LUNG ULTRASOUND IN RULING OUT COVID-19 AMONG HEALTHCARE WORKERS IN TWO ITALIAN EMERGENCY DEPARTMENTS: A MULTICENTER STUDY.

Running title: Lung ultrasound in monitoring for COVID-19 of healthcare professionals.

Roberto Copetti [1], Giulia Amore [1], Caterina Anna Giudice [1], Daniele Orso [2], Silvia Cola [1], Pierpaolo Pillinini [3], Chiara Rocco [3], Dario Cappello [3], Alessia Geneve Dibenedetto [3], Stefano Meduri [4].

[1] Department of Emergency Medicine, ASUFC Latisana Community Hospital, Latisana (UD).

[2] Department of Medicine, University of Udine, & Department of Anesthesia and Intensive Care Medicine, ASUFC University Hospital of Udine.

[3] Department of Emergency Medicine, ASUFC "Sant'Antonio Abate" Tolmezzo Community Hospital, Tolmezzo (UD).

[4] Department of Radiology, ASUFC Latisana Community Hospital, Latisana (UD).

Corresponding Author: Daniele Orso. Department of Medicine, University of Udine and Department of Anesthesia and Intensive Care Medicine, ASUFC University Hospital of Udine, P.le Santa Maria della Misericordia 15, 33100 Udine, Italy. Electronic mail: sd7782.do@gmail.com; Orcid ID: 0000-0001-7136-0343.

DECLARATIONS:

Consent for publication: Each patient included in the study signed informed consent for participation in the study, the collection of personal data and consent to the publication of data obtained from the analysis of clinical data, according to the Declaration of Helsinki.

Availability of data and material: Data are available following a reasoned request.

Competing interests: No conflict of interest must be declared for any of the authors.

Funding: No fund was used for this study.

Authors' contributions: RC designed the study, reviewed the final diagnosis and supervised the final draft; GA reviewed the radiographic images; CG collected the data; DO performed the statistical analysis, drafted the first draft and supervised the final draft; SC designed the study, collected the data, drafted the first draft and supervised the final draft; PPP reviewed the final diagnosis; CR collected the data; DC collected the data; AGD collected the data; SM reviewed the radiographic images.

Acknowledgments: dr. Marcella Montico for her statistical advice.

Abstract:

Purpose: The high percentage of asymptomatic patients and the non-high sensitivity of real-time reverse transcription-polymerase chain reaction (RT-PCR) test on nasopharyngeal swab cause some healthcare workers to be infected but asymptomatic and a source of spread of the epidemic. This study aimed to verify if the lung ultrasound (LUS) had enough high negative predictive value to rule out coronavirus disease 2019 (COVID-19) among a population of healthcare workers operating in the Emergency Department.

Methods: A multicenter prospective observational study was conducted, enrolling healthcare workers among the staff of two Emergency Departments in Northeast Italy. The definitive diagnosis of COVID-19 was established by an adjudication committee, based on the clinical data and RT-PCR on nasopharyngeal swab result.

Results: From March 30, 2020, to April 22, 2020, we enrolled 155 cases. The adjudication committee determined two true positives for COVID-19. Twenty-one healthcare workers presented suggestive symptoms (2 true positives and 19 false positives). The nasopharyngeal swab was positive in one case (1 false-negative case). LUS was suggestive for COVID-19 pneumonia in 4 cases (2 false-positive cases). The diagnostic accuracy of LUS was 98.7% (95% CI 95.4%-99.8%). The sensitivity and the specificity of LUS were 100% (95% CI 15.8% -100%) and 98.7% (95% CI 95.4% - 99.8%), respectively. The negative predictive value was 100% (95% CI 100% -100%).

Conclusion: LUS has a good enough negative predictive value for ruling out COVID-19 in a population of healthcare workers exposed to COVID-19.

Keywords: COVID-19; Lung ultrasound; Accuracy; Healthcare personnel; Emergency Department.

INTRODUCTION

Since the first reports of coronavirus disease 2019 (COVID-19) in Italy, the health authorities have implemented specific protocols to triage the infected patients in the Emergency Department [1]. These protocols, although with variations from the center to center, provide for early isolation of highly suspected patients and the execution of a real-time reverse transcription-polymerase chain reaction **(**RT-PCR) test on samples of body secretions (i.e., in the Emergency Department, a nasopharyngeal swab) and further exams to establish the severity of the disease (i.e., arterial blood gas analysis, chest radiography, lung ultrasound). Health personnel assigned to identify positive COVID-19 patients, as well as wearing personal protective equipment (PPE, i.e., gloves, visor, N95 mask, gown), were subjected to surveillance checks (i.e., nasopharyngeal swab) [2]. The importance of guaranteeing healthcare workers' safety lies in ensuring a good state of health for workers [3] and containing epidemic outbreaks and preventing healthcare personnel from being a source of coronavirus spread [4]. Healthcare professionals have an increased risk of exposure and are potential carriers of subsequent transmission. Over 3,300 healthcare workers have been infected in China (4% of 81,285 reported infections) [5]. In Spain, on March 25, 2020, nearly 6,500 medical staff members were affected (13.6%) of the country's 47,600 total cases, 1% of the healthcare workforce [6]. Therefore, it is essential to identify health care professionals infected with COVID-19 early and rule out healthy staff (who may present suggestive symptoms for COVID-19, as they are not very specific). During this epidemic, the role of lung ultrasound (LUS) became central to both identifying infected patients and monitoring them once they are hospitalized [7,8].

This study aimed to verify whether LUS has a negative predictive value sufficient to be used in the exclusion of COVID-19 from a population of health workers operating in two Emergency Departments.

METHODS

Setting and participants

From March 30, 2020, to April 22, 2020, every health care worker of two rural Emergency Departments in the Northeast of Italy (Hospital of Latisana – of about 25,000 patients/year – and Hospital of Tolmezzo – of about 15,000 patients/year) was asked to participate voluntarily in a LUS screening to establish the presence or absence of findings compatible with COVID-19 pulmonary involvement. The workers considered were 42

physicians, 72 nurses, 21 support workers, and 25 ambulance drivers stationed in the emergency departments. During the period considered, the two emergency departments visited COVID-19-positive patients, with a prevalence of the disease among the patients visited about 20%.

Data Collection, Adjudication, and Follow-Up

Clinical data were registered prospectively. The definitive diagnosis of COVID-19 pneumonia was established by an adjudication committee, based on the clinical data, the result of the RT-PCR test on nasopharyngeal swab performed in the Emergency Room and any subsequent swab test performed during hospitalization, chest x-ray and computed tomography (CT) scan in those patients who required it. The adjudication committee was composed of two experienced emergency physicians and was blinded to the LUS findings. At the end of the review of each case, the adjudication committee expressed a dichotomous opinion: "positive" to COVID-19 or "negative" to COVID-19. An experienced radiologist and another qualified emergency physician reviewed the chest x-rays (or CT scan) images (which were categorized in the final report as "suggestive for COVID-19 pneumonia" or "non-suggestive for COVID-19 pneumonia") before the adjudication committee viewed these images. RT-PCR tests used in this study were Roche Cobas® SARS-CoV-2 essay (F. Hoffmann-La Roche AG Konzern Hauptsitz Grenzacherstrasse 124. CH-4070 Basel Schweiz), and Liaison® MDX DiaSorin (DiaSorin S.p.A., via Crescentino SNC, 13040 Saluggia (VC), Italy).

Healthcare professionals were re-evaluated weekly for up to one month or until SARS-CoV2 infection was confirmed via nasopharyngeal molecular swab if symptoms appeared earlier than one month.

Lung ultrasound diagnosis

Lung ultrasound examinations were performed before the RT-PCR result for SARS-CoV-2. Therefore, the two sonographers were blinded for the report of this exam but not the possible presence of potentially suggestive symptoms. Lung ultrasound examinations were performed by dividing the chest into 12 areas: four anterior right areas, four anterior left areas, two right posterior areas, and two left posterior areas. The images were evaluated by describing the ultrasound findings classified as bilateral B-lines, consolidations, subpleural consolidations, thickening, and irregularities of the pleural line, pleural effusion for every thoracic

area ("LUS findings" on Supplemental Material). For early identification of suggestive cases (and therefore to have the maximum sensitivity of the LUS), the examination was considered positive if even one of these areas showed at least one of the previously mentioned suggestive signs of pulmonary involvement from COVID-19. The undetermined cases were re-discussed by the two sonographers together to categorize them into "positive" or "negative" for COVID-19 pneumonia, according to the International Proposed LUS protocol [7]. The two sonographers were two emergency physicians with more than 15 years of experience in LUS.

Sample Size

A convenience sample of 150 suitable health workers was considered. In calculating the confidence interval's magnitude, we hypothesized that at least 140 operators who would have agreed to participate in the study were negative for COVID-19-related pneumonia on LUS examination. Estimating a negative predictive value for LUS of approximately 98%, we calculated a 95% confidence interval ranging from 96% to 100%. The final sample was 153 health workers examined with LUS for a disease prevalence of 1.3% (95%CI 0.2% to 4.6%). The study has a sufficiently precise confidence interval to detect a reliable negative predictive value.

Statistical analysis

We reported the clinical and demographic characteristics of our population: sex, age, the presence of symptoms suggestive of COVID-19, the result of the LUS examination, the finding of the CT scan.

The categorical variables were expressed in absolute value (and respective percentage). Statistical significance between the two groups was calculated using Pearson's Chi-square test (or exact Fisher's test if appropriated).

Due to their nonparametric distribution, the continuous variables were shown with the median value and interquartile range (1st and 3rd interquartile). Statistical significance between the two groups was calculated using the Kruskal-Wallis test.

Statistical significance was considered for an α error value less or equal to 0.05 (i.e., a two-tail p-value $\leq$ 0.05). A Benjamini-Hochberg correction for multiplicity was applied.

Based on a contingency table (considering the frequencies of the True Positives, False Positives, True Negatives, and False Negatives), sensitivity, specificity, positive and negative likelihood ratios, positive and negative predictive values, and diagnostic accuracy of the LUS were calculated.

All statistical analyses were performed using the open-source R-CRAN project ver. 4.0.0. statistical software. The followed packages were implemented: 'irr', 'caret' and 'compareGroups'.

RESULTS

In the period considered, we enrolled 155 healthcare workers. The median age was 43 years. The characteristics of the enrolled population are described in Table 1.

The adjudication committee established two true positives for COVID-19. The remaining 153 cases were true negative (Figure 1). By dividing the population based on the diagnosis of COVID-19, the presence of symptoms and the nasopharyngeal swab results were significantly differently distributed between the two groups (p-value 0.018 and 0.013, respectively) (Table 1).

Healthcare workers who presented suggestive symptoms were 21 (2 true positives and 19 false positives). The cough was present in 6 cases; cold/rhinitis in 10 cases; fever in 3 cases; sore throat in 2 cases; myalgia/arthralgia in 2 cases; and, finally, one case had exertional dyspnea. The RT-PCR on nasopharyngeal swab was positive in one of the two true positive cases. Neither of the two cases rated "false positives" developed symptoms suspicious of COVID-19 during the month of follow up.

Lung ultrasound was suggestive for COVID-19 pneumonia in 4 cases: two cases were true positives, while two other cases were false positives. The sensitivity and the specificity of LUS in our population were 100% (95% CI 15.8% -100%) and 98.7% (95% CI 95.4% - 99.8%), respectively. Based on these values, the diagnostic accuracy was 98.7% (95% CI 95.4-99.8%). The positive and negative likelihood values were 76.5 (95% CI 19.3-303.1) and 0. Therefore the positive predictive value was 50% (95% CI 20.2-79.8%) and the negative predictive value was 100% (95% CI 100% -100%) (Figure 2).

DISCUSSION

Our study shows that LUS is a useful test for ruling out COVID-19 involvement in a low prevalence population. This result is twofold: first, LUS is useful for quickly verifying the effectiveness of the internal

protocols provided on the isolation pathways of COVID-19 infected patients and the efficiency of PPE. Furthermore, the population we studied is substantially comparable to the population of the countries affected by the COVID-19 pandemic at the time of the reduction of the infectious peak.

The community of health workers represented a population at high risk of exposure, but with a low probability, thanks to the PPE applied [9,10]: it was, in fact, a population with a low prevalence of coronavirus disease. Therefore, it represents the general population well when the peak of SARS-CoV-2 infection is low [11]. In this situation, it becomes essential to identify infected patients and exclude uninfected patients with an adequate margin of safety.

The literature shows that asymptomatic cases can constitute up to 60% of COVID-19 cases [12]. Given the high infectivity of patients, it is essential to exclude with reasonable certainty the cases that, even with symptoms suggestive of COVID-19 (given their non-specificity), have not been infected.

The role of asymptomatic subjects is increasingly emerging in the spread of infection [13,14]. However, there is still no adequate diagnostic screening method: RT-PCR tests have too variable sensitivity (37-71%) and long processing times [15-17], even chest X-ray has insufficient sensitivity (69 %) [18] and chest CT scan, although a high sensitivity (98%), [19] exposes a non-negligible radiological risk. [20] Subjects positive for Covid-19 but asymptomatic may have changes in the lung parenchyma, as demonstrated by the tomographic study of the passengers of the "Diamond Princess" cruise ship [21]. More precisely, 54% had a lung disease pattern: ground glass in 80% and consolidations in 20%. [22] In a retrospective study of 29 COVID-19 patients, Yang and colleagues compared LUS with CT scan [23]. They found that LUS was more sensitive than CT scan: in the diagnosis of regional of alveolar-interstitial pattern (60% vs. 38.5%, $P<0.0001$), alveolar-interstitial syndrome (93.3% vs. 68.9%, p-value =0.001), consolidations (38.9% vs. 3%, p-value <0.0001) and, finally, of pleural effusion (74.4% vs. 15.6%, p-value <0.0001).

The experiences reported so far in the literature indicate a low specificity of ultrasound findings [24,25]. Changes in the pleural line and the presence of multiple B lines are ultrasound findings common to other forms of interstitial lung disease, such as fibrosis, emphysema, or early pulmonary edema. Lung ultrasound is a candidate to be a ruling out tool rather than for the diagnostic confirmation of COVID-19 cases [26]. Our results, conducted on a large population of healthcare professionals, confirm this hypothesis.

The containment strategies adopted by various countries around the world – mostly consisting of the limitation and reduction of physical contacts between people and in the use of masks and gloves – have, in fact, probably allowed to "flatten" the spread curve of the epidemic [27]. However, the re-ignition of infectious foci - in the absence of active prevention tools such as, for example, a vaccine - is possible, especially in places where cases of residual infection can concentrate, such as hospitals [28]. In the uncertainty of the next scenarios, the need for an "intermittent lockdown" is anticipated due to the possibility of new epidemic outbreaks [29]; using a bedside examination, non-invasive and easily repeatable, allows expanding the potentially explorable user base. Since the hypothesis of mass screening seems to be one of the few plausible strategies, LUS has all the characteristics to be an adequate tool in terms of negative predictive value.

Limitations

Our study was conducted in two emergency departments with a relatively low prevalence of COVID-19 among hospitalized patients. We do not have enough data to determine how our results can change in another context. Additionally, the ultrasound diagnosis of COVID-19 has been established by physicians with a long experience in LUS. In other contexts, LUS could have performances that are not comparable to ours.

Furthermore, the main limitation is that we did not have a reference exam that can be considered a gold standard. As we have extensively exposed, RT-PCR from nasopharyngeal swabs does not enjoy optimal sensitivity. We have overcome this difficulty by creating an adjudication committee to evaluate all the clinical documentation and tests available.

Furthermore, due to the nature of the SARS-CoV-2 disease, the two sonographers could not be blinded to the presence of any suggestive symptoms.

Conclusion

Lung ultrasound has a good enough negative predictive value for ruling out pulmonary disease in the suspect of COVID-19 infection in a population of healthcare workers exposed to COVID-19 but with a low prevalence of the disease.

References:

1. Spina S, Marrazzo F, Migliari M, Stucchi R, Sforza A, Fumagalli R. The response of Milan's Emergency Medical System to the COVID-19 outbreak in Italy. Lancet. 2020; 395: e49-e50.
2. Giwa AL, Desai A, Duca A. Novel 2019 coronavirus SARS-CoV-2 (COVID-19): an overview for emergency clinicians. Pediatr Emerg Med Pract 2020; 17: 1-24.
3. Wee LE, Sim XYJ, Conceicao EP, Aung MK, Goh JQ, Yeo DWT, Gan WH, Chua YY, Wijaya L, Tan TT, Tan BH, Ling ML, Venkatachalam I. Containment of COVID-19 cases among healthcare workers: The role of surveillance, early detection, and outbreak management. Infect Control Hosp Epidemiol 2020; 41: 765-771.
4. Wee LE, Hsieh JYC, Phua GC, Tan Y, Conceicao EP, Wijaya L, Tan TT, Tan BH. Respiratory surveillance wards as a strategy to reduce nosocomial transmission of COVID-19 through early detection: The experience of a tertiary-care hospital in Singapore. Infect Control Hosp Epidemiol 2020; 41: 820-825.
5. https://www.businessinsider.com/healthcare-workers-getting-coronavirus-500-infected-2020-2?r=US&IR=T [Accessed 10/06/2020]
6. https://globalnews.ca/news/6729174/coronavirus-spain-death-toll-china/ [Accessed 10/06/2020]
7. Soldati G, Smargiassi A, Inchingolo R, Buonsenso D, Perrone T, Briganti DF, Perlini S, Torri E, Mariani A, Mossolani EE, Tursi F, Mento F, Demi L. Proposal for International Standardization of the Use of Lung Ultrasound for Patients With COVID-19: A Simple, Quantitative, Reproducible Method. J Ultrasound Med 2020; 39: 1413-1419.
8. Vetrugno L, Bove T, Orso D, et al. Our Italian experience using lung ultrasound for identification, grading and serial follow-up of severity of lung involvement for management of patients with COVID-19. Echocardiography 2020; 37: 625−627.
9. Wei XS, Wang XR, Zhang JC, Yang WB, Ma WL, Yang BH, Jiang NC, Gao ZC, Shi HZ, Zhou Q. A cluster of health care workers with COVID-19 pneumonia caused by SARS-CoV-2. J Microbiol Immunol Infect 2020: S1684-1182(20)30107-9. doi: 10.1016/j.jmii.2020.04.013. Epub ahead of print.
10. Jin YH, Huang Q, Wang YY, Zeng XT, Luo LS, Pan ZY, Yuan YF, Chen ZM, Cheng ZS, Huang X, Wang N, Li BH, Zi H, Zhao MJ, Ma LL, Deng T, Wang Y, Wang XH. Perceived infection transmission routes,

infection control practices, psychosocial changes, and management of COVID-19 infected healthcare workers in a tertiary acute care hospital in Wuhan: a cross-sectional survey. Mil Med Res 2020; 7: 24.

11. Xiao N, Abboud S, McCarthy DM, Parekh N. Incidentally discovered COVID-19 in low-suspicion patients-a threat to front line health care workers. Emerg Radiol 2020; 27: 589-595.
12. Qiu J. Covert coronavirus infections could be seeding new outbreaks [published online ahead of print, 2020 March 20]. Nature 2020;10.1038/d41586-020-00822-x.
13. Bai Y, Yao L, Wei T, Tian F, Jin DY, Chen L, Wang M. Presumed Asymptomatic Carrier Transmission of COVID-19. JAMA 2020; 323: 1406-1407.
14. Sakurai A, Sasaki T, Kato S, Hayashi M, Tsuzuki SI, Ishihara T, Iwata M, Morise Z, Doi Y. Natural History of Asymptomatic SARS-CoV-2 Infection. N Engl J Med 2020; 383: 885-886.
15. Li Y, Yao L, Li J, Chen L, Song Y, Cai Z, Yang C. Stability issues of RT-PCR testing of SARS-CoV-2 for hospitalized patients clinically diagnosed with COVID-19. J Med Virol 2020; 92: 903-908.
16. Wang W, Xu Y, Gao R, Lu R, Han K, Wu G, Tan W. Detection of SARS-CoV-2 in Different Types of Clinical Specimens. JAMA 2020; 323: 1843-1844.
17. Ai T, Yang Z, Hou H, Zhan C, Chen C, Ly W, Tao Q, Sun Z, Xia L. Correlation of Chest CT and RT-PCR Testing in Coronavirus Disease 2019 (COVID-19) in China: A Report of 1014 Cases. Radiology 2020; 296: E32-E40.
18. Wong HYF, Lam HYS, Fong AH, Leung ST, Chin TW, Lo CSY, Lui MM, Lee JCY, Chiu KW, Chung TW, Lee EYP, Wan EYF, Hung IFN, Lam TPW, Kuo MD, Ng MY. Frequency and Distribution of Chest Radiographic Findings in Patients Positive for COVID-19. Radiology 2020; 296: E72-E78.
19. Fang Y, Zhang H, Xie J, Lin M, Ying L, Pang P, Ji W. Sensitivity of Chest CT for COVID-19: Comparison to RT-PCR. Radiology 2020; 296: E115-E117.
20. Rubin GD, Ryerson CJ, Haramati LB, Sverzellati N, Kanne JP, Raoof S, Schluger NW, Volpi A, Yim JJ, Martin IBK, Anderson DJ, Kong C, Altes T, Bush A, Desai SR, Goldin J, Goo JM, Humbert M, Inoue Y, Kauczor HU, Luo F, Mazzone PJ, Prokop M, Remy-Jardin M, Richeldi L, Schaefer-Prokop CM, Tomiyama N, Wells AU, Leung AN. The Role of Chest Imaging in Patient Management During the COVID-19 Pandemic: A Multinational Consensus Statement From the Fleischner Society. Chest 2020; 158:106-116.

21. Yamahata Y, Shibata A. Preparation for Quarantine on the Cruise Ship Diamond Princess in Japan due to COVID-19. JMIR Public Health Surveill 2020; 6:e18821.
22. Inui S, Fujikawa A, Jitsu M, Kunishima N, Watanabe S, Suzuki Y, Umeda S, Uwabe Y. Chest CT Findings in Cases from the Cruise Ship "Diamond Princess" with Coronavirus Disease 2019 (COVID-19). Radiol Cardiothorac Imaging. 2020; 2: e200110.
23. Yang Y, Huang Y, Gao F, Yuan L, Wang Z. Lung ultrasonography versus chest CT in COVID-19 pneumonia: a two-centered retrospective comparison study from China. Intensive Care Med 2020; 46: 1761-1763.
24. Hankins A, Bang H, Walsh P. Point of care lung ultrasound is useful when screening for CoVid-19 in Emergency Department patients. medRxiv [Preprint] 2020 Jun 12:2020.06.09.20123836.
25. Vetrugno L, Bove T, Orso D, Bassi F, Boero E, Ferrari G. Lung Ultrasound and the COVID-19 "Pattern": Not All That Glitters Today Is Gold Tomorrow. J Ultrasound Med 2020; 39: 2281-2282.
26. Copetti R, Amore G, Di Gioia CC, Orso D. First comes the A, then the B: what we learned from the COVID-19 outbreak. Eur J Intern Med 2020; 80:108-110.
27. Lonergan M, Chalmers JD. Estimates of the ongoing need for social distancing and control measures post-"lockdown" from trajectories of COVID-19 cases and mortality. Eur Respir J 2020; 56: 2001483.
28. Glauser W. Proposed protocol to keep COVID-19 out of hospitals. CMAJ 2020; 192: E264-E265.
29. Westerhoff HV, Kolodkin AN. Advice from a systems-biology model of the corona epidemics. NPJ Syst Biol Appl 2020; 6: 18.

Tables legend

Table 1. Comparison between the main characteristics of the positive COVID-19 group and the negative COVID-19 group.

Figures Legend

Figure 1. Patient enrollment flow chart. LUS was suggestive for COVID-19 pneumonia in 4 cases (2 false-positive cases).

Figure 2. Likelihood Ratio Nomogram for LUS in diagnosing COVID-19 pneumonia. The positive likelihood ratio was 76.50 (19.31 - 303.12); the negative likelihood ratio was 0.

	Population (n =155)	COVID-19 negative (n=153)	COVID-19 positive (n=2)	Overall p-value
Age (1st and 3rd quartiles)	43 (33-50)	42 (33-50)	39 (32-46)	0.811
Sex (Male)	60 (38.7%)	60	0	0.522
Symptoms (any)	21 (13.5%)	19	2	0.018*
Nasoparhyngeal Swab (+)	1 (0.66%)	0	1	0.013*
LUS (+)	4 (2.58%)	2	2	0.001*

Table 1. Comparison between the main characteristics of the positive COVID-19 group and the negative COVID-19 group.