SHORT COMMUNICATION

SARS-CoV-2 infection in persons living with HIV: A single center prospective cohort

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Abstract

Information about severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection in HIV-infected individuals is scarce. In this prospective study, we included HIV (human immunodefeciency virus)-infected individuals (people living with HIV [PLWHIV]) with confirmed SARS-CoV-2 infection and compared them with PLWHIV negative for SARS-CoV-2. We compared 55 cases of SARS-CoV-2 infection with 69 asymptomatic PLWHIV negative for SARS-CoV-2 reverse transcription-polymerase chain reaction and/or serology. There was no significant difference between SARS-CoV-2 positive or negative patients for age distribution, gender, time with HIV infection, nadir CD4-cell counts, type and number of co-morbidities, current CD4 and CD8 counts and type of anti-HIV therapy. Positive patients presented with a median of three symptoms (interquartile range, 1-3). Most common symptoms were fever (76%), dyspnea (35%), anosmia (29%) non-productive cough (27%), fatigue 22%), and ageusia (20%). Ten patients (18%) were completely asymptomatic. Four (7.2%) subjects died of coronavirus disease 2019. Factors significantly ($P < .05$) associated with death included age and number of co-morbidities, while time from HIV infection and lower current CD4 counts were significant only in univariate analysis. HIV-infected individuals are not protected from SARS-CoV-2 infection or have a lower risk of severe disease. Indeed, those with low CD4 cell counts might have worse outcomes. Infection is asymptomatic in a large proportion of subjects and this is relevant for epidemiological studies.

KEYWORDS
asymptomatic, CD4, CNS, cohort, co-morbidities, COVID-19, HIV, mortality, risk factors, SARS-CoV-2, symptoms

1 | INTRODUCTION

On 31 December 2019 an outbreak of pneumonia was first reported in Wuhan, China, and soon after identified as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2),[1] and the disease due to this infection was named coronavirus disease 2019 (COVID-19). The increasing number of cases of COVID-19 worldwide, induced World Health Organization to declare a pandemic.[2]

Italy is one of the most affected Countries in Europe with 238 499 confirmed cases as of 21 June 2020.[3] The Province of Bergamo with about 1 100 000 inhabitants and 14 120 infected subjects is one of the focal areas of the Italian epidemic.[3]

Very little is known about how persons living with human immunodefeciency virus (PLWHIV) could react to SARS-CoV-2 infection. It has been postulated that human immunodefeciency virus (HIV)-infected individuals might be at an increased risk because

of the presence of comorbidities, lower CD4 cell counts, or unsuppressed virus.[4-6] Conversely, it has been suggested that immunosuppression might prevent or lower the severe cytokine storm observed in COVID-19 reducing its severity.[7,8] Finally, antiretrovirals have been indicated as able to modify the risk of infection with SARS-CoV-2 and its clinical picture in PLWHIV.[8,9]

This uncertainty is due to the paucity of work on the topic, being available data based on small cases series,[4,10-14] or on two larger cohorts either uncontrolled[8] or with controls untested for SARS-CoV-2.[6]

This is the first cohort study in which PLWHIV either symptomatic or not were tested for SARS-CoV-2.

2 | METHODS

2.1 | Study characteristics

This is a cohort, single center, clinical, prospective study performed in a Province of Northern Italy highly hit by the SARS-CoV-2 epidemic. Aim of the study was to identify possible characteristics of PLWHIV that could correlate with the risk of acquiring SARS-CoV-2 infection and, in the case of infection, would influence the outcome.

2.2 | Data collection

Since the beginning of the epidemic, data of all suspected or confirmed COVID-19 cases were recorded in a specific database linked to a common research project authorized by the local Ethical Committee. All patients gave their informed consent. Data for the present cohort study were extracted from this database. They were crosslinked with information from the outpatient clinic electronic health records. Data of patients not admitted to the hospital were obtained by each patient during a visit performed in the first 15 days of June. All these patients subscribed an informed consent, too.

Recorded variables were age, gender, comorbidities, HIV-specific variables, such as year of HIV infection diagnosis, nadir and most recent (eg, within 3 months from 1st March 2020) CD4 cell counts, CD8 cell counts, CD4/CD8 ratios, HIV-RNA plasma levels, current antiretroviral therapy. Clinical characteristics of COVID-19, and outcomes were recorded for SARS-CoV-2 positive patients.

2.3 | Laboratory procedures

During the acute phase of the epidemics, Laboratory diagnosis of SARS-CoV-2 infection was done by reverse transcription-polymerase chain rreaction (RT-PCR) with primer and probes targeting E, RdRp and N genes. Nasopharyngeal swabs or lower respiratory tract aspirates were tested only in individuals admitted to the hospital as public health authorities' regulations did not recommend tests in individuals with mild symptoms not admitted to hospitals. Later on, for specific categories (eg, health-care workers) nasopharyngeal swabs were permitted. In all other cases, the diagnosis was achieved by means of serological tests.

Serological diagnosis was made with VivaDiagTM COVID-19 immunoglobulin M/immunoglobulin G immune-chromatographic assay from VivaChekTM Biotech (China), performed according to manufacturer's instructions.

Blood tests were done according to the clinical needs of each patient by means of routine laboratory procedures. Radiologists performed Chest x-ray assessments.

2.4 | Definitions

Confirmed COVID-19 was defined by positive RT-PCR for SARS-CoV-2 in respiratory samples or a positive serological test. Suspected cases were those in individuals with clinical and radiological findings compatible with COVID-19, but whose RT-PCR results were inconclusive in the absence of any other proven cause (eg, *Pneumocystis jiroveci* pneumonia).

The severity of disease was scored based on the worse type of respiratory support needed (eg, invasive mechanical ventilation, noninvasive mechanical ventilation, oxygen mask).

Confirmed SARS-CoV-2 negative patients were asymptomatic, with either RT-PCR or serology or both tests negative.

2.5 | Statistical analysis

No sample size was calculated given that all known individuals with a diagnosis of COVID-19 were included. Continuous variables are presented as median and interquartile range (IQR). Categorical variables are expressed as number of patients (percentage).

Comparisons were assessed by using the Mann-Whitney U test for continuous variables, whereas categorical variables were assessed by the χ^2 test. We used a binary logistic regression model to explore the factors associated with COVID-19 diagnosis and the risk of death. Statistical significance was defined as a two-sided $P < .05$. All statistics were done with SPSS Statistics for Windows, version 17.0.

2.6 | Role of the funding source

This study has no funder.

3 | RESULTS

At our center, 2898 PLWHIV are currently in active follow-up. Among these, we identified 55 cases of SARS-CoV-2 infection either by RT-PCR test (16 cases, 29%), serology (33 cases, 60%), or clinical grounds (6 cases, 11%) when tests were negative, the clinical picture was highly suggestive and no other explanation was found. We

compared these cases with 69 PLWHIV who tested negative for RT-PCR (16 cases, 23%) or serology (53 cases, 77%).

Baseline characteristics are reported in Table 1. No significant difference was observed between SARS-CoV-2 positive and negative patients for age distribution, gender, time with HIV infection, nadir CD4 cell counts, type and number of co-morbidities, current CD4 and CD8 counts and type of anti-HIV therapy. The only exception was a

barely significant difference in the use of integrase inhibitors (higher in in SARS-CoV-2 tinfected), while other anchor drugs and backbone components were used in similar proportions. Specifically, as the role of tenofovir is debated, we compared subjects receiving or not this drug. Tenofovir was a part of the antiretroviral regimen in 33 (60.0%) of SARS-CoV-2 positive subjects and in 42 (60.8%) of controls. In the positive group, only one patient had detectable HIV-RNA

TABLE 1 baseline characteristics in 55 subjects SARS-CoV-2 positive and 69 SARS-CoV-2 negative

Variable	SARS-CoV-2 positive	SARS-CoV-2 negative	Total	P value
Gender				.401
Male	44 (80.0%)	50 (72.5%)	94 (75.8%)	
Female	11 (20.0%)	19 (27.5%)	30 (24.2%)	
Age, y	54 (49-58)	52 (46-59)	51 (46-59)	.249
Risk factor for HIV				.434
Heterosexual contacts	24 (43.7%)	38 (55.0%)	62 (50.0%)	
MSM	18 (32.7%)	17 (24.7%)	35 (28.2%)	
IVDU	13 (23.6%)	14 (20.3%)	27 (21.8%)	
Years since HIV infection	16 (9-23)	14 (9-23)	13 (8-18)	.757
Nadir CD4 count, cells per μL	281 (37-550)	292 (88-528)	292 (37-547)	.788
Antiretroviral drugs				
NRTIs	47 (85.4%)	56 (81.1%)	103 (83.0%)	.633
NNRTIs	20 (36.4%)	31 (44.9%)	51 (41.1%)	.363
PIs	11 (20.0%)	24 (34.7%)	35 (28.2%)	.075
INIs	32 (58.2%)	27 (39.1%)	59 (47.6%)	.046
Number of ARV drugs	3 (2-3)	3 (3-3)	3 (2-3)	.422
Number of co-morbidities	1 (0-1)	1 (0-1)	1 (0-1)	.642
Major co-morbidities				
Cardiovascular diseases	9 (16.4%)	5 (7.2%)	14 (11.2%)	.154
Hypertension	12 (21.8%)	11 (15.9%)	23 (18.5%)	.487
Gastro-enteric	6 (10.9%)	6 (8.7%)	12 (9.6%)	.764
Malignancies	5 (9.0%)	8 (11.6%)	13 (10.4%)	.772
Neurological	4 (7.3%)	9 (13.0%)	13 (10.4%)	.383
Diabetes	3 (5.5%)	3 (4.3%)	6 (4.8%)	1.000
HBV co-infection	5 (9.0%)	5 (7.2%)	10 (8.0%)	.749
HCV co-infection				.985
Negative	41 (74.5%)	52 (75.3%)	93 (75.0%)	
Cured	13 (23.6%)	16 (23.1%)	29 (23.4%)	
HCV-RNA positive	1 (1.8%)	1 (1.4%)	2 (1.6%)	
Last CD4 count, cells per μL	904 (557-1110)	822 (556-1035)	829 (559-1054)	.486
Last CD8 count, cells per μL	953 (633-1279)	911 (591-1226)	921 (629-1262)	.720
Last CD4/CD8 ratio	0.89 (0.64-1.20)	0.92 (0.56-1.20)	0.96 (0.54-1.2)	.731
Last HIV-RNA < 50 copies/mL	54 (98.1%)	64 (92.7%)	118 (95.2%)	.376

Note: Number and (percentages) or median and (IQR).
Abbreviations: ARV, antiretroviral; HIV, human immunodeficiency virus; INI, integrase inhibitor; IQR, interquartile range; IVDU, intravenous drug users; MSM, men who have sex with men; NRTI, nucleoside reverse transcriptase inhibitor; NNRTI, non-nucleoside reverse transcriptase inhibitor; PI, protease inhibitor; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

(VL 431 000 copies/mL, CD4 count 29 cells/µL). He was diagnosed HIV-positive in February and admitted to the hospital for dizziness, loss of visual acuity and weight loss. Nasal swabs and serology for SARS-CoV-2 were negative. Because of worsening of the clinical picture, he performed a lumbar puncture. RT-PCR was negative for neurotropic viruses, but was positive for SARS-CoV-2. Two months later SARS-CoV-2 was negative in cerebro spinal fluid, but he did not develop an immunological response to the virus. In the control group, four patients showed viral blips (<200 copies/mL) at last control. One subject had an HIV-RNA of 55 100 copies/mL. He was the only patient that, in the negative group, was admitted, in April, to the COVID-19 sub-intensive unit because of fever, respiratory distress, and a patchy ground glass alteration at chest radiology. He tested positive for HIV during the hospital stay, his nasal swabs were negative for SARS-CoV-2 and RT-PCR was negative on bronco-aspirate that, conversely, turned positive for *Pneumocystis jiroveci*. One month later, his SARS-CoV-2 serology was negative, too.

Amongst positive cases only 15 (27%) were admitted to the hospital because of COVID-19. Positive patients presented with a median of three symptoms (IQR, 1-3). The most common symptoms were fever (76% of cases), dyspnea (35%), anosmia (29%) non-productive cough (27%), fatigue 22%), and ageusia (20%). However, 10 patients (18%) were completely asymptomatic.

In positive subjects, we evaluated the severity of diseases. Overall, 11 subjects (20%) required more intensive respiratory support. Factors associated with the need of invasive mechanical ventilation or c-PAP included the number of co-morbidities (P = .02) and age (P = .004). Only 11% of subjects without co-morbidities required invasive mechanical ventilation or c-PAP compared with 18% of those with 1 comorbidity and 41% of those with two or more co-morbidities. Median age in patients requiring at the most oxygen mask was 53 years (IQR, 47-57) and raised to 62 years (IQR, 57-66) in those who needed more intensive support. In the multivariable model, however, only age (P = .023) retained a statistical significance.

COVID-19 had a fatal outcome in four (7.2%) subjects. According to univariate analysis, factors significantly associated with death included age, time from HIV infection, number of current co-morbidities and current CD4 counts (Table 2). However, only age and the number of co-morbidities were statistically associated with the outcome in the multivariate model.

4 | DISCUSSION

Our study addresses some of the unknown aspects of SARS-CoV-2 infection in PLWHIV.

A distinctive aspect of our study is the fact that all included patients were tested for SARS-CoV-2 making it one of the largest cohort of individuals with HIV and demonstrated SARS-CoV-2 co-infection.

Interestingly, none of the classical variables linked to HIV infection, such as nadir CD4 cell counts, time of HIV infection or current CD4 counts were predictive of the risk of acquiring SARS-CoV-2 infection, nor the use of specific antiretrovirals resulted having a protective effect as previously reported.[8,9]

It can be postulated that in PLWHIV, as in the general population, other variables not HIV-related, such as work activities or adherence to lock-down and social-distancing procedures might be prominent in determining the risk of infection.[13]

Previous studies have suggest that immunosuppression and low CD4 cell counts might protect HIV-infected individuals from developing the cytokine storm observed in patients with COVID-19.[7,8,14] In our casuistry, the seriousness of the infection was not influenced by the current immunological situation nor by the previous immune-depressive status as measured by the nadir CD4 cell counts. As a matter of fact, patients with a fatal outcome presented with lower CD4 counts, even if the difference was not statistically significant when analyzed according to a multivariate model. These findings confirm previously reported observations.[6,15]

Most previous studies in PLWHIV concentrated on patients admitted to the hospital because of COVID-19.[6,8,15] In our cohort, most of the included patients were not admitted to the hospital, testifying that, even in HIV-positive subjects, SARS-CoV-2 may present as a mild diseases not requiring

TABLE 2 Variables associated with the risk of death due to COVID-19

Variable	Survivors	Deceased	P value Univariate	P value Multivariate
Age, y	54 (48-58)	65 (59-69)	.025	.044
Years since HIV infection	14 (9-23)	24 (20-32)	.044	.053
Number of co-morbidities			.037	.029
None	27 (100%)	0		
One	15 (93%)	1 (7%)		
Two	5 (83%)	1 (17%)		
More than two	4 (66%)	2 (44%)		
Last CD4 count, cells per µL	913 (557-1119)	514 (427-601)	.001	.187

Note: Number and (percentages) or median and (IQR).
Abbreviations: COVID-19, coronavirus disease 2019; HIV, human immunodeficiency virus; IQR, interquartile range.

hospital admission or intensive care.[16,17] As a matter of fact, the proportion of asymptomatic patients resulted quite high (18%). Such a value counting for almost a fifth of the potentially infected subjects needs to be taken into account in future analysis of the incidence of SARS-CoV-2 in HIV infected subjects[17] and potentially strengths the use of serological tests that allowed us to detect subjects with mild symptoms or even without any symptom.

Our study has several limitations. First, the small number of individuals prevents us from generalizing our results, also because obtained in a highly epidemic area and therefore not necessarily applicable in places with a different prevalence. Second, the small sample does not allow us to definitively establish the role of immune status or the presence of comorbidities in the clinical presentation and outcomes. Third, as stated, some bias may exist in the rate of infection because local recommendations restricted confirmatory testing. Although we included all the HIV-infected individuals with documented SARS-CoV-2 infection, the high rate of asymptomatic patients we found raises the doubt that more unknown subjects could have been infected.

The two specific cases we report in the results stress the risk of misdiagnosis, especially at the beginning of the epidemic and the importance of extended testing even if in extremely immune-depressed patients the immunological response may fail to mount.

In conclusion, none of the parameters classically used to define immune suppression or risk of immune impairment in HIV-positive subjects does correlate with the risk of acquiring SARS-CoV-2 infection. Although low CD4 counts were not associated with the positivity for SARS-CoV-2, relative immunosuppression did seem to affect disease severity, and it might be associated with adverse outcomes. By contrast, there was no evidence that any specific antiretroviral drug affected SARS-CoV-2 infection or COVID-19 severity.

The disease may cover a vast range of clinical pictures, being almost one fifth of infected individuals asymptomatic.

Variables already described for the general population as risk factors for a more severe disease, such as advanced age and the presence of multiple co-morbidities do apply to PLWHIV, too.

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CONFLICT OF INTERESTS
The authors declare that there are no conflict of interests.

DATA AVAILABILITY STATEMENT
Data available on request from the authors.

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REFERENCES
1. Phelan AL, Katz R, Gostin LO. The novel coronavirus originating in Wuhan, China: challenges for global health governance. *JAMA*. 2020; 323:709.
2. WHO. WHO Director-General's opening remarks at the media briefing on COVID-19—11 March 2020. Geneva: World Health Organization. https://www.who.int/dg/speeches/detail/who-directorgeneral-s-opening-remarks-at-the-media-briefing-on-covid-19-11-march-2020. Accessed June 21, 2020.
3. Italian Ministry of Health (Ministero della Salute). Novel coronavirus. http://www.salute.gov.it/portale/nuovocoronavirus/home NuovoCoronavirus.jsp?lingua=english. Accessed June 21, 2020.
4. Zhu F, Cao Y, Xu S, Zhou M. Co-infection of SARS-CoV-2 and HIV in a patient in Wuhan city, China. *J Med Virol*. 2020;92:529-530. https://doi.org/10.1002/jmv.25732
5. Zhu F, Cao Y, Xu S, Zhou M. Reply to comments on "Co-infection of SARS-CoV-2 and HIV in a patient in Wuhan city, China." *J Med Virol*. 2020. https://doi.org/10.1002/jmv.25838
6. Vizcarra P, Pérez-Elías MJ, Quereda C, et al. Description of COVID-19 in HIV-infected individuals: a single-centre, prospective cohort. *Lancet HIV*. 2020. https://doi.org/10.1016/S2352-3018(20)30164-8
7. Hileman CO, Funderburg NT. Inflammation, immune activation, and antiretroviral therapy in HIV. *Curr HIV/AIDS Rep*. 2017;14:93-100.
8. Gervasoni C, Meraviglia P, Riva A, et al. Clinical features and outcomes of HIV patients with coronavirus disease 2019. *Clin Infect Dis*. 2020. https://doi.org/10.1093/cid/ciaa579
9. Jockusch S, Tao C, Li X, et al. Triphosphates of the two components in DESCOVY and TRUVADA are inhibitors of the SARS-CoV-2 polymerase. *bioRxiv*. 2020. https://doi.org/10.1101/2020.04.03.022939
10. Blanco JL, Ambrosioni J, Garcia F, et al. COVID-19 in patients with HIV: clinical case series. *Lancet HIV*. 2020;7:314. https://doi.org/10.1016/S2352-3018(20)30111-9
11. Ridgway JP, Farley B, Benoit J-L, et al. A case series of five people living with HIV hospitalized with COVID-19 in Chicago, Illinois. *AIDS Patient Care STDS*. 2020. https://doi.org/10.1089/apc.2020.0103
12. Zhao J, Liao X, Wang H, et al. Early virus clearance and delayed antibody response in a case of COVID-19 with a history of co-infection with HIV-1 and HCV. *Clin Infect Dis*. 2020. https://doi.org/10.1093/cid/ciaa408
13. Chou R, Dana T, Buckley DI, Selph S, Fu R, Totten AM. Epidemiology of and risk factors for coronavirus infection in health care workers. *Ann Intern Med*. 2020;173:120-136. https://doi.org/10.7326/M20-1632
14. Marimuthu J, Kumar BS, Aravind Gandhi P. HIV and SARS CoV-2 co-infection: a retrospective, record based, case series from South India. *J Med Virol*. 2020. https://doi.org/10.1002/jmv.26271
15. Ruan L, Zhang Y, Luo Y, et al. Clinical features and outcomes of four HIV patients with COVID-19 in Wuhan, China. *J Med Virol*. 2020. https://doi.org/10.1002/jmv.26223
16. Masukume G, Mapanga W, Grinberg S, van Zyl DS. COVID-19 and HIV co-infection an emerging consensus. *J Med Virol*. 2020. https://doi.org/10.1002/jmv.26270
17. Patel RH. Clinical outcomes and prognosis of patients with HIV and SARS-CoV-2 coinfection. *J Med Virol*. 2020. https://doi.org/10.1002/jmv.26177