13.3)
HCWs with at least one PCR result [*n*]	690	2340	2719
[*percent of positivity (CI)*]	21.3% (18.3 – 24.5)	14.1% (12.7 – 15.6)	17.5% (16.1 – 18.9)
PCR per HCW [*mean (SD)***]**	1.10 (0.30)	1.63 (0.90)	1.68 (1.02)
HCWs with at least one serology result [*n*]	2737	417	3014
[*percent of positivity (CI)*]	9.8% (8.7 – 10.9)	27.8% (23.6 – 32.4)	12.3% (11.2 – 13.4)
Serology per HCW [*mean (SD)***]**	1.01 (0.08)	1.04 (0.23)	1.06 (0.25)

CI: Confidence interval; IQR: Interquartile range; HCW: Health-care worker; n: number; SD: Standard deviation;

Figures Legends

Figure 1. PCR and serology results during the first and the second wave of the COVID-19 pandemic. Weekly SARS-CoV-2 RNA PCR results (A.) and SARS-CoV-2-specific IgG serology results (B.) among staff HCWs in *Hôpital Européen Georges Pompidou*, Paris, France, during the first and second waves of the COVID-19 epidemic in 2020. The dashed vertical line indicates the separation between the first and second waves. The shaded areas correspond to the lockdown period decided by the French government. Positive and negative results are in red and blue, respectively.

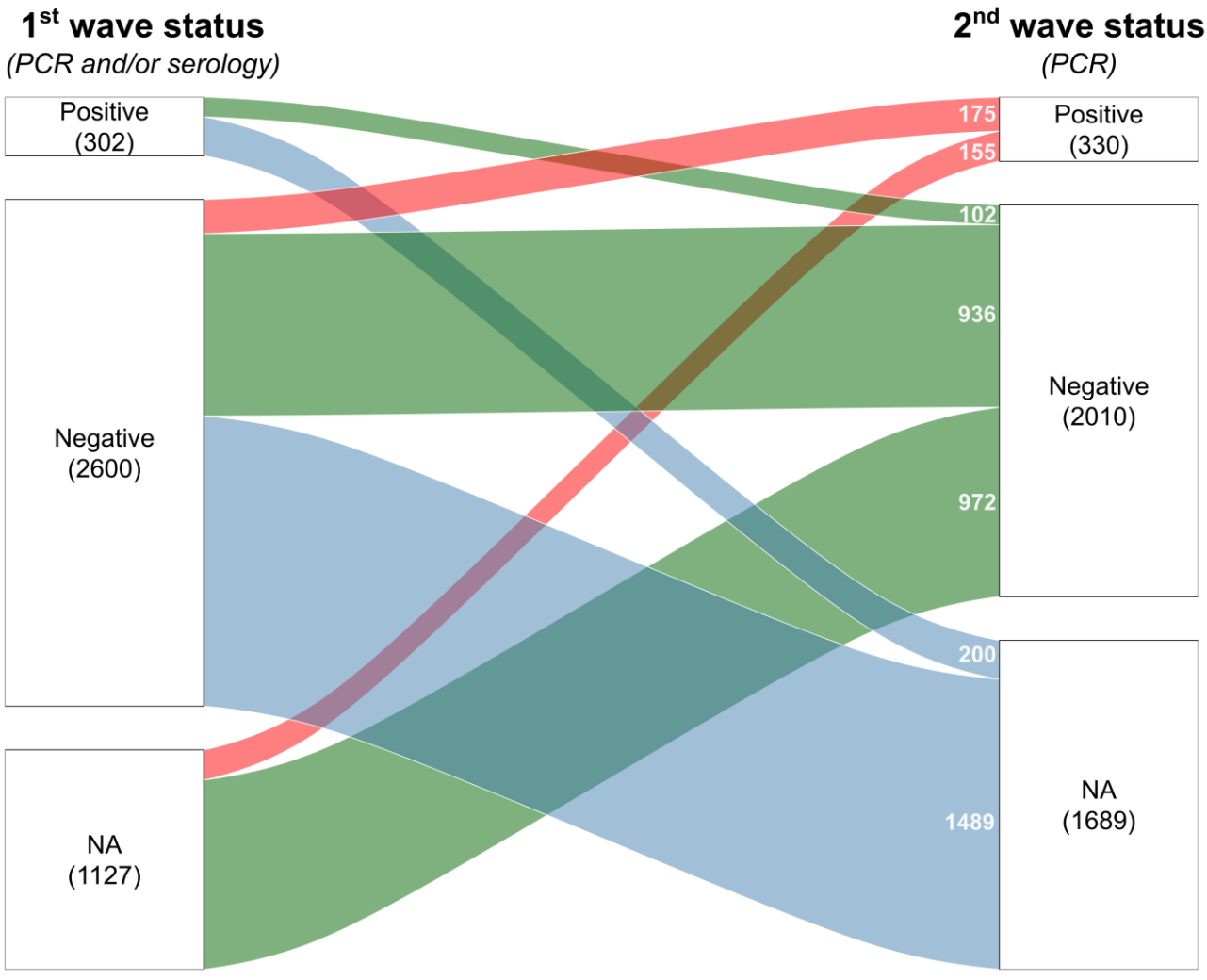

Figure 2. Health-care workers COVID-19 status evolution between the first and the second wave of the COVID-19 pandemic. Sankey diagram showing the evolution of SARS-CoV-2 status among HCWs in *Hôpital Européen Georges Pompidou*, Paris, France, between the first and the second

waves. Boxes on the left represent results either by PCR and/or serology during the first wave of the SARS-CoV-2 epidemic, while those on the right represent results only by PCR during the second wave. The different lanes show the status evolution with lane sizes proportional to the number of HCWs. The blue lanes are for HCWs with no result during the second wave. The green lanes are for HCWs with a negative result during the second wave. The red lanes are for HCWs with a positive result during the second wave. NA: Results not available.