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EDITORIAL

Vaccinations: What's best?

Seasonal influenza and pneumococcal infection, including community-acquired pneumonia, are preventable diseases that are still causing a significant amount of morbidity and mortality worldwide.[1]

Influenza epidemics causes each year from 250.000 to 500.000 and *Streptococcus pneumoniae* nearly 1.5 million deaths worldwide, particularly in frail older patients.[2–4]

Despite the huge number of recommendations issued by international and national Agency and Scientific Societies, promoting vaccinations in frail and vulnerable patients, vaccine coverage is still unacceptably low.[5–9]

Even if the number of countries in which influenza vaccination is recommended in high-risk subjects increased by more than 40% between 2014 and 2018, the vaccine uptake did not show any significant increase in the same time-lag.[10–11] Notably, the European Centre for Disease Prevention and Control highlighted that the median vaccination coverage was lower than 50% for elderly and frail individuals, in spite of the goal of vaccinating at least 75% of at risk subjects.[12] The recent Sars-CoV2 pandemic reopened the discussion on the strategic arrangements for vaccination in the face of spreading infections, highlighting the vulnerability of older adults with comorbidities to infectious diseases and the need for robust healthcare systems to face the emergency.

Froes F et al., in the present issue of Pulmonology, report interesting results of the Vacinómetro® initiative, an eleven-year study that monitored the influenza vaccine uptake in four different at-risk populations in Portugal.[13] The overall figure revealed an increase of vaccination rate in all the four target populations (1.Patients ≥ 65 years old; 2. Patients with chronic conditions; 3. Health care workers (HCWs); 4. Patients 60-64 years old) that reached the coverage target of 75% in the elderly population (group 1).

Free-of-charge vaccination is an important driver for increasing the vaccine uptake and should probably be part of the proactive efforts of any vaccination campaign to provide influenza vaccination to the general population.

The study[13] confirms the key role of physicians in promoting vaccinations and in overcoming the most common barriers to vaccination such as fear of adverse effects, uncertainty about the vaccine's efficacy, and misconceptions about the vaccine and the nature of the infection. On the other end it is interesting that HCWs vaccine uptake was as low as 36% in 2011 and reached 59% in 2020.

These results are of paramount importance because it is well known that vaccinating HCWs against influenza is not only a simple and cost-effective measure to reduce infection among staff but also an excellent tool to prevent morbidity and mortality.[14]

In a survey on attitudes and uptake of H1N1 influenza vaccination among HCWs members of the European Respiratory Society and the European Society of Clinical Microbiology and Infectious Diseases the main reasons for vaccination were to 'avoid virus spread to patients' and to 'protect myself', and safety being the main concern against vaccination.[15] Education of HCWs on vaccination seems to be an important target for improving vaccine uptake both among HCWs and general population. Fig. 1 depicts the major issues concerning vaccinations and the possible proactive interventions.

In conclusion, the article by Froes et al.[13] paves the way to a better design of any future vaccination programs, helping health authorities to target the right at risk groups and implementing educational and economical initiatives to improve vaccines uptake.

Declaration of Competing Interest

The authors declare no conflict of interest concerning this editorial.

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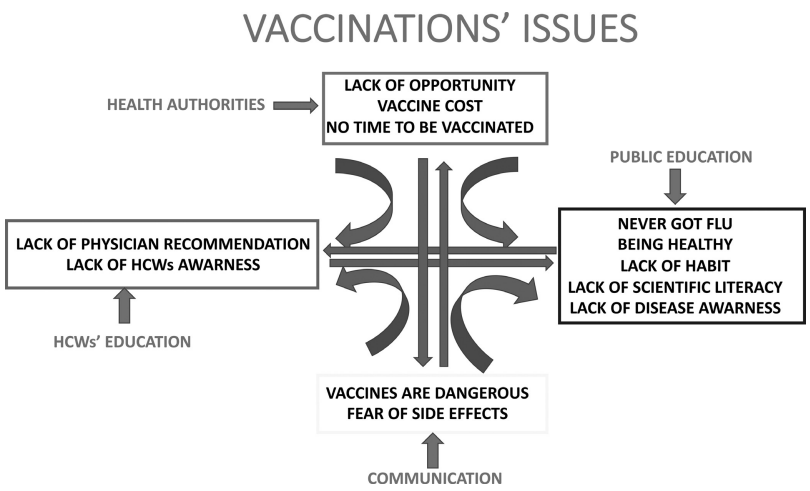

Fig. 1 Major issues on vaccinations and possible interventions.

References

1. Aliberti S, Dela Cruz CS, Amati F, Sotgiu G, Restrepo MI. Community-acquired pneumonia. Lancet. 2021;398(10303):906–19. https://doi.org/10.1016/S0140-6736(21)00630-9.
2. Martínez A, Soldevila N, Romero-Tamarit A, Torner N, Godoy P, Rius C, et al. Surveillance of hospitalized cases of severe influenza in catalonia working group. Risk factors associated with severe outcomes in adult hospitalized patients according to influenza type and subtype. PLoS One. 2019;14(1):e0210353. https://doi.org/10.1371/journal.pone.0210353. eCollection 2019.
3. Torres A, Cillóniz C, Blasi F, Chalmers JD, Gaillat J, Dartois N, et al. Burden of pneumococcal community-acquired pneumonia in adults across Europe: a literature review. Respir Med. 2011;137:6–13. https://doi.org/10.1016/j.rmed.2018.02.007.
4. Falcone M, Blasi F, Menichetti F, Pea F, Violi F. Pneumonia in frail older patients: an up to date. Intern Emerg Med. 2012;7 (5):415–24. https://doi.org/10.1007/s11739-012-0796-7.
5. MacDougall DM, Halperin BA, MacKinnon-Cameron D, Li L, McNeil SA, Langley JM, Halperin SA. The challenge of vaccinating adults: attitudes and beliefs of the Canadian public and healthcare providers. BMJ Open. 2015;5(9):e009062. https://doi.org/10.1136/bmjopen-2015-009062.
6. European Centre for Disease Prevention and Control. Seasonal Influenza Vaccination and Antiviral use in EU/EEA Member States – Overview of Vaccine Recommendations for 2017–2018 and Vaccination Coverage Rates for 2015–2016 and 2016–2017 Influenza Seasons. Stockholm: ECDC; 2018.
7. Doherty M, Schmidt-Ott R, Santos JI, Stanberry LR, Hofstetter AM, Rosenthal SL, Cunningham AL. Vaccination of special populations: protecting the vulnerable. Vaccine. 2016;34(52):6681–90. https://doi.org/10.1016/j.vaccine.2016.11.015.
8. Blasi F, Di Pasquale M, Gramegna A, Viale P, Iacobello C, Gori A, Tumbarello M, et al. A new call for influenza and pneumococcal vaccinations during COVID-19 pandemic in Italy: a SIP/IRS (Italian Respiratory Society) and SITA (Italian Society of Antiinfective therapy) statement. Resp Med. 2021;190:106674. https://doi.org/10.1016/j.rmed.2021.106674.
9. Antonelli-Incalzi R, Blasi F, Conversano M, Gabutti G, Giuffrida S, Maggi S, et al. Manifesto on the value of adult immunization: "we know, we intend, we advocate". Vaccines (Basel). 2021;9 (11):1232. https://doi.org/10.3390/vaccines9111232.
10. Morales KF, Brown DW, Dumolard L, Steulet C, Vilajeliu A, Ropero Alvarez AM, et al. Seasonal influenza vaccination policies in the 194 WHO member states: the evolution of global influenza pandemic preparedness and the challenge of sustaining equitable vaccine access. Vaccine X. 2021;8:100097. https://doi.org/10.1016/j.jvacx.2021.100097.
11. Oakley S, Bouchet J, Costello P, Parker J. Influenza vaccine uptake among at-risk adults (aged 16-64 years) in the UK: a retrospective database analysis. BMC Public Health. 2021;21 (1):1734. https://doi.org/10.1186/s12889-021-11736-2.
12. https://www.ecdc.europa.eu/sites/default/files/documents/seasonal-influenza-antiviral-use-2018.pdf. Accessed July, 22, 2022
13. Froes F, Morais A, Hespanhol V, Nogueira R, Sequeira Carlos J, Jacinto N, et al. The Vacinómetro® initiative: an eleven-year monitorization of influenza vaccination coverage rates among risk groups in Portugal, *Pulmonology*, S2531-0437(22)00078-2, 2022. https://doi.org/10.1016/j.pulmoe.2022.03.005.
14. Carman WF, Elder AG, Wallace LA, McAulay K, Walker A, Murray GD, Stott DJ. Effects of influenza vaccination of health-care workers on mortality of elderly people in long-term care: a randomised controlled trial. Lancet. 2000;355(9198):93–7. https://doi.org/10.1016/S0140-6736(99)05190-9.
15. Blasi F, Palange P, Rohde G, Severin T, Cornaglia G, Finch R. Healthcare workers and influenza vaccination: an ERS-ESCMID Web-based survey. Clin Microbiol Infect. 2011;17(8):1223–5. https://doi.org/10.1111/j.1469-0691.2011.03501.x.

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EDITORIAL

COVID-19 pandemic and tuberculosis: How to ensure adequate care in pediatric age

As expected that the SARS-CoV-2 pandemic played an important role in the prevention and control of tuberculosis status. The investment needed to contain the pandemic has negatively influenced resources to control other infections.[1] The 2020 lockdown has hampered access to health care, potentially delaying the diagnosis and treatment of tuberculosis. Glaziou P. predicted that tuberculosis incidence would slowly decline with a potential rebound by increased duration of infectiousness and decreased case detection during lockdown.[2] The chains of transmission of pediatric tuberculosis would be affected: transmission in home clusters (the main focus of exposure in children) would increase, while transmission in social settings should decrease.[1,3]

There is a network of Tuberculosis Outpatient Centers within the Portuguese National Health Service. Since 2010 there is a Pediatric TB Reference Center in the North of Portugal, the first in the country. The Center has the aim of screening and diagnosing pediatric TB, with a multidisciplinary team with pediatric specialization. The team includes 2 pediatricians with experience in childhood TB, a nurse, a pulmonologist, an infectious disease specialist, a radiology technician and administrative technicians. Teams with experience in pediatric age and pediatric TB should be involved in the care of these patients.[1]

To face the constraints imposed by the pandemic situation, the following measures were implemented:

1. When possible (e.g., pediatric patients being treated for latent infection, cases of surveillance after completing treatment), consultations were conducted by telephone, with parental consent. When there were concerns that warranted additional observation, a face-to-face consultation was scheduled as soon as possible. Although not unknown, telephone consultations were rare before the pandemic.
2. In cases of patients who required blood analysis control, e.g., in cases of side effects, requests were sent via text message to patients' cell phones. The analysis was scheduled in a laboratory with an agreement in the national health system; the results were sent via email to the pediatrician. This facility still exists. In the case of sputum collection, this was carried out in a room at the Center or at the patient's home. Hospitalizations for diagnostic investigation (such as gastric aspirate collection and bronchoalveolar lavage) were kept in the Pediatric Department of the local hospital.
3. The first consultations were made in person to allow for physical examination, chest radiography, and immunological tests (scheduled for the same day). Consultations were also in person when there was a change in the clinical status or during the transition periods from induction to maintenance medication.
4. Referral patients from the emergency department and hospital appointments with suspected or confirmed TB was facilitated as previously, as the pediatricians work at both locations.
5. Chest computed tomography (CT) scans were performed at the local hospital, after a PCR test for SARS-CoV-2. CT interpretation was performed by the radiology team. Before the pandemic, it was easier to perform this imaging test in clinics with an agreement in the national health system. Chest X-ray was performed at the Center and its interpretation was made by the pediatricians' team, as before.
6. Before the pandemic, directly observed treatment (DOT) was in person. In cases of active tuberculosis, DOT was maintained. In bacilliferous cases, the nurse would still visit the patient's home. In the presence of possible poor compliance factors, DOT was maintained by the patient's family nurse. In the remaining cases, DOT was done in person, once a week, and medication was provided for that period, with telephone contact by the nurse to ensure compliance. In children on preventive treatment, medication was provided for a longer period (1 to 3 months).
7. Coordination with public health services was maintained with monthly online meetings to discuss cases and ensure the screening of families or groups exposed. As in the

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past, migrant children are considered at risk and candidates for BCG administration if they come from countries with a high incidence of TB.

A retrospective descriptive analysis was performed, based on medical records, comparing the type of referrals, number of consultations, and the diagnosis of pediatric patients followed up from January 2019 (pre-pandemic year) to December 2020 (pandemic year). There was a total of 721 medical consultations in 2019 compared to 580 in 2020. These numbers represent a 19.6% decrease, with a greater reduction in subsequent in person consultations (-53%). First medical consultations were also reduced (-12.2%). Telephone consultations increased significantly (+87.4%). Patients observed for suspected active disease reduced by 21.1%, and for tuberculosis screening by 10.6%. In 2020, 12 patients started antituberculosis drugs for active tuberculosis, compared to 7 patients in 2019. The diagnosis of latent tuberculosis infection was made in 12 cases in 2020 and in 13 cases in 2019. Fewer missed consultations (-56.8%) were noticed in 2020. There were no treatment abandonments in the two periods. No significant side effects were reported during these years.

Following the covid pandemic restrictions at the beginning of 2020, the team was able to maintain quality services. We verified that the number of diagnosed latent infections and active cases of tuberculosis remained unchanged. The data shows no decrease in referrals to the center. The team believes that it successfully overcame the difficulties in maintaining clinical activity, mostly due to effective digital monitoring and easy accessibility to the Center.

Rodrigues et al recently reported the activity of Outpatient Tuberculosis Centers in Portugal with the adult population during the pandemic. It was observed that there were fewer outpatient visits to the Centers, probably due to a decrease in referrals by other health units. As in our pediatric Center, the DOT strategy was modified. It should be noted that 25% of the Centers reported using DOT only in high-risk patients and 18.7% did not use it at all. Unlike our Center, there was an increase in missed consultations from the beginning of the pandemic. The authors expressed concern about the delays in diagnosing active disease.[4]

The emergence of COVID-19 should not relax the measures and efforts to maintain a reduction in TB incidence. In Portugal, access to Outpatient TB Centers remains free and easy. Follow-up by trained pediatricians in these types of community Outpatient TB Centers allows for better management of pediatric TB disease. Even in times of difficulty, such as during the pandemic, the level response in tuberculosis cases can be maintained if assistance is adapted.

References

1. Starke JR, Erkens C, Ritz N, Kitai I. Strengthening tuberculosis services for children and adolescents in low endemic settings. Pathogens. 2022;11:158. https://doi.org/10.3390/pathogens 11020158.
2. Glaziou P. Predicted impact of the COVID-19 pandemic on global tuberculosis deaths in 2020. MedRxiv. 2020. https://doi.org/ 10.1101/2020.04.28.20079582.
3. Gomes BLM, Carvalho I. Tuberculosis - Reaping benefits from COVID-19 in Portugal. Pulmonology. 2021;27(1):4—6. https:// doi.org/10.1016/j.pulmoe.2020.08.006.
4. Rodrigues I, Aguiar A, Migliori GB, Duarte R. Impact of the COVID-19 pandemic on tuberculosis services. Pulmonology. 2022;28 (3):210—9. https://doi.org/10.1016/j.pulmoe.2022.01.015.

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SPECIAL ARTICLE

High-flow nasal oxygen in individuals with COVID-19 pneumonia and mild hypoxaemia: An independent discussion

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The COVID-19 pandemic has been associated with a storm of information by social media and with an increase in publications with high percentages of retractions.[1,2] There is a clear need for reproducible randomised controlled trials (RCTs) on the topic. A discussion by independent authors with expertise in research methodology has been proposed for probing inferential reproducibility and for addressing issues in discussion sections of evaluated papers.[3]

The pandemic has also triggered unprecedented use of tools supposedly intended to prevent invasive mechanical ventilation among individuals with COVID-19 associated acute respiratory failure.[4,5] Quality data, in particular reproducible RCTs regarding modes of non-invasive respiratory support (RS), are greatly needed.[6] High-flow nasal oxygen (HFNO) is one such tool.[7,8]

In a recently published RCT, the COVID-HIGH trial,[9] individuals with COVID-19 and mild hypoxaemia were randomised to treatment with either HFNO or conventional oxygen therapy (COT).[9] The trial is particularly relevant as HFNO is increasingly being used in enviroments with a lower level of monitoring where such patients are often treated.[10,11] Using a structured independent discussion,[3] two authors with expertise in research methodology consider the findings and the inferential reproducibility of this RCT. Below, original and independent discussions for each section of the paper [9] are compared.[3]

Main findings

Original Discussion: The authors report that individuals with COVID-19 pneumonia and mild hypoxaemia randomised to HFNO versus COT had similar rates of RS escalation within 28 days.[9]

Independent discussion

The study and control groups did not differ in the rate of the primary outcome. The sample size for the study [9] was calculated based on a retrospective study, the best evidence at the time.[12] However, effect size is often inflated in retrospective studies,[13,14] and indeed the observed rate of RS escalation was lower than expected. COVID-HIGH has an 80% power to identify only a 35% relative difference in event rate (i.e. a 15% absolute risk reduction).[9] In order to identify a 27% relative difference, which takes into consideration the actual event rate (i.e. an 11% absolute risk reduction), 580 individuals would need to be recruited. Therefore the study, with 364 participants, only has a 60% power to refute the baseline hypothesis of no difference between the two modes of treatment. The likelihood of a type II error (i.e. a false negative finding) is high.

Commentary: The original and independent discussions are concordant in their interpretation of the main study findings in COVID-HIGH. The independent discussion also highlights that study underpowering may limit the validity of the main study finding.

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Relationship of main findings to previous studies

Original discussion: The authors compare their findings to those of the RECOVERY-RS multicentre trial which showed no difference between HFNO and COT for the primary outcome of intubation or 30-day mortality.[15] They highlight that the study[15] recruited individuals with greater disease severity, with a SpO_2 $\leq$94% despite receiving an inspiratory oxygen fraction (FiO_2) of at least 40% and point out that RECOVERY-RS[15] had continuous positive airway pressure (CPAP) as a third study arm. The authors also cite a RCT by Ospina-Tascon et al.[16] in which HFNO significantly reduced the intubation risk and time to clinical recovery in individuals with FiO_2 <200. They propose that, taken together, the findings of these trials suggest different clinical effects of HFNO versus COT in individuals with different disease severity.[9,15,16]

Independent discussion: Like COVID-HIGH,[9] the RCT by Ospina-Tascon et al., was underpowered for the primary outcome (220 participants).[16] RECOVERY-RS was an adaptive (group-sequential) cohort nested within a pragmatic trial. Hence it should have been adequately powered to show different treatment effects, but was stopped early for futility.[15]

The effect of any intervention depends on several variables, including baseline load of comorbidity, disease severity, treatment timing (late vs. early) and dose. The COVID-HIGH trial[9] included participants with moderate Charlson comorbidity scores. Markers of inflammation were not used as inclusion criteria,[17] but these data are presented and indicate moderate disease.[9] Both Ospina-Tascon et al.[16] and RECOVERY-RS[15] included individuals with worse disease than COVID-HIGH.[9] Ospina-Tascon et al.[16] described comorbidity rates similar to those described in COVID-HIGH.[9] Although the ROX indices[18,19] of individuals under HFNO were higher than those under COT, the levels of inflammation markers suggest more severe disease than in COVID-HIGH.[9] RECOVERY-RS decribes more heart and lung diseases but provides neither the overall weight of comorbidity nor data on inflammation markers.[15]

In COVID-HIGH the time from symptom onset to randomization averaged seven[9] versus eleven days in the Ospina-Tascon trial[16] and nine days in RECOVERY−RS.[15] Finally, in COVID-HIGH[9] the treatment protocol (i.e. "dose") was preset as were the criteria for treatment escalation. The average duration of treatment was three days. Ospina-Tascon[16] also protocolized treatment, including the criteria for intubation. Treatment duration was planned as six days but ultimately averaged only one day. In RECOVERY-RS[15] treatment with HFNO was not protocolized and the duration of treatment was not described.

Commentary: Both discussions refer to the same two studies,[15,16] however, the independent discussion identified more differences between the studies.

Secondary findings

Original discussion: The authors report that the secondary clinical outcomes (i.e. likelihood of clinical recovery, time to first RS escalation, rate of intensive care unit [ICU] admission and 28- and 60-day mortality) did not differ between the treatment arms.[9]

Independent discussion: No difference was found in secondary outcomes between study and controls.

Commentary: The original and independent discussions are concordant in their interpretation of the secondary study findings in COVID-HIGH study.[9]

Relationship of additional (secondary) findings to previous studies

Original discussion: The relationship of secondary findings to previous studies was not discussed by the authors.

Independent discussion: The COVID-HIGH trial was not powered for any of the secondary outcomes examined although these were preplanned and were registered in the study protocol.[9]

Ospina-Tascon[16] noted earlier (but not more) recovery among participants treated with HFNO. ICU admission rates were similarly unaffected by treatment with HFNO in COVID-HIGH[9] and RECOVERY-RS.[15] Thirty day mortality was part of the primary composite outcome of RECOVERY-RS[15] and was not related to treatment. It was a secondary outcome in the Ospina-Tascon trial[16] where it was also unrelated to treatment. Neither trial[15,16] reported the time to first RS escalation or 60 day mortality.

Commentary: HFNO does not seem to have a consistent effect on any of the objective secondary outcomes studied. Indications for ICU admission and length of stay are also dependent on local bed availability and practices. Long term outcomes were not studied in any of the trials but short-term mortality rates (28- or 30- day mortality in all three trials) seem consistently unaffected.[9,15,16]

Limitations

Original discussion: The authors admit their study has several limitations. Due to the nature of interventions, blinding was not possible. However, clinical criteria used to decide on RS escalation were standardised. Subjectivity in clinical judgement could not be excluded. In selected cases, clinicians may have considered HFNO as a form of RS and been less likely to escalate to CPAP/NIV compared with COT. This may partly explain the higher protocol violation rate in the control group. The trial was underpowered. However a clinically meaningful benefit from HFNO in this population could not be definitely ruled out. The COVID-HIGH cohort included 64% male participants, which may limit the generalisability of the findings.[9] However, the adjusted odds-ratio for sex showed no significant effect on the association between occurrence of the primary outcome and study interventions. Due to the multinational and multicentre nature of the study, different pandemic surges may have had different indirect consequences on the care level at study sites. Data on SARS-CoV-2 variants or vaccination status of participants were not registered. Finally, the results of the subgroup analyses should be considered exploratory as positive findings may be attributed to repeated testing.[9]

Independent discussion: Lack of power is the most important study limitation in COVID-HIGH.[9] Despite the difference between the expected and observed RS escalation rates, the investigators chose to close the study with the preplanned number of participants as prolonging the study would have increased population variability (e.g. COVID variants) and

co-treatment effects (e.g. vaccine effects, local practice). The COVID-HIGH investigators provide no data on the use of ancillary respiratory support therapies such as self-proning,[20] physiotherapy and mobilisation.[21] While individuals with limitation of care before randomization were not included, this status may change during treatment. Like previous RCTs, information regarding with-holding/withdrawal of care is missing. Finally, the rates of specific COVID phenotypes may have differred in the two study groups and specific phenotypes may respond differently to different management strategies.[22]

Commentary:

Both discussions agree on the lack of power, the independent discussion highlighted several issues that were not mentioned in the original discussion.

Future directions

Original discussion: The original discussion did not consider future research directions.

Independent discussion: Future physiological work should include comparative data on work of breathing with HFNO versus COT. Clinical data should include adequately powered RCTs with more detailed information on the effects of HFNO, if such exist, in different COVID-19 disease phenotypes and data on the long term effect of HFNO. More data is also required on the human- and health-resource costs of using HFNO and on the risks of caregiver contamination.[23]

Commentary: Only the independent discussion suggests directions for future research and how these may be informed by the findings of COVID-HIGH.

Conclusion

Original discussion: The authors concluded that HFNO did not significantly decrease the escalation of RS compared with COT among hospitalised individuals with COVID-19 pneumonia with mild hypoxaemia.[9]

Independent discussion: The current evidence shows no proof either for or against indiscriminate use of HFNO in hospitalised individuals with COVID-19 pneumonia with mild hypoxaemia.

Commentary: The independent conclusion highlights the need for additional research.

Inferential reproducibility

There was acceptable inferential reproducibility between the two discussions. The independent discussion provides a more detailed description and analysis of the primary and secondary findings in relation to current literature than the original discussion and offers more in-depth explanations for the lack of effect and methodological issues in COVID-HIGH. However, both discussions agree on the key findings and their interpretation.

Declarations of Competing Interest

SE is a Cochrane editor, a member of the Data Use Committee of the American Society of Anesthesiologists, has lectured and chaired panels on noninvasive ventilation (unsupported), has patents with Medtronic and has received funding from Zoll, Astra-Zeneca, Artisanpharma, Eisai and from the Israel Ministry of Health, National Institute for Health Policy Research, and Hebrew University Research and Development authority. SE and NA were members of the COVID-HIGH Trial Oversight Committee without any compensation.

References

1. Carvalho TA, Lima TM, Melani VF, Mendes MF, Pereira LR, Marson FAL. The scientific production during 2009 swine flu pandemic and 2019/2020 COVID-19 pandemic. Pulmonology. 2020;26(6):340—5.
2. Boschiero MN, Carvalho TA, Marson FAL. Retraction in the era of COVID-19 and its influence on evidence-based medicine: is science injeopardy? Pulmonology. 2021;27(2):97—106.
3. Avidan MS, Ioannidis JPA, Mashour GA. Independent discussion sections for improving inferential reproducibility in published research. Br J Anaesth. 2019;122(4):413—20. https://doi.org/10.1016/j.bja.2018.12.010.
4. Winck JC, Scala R. Non-invasive respiratory support paths in hospitalized patients with COVID-19: proposal of an algorithm. Pulmonology. 2021;27(4):305—12. https://doi.org/10.1016/j.pulmoe.2020.12.005.
5. Winck JC, Ambrosino N. COVID-19 pandemic and non invasive respiratory management: Every Goliath needs a David. An evidence based evaluation of problems. Pulmonology. 2020;26(4):213—20. https://doi.org/10.1016/j.pulmoe.2020.04.013.
6. Weerakkody S, Arina P, Glenister J, et al. Non-invasive respiratory support in the management of acute COVID-19 pneumonia: considerations for clinical practice and priorities for research. Lancet Respir Med. 2022;10(2):199—213. https://doi.org/10.1016/S2213-2600(21)00414-8.
7. Vega ML, Pisani L. Nasal high flow oxygen in acute respiratory failure. Pulmonology. 2021;27(3):240—7. https://doi.org/10.1016/j.pulmoe.2021.01.005.
8. Crimi C, Pierucci P, Renda T, Pisani L, Carlucci A. High-flow nasal cannula and COVID-19: a clinical review. Respir Care. 2022;67(2):227—40. https://doi.org/10.4187/respcare.09056.
9. Crimi C, Noto A, Madotto F, et al. High-flow nasal oxygen versus conventional oxygen therapy in patients with COVID-19 pneumonia and mild hypoxaemia: a randomised controlled trial. Thorax. 2022. https://doi.org/10.1136/thoraxjnl-2022-218806. May 17: thoraxjnl-2022-218806Epub ahead of print.
10. Calligaro GL, Lalla U, Audley G, et al. The utility of high-flow nasal oxygen for severe COVID-19 pneumonia in a resource-constrained setting: a multi-centre prospective observational study. EClinicalMedicine. 2020;28:100570. https://doi.org/10.1016/j.eclinm.2020.100570.
11. Sykes DL, Crooks MG, Thu Thu K, et al. Outcomes and characteristics of COVID-19 patients treated with continuous positive airway pressure/high-flow nasal oxygen outside the intensive care setting. ERJ Open Res. 2021;7(4):00318—2021. https://doi.org/10.1183/23120541.00318-2021.
12. Patel M, Gangemi A, Marron R, et al. Retrospective analysis of high flow nasal therapy in COVID-19-related moderate-to-severe hypoxaemic respiratory failure. BMJ Open Respir Res. 2020;7(1):e000650. https://doi.org/10.1136/bmjresp-2020-000650.
13. Dziadkowiec O. Use of statistical power analysis in prospective and retrospective research. J Obstet Gynecol Neonatal Nurs. 2021;50(2):119—21. https://doi.org/10.1016/j.jogn.2021.01.004.
14. Bakdash JZ, Marusich LR, Kenworthy JB, Twedt E, Zaroukian EG. Statistical significance filtering overestimates effects and

impedes falsification: a Critique of. Front Psychol. 2020;11:609647. https://doi.org/10.3389/fpsyg.2020.609647.
15. Perkins GD, Ji C, Connolly BA, Couper K, et al. Effect of noninvasive respiratory strategies on intubation or mortality among patients with acute hypoxemic respiratory failure and COVID-19: the RECOVERY-RS Randomized Clinical Trial. JAMA. 2022;327 (6):546–58. https://doi.org/10.1001/jama.2022.0028.
16. Ospina-Tascón GA, Calderón-Tapia LE, García AF, et al. Effect of high-flow oxygen therapy vs conventional oxygen therapy on invasive mechanical ventilation and clinical recovery in patients with severe COVID-19: a randomized clinical trial. JAMA. 2021;326(21):2161–71. https://doi.org/10.1001/jama.2021. 20714.
17. Bauchner H, Fontanarosa PB. Randomized clinical trials and COVID-19: managing expectations. JAMA. 2020;323 (22):2262–3. https://doi.org/10.1001/jama.2020.8115.
18. Vega ML, Dongilli R, Olaizola G, et al. COVID-19 Pneumonia and ROX index: time to set a new threshold for patients admitted outside the ICU. Pulmonology. 2022;28(1):13–7. https://doi. org/10.1016/j.pulmoe.2021.04.003.
19. Roca O, Messika J, Caralt B, et al. Predicting success of high-flow nasal cannula in pneumonia patients with hypoxemic respiratory failure: the utility of the ROX index. J Crit Care. 2016;35:200–5. https://doi.org/10.1016/j.jcrc.2016.05.022.
20. Li J, Luo J, Pavlov I, et al. Awake prone positioning for non-intubated patients with COVID-19-related acute hypoxaemic respiratory failure: a systematic review and meta-analysis. Lancet Respir Med. 2022;10(6):573–83. https://doi.org/10.1016/ S2213-2600(22)00043-1.
21. Polastri M, Ciasca A, Nava S, Andreoli E. Two years of COVID-19: trends in rehabilitation. Pulmonology. 2022;S2531-0437(22). https://doi.org/10.1016/j.pulmoe.2022.01.012. 00024-1Epub ahead of print.
22. Marini JJ, Gattinoni L. Management of COVID-19 respiratory distress. JAMA. 2020;323(22):2329–30. https://doi.org/10.1001/ jama.2020.6825.
23. Ippolito M, Vitale F, Accurso G, et al. Medical masks and respirators for the protection of healthcare workers from SARS-CoV-2 and other viruses. Pulmonology. 2020;26(4):204–12. https:// doi.org/10.1016/j.pulmoe.2020.04.009.

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ORIGINAL ARTICLE

The Vacinómetro® initiative: an eleven-year monitorization of influenza vaccination coverage rates among risk groups in Portugal

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KEYWORDS
Influenza;
Vaccination;
Seasonal;
Vaccination coverage rate;
Risk groups;
Portugal

Abstract Annual vaccination is fundamental for individual and group protection against seasonal influenza infection. International and Portuguese healthcare organizations have established influenza vaccination coverage rate (VCR) targets for risk groups, namely 75% in people ≥ 65 years old. The Vacinómetro® initiative has been monitoring influenza VCR among target risk groups in Portugal since 2009,: Group 1, ≥ 65 years old; Group 2, patients with chronic conditions; Group 3, healthcare workers in direct contact with patients; and Group 4, 60-64 years old. Besides VCR, social-demographic and health-related variables have been evaluated. During the study period (2009/2010 — 2019/2020), the VCR increased in the 4 target risk groups: from 58.6% to 76.0% in Group 1 (reaching the WHO target); 33.3% to 72.0% in Group 2; 25.0% to 58.9% in Group 3; and 36.6% to 42.8% in Group 4. "Physician recommendation" was the main driver for vaccination whereas "lack of habit" was the main barrier to vaccination. Vacinómetro® data demonstrate that free-of-charge vaccination has a positive impact on VCR. The observed positive trends in influenza VCR demonstrate that public health measures implemented in Portugal to facilitate access to influenza vaccine result in increased vaccine uptake. Strategies to promote population literacy and the physician's awareness should be continued and reinforced. Free-of-charge vaccination criteria extended to more risk groups would also contribute to higher influenza VCR in Portugal.

Abbreviations: WHO, World Health Organization; DGS, Direção-Geral da Saúde (Directorate-General of Health); HCWs, Healthcare Workers; ECDC, European Centre for Disease Prevention and Control; VCR, Vaccination Coverage Rate; EU, European Union; SPP, Sociedade Portuguesa de Pneumologia (Portuguese Society of Pneumology); APMGF, Associação Portuguesa de Medicina Geral e Familiar (Portuguese Association of General Practice and Family Medicine); NUTS, Nomenclatura das Unidades Territoriais para fins eStatísticos (Nomenclature of Territorial Units for Statistics); CATI, Computer-Assisted Telephone Interviewing; LVT, Lisboa e Vale do Tejo (Lisbon and Tagus Valley).

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Introduction

Vaccination is the most effective measure to prevent influenza virus infection.[1] The World Health Organization (WHO) recommends the annual vaccination for the most vulnerable population groups.[2] In the European Union (EU), vaccination coverage rates (VCR) among risk groups is below the recommended levels.[1,3,4] The average VCR of elderly people in EU was 41.8% in 2007-2015, far behind the WHO target of 75%.[5] Low VCR rates are also reported in people with chronic illnesses (50.3%) and healthcare workers (HCWs) (25.7%). The low VCR rates in EU are attributed to several factors, namely: no confidence in the vaccine, low perceived need for vaccination, no recommendation from healthcare providers and, in some countries, out-of-pocket costs of vaccine.[1] In this context, it is critical to monitor VCR trends to evaluate the impact of public health policies. Vacinómetro® is an initiative promoted by the Portuguese Pneumology Society (SPP) and the Portuguese Association of General Practice and Family Medicine (APMGP)[6,7] to monitor the influenza VCR in Portugal since the 2009-2010 flu season. Here we report Vacinómetro® results until the 2019-2020 season.

Methods

Vacinómetro® primary objective is to evaluate VCR in the four target groups defined by the Portuguese Directorate-General of Health (DGS)[8]: Group 1, $\geq$ 65 years old; Group 2, patients with chronic conditions; Group 3, HCWs in direct contact with patients; and Group 4, 60-64 years old. Data collection was based on annual phone surveys. Data was analysed using descriptive statistics. Chi-square test was used to test the linear trend against the null hypothesis of no trend and the trend of each vaccination/no-vaccination motive along seasons. For further information, please refer to the Supplementary Information.

Results

Vacinómetro® study sample increased from 200 subjects in 2009 to 2851 in 2020.[6] Overall, a total of 14832 questionnaires were collected. Fig. 1 summarizes the VCR evolution per target risk group over the years. There was an increasing trend for VCR in all target risk groups ($p<0.001$) – Table 1. There were no statistically significant differences in VCR according to gender (data not shown).

There were differences in overall VCR according to country regions: *North,* 59.3%; *Alentejo,* 54.5%; Centre, 53.7%; Algarve, 51.6%; and Lisbon and Tagus Valley (LVT), 45.1%. There was an increasing trend for VCR along the seasons in North, Centre, LVT, and Alentejo regions ($p\leq0.001$). The islands regions, Azores and Madeira, were included only in 2019-2020 season with reported VCR of 52.4% and 54.8%, respectively.

Reasons for vaccination/no vaccination were analysed in 2016-2020 – see supplementary tables 1 and 2. The main reason for vaccination in Groups 1, 2, and 4 was "physician recommendation" (66.3%) followed by "own initiative" (19.5%), both with statistically significant trends ($p<0.001$). Group 3 (HCWs) reported "part of a workplace initiative" as the main reason for vaccination (68.9%) with an increasing trend ($p<0.001$). Amongst the unvaccinated subjects, "lack of habit" (44.5%), "being healthy" (22.3%), and "never got the flu" (15.7%) were the primary reasons mentioned for no vaccination, all with statistically significant trends ($p<0.001$).

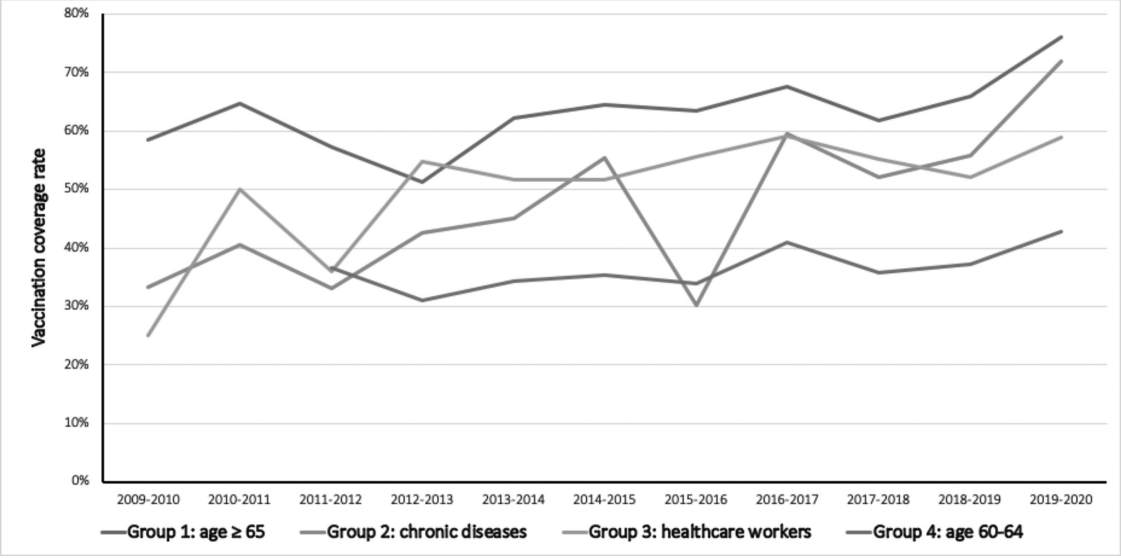

Fig. 1 Evolution of influenza vaccination coverage rates by risk group.

Table 1 Influenza vaccination coverage rates by risk group.

Vaccination Season	2009-2010	2010-2011	2011-2012	2012-2013	2013-2014	2014-2015	2015-2016	2016-2017	2017-2018	2018-2019	2019-2020	p*
Group 1 — age ≥ 65												
n vaccinated	169	153	334	300	601	600	600	759	680	778	1162	<0.001
(%)	58.6%	64.7%	57.2%	51.3%	62.2%	64.5%	63.5%	67.6%	61.8%	65.9%	76.0%	
95% CI	51.2% — 66.0%	57.1% — 72.3%	53.7% — 64.3%	45.6% — 57.0%	58.3% — 66.1%	60.7% — 68.3%	59.6% — 67.4%	64.3% — 70.9%	58.1% — 65.5%	62.6% — 69.2%	73.5% — 78.5%	
Group 2 — chronic diseases												
n vaccinated	27	38	300	300	300	300	300	517	643	699	813	<0.001
(%)	33.3%	40.5%	33.0%	42.7%	45.0%	55.3%	30.3%	59.6%	52.1%	55.8%	72.0%	
95% CI	15.5% — 51.1%	32.9% — 64.7%	27.7% — 38.3%	37.1% — 48.3%	39.4% — 50.6%	49.7% — 60.9%	25.1% — 35.5%	55.4% — 63.8%	48.2% — 56.0%	52.1% — 59.5%	68.9% — 75.1%	
Group 3 — healthcare workers												
n vaccinated	4	10	312	300	300	300	300	313	306	344	705	<0.001
(%)	25.0%	50.0%	35.9%	54.7%	51.7%	51.7%	55.7%	59.1%	55.2%	52.0%	58.9%	
95% CI	0.0% — 67.4%	19.0% — 81.0%	32.3% — 43.1%	49.1% — 60.3%	46.0% — 57.4%	46.0% — 57.4%	50.1% — 61.3%	53.7% — 64.5%	49.6% — 60.8%	46.7% — 57.3%	55.3% — 62.5%	
Group 4 — age 60-64												
n vaccinated			306	300	300	300	300	333	319	306	980	<0.001
(%)			36.6%	31.0%	34.3%	35.3%	34.0%	41.0%	35.7%	37.3%	42.8%	
95% CI			31.2% — 42.0%	25.8% — 36.2%	30.9% — 41.7%	29.9% — 40.7%	28.6% — 39.4%	35.7% — 46.3%	30.4% — 41.0%	31.9% — 42.7%	40.1% — 46.3%	

CI, confidence interval; * Chi-square test for trend.

Place for vaccine acquisition/administration was analysed from 2011. HCWs were vaccinated mostly free-of-charge in their work place (98.7%). Group 1 was mainly vaccinated at primary care centres, free-of-charge (83.7%), while Group 4 vaccines were mostly acquired with reimbursement at a pharmacy (57.3%). Across all study, vaccines acquired in pharmacies were mostly administered there.

In 2019-2020, pregnant women were analysed: 23.5% [95%CI = (20.1% — 6.9%)] of the 609 identified pregnant women were vaccinated, 50.3% of those for the first time and mainly due to "physician recommendation" (84.6%). Among unvaccinated pregnant women, "lack of physician recommendation" was the main reason for no vaccination (63.7%), yet 99.4% reported no intention of being vaccinated. Pregnant women were mostly vaccinated at primary care centres (46.9%).

Discussion

Influenza VCR is an important indicator of assessing the success of health policies promoting influenza vaccination. According to Vacinómetro®, influenza VCR in Portugal has improved in all target risk groups. Of note, in 2019-2020, the UE target of 75% VCR for people ≥ 65 years old[1,9] was achieved in Portugal with a reported VCR of 76%. Indeed, Portugal has been recognized as one of the few EU countries with a positive VCR trend in the risk group of older people.[3,10] A positive trend was also observed among patients with chronic conditions (some of them entitled to free-of-charge vaccination), reaching a VCR maximum of 72.0%. In HCWs, VCR gradually approached 60%.

The lowest VCR was observed in people 60-64 years old, with a maximum VCR of 42.8%. Although vaccination is recommended in this age group, it is not recognized as a risk group and therefore is not eligible for free administration,[8,9] in contrast to ≥ 65 years old who are entitled to free vaccination since 2012. This highlights the importance of free-of-charge influenza vaccination to increase VCR among target risk groups.

Vacinómetro® data has revealed regional differences for VCR, particularly between the two most populated regions in Portugal — North (59.3% VCR) vs. LVT (45.1% VCR). This may result from subtle regional differences in access to vaccination and/or population awareness, which would be interesting to investigate in order to fine-tune public health policies.

Vacinómetro® has demonstrated that physicians have a key role in vaccination promotion, since both vaccinated and unvaccinated subjects affirmed that physician's recommendation or lack of recommendation was a significant reason for their decision. This was particularly relevant among pregnant women, with an 84.6% recommendation rate. Since this group is eligible for free-of-charge vaccination since 2020, VCR will likely increase in the upcoming seasons.

Vacinómetro® has also provided insights on how to improve initiatives aiming to promote vaccination literacy and awareness, as among unvaccinated subjects, "being healthy", "lack of opportunity", and "lack of habit" are still referred as reasons for no vaccination.

In summary, Vacinómetro® has been demonstrated as an important tool to monitor influenza VCR in Portugal among

target risk groups. The observed VCR positive trends illustrate the success of health policies implemented in Portugal to promote and facilitate access to vaccines.

Conflicts of Interest

MM and CG are Sanofi Pasteur employees and may hold shares. The remaining authors did not receive any financial support and have no conflicts of interest to declare for the execution of the study.

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References

1. World Health Organization (WHO). Influenza vaccination coverage and effectiveness [Internet]. Available from: https://www.euro.who.int/en/health-topics/communicable-diseases/influenza/vaccination/influenza-vaccination-coverage-and-effectiveness.
2. World Health Organization (WHO). Vaccines against influenza WHO position paper. Relev Epidemiol Hebd. 2012 Nov;87(47):461–76.
3. Jorgensen P, Mereckiene J, Cotter S, Johansen K, Tsolova S, Brown C. How close are countries of the WHO European REGION to achieving the goal of vaccinating 75% of key risk groups against influenza? Results from national surveys on seasonal influenza vaccination programmes 2008/2009 to 2014/2015. Vaccine. 2018 Jan;36(4):442–52 https://linkinghub.elsevier.com/retrieve/pii/S0264410X17317620.
4. Finnegan G. Flu vaccine: did your country hit its target. Vaccines Today. 2020. Available from: https://www.vaccinestoday.eu/stories/flu-vaccine-did-your-country-hit-its-target/.
5. European Centre for Disease Prevention and Control. Seasonal influenza vaccination and antiviral use in EU/EEA Member States. 2018.
6. Sociedade Portuguesa de Pneumologia (SPP). Vacinómetro [Internet]. 2020. Available from: https://www.sppneumologia.pt/projetos-e-iniciativas/vacinometro
7. Associação Portuguesa de Medicina Geral e Familiar (APMGF). Vacinómetro [Internet]. 2020. Available from: https://apmgf.pt/2020/12/23/vacinometro-2020-2021-comprova/
8. DGS – Direção Geral de Saúde. Vacinação contra a gripe com a vacina trivalente na época 2011/2012 – alargamento dos Grupos de risco rara vacinação gratuita. 2011. Available from: https://www.spp.pt/UserFiles/file/EVIDENCIAS%20EM%20PEDIATRIA/ORIENTACAO%20DGS_031.2011%20DE%20SET.2011%20ACT.%20OUT.2011.pdf
9. Mereckiene J. Seasonal influenza vaccination in Europe – vaccination recommendations and coverage rates in the EU member states for eight influenza seasons –2007-2008 to 2014-2015 [Internet]. 2017. Available from: https://www.ecdc.europa.eu/sites/portal/files/documents/influenza-vaccination-2007–2008-to-2014-2015.pdf
10. Guedes L, Figueiredo A, Afonso A, Couto M, Natividade A, Gouveia A, et al. Increase in the influenza vaccination rates in Portugal: comparing 2020-2021 to the last 10 years. Eur J Public Health. 2021 Oct 20;31(Supplement_3). Available from https://academic.oup.com/eurpub/article/doi/10.1093/eurpub/ckab165.478/6406350.

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ORIGINAL ARTICLE

COPD: How can evidence from randomised controlled trials apply to patients treated in everyday clinical practice?

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KEYWORDS
COPD;
Randomised controlled trials;
Generalisability;
Inclusion criteria;
Exclusion criteria.

Abstract
Objectives: To evaluate the degree to which evidence from large clinical trials can be applied to patients treated in a local hospital cohort of COPD outpatients.
Methods: The authors selected seventeen RCTs identified in a systematic way from GOLD 2019 consensus document, and applied their inclusion and exclusion criteria to a real-world cohort of a previous cross-sectional study of 303 COPD outpatients included consecutively.
Results: When the inclusion criteria of the 17 RCTs were applied to a real-world cohort of COPD outpatients, only a small portion of them were eligible to participate in the referred trials, from 4.29% to 60.07%. However, when both the inclusion and the exclusion criteria were applied, only as little as 3.63% to as much as 40.59% of patients were eligible to participate. Hence, only a small fraction of patients from this cohort could benefit from the findings of these RCTs.
Conclusions: There is a need to complement the efficacy evidence provided by large RCTs according to the extent to which their results, designed to target significant patient populations, can be applied to typical patients treated in routine clinical practice.

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Background and objectives

Evidence-based medicine (EBM) has significantly changed the practice of medicine since the early 1990s. Well-designed randomized controlled trials (RCTs), together with systematic reviews and meta-analysis, remain the cornerstone of clinical research, and provide the basis for guideline recommendations.[1] Guidelines reduce uncertainty and promote the standardisation of clinical practice.[2] A number of significant RCTs, published in the current decade, have contributed extensively to the scientific evidence related to the treatment of COPD patients. RCTs are the gold-standard for safety and efficacy evaluations of new drugs. They present strong internal validity, with narrow inclusion criteria and demanding exclusion criteria, to reduce bias. Their homogeneous highly-selected populations ensure that only exposure to treatment differs between the arms of the study.[3] Therefore, patients enrolled in RCTs significantly differ from those treated in everyday clinical practice, reducing the possibility of extrapolation of their findings to unselected patient populations.[4] Pragmatic trials and other real-world studies can be performed to complement RCTs, providing evidence of drug effectiveness in the heterogeneous populations of clinical practice.[5] There is no robust knowledge of how representative patients enrolled in large RCTs are of a typical patient population for whom they intend to develop their findings and conclusions.[6] Furthermore, it was not known how representative these patients are of a Portuguese population of patients with COPD. The objective of the present study was to evaluate the degree to which evidence from RCTs can be applied to a cohort of patients treated in the out-patient clinic of Hospital de Guimarães.

Table 1 Demographic, clinical and functional characteristics of COPD patients.

Characteristics	n = 303
Male gender	241 (79.5)
Mean age (years)	67.5 ± 10.2
Age ≥ 65 Years	186 (61.4)
Education level ≤ 3 school years	89 (29.4)
Very low monthly income (< 530 Euros)	197 (65.7)
Mean smoking amount (pack/years)	49.3 ± 32.4
Current smokers / ex-smokers	224 (73.9)
Never smokers or < 10 pack/year	90 (29.7)
Occupational exposure	164/295 (55.5)
Indoor exposure in women	37/58 (63.8)
Alfa-1-AT deficiency / ZZ genotype	12 (3.9) / 7 (2.3)
Previous history of asthma	82/299 (27.4%)
mMRC grade ≥ 2	185 (61.1)
CAT score ≥ 10	152 (72.4)
Frequent ECOPD (≥ 2 / previous year)	115 (38.0)
GOLD stage:	
I	30 (9.9)
II	127 (41.9)
III	106 (35.0)
IV	40 (13.2)
Gold 2017 classification:	
A	70 (23.1)
B	120 (39.6)
C	7 (2.3)
D	106 (35.0)

Data shown as n (%); many different exposures relevant for COPD overlapped in the same patients.
Abbreviations: Occupational exposure, self-reported occupational exposure to gas, fumes and dust relevant to COPD; Indoor exposure, sustained indoor-exposure to household air pollution from coal and biomass fuel combustion in women; Alfa-1-AT, alfa-1-antitrypsin; mMRC, Medical Research Council Dyspnea Questionnaire; CAT, COPD Assessment Test; ECOPD, COPD exacerbations; GOLD, Global Initiative for Chronic Obstructive Lung Disease.

Methods

Study design

This is a retrospective analysis, using a pre-existing cohort of COPD out-patients. We applied the inclusion and exclusion criteria of the selected RCTs to COPD patients observed in our routine clinical practice, and diagnosed according to GOLD criteria.[7] From March 2016 to May 2017, participants were recruited consecutively in the outpatient pulmonary clinic of Guimarães Hospital, a middle-sized public hospital, treating patients from an urban and rural background.[8] The only exclusion criteria were refusal to participate and inability to understand simple questionnaires.

Selection of RCTs

The GOLD strategy has a worldwide influence in the management of COPD patients, and we searched for RCTs with a significant impact on the pharmacologic management of COPD. They had to fulfil the following conditions:

- Cited in the GOLD 2019 consensus document,[9] in chapter 3, ''Evidence supporting prevention and maintenance therapy'', section on ''Pharmacological therapy for stable COPD'', and chapter 4 ''Management of stable COPD'', section on ''Treatment of stable COPD: pharmacological treatment''.
- Related to inhaled corticosteroids, long-acting β_2-agonists or long-acting anticholinergic therapy.
- Related to single or combination inhaled therapy.
- RCTs published in the present decade (from 2010 to 2019).
- RCTs with at least 400 COPD patients at randomisation, and lasting 52 weeks or more.
- RCTs studying only COPD patients diagnosed according to GOLD criteria, or having a history of COPD as defined by the American Thoracic Society or the European Respiratory Society.

RCTs were identified in a systematic way, and the flow diagram related to the extraction of RCTs from GOLD 2019 strategy is described in Fig. 1.

The inclusion and exclusion criteria were obtained from published papers and/or described in the on-line supplementary appendixes.[10–26] Secondary analysis, as systematic

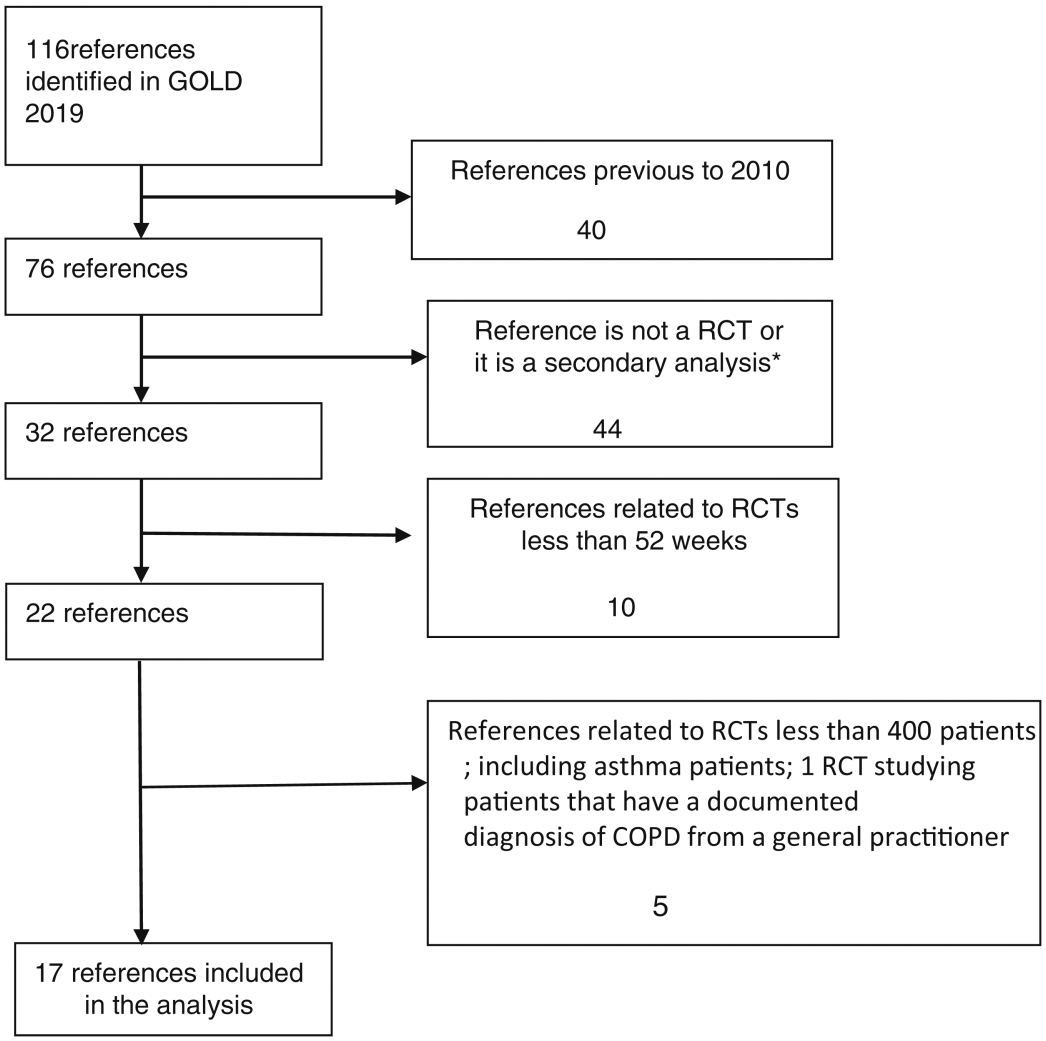

Fig. 1 Flow diagram for selection of RCTs from GOLD 2019 strategy.
Note: *All secondary analysis, as systematic reviews, with or without meta-analysis, post hoc analysis and pooled analysis were excluded.

reviews, with or without meta-analysis, post hoc analysis and pooled analysis were excluded, because they shared the same populations studied.

Data source

The main demographic, clinical and functional characteristics of 303 COPD patients are described in Table 1.

Only 9.5% of patients reported no comorbid conditions, and the most prevalent are presented in Fig. 2.

The mean post-bronchodilator FEV_1% was 53.2 ± 19.7, referenced according to the Global Lung Function Initiative predict equations (GLI 2012).[27] A comprehensive report of patients' characteristics is described and published elsewhere.[8] The statistical analysis was performed with IBM SPSS Statistics for Windows, version 23.0, Armonk, NY: IBM Corp.

Results

Table 2 describes the largest possible number and percent of the 303 patients from the Guimarães Hospital Cohort (HGC) who met the 17 RCTs inclusion and exclusion criteria.

Some data, related to the described RCTs inclusion and inclusion criteria and their patients mean age, are described in Table 3, 4 and 5. Patients of HGC were significantly older than those participating in all of the referred trials.

Discussion

Key results

When the inclusion criteria of the referred RCTs were applied to HGC, only a small portion of them could be eligible to participate in the referred trials, from 4.29% in the SUMMIT to

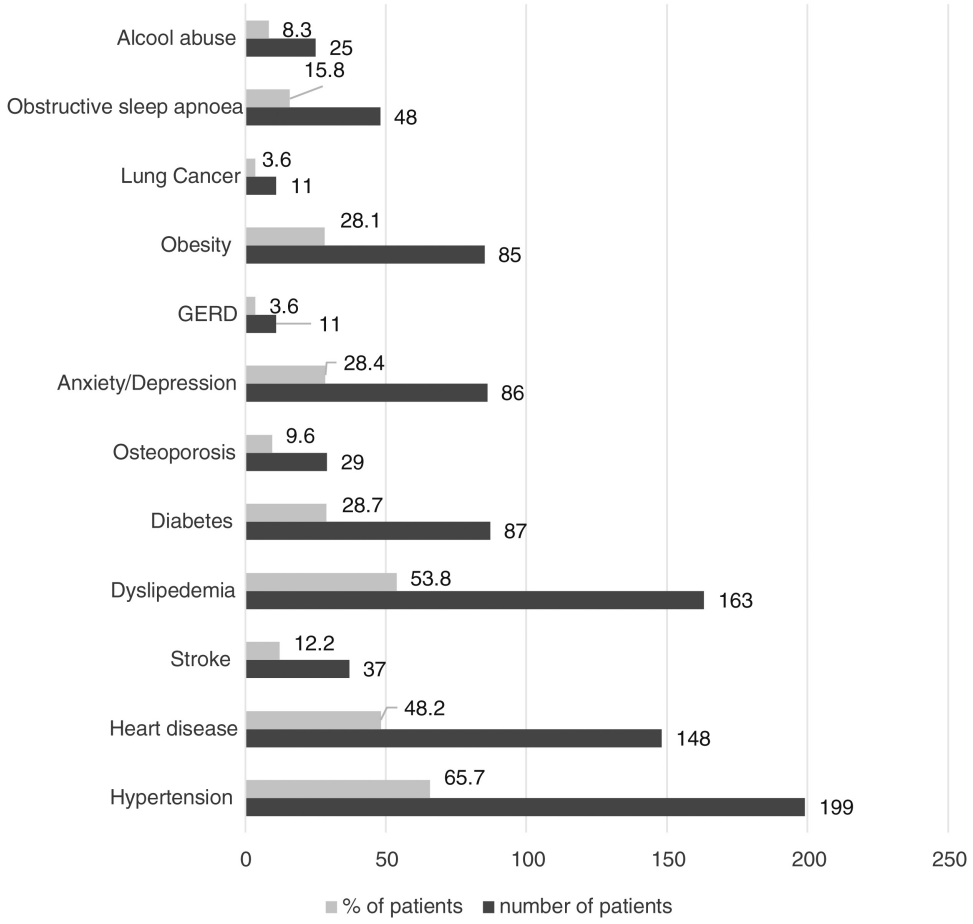

Fig. 2 Common comorbid conditions in 303 patients.
Notes and abbreviations: Heart disease: ischemic heart disease, heart failure or atrial fibrillation; GERD, gastroesophageal reflux disease; Stroke, history of stroke. Obesity: BMI $\geq$ 30 Kg/m^2.

Table 2 Patients from the HGC who could participate in the referred RCTs.

RCTs[REF] (2010 / 2018)	Patients from HGC meeting the inclusion criteria[a]	Patients from HGC meeting both the inclusion and exclusion criteria[a]
Indacaterol/formoterol[9]	165 (54.46)	<123 (40.59)
POET[10]	90 (29.70)	<67 (22.11)
SPARK[11]	70 (23.10)	<39 (12.87)
INVIGORATE[12]	48 (15,84)	<41 (13.53)
Fluticasone + vilanterol/ vilanterol[13]	90 (29.70)	<53 (17.49)
WISDOM[14]	70 (23.10)	<47 (15.51)
Crim C et al.[15]	90 (29.70)	<50 (16.50)
FLAME[16]	61 (20.13)	<31 (10.23)
SUMMIT[17]	13 (4.29)	<11 (3.63)
TRILOGY[18]	47 (15.51)	<24 (7.92)
Tie-COPD[19]	150 (49.50)	<104 (34.32)
TONADO[20]	182 (60.07)	<110 (36.30)
TRINITY[21]	47 (15.51)	<24 (7.92)
FULFIL[22]	106 (34.98)	106 (34.98)
TRIBUT[23]	47 (15.51)	<29 (9.57)
DYNAGITO[24]	83 (27.39)	<67 (22.11)
IMPACT[25]	54 (17.82)	<44 (14.52)

[a] Data shown as number (% of total -303- patients). HGC, Guimarães Hospital Cohort; RCTs, randomized controlled trials.

Table 3 RCTs inclusion and exclusion criteria and patients mean age.

Study/reference Year of publication	Indacaterol/ Formoterol[9] (2010)	POET[10] (2011)	SPARK[11] (2013)	INVIGORATE[12] (2013)	Fluticasone + Vilanterol / Vilanterol[13] (2013)	WISDOM[14] (2014)
Patients' age (years); mean age	63.0 64.0 (median)	62.8 (9.0) to 62.9 (9.0)	63.1 (8.0) to 63.6 (7.8)	40-91 64.0 (Not reported)	63.5 (8.8) to 64.0 (9.3)	63.8 (8.5)
Inclusion of never-smokers	No	No	No	No	No	No
Inclusion if smoking amount < 10 pack/years	No	No	No	No	No	No
Inclusion of GOLD stage I patients	No	No	No	No	No	No
Inclusion referred to FEV_1%	<80 and ≥30	≤ 70	<50	≥30 and <50	≤ 70	<50
Inclusion if mMRC<2 or CAT<10	Yes	Not reported	Not reported	Yes	Yes	Not reported
Inclusion of patients without previous ECOPD	Yes	No	No	No	No	No
Exclusion if previous asthma	Yes	Yes	Yes	Yes	No	No
Exclusion if current asthma	Yes	Yes	Yes	Yes	Yes	Yes
Exclusion of patients in LTOT	No	Not reported	Yes	No	Yes	Yes
Exclusion if Alfa-1-AT deficiency	No	Not reported	Yes	No	Yes	Not reported
Exclusion of malignancy / significant pulmonary comorbidities / unstable pulmonary disease	No	Not reported	Yes	No	Yes	Yes
Exclusion if clinical bronchiectasis	No	No	Yes	No	Yes	Yes
Exclusion of comorbidities**	No	Yes	Yes	No	Yes	Yes

RCTs, randomized controlled trials; mMRC, Medical Research Council Dyspnoea Questionnaire; CAT, COPD Assessment Test; LTOT, long-term Oxygen therapy; Alfa-1-AT, alfa-1-antitrypsin deficiency; * Mean age unless stated otherwise; **comorbidities, current significant disease which can influence the results of the studies or the patients' ability to participate in the trials.

60.07% in the TONADO study. When both the inclusion and the exclusion criteria were applied, only as little as 3.63% in the SUMMIT, to as much as 40.59% in the study comparing formoterol to indacaterol were eligible to participate. The lack of representativeness of clinical trials in real patient populations seems to be common knowledge. In this study we quantify their magnitude in a local population of COPD outpatients followed in a medium-sized hospital. To the best of our knowledge this is the first study with such characteristics developed in a Portuguese population of COPD patients.

In the HGC study, 90 participants were never-smokers, or the smoking amount was < 10 pack/years, totalising 29.70% patients. They were not represented and could not benefit from the findings of 15 of the 17 referred studies. However, never-smokers can account for 31.7–45% of COPD patients in different studied populations,[28,29] and a significant proportion of COPD patients around the world are known to be never-smokers.[30] Nor were the findings applicable to 47 (75.80%) of the 62 women of HGC, again because they were never-smokers. It seems that the evidence from large RCTs is not applicable to never-smoking COPD patients. Asthma is a well-known risk factor for COPD, but because of the possibility of confusion between COPD and asthma, these patients are excluded from many RCTs.[31] In the HGC 82 patients (27.42%) referred to a previous history of asthma under the age of 40, usually in childhood. The findings of nine of the

Table 4 RCTs inclusion and exclusion criteria and patients mean age (cont.).

Study/reference Year of publication	Crim C et al[15] (2014; ref 16)	FLAME[16] (2016; ref 17)	SUMMIT[17] (2016; ref 18)	TRILOGY[18] (2016; ref 19)	Tie-COPD[19] (2017; ref 20)	TONADO[20] (2017; ref 21)
Patients' age (years); mean age	63.6 (9.4) to 63.8 (9.2)	64.6 (7.8)	65 (8)	63.3 (7.9) to 63.8 (8.2)	63.9 (8.6) to 64.2 (8.2)	63.8 (8.3) to 66.2 (8.0)
Inclusion of never-smokers	No	No	No	No	Yes	No
Inclusion if smoking amount < 10 pack/years	No	No	No	No	Yes	No
Inclusion of GOLD stage I patients	No	No	No	No	Yes	No
Inclusion referred to FEV_1%	$\leq$ 70%	$\geq$25 and <60	>50 and $\leq$70	<50	$\geq$ 50	< 80
Inclusion if mMRC<2 or CAT<10	Yes	No	No	No	Yes	Yes
Inclusion of patients without previous ECOPD	No	No	Yes	No	Yes	Yes
Exclusion if previous asthma	No	Yes	No	Yes	Yes	Yes
Exclusion if current asthma	yes	Yes	Yes	Yes	Yes	Yes
Exclusion of patients in LTOT	Yes	Yes	Yes	Yes	Not applicable	Yes
Exclusion if Alfa-1-AT deficiency	Yes	Yes	Not reported	Yes	Not reported	Not reported
Exclusion of malignancy / significant pulmonary comorbidities / unstable pulmonary disease	Yes	Yes	Yes	Yes	Yes	Yes
Exclusion if clinical bronchiectasis	Yes	Yes	Yes	Yes	Not reported	Yes
Exclusion of comorbidities*	Yes	Yes	Not applicable	Yes	Yes	Yes

RCTs, randomized controlled trials; mMRC, Medical Research Council Dyspnoea Questionnaire; CAT, COPD Assessment Test; LTOT, long-term Oxygen therapy; Alfa-1-AT, alfa-1-antitrypsin deficiency; *comorbidities, current significant disease which can influence the results of the studies or the patients' ability to participate in the trials.

17 RCTs were not applicable to them. This matches previous papers.[32] Twelve patients of HGC reported alfa-1-antitrypsin deficiency and 7, presenting a ZZ genotype, fulfilled criteria for augmentation therapy.[33] They could not benefit from the evidence of at least eight RCTs. Thirty patients (9.90%) classified as GOLD stage I are represented in only one of the trials mentioned. Seventy COPD patients (23.10%) were classified as group A for therapeutic purposes. The findings of at least seven RCTs could not apply to them. The findings of eleven RCTs could also not apply to a significant number of the 45 patients benefiting from long-term oxygen therapy. From the 303 patients of HGC, 145 (47.85%) had not reported any ECOPD in the previous year. The findings of 12 of the 17 RCTs could not be applied to them. In the HGC the patients' mean ages were significantly higher than reported in all RCTs. The process of aging is often associated with an increased number of comorbidities and prescribed medication. It is well-known that there is a low rate of adverse effects when using inhaled medication in COPD patients. Nevertheless, by excluding patients with significant comorbidities, needing several chronic medications, RCTs promise a high rate of tolerability, which can be different in daily clinical practice. This may also change the way the results of RCTs apply to patients treated in routine clinical practice.

We acknowledge that the results of clinical RCTs will not be relevant to all COPD patients, and clinicians should select the patients to whom the results can be applied.[32] However, a general lack of reliability must thus be assumed in the generalisation of conclusions from a significant number of RCTs to never-smokers, to patients with an asthma background, to patients suffering from alfa-1-antitrypsin deficiency, to patients GOLD stage I, to those not reporting previous exacerbations, and mainly to patients with significant comorbid conditions. Moreover, the inclusion criteria referred to by

Table 5 RCTs inclusion and exclusion criteria and patients mean age (cont.).

Study/reference; Year of publication	TRINITY[21] (2017; ref 22)	FULFIL[22] (2017; ref 23)	TRIBUTE[23] (2018; ref 24)	DYNAGITO[24] (2018; ref 25)	IMPACT[25] (2018; ref 26)
Patients' age (years); mean age	62.6 (8.9) to 63.4 (8.7)	63.9 (8.6)	64.4 (7.7) to 64.5 (7.7)	66.3 (8.5) to 66.5 (8.4)	65.3 (8.3)
Inclusion of never-smokers	No	Yes	No	No	No
Inclusion if smoking amount < 10 pack/years	No	Yes	No	No	No
Inclusion of GOLD stage I patients	No	No	No	No	No
Inclusion referred to FEV_1%	<50	<80	<50	<60	<80
Inclusion if mMRC<2 or CAT<10	No	No	No	Not reported	No
Inclusion of patients without previous ECOPD	No	Yes	No	No	No
Exclusion if previous asthma	Yes	No	No	No	No
Exclusion if current asthma	Yes	Yes	Yes	Yes	Yes
Exclusion of patients in LTOT	Yes	No	Yes	No	Yes (if $\geq$ 3 L)
Exclusion if Alfa-1-AT deficiency	Yes	Not reported	Yes	Not reported	Yes
Exclusion of malignancy / significant pulmonary comorbidities / unstable pulmonary disease	Yes	Not reported	Yes	Yes	Yes
Exclusion if clinical bronchiectasis	Yes	Not reported	Yes	Not reported	Yes
Exclusion of comorbidities*	Yes	No	Yes	Not reported	Yes

RCTs, randomized controlled trials; mMRC, Medical Research Council Dyspnoea Questionnaire; CAT, COPD Assessment Test; LTOT, long-term Oxygen therapy; Alfa-1-AT, alfa-1-antitrypsin deficiency; *comorbidities, current significant disease which can influence the results of the studies or the patients' ability to participate in the trials.

many RCTs significantly differ from normal clinical practice, where the ABCD assessment tool is usually used for therapeutic purposes.

Comparison with previous studies

Our findings agree with previous literature. In the respiratory field, as little as 5% of the target population was represented in the recruited populations of RCTs,[34] Travers et al. demonstrated that only one in 20 patients with COPD, identified from a large general population survey in New Zealand, would have met the inclusion criteria for the major RCTs informing guidelines in COPD.[35] Using data from a large European COPD primary care database, Kruis et al. found that 58–83 % of COPD patients in primary care would not serve as candidates for inclusion, in significant RCTs.[36] Costa et al. related that only 7.4% of primary care patients could met the inclusion criteria used in 4 RCTs for allergic rhinitis, [6] and in a Norwegian study only a very small fraction of patients with asthma or COPD were shown to be represented in a typical clinical trial.[4] A previous study,[37] combining an extensive RCTs selection with a very representative COPD population, found that only around a quarter of community patients with COPD were eligible from RCTs participation.

Strengths and limitations of the study

The present study was conducted by independent researchers, and the described RCTs were selected in a systematic manner. They have extensively contributed to the scientific evidence related to the treatment of COPD patients, and their conclusions are embodied in the Global

Initiative for Chronic Obstructive Lung Disease (GOLD) report.

However, a significant number of exclusion criteria referred to by RCTs could not be identified in the HGC. In the HGC, there was no information related to established peripheral vascular disease and to diabetes mellitus with target organ disease. It was also not possible to specifically identify patients with established coronary artery disease and previous myocardial infarction, in the 148 patients referred to as suffering from heart disease. Because of that, only 13 patients of HGC (4.29%) meet the inclusion criteria of the SUMMIT study. The presence of a significant current disease, other than COPD, which can influence the results of the studies or the patients' ability to participate in the trials, was a common exclusion criterion, in many of the referred RCTs. However, it was not considered in the present analysis, because it was a very subjective issue. A clinical diagnosis of bronchiectasis was also referred as exclusion criteria in the majority of these studies. Likewise, it was not considered in the present analysis, because bronchiectasis' symptoms significantly overlap with those of COPD. Therefore, the number of patients who could benefit from the findings of these studies could be significantly lower.

Conversely, the HGC presented some specific characteristics that can partially justify the small fraction of patients that could benefit from the results of the referred RCTs: a significant rate of patients referring a previous history of asthma; the high prevalence of alfa-1-antitrypsin deficiency; the substantial number of individuals with low monthly income and low education level; a significant number of never-smokers COPD patients, mainly women, referring exposure to household air pollution from coal and biomass fuel combustion.

All of the referred above can significantly limit the generalisation of the results to other populations with different characteristics.

Conclusions

Randomised controlled trials are performed to provide a significant level of evidence on treatments efficacy. However, there is a significant lack of representativeness of RCTs in real populations of COPD patients. There is a need to complement the evidence provided by large RCTs, according to the extent to which their results, designed to target significant patients' populations, can be applied to typical patients treated in routine clinical practice. Pragmatic trials and other real-life studies must provide additional evidence about treatment effectiveness, in the heterogeneous populations of clinical practice.

Author contributions

Duarte-de-Araújo conceived and developed the study, carried out selection of bibliography, wrote the first draft and collaborated in the final writing. Pedro Teixeira carried out the statistical analysis and reviewed the final draft. Venceslau Hespanhol reviewed the final draft. Jaime Correia-de-Sousa reviewed all the drafts and collaborated in the final writing. All the authors approved the final manuscript.

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Conflicts of interest

The authors have no conflict of interest to declare regarding the present study.

References

[1]. Shrimanker R, Beasley R, Kearrns C. Letting the right one in: evaluating the generalisability of clinical trials. Eur Respir J. 2018;52:1802218.

[2]. Mercury M, Sherbino J, Sedran R, Frank J, Gafni A, Norman G. When Guidelines Don't Guide: The Effect of Patient Context on Management Decisions Based on Clinical Practice Guidelines. Acad Med. 2015;90(2):191–6.

[3]. Cohen A, Goto S, Schreiber K. Why do we need observational studies of everyday patients in the real-life setting? Eur Heart J. 2015;17 Suppl D:D2–8.

[4]. Herland K, Akselsen J, Skjonsberg O, Bjermer L. How representative are clinical study patients with astma or COPD for a larger 'real life' population of patients with obstructive lung disease? Resp Med. 2005;99:11–9.

[5]. Blonde L, Fhunti K, Harris S. Interpretation and Impact of Real-World Clinical Data for the Practing Clinician. Adv Ther. 2018;35:1763–74.

[6]. Costa D, Amouyal M, Lambert P, Ryan D, Schunemann H, Daures J, et al. How representative are clinical study patients with allergic rhinitis in primary care? J All Clin Immunol. 2011;127(4):920–7.

[7]. The Global Initiative for Chronic Obstructive Lung Disease (GOLD), updated; 2017. Available from http://goldcopd.org/gold-2017-global-strategy-diagnosis-management-prevention-copd/

[8]. Duarte-de-Araújo A, Teixeira P, Hespanhol V, Correia-de-Sousa J. Characterisation of morbidity in a COPD hospital cohort. Pulmonology. 2019;25(4):200–7.

[9]. The Global Initiative for Chronic Obstructive Lung Disease (GOLD); 2019, report. Available from https://goldcopd.org/wp-content/uploads/2018/11/GOLD-2019-v1.7-FINAL-14Nov2018-WMS.pdf.

[10].Dahl R, Chung K, Buhl R, Magnussen H, Nonikov V, Jack D, et al. Efficacy of a new once-daily long-acting inhaled β_2-agonist indacaterol versus twice-daily formoterol in COPD. Thorax. 2010;65:473–9.

[11].Volgelmeier C, Hederer B, Glaab T, Schmidt H, Molken M, Beeh K, et al. Tiotropium versus Salmeterol for the Prevention of Exacerbations of COPD. N Engl J Med. 2011;364(12):1093–103.

[12].Wedzicha J, Decramer M, Ficker J, Niewoehner D, Sandstrom T, Taylor A, et al. Analysis of chronic obstructive pulmonary disease exacerbations with the dual bronchodilator QVA149 compared with glycopyrronium and tiotropium (SPARK): a randomized, double-blind, parallel-group study. Lancet Respir Med. 2013;1(3):199–209.

[13].Decramer M, Chapman K, Dahl R, Frith P, Devouassoux G, Fritscher C, et al. Once-daily indacaterol versus tiotropium for patients with severe chronic obstructive pulmonary disease (INVIGORATE): a randomized, blinded, parallel-group study. Lancet Respir Med. 2013;1:524–33.

[14].Dransfield M, Bourbeau J, Jones P, Hanania N, Mahler D, Vestbo J, et al. Once-daily inhaled fluticasone furoate and vilanterol versus vilanterol only for prevention of exacerbations of COPD: two replicate double-blind, parallel-group, randomized controlled trials. Lancet Respir Med. 2013;1:210–23.

[15].Magnussen H, Disse B, Rodriguez-Roisin R, Kirsten A, Watz H, Tetzlaff K, et al. Withdrawal on Inhaled Glucocorticoids and Exacerbations of COPD. N Engl J Med. 2014;371(14):1285–94.
[16].Crim C, Dransfield M, Bourbeau J, Jones P, Hanania N, Mahler D, et al. Pneumonia Risk with Inhaled Fluticasone Furoate and Vilanterol Compared With Vilanterol Alone in Patients with COPD. . Ann Am Thors Soc. 2015;12(1):27–34.
[17].Wedzicha J, Banergi D, Chapman K, Vestbo J, Roche N, Ayers T, et al. Indacaterol-Glycopyrronium versus Salmeterol-Fluticasone for COPD. N Engl J Med. 2016;374(23):2222–34.
[18].Vestbo J, Anderson J, Brook R, Calverley P, Celli B, Crim C, et al. Fluticasone furoate and vilanterol and survival in chronic obstructive pulmonary disease with heightened cardiovascular risk (SUMMIT): a double-blind randomised controlled trial. Lancet. 2016;387:1817–26.
[19].Singh D, Papi A, Corradi M, Pavlisová I, Montagna I, Francisco C, et al. Single inhaler therapy versus inhaled corticosteroid plus long-acting β_2-agonist therapy for chronic obstructive pulmnonary disease (TRILOGY): a double-blind, parallel group, randomised controlled trial. Lancet. 2016;388: 963–73.
[20].Zhou Y, Zhong N, Xiaochen Li, Chen S, Zheng J, Zhao D, et al. Tiotropium in Early-Stage Chronic Obstructive Pulmonary Disease. N Engl J Med. 2017;377:923–35.
[21].Bai C, Ichinose M, Lee S, Lee K, Jons O, Botchner U, et al. Lung function and long-term safety of tiotropium/olodaterol in East Asian patients with chronic obstructive pulmonary disease. Int JCOPD. 2017;12:3329–39.
[22].Vestbo J, Papi A, Corradi M, Blazhko V, Montagna I, Francisco C, et al. Single inhaler extrafine triple therapy versus long-acting muscarinic antagonist therapy for chronic obstructive pulmonary disease (TRINITY): a double-blind, parallel group, randomised controlled trial. Lancet. 2017;389:1919–29.
[23].Lipson D, Barnacle H, Birk R, Brealey N, Locantore N, Lomas D, et al. FULFIL Trial: Once-daily Triple Therapy for Patients with Chronic Obstructive Pulmonary Disease. Am J Respir Crit Care Med. 2017;196(4):438–46.
[24].Papi A, Vestbo J, Fabbri L, Corradi M, Prunier H, Cohuet G, et al. Extrafine inhaled triple therapy versus dual bronchodilator therapy in chronic obstructive pulmonar disease (TRIBUTE): a double-blind, parallel group, randomised controlled trial. Lancet. 2018;391:1076–84.
[25].Calverley P, Anzueto A, Carter K, Gronke L, Hallmann C, Jenkins C, et al. Tiotropium and olodaterol in the prevention of chronic obstructive pulmonary disease exacerbations (DYNAGITO): a double-blind, randomised, parallel-group, active-controlled troal. Lancet Respir Med. 2018;6:337–44.
[26].Lipson D, Barnhart F, Brealey N, Brooks J, Criner G, Day N, et al. Once-Daily Single-Inhaler Triple versus Dual Therapy in Patients with COPD. N Engl J Med. 2018;378(18):1671–80.
[27].Quanjer P, Stanojevic S, Cole T, et al. Multi-ethnic reference values for spirometry for the 3-95-year age range: the global lung function 2012 equations: report of the Global Lung Function Initiative (GLI). ERS task force to establish improved lung function reference values. Eur Respir J. 2012;40(6):1324–43.
[28].Denguesli M, Daldoul H, Harrabi I, Gnatiuc L, Coton S, Burney P, et al. COPD in non-smokers: report from the Tunisian Population-based Burden of Obstructive Lung Disease. Plos One. 2016;11(3):e0151981.
[29].Lee S, Kim S, Kong K, Ryu Y, Lee J, Chang J. Risk factors for chronic obstructive pulmonary disease among never-smokers in Korea. Int J COPD. 2015;10:497–506.
[30].Zeng G, Sun B, Zhong N. Non-smoking-related chronic obstructive pulmonary disease: a neglected entity? Respirology. 2012;17:908–12.
[31].Cullinan P. Occupation and chronic obstructive pulmonary disease (COPD). Br Med Bull. 2012;104:143–61.
[32].Rothwell P. Factors That Can Affect the External Validity of Randomised Controlled Trials. PLoS Clin Trials. 2006;1(1):e9.
[33].Lopes A, Mineiro M, Costa F, Gomes J, Santos C, Antunes C, et al. Portuguese consensus document for the management of alfa-1-antitrypsin deficiency. Pulmonology. 2018;24(S1):1–21.
[34].Roche N, Reddel H, Martin R, Brusselle G, Papi A, Thomas M, et al. Quality Standards for Real-World Research Focus on Observational Database Studies of Comparative Effectiveness. Ann Am Thor Soc. 2014;11(2):S99–104.
[35].Travers J, Marsh S, Caldwell B, Williams M, Aldington S, Weatherall M, et al. External validity of randomized controlled trials in COPD. Respir Med. 2007;101(6):1313–20.
[36].Kruis A, Stallberg B, Jones R, Tsiligianni I, Lisspers K, Molen T, et al. Primary Care COPD Patients Compared with Large Pharmaceutically-Sponsored COPD Studies: Na UNLOCK Validation Study. Plos One. 2014;9(3):e90145.
[37].Halpin D, Kerkhof M, Soriano J, Mikkelsen H, Price D. Eligibility of real-life patients with COPD for inclusion in trials of inhaled long-acting bronchodilator therapy. Respir Res. 2016;17(120).

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ORIGINAL ARTICLE

Dynamic hyperinflation, chronotropic intolerance and abnormal heart rate recovery in non-severe chronic obstructive pulmonary disease patients-reflections in the mirror

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KEYWORDS
Heart rate recovery;
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Abstract
Background: The presence of abnormal heart rate recovery (HRR) and chronotropic incompetence (CI) suggests autonomic dysfunction (AD) and is associated with diminished physical activity and increased cardio-vascular (CV) risk.
Aim: Our aim is to analyse the correlation between AD and airflow obstruction - forced expiratory volume in 1 s (FEV1), dynamic hyperinflation (DH) and disease prognosis - the BODE – index (BMI; Obstruction – FEV1;Dyspnea – mMRC;E – exercise capacity) in non-severe COPD patients without overt CV comorbidities.
Methods: We used cardio-pulmonary exercise testing (CPET) with 67 subjects. Inspiratory capacity (IC) manouevres were performed for DH assessment. Echocardiography was executed before CPET and 1—2 min after peak exercise. Stress left ventricular diastolic dysfunction (LVDD) was assumed if stress E/e' > 15.Wilkoff method calculated the metabolic-chronotropic relationship (MCR). Chronotropic incompetence (CI) and abnormal HR recovery (HRR) were determined.
Main results: CI was detected in 44% of the mild and 65% of the moderate COPD patients. Abnormal HRR was present in 75% of the mild and 78% of the moderate COPD subjects. Multivariate regression analysis showed no association between FEV1, CPET parameters, BODE index, stress LVDD and AD. DH was the only independent predictor for both abnormal HRR and CI.

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Conclusion: Evaluation of AD during incremental CPET unravels lung hyperinflation as a potential mechanism of attenuated HR response and diminished physical activity in non-severe COPD free of overt CV comorbidities. This multifaceted approach to dyspnea may facilitate the discrimination of its pathogenesis and improve its proper clinical management.

Introduction

Chronotropic incompetence (CI) – the inability to reach the target heart rate (HR) during exercise is believed to represent an impaired sympathetic response and is an independent predictor of cardiovascular diseases.[1,2] Heart rate recovery (HRR) – the rate of restoration of the heart rate (HR) during the first minute after exercise – the recovery phase, represents the parasympathetic response. Its abnormal delay (a decline of HR < 12 beats/min) implies parasympathetic dysfunction.[3,4] Abnormal HRR and CI indicate the presence of autonomic dysfunction (AD). Being established as independent predictors of cardiac mortality, these two types of abnormal physiological responses may find application in cardiovascular risk stratification.[5,6]

Though not systematically investigated, both CI and HRR have been described in either hypoxemic, or normoxemic COPD patients.[7,8] Their prevalence is variable within the range of 56–86% among studies. Even more controversial is the data regarding the relation between AD and COPD stages.[8–11] This can be explained by the fact that different COPD stages have been investigated.

Considered a major contributor to exercise intolerance and dyspnea, dynamic hyperinflation (DH) has additional negative consequences in COPD.[12,13] DH is a result of expiratory airflow limitation and occurs when ventilatory demand increases, leaving less time for expiration. The end-expiratory lung volume (EELV) increases, altering the intrathoracic pressure gradients. Cardio-vascular consequences follow – the right and left ventricle preload diminish, while the left ventricle afterload increases. This impairs LV filling, compliance and cardiac output.[12,13] DH deters not only the stroke volume, but also blunts HR surge. It has been recently observed that it is associated with AD, and correlates to impaired metabolic-chronotropic relationship (CI) independent of age, static lung volumes and airway obstruction.[14] The autonomic abnormalities, associated with DH are much more constant than the haemodynamic heart-lung effects in the different COPD stages.

Assuming this we set the following aims: (1) to detect the prevalence of CI and abnormal HRR in non-severe COPD patients free of overt CV diseases; (2) to analyse their association with disease severity – forced expiratory volume in 1 s (FEV1), dynamic hyperinflation; (3) and disease prognosis the BODE – index (BMI; Obstruction – FEV1;Dyspnea – mMRC scale;E – exercise capacity).

Materials and methods

Patients and study protocol

It was a prospective study which was performed in 224 clinically stable outpatients, diagnosed with COPD at the University Hospital for Respiratory Diseases ''St. Sophia'', Sofia. Only 163 of them met the inclusion criteria: (1) non-severe COPD (post bronchodilatator FEV1/FVC < 70%; FEV1/ > 50%); (2) preserved left ventricular systolic function LVEF > 50%; (3) lack of overt cardiovascular disease; (4) exertional dyspnea. A total of 67 patients were considered eligible, assuming the exclusion criteria. The recruitment period was between April 2018–April 2019, and was approved by the local Ethical Committee (protocol 5/12.03.2018). All the patients signed informed consent before their participation. They were previously acquainted with the aim of the study, its scientific value and the potential presentation of data at different forums. Dyspnea was rated applying the modified Medical Research Council (mMRC) scale. Exercise capacity was assessed using the 6 min walking test (6MWT). Body mass index, airflow Obstruction, Dyspnea and Exercise capacity – (BODE) index was calculated.[15] Inhaled bronchodilators (β2 agonists and anti-cholinergics) were withdrawn 24 h before investigation.

The following exclusion criteria were considered: (1) LVEF < 50%; (2) LVDD at rest more than first grade; (3) echocardiographic signs of systolic pulmonary arterial hypertension; (4) valvular heart disease; (5) documented cardiomyopathy; (6) severe uncontrolled hypertension (systolic blood pressure > 180 mmHg and diastolic blood pressure >90 mmHg); (7) atrial fibrillation or malignant ventricular arrhythmia; (8) the intake of β-blockers; (9) ischaemic heart disease; (10) anaemia; (11) diabetes mellitus; (12) cancer; (13) chronic kidney disease; (14) recent chest or abdominal surgery; (15) recent exacerbation (during the last three months); (16) recent change (during the last three months) in medical therapy; (17) none of the subjects had non-invasive positive-pressure ventilation support or long-term ambulatory O2 therapy; (18) previous rehabilitation procedures.

Procedures

Pulmonary function testing and body plethysmography at rest

All subjects underwent preliminary clinical examination which included chest X-ray, spirometry, electrocardiogram,

and echocardiography. Those eligible for the study performed spirometry and exercise stress test. Both tests were performed on Vyntus, Carefusion, Germany following the guidelines. Spirometry was performed after bronchodilatation test – application of (400 μkg) of salbutamol. Following the ERS guidelines a post-bronchodilatation ratio of FEV1/FVC < 70% was assumed for the diagnosis of COPD.[16] Only patients with mild/moderate airway obstruction (FEV1 > 50%) were selected. The severity of COPD was staged according to the GOLD criteria.[17] Static lung volumes - residual volume (RV), total lung capacity (TLC), inspiratory lung volume and inspiratory capacity (IC) were measured by body plethysmography (Vyntus, body plethysmograph, CareFusion, Germany). Static hyperinflation has been assumed if FRC was above the upper lower limit of normal. Static lung volume measurements and interpretation of data has been performed following the guidelines.[18,19] The ECCS/ERS equations have been used for lung volume analysis.[20]

Exercise tests

Six-minute walk test (6-MWT)

Six-minute walking test was performed in accordance with ATS guidelines.[21] It was done on a separate day after the initial visit for study eligibility criteria and after the performance of the exercise stress test and stress echocardiography. Subjects were instructed and encouraged to walk through 30 m preliminary measured distance in a hospital corridor. SpO2, heart rate and arterial blood pressure were obtained before and during the recovery period.

Cardio-pulmonary exercise testing (CPET) – stress test protocol

All the patients underwent symptom limited incremental exercise stress test following the guidelines.[22] It was performed in an upright position on a bicycle after the clinical examination and spirometry. Subjects respired through an oro-nasal mask (Hans Rudolf 7450 SeriesV2™ Mask, CareFusion). Breath-by-breath cardiopulmonary data (Vyntus, CareFusion) were measured at rest, warm up and incremental exercise testing. Gas and flow sensors were calibrated before each test. Clinical monitoring of the patients included standard electrocardiography through the whole exercise test; manual blood pressure measurements, and heart rate recordings at the end of every stage.

A continuous ramp protocol was applied. After two minutes of unloaded pedaling (rest phase-0 W), a three minute warm-up phase (20 W) followed. The test phase included 20 W/2 min load increments. Patients were instructed to pedal with 60–65 rotations per minute. Patients' effort was considered to be maximal if two of the following criteria emerged: predicted maximal HR is achieved; predicted maximal work is achieved; V'E/V'O2 > 45, RER > 1.10, lactate level > 6 mmol L^{-1}, and pH drop > 0.06, as recommended by the ATS/ACCP.[23] Arterial blood (240 μL) was sampled at rest and at peak exercise and immediately analyzed using a blood gas analyzer/cooximeter (ABL700, Radiometer, France).

A breath-by-breath analysis was used for expiratory gas evaluation. Oxygen uptake ('VO2 (mL/kg/min)), carbon dioxide production ('VCO2 (L/min)), minute ventilation ('VE (L/min)) and end-tidal CO2 pressure (PetCO2 (mm Hg)) were collected continuously at rest and throughout the exercise test. Peak values of oxygen consumption and carbon dioxide production were presented by the highest 30-second average value, obtained during the last stage of the exercise test. Peak respiratory exchange ratio was the highest 30 s averaged value between'VO2 and' VCO2 during the last stage of the test. Ten-second averaged' VE and' VCO2 data, from the initiation of exercise to peak, were used to calculate the' VE/'VCO2 slope via least squares linear regression.[24] A dual approach for the measurement of the anaerobic threshold (AT) was applied. Both V-slope method and the ventilatory equivalents method for 'VO2 and' VCO2 were used. The modified Borg scale was applied for peak dyspnea and leg discomfort.

The maximum HR (MHR) was calculated (MHR = 220 – age). The target HR (THR) was set at 80% of MHR. A cut-off point of 12 beats was taken as an abnormal HRR.[4] The chronotropic response index was calculated.[2] The metabolic-chronotropic relationship (MCR) was calculated by Wilkoff's formula.[25] CI was assumed if MCR < 0.80. Breathing reserve (BR) was calculated as MVV - peak V'E/MVV x100 where MVV is maximal voluntary ventilation estimated as FEV1 multiplied by 35.

Dynamic hyperinflation (DH) during CPET

Changes in operational lung volumes were derived from measurements of dynamic inspiratory capacity (IC), assuming that total lung capacity (TLC) remained constant during exercise.[26,27] This has been found to be a reliable method of tracking acute changes in lung volumes.[26,27] IC was measured at the end of a steady-state resting baseline, at 2 min intervals during exercise, and at end exercise. End-expiratory lung volume (EELV) was calculated from IC maneuvers at rest, every 2 min during exercise and at peak exercise (Vyntus).In these maneuvers, after EELV was observed to be stable over 3–4 breaths, subjects were instructed to inspire maximally to TLC. For each measurement, EELV was calculated as resting TLC minus IC, using the plethysmographic TLC value. Dynamic IC (ICdyn) was defined as resting IC minus IC at peak exercise.[28] Dynamic hyperinflation (DH) was defined as a decrease in IC from rest of more than 150 mL or 4.5% pred at any time during exercise.[29]

Stress echocardiographic methods and CPET

After exercise cessation patients were put on a bed, near the ergometer. Stress echocardiography was performed in a supine position on a patient lying down1—2 min after peak exercise.

Routine structural and haemodynamic indices of both chambers at rest were measured following the guidelines.[30] The systolic function of the left ventricle was defined by Simpson's modified rule. The diastolic function of both ventricles was evaluated by the E/A ratio at rest.[31] As a more precise approach for diastolic dysfunction detection, tissue Doppler analysis was used. We used e' value as the average of medial and the lateral measurements for the mitral annulus. The peak of the average E/e' ratio > 15 was considered as a marker for stress induced left ventricular diastolic dysfunction.[30]

Statistical analysis

Descriptive statistics was used for demographic and clinical data presentation. The Kolmogorov-Smirnov test was used to explore the normality of distribution. Continuous variables were expressed as median and interquartile range when data was not normally distributed and with mean ± SD if normal distribution was observed. Categorical variables were presented as proportions. Data were compared between patients with GOLD I and GOLD II. An unpaired Student's t test was performed for normally distributed continuous variables. Mann–Whithney-U test was used in other cases. Categorical variables were compared by the $\chi 2$ test or the Fisher exact test. Univariate logistic regression analysis was applied to determine the ventilatory, echocardiographic and CPET parameters, associated with CI and abnormal HRR. Age, FEV1, body mass index, ICdyn, LV E/e' at rest, stress LV E/e' > 15 were taken as covariates in multivariate logistic regression analysis. In all cases a p value of less than 0.05 was considered significant as determined with SPSS® 13.0 Software (SPSS, Inc, Chicago, Ill) statistics.

Results

Echocardiographic, ventilatory and cardiovascular parameters of GOLD I and GOLD II patients at rest

Subjects enrolled in the study were Caucasians with a mean age of 62.9 ± 7.5. Subjects are divided into two groups based on the GOLD stages. The demographic and clinical data of the patients is presented in Table 1. Though not of statistical significance, there is a higher prevalence of DH, CI and abnormal HRR in moderate, as compared to mild, COPD patients. The echocardiographic characteristics (Table 2) were similar between the patients with GOLD I and GOLD II, but those with moderate disease demonstrated a higher incidence of stress LVDD. The ventilatory, cardiovascular and cardio-pulmonary exercise testing parameters of the two groups at peak exercise are given in Tables 3 and 4.

Cardio-vascular parameters of GOLD I and GOLD II patients at peak exercise

The patients with mild COPD achieved significantly higher peak HR and performed with higher MCR (Table 4). CI was met in seven (44%) of them with median heart rate reserve utilization – 78.53 (69.61–89.42) Abnormal heart rate recovery was established in 12 (75%) of the mild COPD.

The moderate COPD subjects demonstrated much lower MCR and chronotropic intolerance was met in 33 (65%) of these patients; they reached much lower peak HR and had a lower median heart rate reserve utilization 57.08 (41.74–81.12). Abnormal HRR was present in 40 (78%) of the GOLD II patients.

Ventilatory parameters of GOLD I and GOLD II patients at peak exercise

The analysis of the ventilatory parameters at peak exercise showed that mild COPD patients had higher minute ventilation and higher breathing reserve in comparison to those with moderate one. None of the patients in the studied group demonstrated static hyperinflation, but 32(46%) showed DH. There is a predominant prevalence of hyperinflators – 27(53)% among the patients with GOLD II in comparison to GOLD I – 5(31)% (Table 1).

Cardio-respiratory parameters of GOLD I and GOLD II patients at peak exercise

According to the objective ATS/ACCP criteria, exercise was considered maximal in all patients. The mild COPD patients achieved a higher peak load, higher V'O2 at peak and at anaerobic threshold. They performed with higher oxygen pulse and lower VE/VCO2 slope (Table 4). Dyspnea was the predominant limiting factor in 5 (31%) of GOLD I patients, but exhausted breathing reserve was detected in only 2 (13%) of them; in comparison 46 (90%) of the GOLD II patients complained of dyspnea and exhausted breathing reserve was the limiting factor in 18 (35%) of them. Leg fatigue was the reason for exercise cessation in 11 (69%) of GOLD I subjects, while 5 (10%) of the patients with GOLD II mntioned it (Table 4).

Association between ventilatory, cardio-pulmonary and echocardiographic parameters and AD

Table 5 demonstrates univariate logistic regression analysis for predictors of AD. Among the studied ventilatory, cardio-pulmonary and echocardiographic parameters only ICdyn and stress E/e' > 15 presented as predictors for both CI and abnormal HRR. Multivariate logistic regression analysis was also performed. The following covariates – age, BMI, FEV1, RV, RV/TLC, IC/TLC were taken in assumption. The multivariate regression analysis did not show association between the CI or abnormal HRR or any of the tested ventilatory, echocardiographic, CPET parameters or the BODE index Table 5. DH is the only independent predictor of CI and HRR in non-severe COPD patients.

Discussion

The major findings of our study are: (1) we demonstrate a high prevalence of abnormal HRR (76.5%) and CI (54.5%) in non-severe COPD patients who complain of exertional dyspnea and are free of overt cardiovascular diseases; (2) DH was the only independent predictor for AD parameters (CI and/or abnormal HRR); (3) the prevalence of HRR and CI are independent from the FEV1, LV cardiac function and the BODE index.

Chronotropic regulation of the cardiac function is responsible for HR response to exercise, HR recovery and HR variability.[32,33] AD was first reported in advanced COPD with respiratory failure and later in normoxemic patients.[7,34] Data regarding the prevalence of CI in COPD patients is even more controversial. The reasons for this are different criteria for CI definition, different COPD stages and study protocols.[35,36] Abnormal HRR has also been demonstrated in COPD independently of exercise intensity, peak and resting

Table 1 Anthropometric, clinical and autonomic dysfunction parameters of the patients with GOLD I and GOLD II.

	Patients with GOLD I (n, 16)	Patients with GOLD II (n, 51)	p-Value
Demographic data			
Age, year,	57.19 ± 6.13	61.89 ± 5.79	0.769*
Male, n	11	48	0.378‡
Duration of COPD	3.78 (2.86–5.12)	9.87 (6.73–11.58)	0.619†
Pack, years	25.98 (23.79–31.81)	29.19 (30.43 ± 38.87)	0.391†
Body mass index, kg/m^2	28.57 (25.49–32.65)	26.31 (20.86–33.48)	0.717†
Discharge medication			
LABA, n (%)	9 (56%)	18 (35%)	0.187‡
LAMA, n (%)	7 (44%)	12 (24%)	0.097‡
LABA + LAMA, n (%)	–	21 (41%)	0.319‡
Angiotensin converting enzyme Inhibitors, n (%)	11 (69%)	32 (63%)	0.762‡
Diuretics, n (%)	7 (44%)	29 (57%)	0.224‡
mMRC	1.5 (0.8–1.7)	1.8 (1.1–2.0)	0.721
6 MWT			
Distance walked, m	486.05 (451.38–499.21)	465.84 (436.73–481.19)	0.437‡
AD prevalence			
Chronotropic incompetence, n (%)	7 (44%)	33 (65%)	0.635‡
Abnormal HRR at 1 min, bpm	12 (75%)	40 (78%)	0.721‡
Dynamic hyperinflation			
IC_{dyn} > 150 mL	5 (31%)	27 (53%)	0.613‡
Stress LVDD			
Stress E/e' > 15	7 (44%)	39 (76%)	0.247‡

Abbreviations: GOLD – Global Initiative On Obstructive Lung Disease; LABA – long acting β2 agonists; LAMA – long acting muscarine antagonist; AD – autonomic dysfunction;HRR –heart rate recovery;bpm – beats per minute; $IC_{dyn} = IC_{rest}$-$IC_{peak;}$ LVDD – left ventricular diastolic dusfunction; LV – left ventricle; mMRC – modified Medical Research Council; 6MWT – 6 min walking test.

* Unpaired t test.

† Mann–Whitney U test.

‡ Chi square test.

Table 2 Echocardiographic parameters of the patients with GOLD I and GOLD II.

	Patients with GOLD I (n, 16)	Patients with GOLD II (n, 51)	p-Value
LV structural parameters			
LAVI, ml/m2	28.34 (26.58–31.29)	29.18 (27.61–32.83)	0.286*
Septum, mm	11.00 (10–13)	12.00 (11–13)	0.887*
PW, mm	11.05 (10.75–12)	12.00 (11–13)	0.921*
LV functional parameters at rest			
LVEF, %, Simpson	64.50 (61–66)	61.00 (57–66)	0.653*
E/A ratio	0.89 (0.75–1.25)	0.75 (0.65–0.90)	0.520*
E/e' aver ratio	6.46 (6.05–8.33)	6.97 (5.76–9.15)	0.456*
TR jet velocity, m/s	2.16 (1.98–2.31)	2.34 (2.04–2.42)	0.618*
Dec-E, ms	210.5 (201.32–211.29)	211.8 (212.61–221.83)	0.374*
IVRT, ms	89.6 (82.58–94.29)	91.2 (84.61–96.83)	0.895*
LV functional parameters after exercise stress test			
E/A ratio	1.25 (0.8–1.50)	1.53 (1.25–1.86)	0.043*
E/e' aver	8.07 (6.7–9.6)	17.43 (15.71–19.46)	0.036*
e' aver	10.03 (7.8–12.1)	7.8 (6.32–9.11)	0.012*
TR jet velocity, m/s	2.63 (2.22–2.92)	2.76 (2.50–3.12)	0.709*

Abbreviations: GOLD – Global Initiative On Obstructive Lung Disease; LV – left ventricular; LAVI – left atrium volume index; PW – posterior wall; LVEF – left ventricular ejection fraction; TR – tricispidal; Dec-decelaration; IVRT – isovolumic relaxation time.

* Mann–Whitney U test.

HR.[37,38] It was associated with increased risk of all-cause mortality especially among subjects with FEV1 < 50%.[39]

Our data supports previous findings. We observed both abnormal HRR (assuming parasympathetic dysfunction) and CI (an indicator of impaired sympathetic response) in COPD patients, which implies that both limbs of the autonomic cardiac regulation may be affected in mild/moderate COPD.

Table 3 Cardio-vascular and ventilatory parameters at rest and at peak exercise of the patients with GOLD I and GOLD II.

	Patients with GOLD I (n, 16)	Patients with GOLD II (n, 51)	p-Value
Cardio-vascular parameters			
HR at rest, bpm	78 (72–86)	89 (82–98)	0.502†
Peak HR, bpm	138 (110–143)	117 (103–121)	0.042†
HR max, %	86.7 (82.48—93.69)	77.84 (71.24—88.72)	0.014†
Heart rate reserve use, %	78.53 (69.61—89.42)	57.08 (41.74—81.12)	0.019†
MCR	0.73 (0.61—0.78)	0.54 (0.48—0.72)	0.032†
Post-exercise			
HRR at 1 min, bpm	11 (5—12)	10 (3—12)	0.982†
Blood pressure at rest			
Systolic blood pressure, mmHg	126.73 ± 8.41	128.87 ± 11.32	0.803‡
Diastolic blood pressure, mmHg	80.43 ± 4.14	82.31 ± 6.56	0.451‡
Blood pressure at peak exercise			
Systolic blood pressure, mmHg	161.81 ± 9.18	169.62 ± 10.84	0.219‡
Diastolic blood pressure, mmHg	85.37 ± 7.65	87.19 ± 7.32	0.398‡
Ventilatory parameters at rest			
FVC, l	4.68 (4.56—5.72)	3.72 (3.09–4.98)	0.021†
FVC, % pred	83.42%	84.11%	0.036†
FEV 1, l	3.06 (3.02—3.39)	2.32 (1.56—3.21)	0.033†
FEV1, % pred	81.02 (80.56—89.21)	66.18 (59.21—72.65)	0.040†
FEV1/FVC %	65.38(59.26—66.22)	62.30 (50.48—64.45)	0.046†
IC,l	3.19 (3.02—4.43)	2.87 (2.40—3.32)	0.216†
TLC,l	7.48 (6.72—8.09)	6.14 (5.59—7.28)	0.187†
TLC,% pred	91 (88–102)	94 (86–109)	0.709†
RV,l	2.38 (2.33—2.89)	2.81 (1.82—3.39)	0.283†
RV, % pred	92 (78–102)	94 (87–111)	0.421†
IC/TLC,%	45.62 (41.08—52.88)	41.57 (38.89—47.31)	0.179†
BODE index	1.05 (0.67—1.32)	2.64 (1.47—3.81)	0.016†
Ventilatory parameters at peak exercise			
$V_{t,}$ l	2.28 (1.79—3.34)	1.79 (1.57—2.02)	0.212†
VE, l/min	72.38 (60.87—84.58)	54.97 (46—62)	0.031†
BR, %	33.89 (21.68—39.83)	27.94 (20.87—33.38)	0.043†
EELV/ TLC, %	53 (50—59)	61 (54—70)	0.047†

Abbreviations: HFpEF – heart failure with preserved ejection fraction; HR – heart rate; HRR – heart rate recovery; CRI – chronotropic response index;bpm – beats per minute; MCR – metabolic-chrontropic relationship; IC – inspiratory capacity; TLC – total lung capacity; FRC – functional residual capacity; RV – residual volume; V_t – tidal volume; VE – minute ventilation; EELV – end expiratory lung volume; BR – breathing reserve.

† Mann–Whitney U test.

‡ Chi square test.

To the best of our knowledge, we first to describe AD in mild/moderate COPD patients who complain of exertional dyspnea and are free of overt cardio-vascular diseases. The design of the study takes into consideration the intake of medication that may influence the autonomic nervous system response. The intake of β-blockers was an exclusion criteria, and patients had β2-agonists and anticholinergic medication withdrawn 24 h before CPET. Though HRR and CI grew with the increase of BODE index and FEV1, none of the respiratory or CPET parameters correlated with them. Remembering that AD is associated with increased cardio-vascular mortality in the general population, HRR and CI may be useful independent markers for cardio-vascular risk stratification in COPD.[40]

Data regarding the relation between AD and FEV1 is controversial among studies. Chick et al., demonstrate delayed HRR in COPD patients, independently of their FEV1.[9] In contrast, Schedira et al., claim a higher prevalence of HRR as FEV1 decreases.[10] Hulo et al., find similar trends regarding CRI.[14] Gupta et al., describe that both HRR and CRI are becoming more prevalent in advanced COPD stages.[8] The controversies regarding the association between AD and COPD progression is probably due to the different study designs and protocol performance.

It is routinely assumed that AD is secondary to chronic sympathetic system overactivation.[41,42] Lung hyperinflation induces compression of the pulmonary vessels and the heart; the stroke volume and HR decrease.[43,44] It is therefore likely that lung hyperinflation in COPD blunts the cardiac chronotropic response and increases the sympathetic overactivation.[45,46] In normal subjects, sympathetic nerve activity is generally synchronized with the central inspiratory motor activity. The degree of lung volume inflation during inspiration activates pulmonary vagal afferents that in turn inhibits sympathetic nerve discharge.[47] The balance between these mechanisms determines the effect of respi-

Table 4 Cardio-pulmonary exercise testing parameters at peak exercise of the patients with GOLD I and GOLD II.

	Patients with GOLD I (n, 16)	Patients with GOLD II (n, 51)	p-Value
Peak Load, W	98 (100—140)	90 (80—100)	0.018†
Peak Load, % pred	89.4 (78.56—97.14)	83.18 (58.54—88.34)	0.044†
Exercise duration	525.8 + 136.8	484.3 + 129.1	0.312†
Peak VE, l/min	72.38 (60.87—84.58)	54.97 (46—62)	0.031†
Peak V'O2, ml/kg/min	23.81 (22.06—25.18)	18.46 (15.86—19.98)	0.028†
Peak V'O2, % pred	77.4 (62.8–80.9)	70.0 (55.0—80.0)	0.036†
V'O2 at AT, ml/kg/min	16.0 (15.0—16.6)	12.1 (10.6—14.1)	0.047†
O_2 pulse, ml/beat	11.80 (10.15—12.19)	10.90 (10.00—13.04)	0.076†
Peak RER	1.2 (1.1—1.3)	1.2 (1.1—1.3)	0.758†
VE/VCO2 slope	26.76 (24.02—30.58)	31.17 (27.15—34.68)	0.035†
Peak Sat%	95.00 (94.02—95.67)	94.9 (94.4—95.25)	0.089†
Borg dyspnea score	4 (3—7)	4 (3—5)	0.621†
Borg leg discomfort score	4 (4—7)	4 (3—5)	0.098†

Abbreviations: HFpEF- heart failure with preserved ejection fraction; RER – respiratory exchange ratio;AT anaerobic threshold; VE – minute ventilation;V'O2 – oxygen uptake.
‡Chi square test.
† Mann–Whitney U test.

Table 5 Univariate and multivariate logistic regression analysis between respiratory, cardio-pulmonary and echocardiographic parameters and AD parameters.

	CI			Abnormal HRR		
Univaraiate analysis	p-Value	OR	95% CI	p-Value	OR	95% CI
FEV 1, l/min	0.079	1.008	0.990—1.623	0.651	0.924	0.626—1.397
FRC,l	0.064	0.617	0.327—1.324	0.231	0.712	0.478—0.909
RV,l	0.073	1.562	0.998—2.864	0.097	1.023	0.096—1.241
IC/TLC,%	0.082	0.911	0.614—1.206	0.457	0.876	0.418—1.259
RV/TLC,%	0.301	0.921	0.689—1.223	0.793	0.658	0.395—0.968
Vt, l	0.090	1.002	0.831—1.948	0.151	0.918	0.549—1.328
VE, l	0.319	0.954	0.788—1.923	0.421	0.683	0.264—1.014
BR, %	0.065	0.958	0.779—1.561	0.059	0.917	0.692—1.218
ICdyn > 150 mL	0.036	18.9	4.521—32.418	0.027	19.3	3.804—27.613
Peak Load, W	0.071	1.107	0.604 -1.221	0.109	2.401	1.013 -5.411
Peak V'O2, mL/min/kg	0.623	0.769	0.412—0.965	0.398	1.023	0.587- 3.102
Peak O2 pulse mL/min/kg	0.126	0.989	0.674—1.003	0.812	0.911	0.634—2.121
VE/VCO2 slope	0.074	0.830	0.987—1.871	0.231	1.076	0.754—3.812
BODE index	0.152	0.013	0.000—0.938	0.473	0.811	0.432—3.089
LV E/e' at rest	0.078	0.542	0.423—0.897	0.067	1.241	0.932—1.968
Stress LV E/e' >15	0.023	3.673	1.418—6.924	0.048	2.037	1.806—5.473
Multivaraiate analysis						
ICdyn	0.042	11.21	3.862—27.851	0.039	12.06	2.653—19.087

Abbreviations: FEV1 – forced expiratory volume in 1 s; IC – inspiratory capacity; TLC – total lung capacity; RV – residual volume; FRC – functional residual capacity; V_t – tidal volume; VE – minute ventilation; BR – breathing reserve; EELV – end expiratory lung volume; HRR – heart rate recovery; CI – chronotropic incompetence.

ratory modulation on the autonomic nervous system, which in COPD patients is likely to become in favor of sympathetic activation.[45,46] Although hyperinflation correlates to sympathetic overactivation in COPD, in most of the studies its role has been overlooked. It is unclear whether DH is associated with AD. CI has been most commonly diagnosed when HR fails to reach 80%–85% of the HRmax.[33]

In order to objectively evaluate CI in COPD, we, for the first time, employed the Wilkoff formula – the relationship between HR and V'O2 during exercise.[25] The advantage of using the formula and the metabolic-chronotropic relationship as a marker for CI, is that MCR is adjusted for age, physical fitness, functional capacity and is unaffected by the exercise testing mode or protocol.[48,49] This metabolic-chronotropic relationship approach allowed us to define an association between AD and DH. We have shown that CI evaluated by the metabolic-chronotropic relationship is highly prevalent in a cohort of COPD patients. Adding evaluation of CI to standard pulmonary function parameters at rest and during incremental exercise let us determine DH as a

potential mechanism of attenuated HR response even in mild/moderate COPD. The results of the study are strengthened by the fact that none of the patients had LV systolic dysfunction.

In summary, abnormal HRR and CI are prevalent in non-severe COPD even in the absence of overt CV comorbidities. Neither abnormal HRR, nor CI are associated with the degree of airflow limitation (FEV1), LV cardiac function or the BODE index. DH is the only independent predictor for them. Evaluation of AD during incremental CPET unravels lung hyperinflation as a potential mechanism of attenuated HR response and diminished physical activity in non-severe COPD free of overt CV comorbidities. This multifaceted approach to dyspnea may facilitate the discrimination of its pathogenesis and improve its proper clinical management.

Our study confirms the versatile clinical presentation of a disease, affecting the lungs and going beyond them in its pathophysiological sequelae. The contemporary phenotype/endotype approach in COPD is undoubtedly demanding.[50] It is unfortunately obvious that the signs and symptoms, the CT and spirometry parameters, are insuffucient to cluster the patients and to facilitate the establishment of biomarkers, useful for precise pharmacotherapy.[50] The pathophysiological phenotypisation may propel the laborious ambition of understanding the heterogeneous multimorbidity of COPD.

Study limitations

The main limitations of this study are: (1) the relatively small sample size and the multiple tests may lead to false commission or omission results (type I/type II error); (2) coronary artery disease may not be excluded as neither invasive (coronary angiography), nor sophisticated imaging modalities (exercise single photon emission computed tomography (SPECT) – myocardial perfusion imaging (MPI)) were performed; (3) most of our patients had arterial hypertension, but it was controlled by optimal medical treatment, not clinically overt; (4) COPD patients experience enhanced pressure swings during the respiratory cycle and measurements were performed at the end of expiration, which may have influenced the results; (5) we do not have invasive measurement of sPAP; (6) measurements were acquired in the early recovery period (approximately 2 min) after symptom-limited exercise. The timeline of changes in the pulmonary and intrathoracic pressures during the brief time interval from peak exercise to their measurement in early recovery is not well known and underestimation is possible.

Conflicts of interest

The authors have no conflicts of interest to declare.

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References

1. Lauer MS, Okin PM, Larson MG, Evans JC, Levy D. Impaired heart rate response to graded exercise: prognostic implications of chronotropic incompetence in the Framingham heart study. Circulation. 1996;93:1520–6.
2. Lauer MS, Francis GS, Okin PM, Pashkow FJ, Snader CE, Marwick TH. Impaired chronotropic response to exercise stress testing as a predictor of mortality. JAMA. 1999;281:524–9.
3. Kannankeril PJ, Le FK, Kadish AH, Goldberger JJ. Parasympathetic effects on heart rate recovery after exercise. J Investig Med. 2004;52:394–401.
4. Savonen KP, Lakka AT, Laukkanen JA, Halonen PM, Rauramaa TH, Salonen JT, et al. Heart rate response during exercise test and cardiovascular mortality in middle-aged men. Eur Heart J. 2006;27(5):582–8.
5. Curtis BM, O'Keefe JH Jr. Autonomic tone as a cardiovascular risk factor: the dangers of chronic fight or flight. Mayo Clin Proc. 2002;77:45–54.
6. Jouven X, Empana JP, Schwartz PJ, Desnos M, Courbon D, Ducimetière P. Heart rate profile during exercise as a predictor of sudden death. N Engl J Med. 2005;352:1951–8.
7. Stewart AG, Waterhouse JC, Howard P. Cardiovascular autonomic nerve function in patients with hypoxaemic chronic obstructive pulmonary disease. Eur Respir J. 1991;4:1207–14.
8. Gupta M, Bansal V, Chhabra S. Abnormal heart rate recovery and chronotropic incompetence on exercise in chronic obstructive pulmonary disease. Chron Respir Dis. 2013;10(3):117–26.
9. Chick TW, Cagle TG, Vegas FA, Poliner JK, Murata GH. Recovery of gas exchange variables and heart rate after maximal exercise in COPD. Chest. 1990;97:276–9.
10. Seshadri N, Gildea TR, McCarthy K, Pothier C, Kavuru MS, Lauer MS. Association of an abnormal exercise heart rate recovery with pulmonary function abnormalities. Chest. 2004;125:1286–91.
11. Schwartz PJ, La Rovere MT, Vanoli E. Autonomic nervous system and sudden cardiac death: experimental basis and clinical observations for post-myocardial infarction risk stratification. Circulation. 1992;85:177–91.
12. Pelà G, Calzi M, Pinelli S, Andreoli Roberta, Sverzellati Nicola, Bertorelli Giuseppina, et al. Left ventricular structure and remodeling in patients with COPD. Int J Chron Obstruct Pulmon Dis. 2016;11:1015–22.
13. Phan TT, Shivu GN, Abozguia K, Davies C, Nassimizadeh M, Jimenez D, et al. Impaired heart rate recovery and chronotropic incompetence in patients with heart failure with preserved ejection fraction. Circ Heart Fail. 2010;3:29–34.
14. Hulo S, Inamo J, Dehon A, Rouzic O, Edme J, Neviere R. Chronotropic incompetence can limit exercise tolerance in COPD patients with lung hyperinflation. Int J Chron Obstruct Pulmon Dis. 2016;11:2553–61.
15. Celli BR, Cote CG, Marin JM, Casanova C, Montes de Oca M, Mendez RA, et al. The body-mass index, airflow obstruction, dyspnoea, and exercise capacity index in chronic obstructive pulmonary disease. New Engl J Med. 2004;350:1005–12.
16. Miller MR, Hankinson J, Brusasco V, Burgos F, Casaburi R, Coates A, et al. Standardisation of spirometry. Eur Respir J. 2005;26:319–38.
17. GOLD strategy for diagnosis, management and prevention of chronic obstructive pulmonary disease. Executive summary update 2018. Available at: www.goldcopd.org.

18. Wanger J, Clausen JL, Coates A, Pedersen OF, Brusasco V, Burgos F, et al. Standardisation of the measurement of lung volumes. Standardisation of the measurement of lung volumes. Eur Respir J. 2005;26:511–22.
19. Pellegrino R, Viegi G, Brusasco V, Crapo R, Burgos F, Casaburi R, et al. Interpretative strategies for lung function tests. European Respir J. 2005;26:948–68.
20. Kuster SP, Kuster D, Schindler MK, Rochat J, Braun L, Brandli O. Reference equations for lung function screening of healthy never-smoking adults aged 18–80 years. Eur Respir J. 2008;31:860–8.
21. ATS Committee on Proficiency Standards for Clinical Pulmonary Function Laboratories. ATS statement: guidelines for the six-minute walk test. Am J Respir Crit Care Med. 2002;166:111–7.
22. Guazzi M, Arena R, Halle M, Piepoli M, Myers J, Lavie C. 2016 focused update: clinical recommendations for cardiopulmonary exercise testing data assessment in specific patient populations. Circulation. 2016;133:694–711.
23. American Thoracic Society; American College of Chest Physicians. ATS/ACCP Statement on cardiopulmonary exercise testing. Am J Respir Crit Care Med. 2003;167(2):211–77.
24. Arena R, Myers J, Guazzi M. The clinical and research applications of aerobic capacity and ventilatory efficiency in heart failure: an evidence-based review. Heart Fail Rev. 2008;13:245–69.
25. Wilkoff BL, Miller RE. Exercise testing for chronotropic assessment. Cardiol Clin. 1992;10(November (4)):705–17.
26. Donnell DE, Lam M, Webb KA. Measurement of symptoms, lung hyperinflation, and endurance during exercise in chronic obstructive pulmonary disease. Am J Respir Crit Care Med. 1998;158:1557–65.
27. O'Donnell DE, Revill SM, Webb KA. Dynamic hyperinflation and exercise intolerance in chronic obstructive pulmonary disease. Am J Respir Crit Care Med. 2001;164:770–7.
28. Stubbing DG, Pengelly LD, Morse JLC, Jones NL. Pulmonary mechanics during exercise in normal males. J Appl Physiol. 1980;49:506–10.
29. Yan S, Kaminski D, Sliwinski P. Reliability of inspiratory capacity for estimating end-expiratory lung volume changes during exercise in patients with chronic obstructive pulmonary disease. Am J Respir Crit Care Med. 1997;156:55–9.
30. Nagueh SF, Smiseth OA, Appleton CP, Byrd BF, Dokainish H, Edvardsen T, et al. Recommendations for the Evaluation of left ventricular diastolic function by echocardiography: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. Eur Heart J Cardiovasc Imaging. 2016;17:1321–60.
31. Lancellotti P, Moura L, Pierard LA, Agricola E, Popescu BA, Tribouilloy C, et al. European Association of Echocardiography European Association of Echocardiography recommendations for the assessment of valvular regurgitation. Part 2: mitral and tricuspid regurgitation (native valve disease). Eur J Echocardiogr. 2010;11:307–32.
32. Fisher JP, Young CN, Fadel PJ. Autonomic adjustments to exercise in humans. Compr Physiol. 2015;5(2):475–512.
33. Coote JH. Recovery of heart rate following intense dynamic exercise. Exp Physiol. 2010;95(3):431–40.
34. Stein PK, Nelson P, Rottman JN, Howard D, Ward SM, Kleiger R, et al. Heart ratevariability reflects severity of COPD in PiZ alpha1-antitrypsin deficiency. Chest. 1998;113:327–33.
35. Jae SY, Fernhall B, Heffernan KS, Kang M, Lee MK, Choi YH, et al. Chronotropic response to exercise testing is associated with carotid atherosclerosis in healthy middle-aged men. Eur Heart J. 2006;27:954–9.
36. Brubaker PH, Kitzman DW. Chronotropic incompetence:causes, consequences, and management. Circulation. 2011;123:1010–20.
37. Nishime EO, Cole CR, Blackstone EH, Pashkow FJ, Lauer MS. Heart rate recovery and treadmill exercise score as predictors of mortality in patients referred for exercise ECG. JAMA. 2000;284:1392–8.
38. Chen MS, Blackstone EH, Pothier CE, Lauer MS. Heart rate recovery and impact of myocardial revascularization on long term mortality. Circulation. 2004;110:2851–7.
39. Lacasse M, Maltais F, Poirier P, Lacasse Y, Marquis K, Jobin J, et al. Post-exercise heart rate recovery and mortality in chronic obstructive pulmonary disease. Respir Med. 2005;99:877–86.
40. De Sutter J, Van de Veire N, Elegeert I. Chronotropic incompetence: are the carotid arteries to blame? Eur Heart J. 2006;27:897–8.
41. Bristow MR, Hershberger RE, Port JD, Gilbert EM, Sandoval A, Rasmussen R, et al. Beta-adrenergic pathways in nonfailing and failing human ventricular myocardium. Circulation. 1990;82 2 Suppl:I12–25.
42. Colucci WS, Ribeiro JP, Rocco MB, Quigg RJ, Creager MA, Marsh JD, et al. Impaired chronotropic response to exercise in patients with congestive heart failure. Role of postsynaptic beta-adrenergic desensitization. Circulation. 1989;80(2):314–23.
43. Breskovic T, Steinback CD, Salmanpour A, Shoemaker JK, Dujic Z. Recruitment pattern of sympathetic neurons during breath-holding at different lung volumes in apnea divers and controls. Auton Neurosci. 2011;164(1–2):74–81.
44. Dempsey JA, Sheel AW, St Croix CM, Morgan BJ. Respiratory influences on sympathetic vasomotor outflow in humans. Respir Physiol Neurobiol. 2002;130(1):3–20.
45. Andreas S, Anker SD, Scanlon PD, Somers VK. Neurohumoral activation as a link to systemic manifestations of chronic lung disease. Chest. 2005;128(5):3618–24.
46. van Gestel AJ, Kohler M, Clarenbach CF. Sympathetic overactivity and cardiovascular disease in patients with chronic obstructive pulmonary disease (COPD). Discov Med. 2012;14(79):359–68.
47. Jouett NP, Watenpaugh DE, Dunlap ME, Smith ML. Interactive effects of hypoxia, hypercapnia and lung volume on sympathetic nerve activity in humans. Exp Physiol. 2015;100(9):1018–29.
48. Da Cunha FA, Farinatti Pde T, Midgley AW. Methodological and practical application issues in exercise prescription using the heart rate reserve and oxygen uptake reserve methods. J Sci Med Sport. 2011;14(1):46–57.
49. Mezzani A, Corrà U, Giordano A, Cafagna M, Adriano EP, Giannuzzi P. Unreliability of the %VO2 reserve versus %heart rate reserve relationship for aerobic effort relative intensity assessment in chronic heart failure patients on or off beta-blocking therapy. Eur J Cardiovasc Prev Rehabil. 2007;14(1):92–8.
50. Corlateanua A, Mendez Y, Wang Y, Jesus R, Garnica A, Botnaru V, et al. Chronic obstructive pulmonary disease andphenotypes: a state-of-the-art. Pulmonology. 2020;26(2):95–100.

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ORIGINAL ARTICLE

Diagnostic accuracy of TB-LAMP assay in patients with pulmonary tuberculosis–a case-control study in northern India

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KEYWORDS
Tuberculosis;
Diagnosis;
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Abstract
Setting: A tertiary care hospital in North India.
Objective: Tuberculosis (TB) remains a major public health problem in developing countries. The diagnosis of tuberculosis is still challenging in primary care settings in endemic countries like India. WHO has endorsed loop mediated isothermal amplification assay (LAMP) for TB as a replacement for smear microscopy for peripheral settings, however, more data is required to establish the specificity of this modality for the diagnosis of TB. In this study we aim to determine the diagnostic accuracy of the TB-LAMP assay in pulmonary tuberculosis.
Design: A total of 236 patients (117 cases suspected of TB and 119 patients with non-TB pulmonary disease) were enrolled between February to July, 2018. Microbiological workups consisting of mycobacterial smear microscopy, culture, Xpert MTB/Rif and TB-LAMP were performed.
Results: From 236 samples, 18 (7.6%) were excluded from the study. TB-LAMP and Xpert MTB/RIF were positive in 46 (21.1%) and 49 (22.5%) of the samples, respectively. The sensitivity of Xpert MTB/RIF and TB-LAMP, when culture was taken as a reference standard, was 90% (95%CI: 78.2-96.7) and 82% (95%CI: 68.6—91.4), respectively. The specificity, positive predictive value (PPV), and negative predictive value (NPV) of TB-LAMP assay were 96.8% (95%CI: 92.8—98.9), 89.1% (95%CI: 77.4—95.2), and 94.4% (95%CI: 90.4—96.5), respectively.
Conclusion: The TB-LAMP assay showed a good specificity and sensitivity for detection of*M. tuberculosis* in adults, however, for programmatic implementation, more studies are required to be conducted at peripheral level healthcare settings.

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Introduction

Tuberculosis (TB) poses a colossal challenge for national tuberculosis control programmes owing to high morbidity and mortality. In 2018, WHO projected a total of ten million new cases, with a mortality of 1.5 million cases across the globe. Thirty endemic countries account for 87% of the TB cases, of which India contributes a total of 2,155,894 notified cases, in spite of stringent elimination programs in place.[1] One of the major contributing factor is high prevalence of affected but hidden cases. Many cases remain undiagnosed due to the inaccessibility to diagnostic tests at peripheral level or due to poor performance of the available tests. The gold standard for the diagnosis of TB remains the culture. However, it is labor-intensive and the results are available quite late. Moreover, the primary test in most of the settings is smear microscopy with low reproducibility and sensitivity as low as 50–70%.[2,3] Nucleic acid amplification tests (NAATs) are useful for rapid diagnosis of the cases, however, the cost and pre-requisite of sophisticated laboratory settings are the main impediments in the peripheral and rural areas. To address this limitation, World Health Organization (WHO) has endorsed cartridge based NAATs (CB-NAAT), the Xpert MTB/RIF assay, for uniformity in diagnosis and detection of drug resistance to rifampicin.[4] Xpert MTB/RIF assay is easy with a shorter turnaround time (TAT). The overall sensitivity and specificity of Xpert MTB/RIF assay in respiratory samples has been reported to be 88% and 98%, respectively. However, the sensitivity and specificity for diagnosis of extra-pulmonary cases was lower but not inferior, when compared to culture as a reference standard. Recently, the newer generation Xpert, i.e. Xpert MTB/RIF Ultra has also been endorsed by WHO with a higher sensitivity (3–5% more than Xpert MTB/RIF assay) but decreased specificity.[5]

Another molecular assay endorsed by WHO as an alternative to smear microscopy for diagnosis of pulmonary TB in symptomatic adults is Tuberculosis- Loop Mediated Isothermal Amplification (TB-LAMP) assay, which has shown a pooled sensitivity of 78% in clinical validation by WHO-FIND. The endorsement of TB-LAMP is another significant step by WHO for accomplishing the goal of ''End TB'' strategy. TB-LAMP is comparatively easier, having strand-displacement along with replication activity, does not require stringent temperature conditions for the reaction, is less labor-intensive, has a higher specificity and sensitivity compared to smear microscopy of 69-100% and has a shorter TAT of one hour.[6] The validation has been based upon studies conducted in limited settings, and needs more extensive research before implementation of this test at the peripheral level. All the available studies have been conducted on the TB suspect samples without including any control arm. Hence, the present study was designed to determine the diagnostic accuracy of TB-LAMP assay in TB suspects as well as in patients with pulmonary diseases other than TB.

Methodology

Study setting

The study was carried out in the Department of Medical Microbiology, Postgraduate Institute of Medical education and Research (PGIMER), Chandigarh, India. The laboratory is accredited by NABL:ISO and Central TB Division, India, for routine diagnosis and drug resistance of tuberculosis. The sputum samples from pulmonary diseases other than TB were collected from patients attending the outpatient department of Pulmonary Medicine, PGIMER, Chandigarh. The sputum samples of suspected TB cases were received in the laboratory from different directly observed treatment, short-course (DOTS) centers under Revised National Tuberculosis Control Program (RNTCP) program in Chandigarh. Ethical clearance was taken from Institute Ethics Committee, PGIMER, Chandigarh.

Clinical samples

A total of 236 patients were enrolled from February to July, 2018. Of the 236 patients, 117 were suspected TB cases and 119 patients had pulmonary disease other than TB. One sputum sample from each patient was taken and transferred to the laboratory immediately. Microbiological investigations smear microscopy, mycobacterial culture, Xpert MTB/RIF and TB-LAMP were performed. The demographic details of all the patients were noted from RNTCP request forms and the patient's proforma.

Smear and mycobacterial culture

The smears were prepared directly from the sputum samples. Ziehl-Neelsen(ZN) staining was performed and the smears were reported as per RNTCP guidelines. Subsequently, the samples were decontaminated by NALC-NaOH method and 500 μL of the processed sample was inoculated in the mycobacterial growth indicator tube (MGIT).[7] The tubes were placed in the MGIT 960 instrument (BD, USA) and incubated for 42 days. The positive tubes were confirmed for *M. tuberculosis* using SD bioline MPT64 Ag kit (Abbott, USA).

Xpert MTB/RIF assay

The Xpert MTB/RIF assay (Cepheid, USA) was performed as per the manufacturer's instructions. The buffer was mixed in double amount with the sample and incubated at room temperature for 15 min. Subsequently, the sample was transferred into cartridge and placed in Xpert machine. The results were interpreted either as *M. tuberculosis* detected with or without rifampicin resistance, or no target detected.[8]

TB-LAMP assay

The TB LAMP assay was performed using Loopamp MTBC Detection Kit (Eiken, Japan). The sample (60 μL) was transferred into the heating tube and incubated at 95 °C for 5 min. Then, the heating tube was screw-capped with absorbent tube, mixed and transferred into reaction mix tubes. The reaction mix tube was incubated at 65 °C and the final results, in terms of fluorescence, were interpreted.[9]

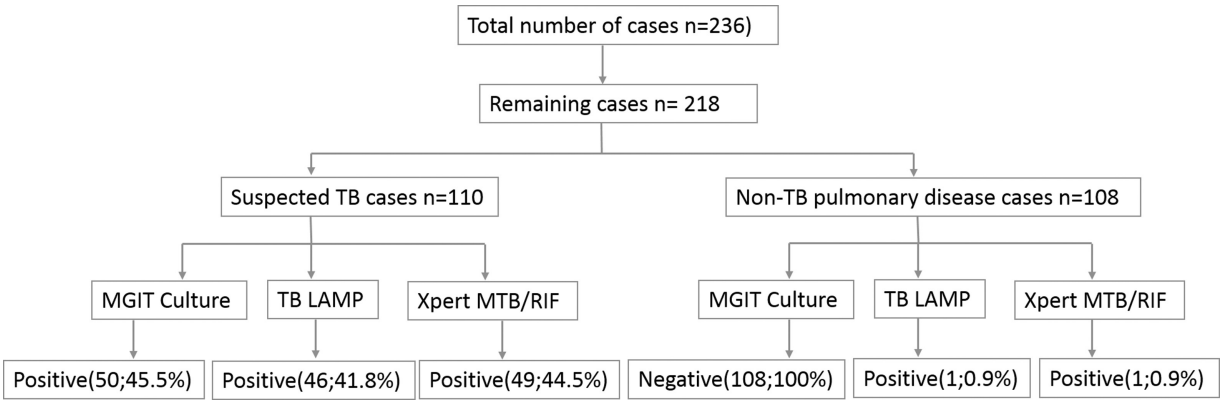

Figure 1 Workflow of the study.

Sensitivity and specificity calculation

The *M. tuberculosis* culture was taken as a reference standard for the calculation of sensitivity, specificity, positive predicted value (PPV) and negative predicted value (NPV). All the parameters were calculated using Medcalc online software.

Results

Of the 236 samples, 17 samples showed culture contamination and one sample had invalid Xpert result, so 18 (7.6%) samples were excluded from the study. Of the remaining 218 samples, there were 128 (58.7%) males and 90 (41.3%) females. The age of the patients was between 18 to 80 years, with a median of 47 years. A total of 110 (50.4%) samples were from suspected TB patients and 108 (49.5%) from pulmonary disease other than TB. The patients of pulmonary disease other than TB included patients with carcinoma (30, 13.7%), allergic bronchopulmonary aspergillosis (8, 3.7%), asthma (11, 5%), bronchiectasis (13, 5.9%), chronic obstructive pulmonary disease (12, 5.5%), chronic pulmonary aspergillosis (9, 4.1%), cough and chest pain (7, 3.2%), shortness of breath (4.1%), and others (9, 4.1%; allergic rhinitis, hilar mass, primary lung mass, metastatic lesions, fibrosis, sarcoidosis, GERD, pneumothorax, and seasonal allergy). In patients with suspected TB, the culture, Xpert MTB/RIF assay and TB-LAMP was positive in 50 (22.9%), 49 (22.5%) and 46 (21.1%) samples, respectively (Fig. 1).

Diagnostic accuracy of TB LAMP and Xpert MTB/RIF

The sensitivity of TB-LAMP, Xpert MTB/RIF and smear was noted to be 82% (95% CI: 68.6-91.4%), 90% (95% CI: 78.2-96.7%) and 64% (95% CI: 49.2-79.1%), respectively, using culture as a reference standard. The specificity, PPV and NPV of TB-LAMP assay was found to be 96.8% (95% CI: 92.8-98.9%), 89.1% (95% CI: 77.4-95.2%) and 94.4% (95% CI: 90.4-96.5%), respectively. On the contrary, the specificity, PPV and NPV of Xpert MTB/RIF was noted to be 97.5% (95% CI: 93.6-99.3%), 91.8% (95% CI: 80.9-96.5%) and 96.8% (95% CI: 93.1- 98.6%), respectively, which is more than TB-LAMP. However, the sensitivity and specificity of TB LAMP and Xpert MTB/RIF in smear negative cases was 55.6% (95% CI: 30.8-78.5%) and 75% (95% CI: 50.9-91.3%) or 97.5% (95% CI: 93.8-99.3%) and 98.1% (95% CI: 94.5-99.6%), respectively. The specificity and NPV of TB-LAMP assay in non-TB cases was found to be 99.1% (95% CI: 94.9- 99.9%) and 100%, respectively (Table 1). The area under curve (AUC) of receiver operating characteristic (ROC) curve for TB-LAMP and Xpert MTB/RIF was 0.895 and 0.938, respectively (Fig. 2).

Discussion

WHO has endorsed TB-LAMP to strengthen diagnostic testing in peripheral health centers to achieve the WHO ''End TB'' strategy.[10,11] The Xpert MTB/RIF assay has been evaluated in many studies across the globe, however, the same does not hold true for TB-LAMP assay, for which very limited studies have been conducted in different geographical regions. In this study, we appraised the sensitivity and specificity of TB LAMP assay in suspected TB as well as non-TB pulmonary disease cases.

In the present study, the overall specificity of TB-LAMP assay was noted to be 96.8%, which is in concordance to other studies that have shown specificity of 96.7–98.7%.[12,13] The overall specificity of Xpert MTB/RIF assay in our study was 97.5%, which was higher than the TB-LAMP assay. This is in concordance with other studies, most of which have revealed a specificity of 97.2–99.3%. The sensitivity of TB LAMP assay was 82%, which is comparable to other studies in which have shown sensitivity of 92–100% respectively.[9,14–16] On the contrary, the studies conducted in China, India and Vietnam revealed sensitivity as low as 70.6–79.6%.[12,17] The overall sensitivity of the TB-LAMP assay was higher than the smear (82% vs. 64%). In smear negative and smear positive TB cases, it was 55.6% and 96.8%, respectively. However, the previous studies had also shown the similar sensitivities in smear negative cases, e.g. Kim et al. and Pham et al. (46.6–58.8%).[12,15] The indeterminate results with TB-LAMP were exceptionally rare[18] and the same was noted in the present study.

The sensitivity of Xpert MTB/RIF assay was 90% in our study, which was better than TB-LAMP assay. This could be because of the larger volume of sample used in Xpert MTB/RIF as compared to TB-LAMP. A meta-analysis to ana-

Table 1 Diagnostic accuracy of TB LAMP and Xpert MTB/RIF assay in different groups.

Patient group	Diagnostic assay	Sensitivity % (95% CI)	Specificity% (95% CI)	Positive predictive value % (95% CI)	Negative predictive value % (95% CI)
Overall	Xpert MTB/RIF	90 (78.2—96.7)	97.5 (93.6—99.3)	91.8 (80.9—96.5)	96.8 (93.1—98.6)
	TB LAMP	82 (68.6—91.4)	96.8 (92.8—98.9	89.1 (77.4—95.2)	94.4(90.4—96.5)
	Smear	64 (49.2—79.1)	99.4 (96.5—99.9)	96.9 (81.8—99.6)	89.7(85.8—92.7)
Smear Negative cases	Xpert MTB/RIF	75 (50.9—91.3)	98.1 (94.5—99.6)	83.3(61.3—94)	96.8(93.5—98.5)
	TB LAMP	55.6 (30.8—78.5)	97.5(93.8—99.3)	71.4(46.6—87.8)	95.2(92.1—97.1)
Smear Positive cases	Xpert MTB/RIF	100 (88.8-100)	98.2 (93.5-99.8)	93.9 (79.7-98.4)	100
	TB LAMP	96.8 (83.3—99.9)	96.4 (91—99)	88.2 (74.1—95.2)	99.1 (93.9—99.9)
Non-TB pulmonary disease cases	Xpert MTB/RIF	–	99.1% (94.9–99.9%)	–	100
	TB LAMP	–	99.1% (94.9-99.9%)	–	100

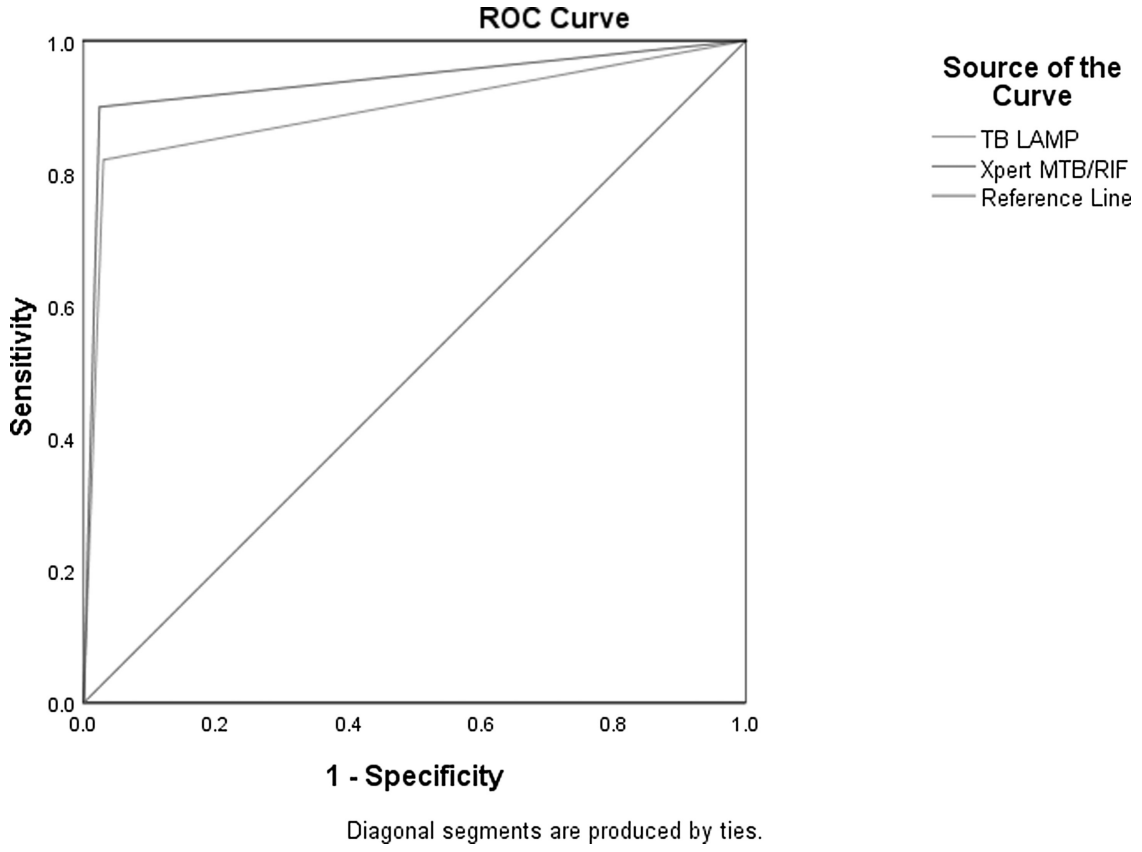

Figure 2 Receiver Operating Characteristic (ROC) curve for detection of *M tuberculosis* by TB LAMP and Xpert MTB/RIF assay.

lyze the diagnostic accuracy of TB-LAMP has concluded that TB-LAMP has moderate sensitivity (77.7%) and high level of specificity (98.1%). The pooled sensitivity and specificity of TB-LAMP is comparable to Xpert MTB/RIF in all, except HIV-positive individuals, in whom the pooled sensitivity falls down to 63.8%. This can be attributed to a high proportion of smear-negativity amongst HIV-positive individuals. The major advantage of Xpert MTB/RIF over TB-LAMP is the detection of rifampicin resistance along with *M. tuberculosis*. The rifampicin resistance detection by Xpert MTB/RIF helps in better management of RR/MDR-TB where the prevalence of rifampicin resistance is high. However, in this study we did not find any resistance to rifampicin by Xpert MTB/RIF and phenotypic DST was also not performed.

We have included sputum samples from pulmonary illness other than TB to evaluate the specificity of this assay which is major strength of this study. Of these 108 samples, only one was detected as false positive by both TB-LAMP and Xpert MTB/RIF assays. The specificity of this assay in pulmonary disease other than TB has not been previously

explored to the best of our knowledge. Other than routine healthcare settings, this assay could also prove to be a useful tool for screening TB in special high-risk settings such as prisons, immigrants and close contact settings. Linhas R et al. discussed the problem of TB screening in immigrants in Portugal and highlighted the importance of the rapid, accurate and economic test for TB screening.[18] The limitation of this study lies in small number of samples in both groups (suspected TB and non-TB pulmonary disease) and that phenotypic DST was not performed for rifampicin.

The present study concluded that TB-LAMP could be used as an ancillary test to sputum microscopy or as a follow-up alternative to the same, since it fulfills the WHO criteria of an alternative test.[19] Though the sensitivity and specificity in our study was inferior to the Xpert MTB/RIF, there was no significant difference for the same, as has also been reported earlier.[20] Moreover, the end-user profile is different for both the tests and the cost effectiveness study conducted by WHO determined low cost per test for TB-LAMP with low operational, budgetary and incremental costs,[6] thus facilitating its use as an alternative highly sensitive test in peripheral settings. Therefore, TB-LAMP is a viable alternative to smear microscopy in resource-poor settings with insufficient infrastructure for Xpert MTB/RIF.

Conflicts of interest

The authors have no conflicts of interest to declare.

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References

1. World Health Organization. Tuberculosis. www.who.int; 2019 https://www.who.int/tb/publications/global_report/en/
2. Pai M, Behr MA, Dowdy D, Dheda K, Divangahi M, Boehme CC, et al. Tuberculosis. Nat Rev Dis Primers. 2016;27:16076.
3. Steingart KR, Henry M, Ng V, Hopewell PC, Ramsay A, Cunningham J, et al. Fluorescence versus conventional smear microscopy for tuberculosis: a systematic review. Lancet Infect Dis. 2006;6:570–81.
4. World Health Organization, Tuberculosis. Using the Xpert MTB/RIF assay to detect pulmonary and extrapulmonary tuberculosis and rifampicin resistance in adults and children; 2013 https://www.who.int/tb/publications/xpert-mtb-rif-assay-diagnosis-meeting-report/en/
5. World Health Organization, Tuberculosis. Next-generation Xpert® MTB/RIF Ultra assay recommended by WHO; 2017. WHO, 2017 https://www.who.int/tb/featuresarchive/Xpert-Ultra/en/
6. World Health Organization, Tuberculosis. The use of loop-mediated isothermal amplification (TB-LAMP) for the diagnosis of pulmonary tuberculosis; 2016 https://apps.who.int/iris/bitstream/handle/10665/249154/9789241511186-eng.pdf?sequence=1
7. Levin W, Brandon GR, McMillen S. The culture method of laboratory diagnosis of tuberculosis. Am J Public Health Nations Health. 1950;40:1305–10.
8. Boehme CC, Nabeta P, Hillemann D, Nicol MP, Shenai S, Krapp F, et al. Rapid molecular detection of tuberculosis and Rifampin resistance. N Engl J Med. 2010;363:1005–15.
9. Yadav R, Sharma N, Khaneja R, Agarwal P, Kanga A, Behera D, et al. Evaluation of the TB-LAMP assay for the rapid diagnosis of pulmonary tuberculosis in Northern India. Int J Tuberc Lung Dis. 2017;21(10):1150–3.
10. Furin J, Cox H, Pai M. Tuberculosis. Lancet. 2019;393(10181):1642–56.
11. WHO End TB Strategy. World Health Organization; https://www.who.int/tb/post2015_ strategy/en/.
12. Pham TH, Peter J, Mello FCQ, Parraga T, Lan NTN, Nabeta P, et al. Performance of the TB-LAMP diagnostic assay in reference laboratories: results from a multicentre study. Int J Infect Dis. 2018;68:44–9.
13. Sharma G, Tewari R, Dhatwalia SK, Yadav R, Behera D, Sethi S. A loop-mediated isothermal amplification assay for the diagnosis of pulmonary tuberculosis. Lett Appl Microbiol. 2019;68(3):219–25.
14. Bojang AL, Mendy FS, Tientcheu LD, Out J, Antonio M, Kampmann B, et al. Comparison of TB-LAMP, GeneXpert MTB/RIF and culture for diagnosis of pulmonary tuberculosis in The Gambia. J Infect. 2016;72(3):332–7.
15. Kim C, Cho EA, Shin DM, Choi SW, Shin SY. Comparative evaluation of the loop-mediated isothermal amplification assay for detecting pulmonary tuberculosis. Ann Lab Med. 2018;38(2):119–24.
16. N'guessan Kouassi K, Riccardo A, Dutoziet Christian C, André G, Férilaha C, Hortense S-A, et al. Genotyping of mutations detected with GeneXpert. Int J Mycobacteriol. 2016;5(2):142–7.
17. Ou X, Li Q, Xia H, Pang Y, Wang S, Zhaoet B, et al. Diagnostic accuracy of the PURE-LAMP test for pulmonary tuberculosis at the county-level laboratory in China. PLoS One. 2014;9(5), e94544 2014.
18. Linhas R, Oliveira O, Meireles P, Oliveira P, de Melo MB, Lourenço J, et al. Immigrants' access to health care: problems identified in a high-risk tuberculosis population. Pulmonology. 2019;25(1):32–9.
19. Shete PB, Farr K, Strnad L, Gray MC, Cattamanchi A. Diagnostic accuracy of TB-LAMP for pulmonary tuberculosis: a systematic review and meta-analysis. BMC Infect Dis. 2019;19(1):268.
20. World Health Organization. WHO. High-priority target product profiles for new tuberculosis diagnostics; 2014 https://www.who.int/tb/publications/tpp_report/en/

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ORIGINAL ARTICLE

Predicting lung nodules malignancy

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Abstract
Background: It is critical to developing an accurate method for differentiating between malignant and benign solitary pulmonary nodules. This study aimed was to establish a predicting model of lung nodules malignancy in a real-world setting.
Methods: The authors retrospectively analysed the clinical and computed tomography (CT) data of 121 patients with lung nodules, submitted to percutaneous CT-guided transthoracic biopsy, between 2014 and 2015. Multiple logistic regression was used to screen independent predictors for malignancy and to establish a clinical prediction model to evaluate the probability of malignancy.
Results: From a total of 121 patients, 75 (62%) were men and with a mean age of 64.7 years old. Multivariate logistic regression analysis identified six independent predictors of malignancy: age, gender, smoking status, current extra-pulmonary cancer, air bronchogram and nodule size ($p<0.05$). The area under the curve (AUC) was 0.8573.
Conclusions: The prediction model established in this study can be used to assess the probability of malignancy in the Portuguese population, thereby providing help for the diagnosis of lung nodules and the selection of follow-up interventions.

Introduction

Pulmonary nodules are a common finding on chest computed tomography (CT) and present as a challenge to clinicians. The majority are not malignant and usually have no clinical significance.[1–4] Fleischner guidelines[5] pertain to the follow-up and management of pulmonary nodules detected

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incidentally on chest CT. However, their malignant potential is mostly unknown at the time of detection.

Lung cancer is the most significant cause of death from cancer in the world. It is usually diagnosed in an advanced stage, resulting in a 5-year survival rate of 17.4%.[6] Its complexity from both histopathological and biological perspectives, perhaps having multiple preneoplastic pathways, poses an enormous challenge for an early diagnosis.[7] Thus, to reduce the mortality rate, the ideal would be developing strategies to diagnose lesions in the pre-invasive state. In 2011, the National Lung Screening Trial (NLST) showed a reduction in lung cancer mortality of 26% in men, 39–61% in women and 20% globally, for annual screening with low-dose CT scanning, compared with no screening or with chest radiography.[1] The recent publication of the Dutch-Belgian Randomized Lung Cancer Screening (Nelson) Trial, a population-based randomized controlled trial, showed that volume low-dose CT screening had lower lung cancer mortality than no screening, in high-risk patients.[8] However, the main challenge in CT screening is the high prevalence of pulmonary nodules and the relatively low incidence of lung cancer.[1–5] Several studies have shown that the most important predictors of malignancy for pulmonary nodules include size, appearance and growth rate (volume doubling time <400 days), patient's age, smoking and extrapulmonary tumour history.[9–15] Although those factors are supported by clinical experience, they do not seem to be enough to choose a course of action.[5,10,12,16,17]

Sometimes the question is raised: what is the best clinical guidance after identification of a pulmonary nodule? Percutaneous CT-guided transthoracic biopsy is frequently used for the diagnosis of lung nodules, particularly for peripheral or bronchoscopic inaccessible lung lesions.[18,19] Although an effective approach in experienced hands, it has limitations with smaller nodules and ground-glass lesions and, a considerable complication rate (pneumothorax range for 4–40% and 1–7% for haemoptysis).[18,20–22] On the other hand, choosing a follow-up strategy increases stress and exposure to radiation from numerous CT scans and allows possibly malignant nodules to evolve, delaying cancer diagnosis and treatment.[9,23] Consequently, clinicians must carefully weigh up whether the risk of cancer of a lung nodule justifies the potential harm of a biopsy.

Objectives

In this paper, the authors aimed to define which clinical and radiological characteristics could suggest malignancy, in a real-world setting, and therefore better correlate with the decision for a biopsy. Finally, the authors built a model to assist in the decision-making process for an invasive diagnosis.

Methods

Study population

Study cohort included patients who underwent percutaneous CT-guided transthoracic biopsy for one year, at Centro Hospitalar Universitário de São João (Porto, Portugal). Only cases where the biopsy target was less than 3 cm diameters in initial CT evaluation were included. Patients with a clinical record of interstitial lung disease were excluded. Written informed consent, before the chest CT biopsy, was obtained from all patients.

The study was approved by Centro Hospitalar Universitário de São João Ethics Committee.

Data collection

Clinical and socio-demographic data were collected from patients' electronic records. The data included age, sex, smoking status (non-smoker, active smoker and former smoker), symptoms at detection and history of tuberculosis. Cancer history was also considered, divided into pulmonary and extra-pulmonary tumours, and subsequently into current or previous illness. Other medical backgrounds, like chronic obstructive pulmonary disease, bronchiectasis, obstructive sleep apnoea syndrome, pulmonary aspergillosis, was also recorded. Later, the histologic result was documented.

Chest CT analysis

Chest CT-scan were obtained from the hospital's electronic records and analyzed by two Radiology assistants with more than 15 years of experience. The biopsy CT-scan was assessed, retrospectively, for tumour characteristics by a first Radiologist, who was blinded to the clinical and histological findings. The cases were later reviewed and approved by a second Radiologist. Radiological features recorded included the tumour shape (round, ovoid, bilobed or irregular), sphericity and attenuation (pure ground glass, semisolid or solid), tumour location (central or peripheral), margins (smooth, spiculated or lobulated), presence of internal air bronchogram, cavitation, single nodule and pleural contact. The size was obtained from the first CT available. Measurements were made to the nearest centimetre using manually placed computer electronic callipers considering the nodule biggest axis.

Statistical analysis

Categorical variables are presented as frequencies and percentages and were compared with the use of the Chi-square test. Continuous variables are presented as means and standard deviations and were compared with the use of the *t*-test. The interaction of these variables with the biopsy result was expressed as risk ratios (RRs). A logistic regression model was constructed to assess the association between the outcome (benign/malignant) and the pre-biopsy characteristics. Individual malignancy probability could be obtained by getting the sum of the products of the coefficient of each independent variable ($\beta_1 - n$) included in the logistic model and their code ($X_1 - n$) – according to the table score – and replacing it in the formula:

$$\frac{1}{1 + e^{-}(Constant - \sum(\beta_1 X_1 + \beta_2 X_2 + \cdots \beta_n X_n))}$$

Odds ratio (OR) and 95% confidence interval (CI) were calculated for the model variables. The significance level was set at $p < 0.05$ (two-sided). IBM SPSS Statistics 24, STATA Statistical Data Analysis 9.0 and BiostatXL MIX 2.0 were used to compute all these estimates.

Results

During the study period, 121 patients were eligible. The mean age was 64.7 ± 12.3 years, and 75 (62%) of the patients were male. The majority (84.5%) were discovered by accident. Table 1 shows the patients' demographic and clinical characteristics. Malignant nodules were observed in sixty-four (53%) patients. The majority of those were lung adenocarcinoma ($n = 35$, 54.7%). Other malignancies included carcinoid tumour ($n = 11$, 17.2%), extrathoracic tumours ($n = 7$, 11%), squamous-cell carcinoma ($n = 4$, 6.3%), large-cell carcinoma ($n = 2$, 3.1%), small-cell lung carcinoma ($n = 2$, 3.1%), lymphoma ($n = 2$, 3.1%) and adenosquamous carcinoma ($n = 1$, 1.6%). Twenty-three (40.4%) patients, with benign lung nodules, had no specific diagnosis (histology with no signs of malignancy), and the remainder included chondroid hamartoma ($n = 13$, 22.8%), benign neoplasms ($n = 11$, 19.1%) and infectious process ($n = 10$, 17.5%).

Patients with malignant nodules were significantly older than patients with benign nodules ($\geq$70 years: 64.6% vs. 35.4%, $p = 0.037$, RR = 1.43) and more likely to have current extra-pulmonary cancer (81.3% vs. 18.8%; $p = 0.015$, RR = 1.64); however, these association were not identified in the previous history of extra-pulmonary cancer. Radiological characteristics were identified with significant associations between malignancy and a central location ($p = 0.008$, RR = 1.85), lobulated ($p = 0.012$, RR = 1.54) and spiculated ($p = 0.025$, RR = 1.48) margins, air bronchogram ($p = 0.014$, RR = 1.73), pleural contact ($p = 0.003$, RR = 1.68), size ($p < 0.001$). Neither the shape, predominant margins, calcification, single nodule nor attenuation differences were statistically associated with a malignant nodule. Table 1 and Fig. 2 show the interaction between some of the patients' clinical and radiological characteristics and the final biopsy result.

Clinical and radiological characteristics were then used to build a logistic model to predict the probability of nodule malignancy. Of those, age, gender, smoking status, current extra-pulmonary cancer, nodule size and presence of air bronchogram were independent risk factors for malignancy (Table 2). Patients $\geq$70 years old (OR 4.77; 95% CI: 1.65–13.76) and female patients (OR 6.51; 95%CI: 1.50–28.10), for instance, were more likely to have malignant nodules. The likelihood of malignancy also increased with every 1-mm increase in diameter (OR 1.25; 95% CI: 1.12–1.38). The clinical prediction model is described as follow: $-8.61 + (1.56 \times \text{Age Category}) + (1.87 \times \textit{Gender}) + (1.26 \times \textit{Smoking Status}) + (1.28 \times \textit{Current ExtraPulmonary Cancer}) + (1.27 \times \textit{Air Bronchogram}) + (0.22 \times \textit{Nodule Size})$–Fig. 1 and Table 3.

Thus, for example, the probability of a 60 year-old male patient, active smoker, with no current extra-pulmonary cancer and a 10 mm nodule with air bronchogram, having a malignant nodule could be computed by: $-8.61 + (1.56 \times 0) + (1.87 \times 1) + (1.26 \times 1) + (1.28 \times 0) + (1.27 \times 1) + (0.22 \times 10) = -2.01$ which indicates a low probability of malignancy -2.5 to 27%. However, a $\geq$70 year-old female patient, former smoker, with no current extra-pulmonary cancer and with a 15 mm nodule with air bronchogram: $-8.61 + (1.56 \times 1) + (1.87 \times 2) + (1.26 \times 0) + (1.28 \times 0) + (1.27 \times 1) + (0.22 \times 15) = 6.04$ has a malignancy probability of 87 to 99%.

The accuracy of the final model was good, with an area under the curve (AUC) of 0.8573. Its sensitivity was 78%, specificity was 85%, the positive predictive value was 85.2%, and the negative predictive value was 78% (Supplementary material). The area under ROC curve of our model [AUC = 0.8573 (95% CI, 0.778–0.919)] was significantly higher than the Brock model [AUC = 0.7384 (95% CI, 0.646–0.813)], $p = 0.005$. ROC curve of our proposed model and the Brock model are displayed in Fig. 3.

Discussion

The authors investigated a sample of 121 Portuguese patients to establish which characteristics would identify a malignant lung nodule. The final model identified four clinical indicators (age, gender, smoking status and current extra-pulmonary cancer) and two imaging indices (maximum nodule diameter and presence of air bronchogram) relevant to estimating the probability of malignancy and help guide follow-up decision. For the most part, these findings were consistent with previous studies. Older age,[10,16,24–26] gender,[27] smoking history[10,24–27] and maximum nodule diameter[10,16,24–26,28] were already referred to as lung cancer predictors. Furthermore, this study identified air bronchogram and current extra-pulmonary cancer as relevant in the decision-making process. In contrast to the BTS guidelines,[15] which recommends the application of the Brock model,[17] our study did not find the presence of spiculation or a predominant spiculated margin and part-solid nodules to be relevant in estimating the probability of malignancy.

The need to distinguish benign and malignant nodules makes the clinical management of pulmonary nodules challenging. The first recommendations, as a standard practice, regarded all noncalcified pulmonary nodules as potentially malignant lesions, requiring follow-up CT screening, until proven stable, for a period of 2 years.[29,30] Later, the 2017 Fleischner Society Guidelines[5] increased the size threshold for routine follow-up of solid nodules to 6 mm, because of data from several screening trials which indicated that the risk of lung cancer in nodules <6 mm is considerably less than 1%, even in patients at high-risk. However, these individuals may warrant follow-up at 12 months, if they have a suspicious morphology, upper lobe location, or both. Solid nodules measuring 6–8 mm in patients with low clinical risk are recommended for follow-up at 6–12 months, extending to 24 months depending on morphology and if stability is uncertain. In high-risk patients, the initial follow-up examination is at 6–12 months and always extends to 24 months. For nodules larger than 8 mm in diameter, both invasive and non-invasive management options are included. Despite these recommendations, some studies[31–33] have shown that the management of pulmonary nodules does not always receive follow-up concordant with Fleischner Society Guidelines.

Table 1 Clinical data description and relationship between these and final biopsy results; TB – tuberculosis.

Population characteristics	Total, *n* (%)	Malignant Nodule, *n* (%)	Benign Nodule, *n* (%)	Missing, *n* (%)	*p* Value
Age					**0.037**
<70 years	73 (60.3)	33 (45.2)	40 (54.8)		
≥70 years	48 (39.7)	31 (64.6)	17 (35.4)		
Gender					0.531
Male	75 (62)	38 (50.7)	37 (49.3)		
Female	46 (38)	26 (56.5)	20 (43.5)		
Smoking status				5 (4.1)	0.505
Not smoker	39 (33.6)	21 (53.8)	18 (46.2)		
Current smoker	31 (26.7)	14 (45.2)	17 (54.8)		
Former smoker	46 (39.7)	27 (58.7)	19 (41.3)		
Current extra-pulmonary cancer history	16 (13.9)	13 (81.3)	3 (18.8)		**0.015**
Pulmonary cancer history	5 (4.3)	4 (80.0)	1 (20.0)	6 (5.0)	0.217
Previous TB	7 (6.0)	3 (42.9)	4 (57.1)	5 (4.1)	0.562
Accidental finding	98 (84.5)	53 (54.1)	45 (45.9)	5 (4.1)	0.452
Central localization	11 (9.1)	10 (90.9)	1 (9.1)		**0.008**
Sphericity	112 (92.6)	61 (54.5)	51 (45.5)		0.222
Margins					
Smooth	108 (89.3)	55 (50.9)	53 (49.1)		0.212
Lobulated	55 (45.5)	36 (65.5)	19 (34.5)		**0.012**
Spiculated	64 (52.9)	40 (62.5)	24 (37.5)		**0.025**
Calcification	3 (2.5)	1(33.3)	2 (66.6)		0.492
Air bronchogram	89 (73.6)	53 (59.6)	36 (40.4)		**0.014**
Cavitation	9 (7.4)	5(55.6)	4 (44.4)		0.869
Single nodule	66 (54.5)	39(59.1)	27 (40.9)		0.135
Pleural contact	35 (28.9)	26 (74.3)	9 (25.7)		**0.003**
Size, mean (range) mm	14.21 (3–29)	16.45 (5–29)	11.68 (3–23)		**<0.001**

Table 2 Multivariate regression analysis of independent risk factors for malignancy.

Variables	Odds ratio	*p* Value	95% Confidence interval
Age <70/≥70 years	4.77	0.004	1.65–13.76
Gender	6.51	0.012	1.51–28.11
Smoking status	3.53	0.003	1.52–8.17
Size	1.25	<0.001	1.12–1.38
Air bronchogram	3.59	0.030	1.13–11.45
Current extra-pulmonary cancer	8.94	0.009	1.72–46.44

Furthermore, the increasing volume of chest imaging and improved image technology is going to create a burden of patients with nodules that need to be managed.[31] Accordingly, there is a growing recognition of the potential utility of risk models to predict lung cancer in patients with pulmonary nodules and allowing more subjects to be monitored with low-dose CT imaging rather than needing invasive procedures. Al-Ameri et al.[4] aimed to validate four models in a UK population – three models based on clinical and CT characteristics (Mayo Clinic,[25] Veterans Association,[10] Brock University[17]), and a fourth model (Herder[34]) additionally incorporating 18Fluorine-Fluorodeoxyglucose (FDG) avidity on positron emission tomography-computed tomography (PET–CT). Both the Mayo (AUC = 0.752) and Brock (AUC = 0.878) models perform well in routine clinical practice. For small pulmonary nodules, the highest AUC value was seen for the Brock model, although there was no significant difference compared to the Mayo model. For patients who underwent PET–CT for nodule evaluation, the Herder prediction model had the highest accuracy. Several other prediction models have been created using clinical and radiological criteria to assist clinicians to discriminate malignant from benign nodules. Older age, smoking history, maximum nodule diameter and spiculation in chest CT appear most frequently as predictors of lung cancer in most of the final models.[16,24,26,35–37]

Overall, our model discriminated well with an excellent overall performance. Its values for discrimination and

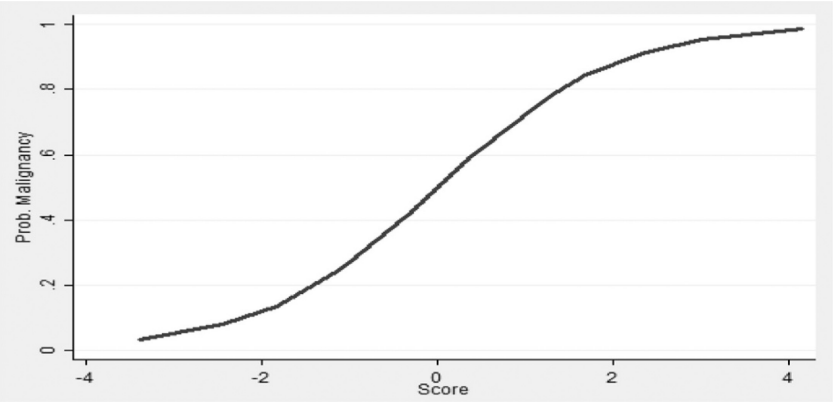

Fig. 1 and Table 3 Final model to calculate malignancy probability: its calculation is made by multiplying each weight by its score. Sum all the values to get the total score for an individual patient. Refer to the second table to get each nodule probability of malignancy.

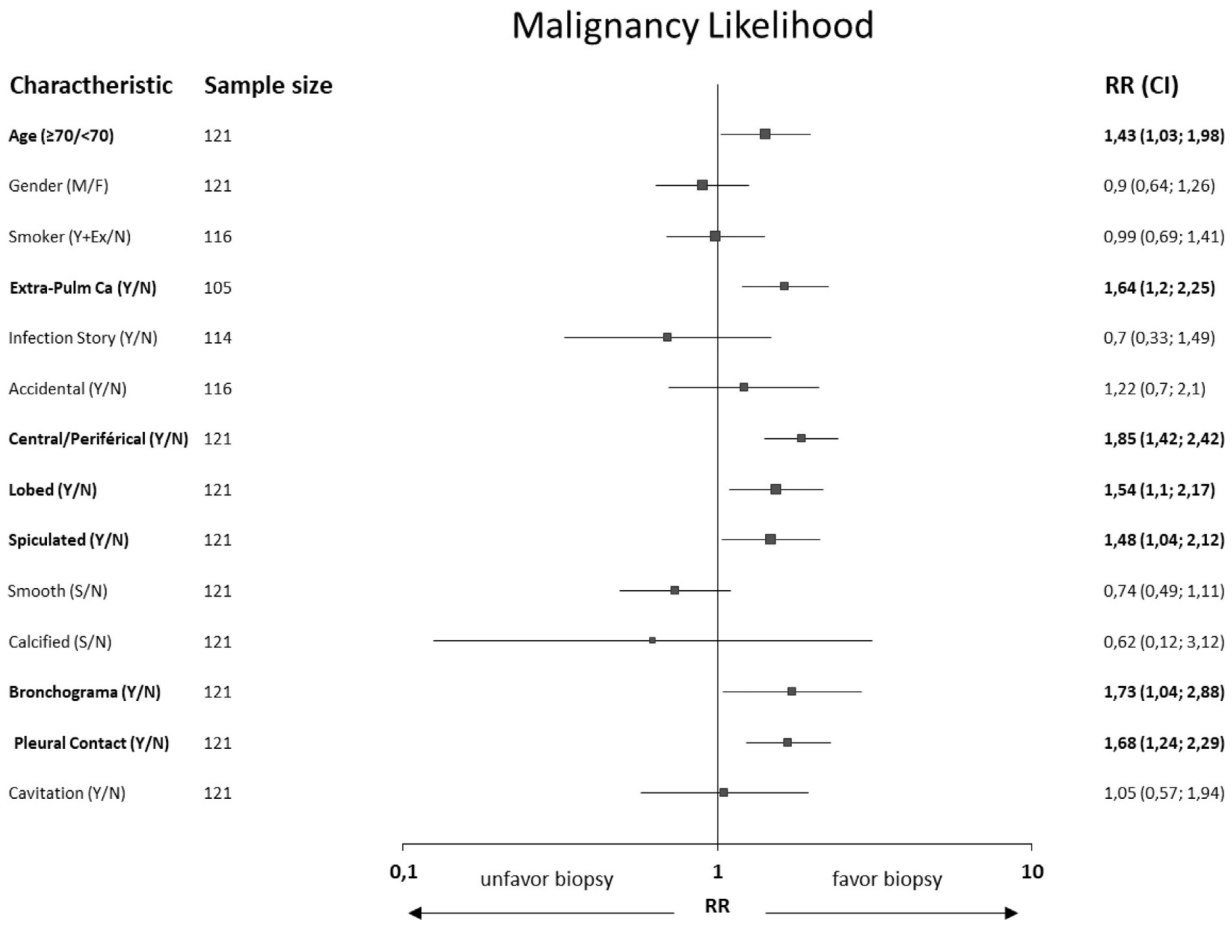

Fig. 2 Risk ratios (RRs) of nodule malignancy according to the presence of patient characteristics.

calibration were comparable to the Brock model[17] (recommended in the BTS guidelines). A direct comparison, using our population, is not entirely accurate, as family history of cancer was not collected in our study (in terms of comparison, it was considered absent when applied to the Brock model). Our model classified five malignant nodules with a low likelihood of malignancy, and those had, also, a lower probability calculated by the Bock model (mean ± standard deviation: 8.2% ± 6%). Moreover, forty-three malignant nodules, that had a high or very high probability of malignancy with our model had a mean probability by the Bock model of 33.6% ± 19%, with more dispersed values (minimum-maximum: 4.6–76.1%). Conversely, forty-five non-malignant nodules classified with low or fair

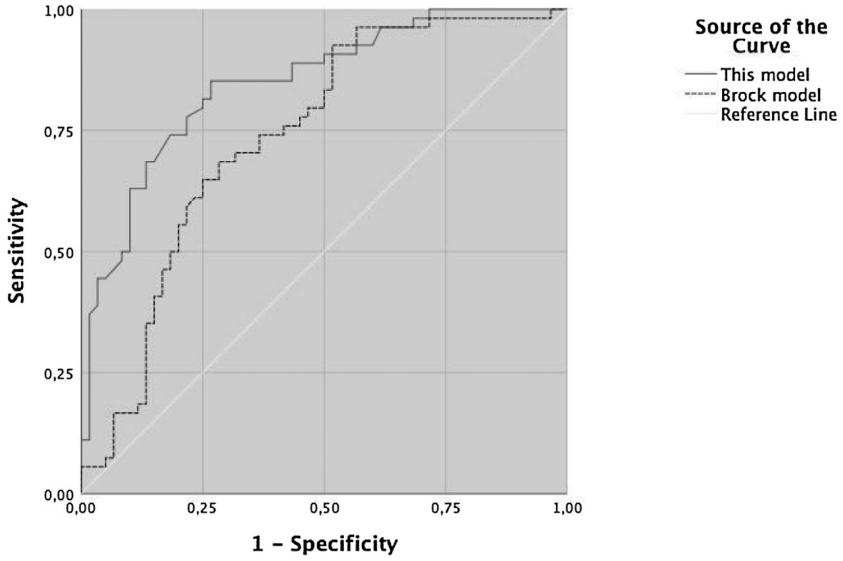

Fig. 3 ROC curve of our proposed model and the Brock model. The area under ROC curve of our model is significantly higher than the Brock model [AUC = 0.8573 (95% CI, 0.778–0.919) vs. AUC = 0.7384 (95% CI, 0.646–0.813)], $p = 0.005$.

probability of malignancy, also had lower mean probability by the Bock model (10.1% ± 7.7%). Furthermore, the AUC of our model performed significantly better than the Brock model, demonstrating that, in a real clinical setting, our model had a similar prediction ability to the Brock model. It is crucial to emphasize that the role of a prediction model is to guide intervention; applying it can enable timely diagnosis and treatment of malignant nodules, prevent unnecessary invasive examinations and surgery for benign nodules, but can never substitute the physician's decision.

However, our study has some limitations. A retrospective study, with a sample of patients who had undergone biopsy, may overestimate the prevalence of malignancy. Moreover, it is a geographically limited group, therefore lacking external validation of this model. Some potentially relevant data was not collected, like pack-years, time since quitting smoking, family history of cancer, variables previously indicated as independent factors for malignancy.

In conclusion, a combination of risk factors for malignancy (age, gender, smoking status, current extrapulmonary cancer, maximum nodule diameter and presence of air bronchogram) can enable accurate differentiation of malignancy from benignancy in lung nodules. To the best of our knowledge, this is the first model pertaining to a Portuguese population and, additionally, with good discrimination, with an AUC value similar to other validated prediction models. This model can help decide the need for a lung biopsy and, thus reducing useless invasive techniques. Although the mathematical models provide an objective basis for judging the character of SPN, we need to emphasize that this prediction model cannot take the place of pathological diagnosis.

Conflicts of interest

The authors have no conflicts of interest to declare.

Appendix A. Supplementary data

Supplementary material associated with this article can be found in the online version available at doi:10.1016/j.pulmoe.2020.06.011.

References

1. Aberle DR, Adams AM, Berg CD, Black WC, Clapp JD, Fagerstrom RM, et al. Reduced lung-cancer mortality with low-dose computed tomographic screening. N Engl J Med. 2011;365:395–409, http://dx.doi.org/10.1056/NEJMoa1102873.
2. Horeweg N, van Rosmalen J, Heuvelmans MA, van der Aalst CM, Vliegenthart R, Scholten ET, et al. Lung cancer probability in patients with CT-detected pulmonary nodules: a prespecified analysis of data from the NELSON trial of low-dose CT screening. Lancet Oncol. 2014;15:1332–41, http://dx.doi.org/10.1016/s1470-2045(14)70389-4.
3. van Klaveren RJ, Oudkerk M, Prokop M, Scholten ET, Nackaerts K, Vernhout R, et al. Management of lung nodules detected by volume CT scanning. N Engl J Med. 2009;361:2221–9, http://dx.doi.org/10.1056/NEJMoa0906085.
4. Al-Ameri A, Malhotra P, Thygesen H, Plant PK, Vaidyanathan S, Karthik S, et al. Risk of malignancy in pulmonary nodules: a validation study of four prediction models. Lung Cancer (Amsterdam, Netherlands). 2015;89:27–30, http://dx.doi.org/10.1016/j.lungcan.2015.03.018.
5. MacMahon H, Naidich DP, Goo JM, Lee KS, Leung ANC, Mayo JR, et al. Guidelines for management of incidental pulmonary nodules detected on CT images: from the Fleischner Society 2017. Radiology. 2017;284:228–43, http://dx.doi.org/10.1148/radiol.2017161659.
6. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2018;68:394–424, http://dx.doi.org/10.3322/caac.21492.
7. Wistuba II, Gazdar AF. Lung cancer preneoplasia. Annu Rev Pathol. 2006;1:331–48, http://dx.doi.org/10.1146/annurev.pathol.1.110304.100103.

8. de Koning HJ, van der Aalst CM, de Jong PA, Scholten ET, Nackaerts K, Heuvelmans MA, et al. Reduced lung-cancer mortality with volume CT screening in a randomized trial. N Engl J Med. 2020;382:503–13, http://dx.doi.org/10.1056/NEJMoa1911793.
9. Vachani A, Tanner NT, Aggarwal J, Mathews C, Kearney P, Fang KC, et al. Factors that influence physician decision making for indeterminate pulmonary nodules. Ann Am Thorac Soc. 2014;11:1586–91, http://dx.doi.org/10.1513/AnnalsATS.201405-197BC.
10. Gould MK, Ananth L, Barnett PG. A clinical model to estimate the pretest probability of lung cancer in patients with solitary pulmonary nodules. Chest. 2007;131:383–8, http://dx.doi.org/10.1378/chest.06-1261.
11. Cummings SR, Lillington GA, Richard RJ. Estimating the probability of malignancy in solitary pulmonary nodules. A Bayesian approach. Am Rev Respir Dis. 1986;134:449–52, http://dx.doi.org/10.1164/arrd.1986.134.3.449.
12. Yonemori K, Tateishi U, Uno H, Yonemori Y, Tsuta K, Takeuchi M, et al. Development and validation of diagnostic prediction model for solitary pulmonary nodules. Respirology (Carlton, Vic). 2007;12:856–62, http://dx.doi.org/10.1111/j.1440-1843.2007.01158.x.
13. Larici AR, Farchione A, Franchi P, Ciliberto M, Cicchetti G, Calandriello L, et al. Lung nodules: size still matters. Eur Respir Rev. 2017;26, http://dx.doi.org/10.1183/16000617.0025-2017.
14. Xu DM, Gietema H, de Koning H, Vernhout R, Nackaerts K, Prokop M, et al. Nodule management protocol of the NELSON randomised lung cancer screening trial. Lung cancer (Amsterdam, Netherlands). 2006;54:177–84, http://dx.doi.org/10.1016/j.lungcan.2006.08.006.
15. Callister ME, Baldwin DR, Akram AR, Barnard S, Cane P, Draffan J, et al. British Thoracic Society guidelines for the investigation and management of pulmonary nodules. Thorax. 2015;70 Suppl. 2:ii1–54, http://dx.doi.org/10.1136/thoraxjnl-2015-207168.
16. Li Y, Chen KZ, Wang J. Development and validation of a clinical prediction model to estimate the probability of malignancy in solitary pulmonary nodules in Chinese people. Clin Lung Cancer. 2011;12:313–9, http://dx.doi.org/10.1016/j.cllc.2011.06.005.
17. McWilliams A, Tammemagi MC, Mayo JR, Roberts H, Liu G, Soghrati K, et al. Probability of cancer in pulmonary nodules detected on first screening CT. N Engl J Med. 2013;369:910–9, http://dx.doi.org/10.1056/NEJMoa1214726.
18. Manhire A, Charig M, Clelland C, Gleeson F, Miller R, Moss H, et al. Guidelines for radiologically guided lung biopsy. Thorax. 2003;58:920–36, http://dx.doi.org/10.1136/thorax.58.11.920.
19. Callister ME, Baldwin DR. How should pulmonary nodules be optimally investigated and managed? Lung Cancer (Amsterdam, Netherlands). 2016;91:48–55, http://dx.doi.org/10.1016/j.lungcan.2015.10.018.
20. Wiener RS, Schwartz LM, Woloshin S, Welch HG. Population-based risk for complications after transthoracic needle lung biopsy of a pulmonary nodule: an analysis of discharge records. Ann Intern Med. 2011;155:137–44, http://dx.doi.org/10.7326/0003-4819-155-3-201108020-00003.
21. Laspas F, Roussakis A, Efthimiadou R, Papaioannou D, Papadopoulos S, Andreou J. Percutaneous CT-guided fine-needle aspiration of pulmonary lesions: results and complications in 409 patients. J Med Imaging Radiat Oncol. 2008;52:458–62, http://dx.doi.org/10.1111/j.1440-1673.2008.01990.x.
22. Hiraki T, Mimura H, Gobara H, Shibamoto K, Inoue D, Matsui Y, et al. Incidence of and risk factors for pneumothorax and chest tube placement after CT fluoroscopy-guided percutaneous lung biopsy: retrospective analysis of the procedures conducted over a 9-year period. Am J Roentgenol. 2010;194:809–14, http://dx.doi.org/10.2214/ajr.09.3224.
23. Lederlin M, Revel MP, Khalil A, Ferretti G, Milleron B, Laurent F. Management strategy of pulmonary nodule in 2013. Diagn Interv Imaging. 2013;94:1081–94, http://dx.doi.org/10.1016/j.diii.2013.05.007.
24. Zhang M, Zhuo N, Guo Z, Zhang X, Liang W, Zhao S, et al. Establishment of a mathematic model for predicting malignancy in solitary pulmonary nodules. J Thorac Dis. 2015;7:1833–41, http://dx.doi.org/10.3978/j.issn.2072-1439.2015.10.56.
25. Swensen SJ, Silverstein MD, Ilstrup DM, Schleck CD, Edell ES. The probability of malignancy in solitary pulmonary nodules. Application to small radiologically indeterminate nodules. Arch Intern Med. 1997;157:849–55.
26. Dong J, Sun N, Li J, Liu Z, Zhang B, Chen Z, et al. Development and validation of clinical diagnostic models for the probability of malignancy in solitary pulmonary nodules. Thorac Cancer. 2014;5:162–8, http://dx.doi.org/10.1111/1759-7714.12077.
27. Marcus MW, Duffy SW, Devaraj A, Green BA, Oudkerk M, Baldwin D, et al. Probability of cancer in lung nodules using sequential volumetric screening up to 12 months: the UKLS trial. Thorax. 2019, http://dx.doi.org/10.1136/thoraxjnl-2018-212263.
28. Shi CZ, Zhao Q, Luo LP, He JX. Size of solitary pulmonary nodule was the risk factor of malignancy. J Thorac Dis. 2014;6:668–76, http://dx.doi.org/10.3978/j.issn.2072-1439.2014.06.22.
29. Tan BB, Flaherty KR, Kazerooni EA, Iannettoni MD. The solitary pulmonary nodule. Chest. 2003;123:89S–96S, http://dx.doi.org/10.1378/chest.123.1_suppl.89S.
30. Ost D, Fein AM, Feinsilver SH. Clinical practice, the solitary pulmonary nodule. N Engl J Med. 2003;348:2535–42, http://dx.doi.org/10.1056/NEJMcp012290.
31. Sethi S, Parrish S. Incidental nodule management-should there be a formal process? J Thorac Dis. 2016;8 Suppl. 6:S494–7, http://dx.doi.org/10.21037/jtd.2016.05.48.
32. Callen JL, Westbrook JI, Georgiou A, Li J. Failure to follow-up test results for ambulatory patients: a systematic review. J Gen Intern Med. 2012;27:1334–48, http://dx.doi.org/10.1007/s11606-011-1949-5.
33. Blagev DP, Lloyd JF, Conner K, Dickerson J, Adams D, Stevens SM, et al. Follow-up of incidental pulmonary nodules and the radiology report. J Am Coll Radiol. 2016;13 Suppl.:R18–24, http://dx.doi.org/10.1016/j.jacr.2015.12.008.
34. Herder GJ, van Tinteren H, Golding RP, Kostense PJ, Comans EF, Smit EF, et al. Clinical prediction model to characterize pulmonary nodules: validation and added value of 18F-fluorodeoxyglucose positron emission tomography. Chest. 2005;128:2490–6, http://dx.doi.org/10.1378/chest.128.4.2490.
35. Zheng B, Zhou X, Chen J, Zheng W, Duan Q, Chen C. A modified model for preoperatively predicting malignancy of solitary pulmonary nodules: an Asia cohort study. Ann Thorac Surg. 2015;100:288–94, http://dx.doi.org/10.1016/j.athoracsur.2015.03.071.
36. Reid M, Choi HK, Han X, Wang X, Mukhopadhyay S, Kou L, et al. Development of a risk prediction model to estimate the probability of malignancy in pulmonary nodules being considered for biopsy. Chest. 2019, http://dx.doi.org/10.1016/j.chest.2019.01.038.
37. Kim H, Goo JM, Park CM. A simple prediction model using size measures for discrimination of invasive adenocarcinomas among incidental pulmonary subsolid nodules considered for resection. Eur Radiol. 2019;29:1674–83, http://dx.doi.org/10.1007/s00330-018-5739-x.

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REVIEW

Bronchoscopic sampling techniques in the era of technological bronchoscopy

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Bronchoalveolar lavage

Abstract Flexible bronchoscopy is a key diagnostic and therapeutic tool. New endoscopes and technologically advanced navigational modalities have been recently introduced on the market and in clinical practice, mainly for the diagnosis of mediastinal lymph adenopathies and peripheral lung nodules. Bronchoscopic sampling tools have not changed significantly in the last three decades, with the sole exception of cryobiopsy.

We carried out a non-systematic, narrative literature review aimed at summarizing the scientific evidence on the main indications/contraindications, diagnostic yield, and safety of the available bronchoscopic sampling techniques.

Performance of bronchoalveolar lavage, bronchial washing, brushing, forceps biopsy, cryobiopsy and needle aspiration techniques are described, focusing on indications and diagnostic accuracy in the work-up of endobronchial lesions, peripheral pulmonary abnormalities, interstitial lung diseases, and/or hilar-mediastinal lymph adenopathies. Main factors affecting the diagnostic yield and the navigational methods are evaluated.

Preliminary data on the utility of the newest sampling techniques (*i.e.*, new needles, triple cytology needle brush, core biopsy system, and cautery-assisted transbronchial forceps biopsy) are shown.

Abbreviations: ACCP, American College of Chest Physicians; CLM, confocal laser microscopy; CT, computed tomography; cTBNA, conventional transbronchial needle aspiration; BW, bronchial washing; BAL, bronchoalveolar lavage; EBB, endobronchial forceps biopsy; EBNA, endobronchial needle aspiration; EBUS-TBNA, endobronchial ultrasound transbronchial needle aspiration; EUS-B-FNA, endoscopic ultrasound (with bronchoscope) fine needle aspiration; EBUS-ca-TBFB, endobronchial ultrasound guided cautery-assisted transbronchial forceps biopsy; EMN, electromagnetic navigation bronchoscopy; IPF, idiopathic pulmonary fibrosis; rEBUS, radial probes endobronchial ultrasound; PPL, peripheral lung lesion; ROSE, rapid on-site evaluation; SLB, surgical lung biopsy; TBNA, transbronchial needle aspiration; TBLC, transbronchial lung cryobiopsy; TB, tuberculosis; TBB, transbronchial biopsy; UIP, usual interstitial pneumonia.

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Take Home Message: A deep knowledge of bronchoscopic sampling techniques is crucial in the era of technological bronchoscopy for an optimal management of respiratory diseases.

Introduction

Flexible bronchoscopy represents an essential diagnostic and therapeutic tool when managing patients with complicated and difficult-to-treat respiratory diseases.[1] After the introduction of the first fiberoptic instruments in 1967, new types of endoscopes were developed: video-bronchoscopes (*i.e.*, endoscopes with a video camera at the distal tip) can significantly improve the quality of the images, ultrathin instruments (*i.e.*, diameter size <3 mm) can explore distal airways beyond segmental bronchi, echo-bronchoscopes can significantly improve the diagnostic accuracy for mediastinal lymph adenopathies.[2–4]

The widespread use of sensitive computed tomography, magnetic resonance imaging, and positron emission tomography have broadened the clinical indications of bronchoscopy and have provided an accurate guide for endoscopic samplings.[3,5,6]

Flexible bronchoscopy is usually recommended for the diagnosis and staging of lung cancer, diagnosis of respiratory tract infections (both in immunocompetent and immunocompromised patients) and of interstitial lung diseases. Furthermore, its use is required for patients with hemoptysis, with unexplained cough and stridor/wheezing, and staging of thoracic malignancies.[1,3,7–11]

Flexible bronchoscopy, performed under conscious sedation and with topical anesthesia, is safe in all age groups, including the elderly, with serious complications and mortality occurring in 1.1% and 0.04% of the cases, respectively.[1,3,12,13]

Bronchoscopic procedures comprehensively assess endobronchial abnormalities (*e.g.*, airway stenosis, bleeding, secretions, *etc.*) and frequently are adopted to collect specimens for microbiological and/or pathological exams,[1,3] quality and quantity of which is key to increase diagnostic accuracy (*e.g.*, idiopathic pulmonary fibrosis, IPF, and lung cancer).[14,15]

New endoscopes and technologically advanced navigational modalities have been recently introduced, mostly for the diagnosis of mediastinal lymph adenopathies and peripheral lung nodules.[16]

With the sole exception of cryobiopsy, bronchoscopic sampling tools have not changed significantly in the last three decades.[16]

The aim of this review is to summarize the scientific evidence on the main indications/contraindications, diagnostic yield, and safety of the available bronchoscopic sampling techniques.

Methods

We carried out a non-systematic, narrative literature review. The search engine Pubmed was used to retrieve the most relevant articles on the above-mentioned topic. The search was conducted without any time restrictions. Only epidemiological studies performed on adult human beings and written in English were selected. The following keywords were combined to address our research question: bronchoscopy; sampling methods; bronchoscopic tools; needle aspiration; biopsy techniques; bronchoalveolar lavage; bronchial washing.

Results

Bronchoalveolar lavage

Bronchoalveolar lavage (BAL) is a safe and minimally invasive bronchoscopic sampling method recommended for patients with several lung medical conditions (*e.g.*, immune-mediated, inflammatory, and infectious diseases). It can provide specimens for cytological and microbiological exams (Table 1).[17]

It is contraindicated in patients with cardiopulmonary instability and/or with a severe haemorrhagic diathesis and It could rarely exacerbate interstitial lung diseases (ILD).[18,19] Transient hypoxemia and low-grade fever within the first 24 h after lavage are the most frequent adverse events.[1,6]

BAL is performed after the assessment of the tracheobronchial tree and before any biopsies.[15,17,20]

The bronchoscope should advance as far as possible to the complete occlusion of the bronchial lumen of a third or fourth bronchial subsegment, in a wedged position. Room temperature sterile saline is employed: 100—300 ml, divided into three to five aliquots, are introduced through the suction channel of the bronchoscope. A volume higher than 5% of the original one (ideally >30%) is collected using a negative suction pressure (<100 mm Hg) avoiding airway collapse.

Interstitial lung diseases

BAL is helpful in patients with suspected ILD[21] both for the diagnosis itself and the differential ascertainment. A high resolution chest CT should be performed within 6 weeks for the optimal identification of the sampling anatomical area.[17]

A differential cellular count for the identification of the inflammatory pattern (*i.e.*, lymphocytic, neutrophilic, eosinophilic, and mast cellular), may be useful in the dif-

Table 1 Summary of bronchoscopic sampling techniques and methods of guidance according to the target lesion.

Endobronchial lesions	Peripheral pulmonary lesions	Hilar and mediastinal lymph adenopathies	Interstitial lung diseases
BW	BAL	TBNA	BAL
EBB	TBB (fluoroscopy or newer navigational modalities-guided)	EBUS-TBNA	TBB (fluoroscopy-guided)
EBNA	TBNA (fluoroscopy or newer navigational modalities-guided)	EUS-B-FNA	Cryobiopsy (fluoroscopy, rEBUS-, CLM- and Cone Beam CT-guided)
Brushing	Brushing (fluoroscopy or newer navigational modalities-guided)	EBUS-ca-TBFB	
Cryobiopsy	Criobiopsy (EMN and rEBUS-guided) Triple brush (EMN and Cone Beam-guided) GenCut Core Biopsy (EMN and Cone Beam-guided)		

BW: bronchial washing; BAL: bronchoalveolar lavage; CLM: confocal laser microscopy; EBB: endobronchial forceps biopsy; EBNA: endobronchial needle aspiration; TBB: transbronchial biopsy; TBNA: conventional transbronchial needle aspiration; rEBUS: radial probes endobronchial ultrasound; EBUS-TBNA: endobronchial ultrasound transbronchial needle aspiration; EUS-B-FNA: endoscopic ultrasound (with bronchoscope) fine needle aspiration; EBUS-ca-TBFB: endobronchial ultrasound guided cautery-assisted transbronchial forceps biopsy.

ferential diagnosis of interstitial lung diseases. A minimal volume of 5 mL of a pooled BAL sample is needed for BAL cellular analysis (the optimal volume is 10–20 ml).

Bloody fluid, with increasing colour intensity in sequential aliquots, can suggest a diffuse alveolar haemorrhage[22] (microscopic diagnosis supported by hemosiderin-laden macrophages).[17] Cloudy (*i.e.*, milky or light brown-beige colour) fluid with flocculent material settling by gravity within 15—20 min and PAS-positive amorphous debris suggests a pulmonary alveolar proteinosis (PAP).

An increased number of CD-1a cells (>5% of BAL cells) strongly suggests pulmonary Langerhans cell histiocytosis.[23]

BAL cellular pattern may help discriminate IPF from eosinophilic pneumonia (eosinophilia >25%), sarcoidosis (high proportion of lymphocytes and CD4/CD8 ratio), and infections.[15]

In patients with a fibrotic interstitial lung disease BAL lymphocytosis of at least 30% may suggest nonspecific interstitial pneumonia and extrinsic allergic alveolitis.[24]

A recent retrospective study that aimed to study the role of bronchoscopy in acute respiratory failure related to ILD, failed to demonstrate a different management and mortality between patients with positive and negative BAL findings.[25]

Peripheral pulmonary lesions

BAL should be used for patients with slowly resolving/non-resolving pneumonia (sensitivity >70%).[1,26] BAL can play a key role in the TB diagnosis for sputum smear-negative patients or in those in whom sputum cannot be collected.[27] BAL diagnosis of pulmonary TB relies on smear microscopy (sensitivity range: 4.7–58.0%), nucleic acid amplification techniques (sensitivity: 31.3–83.8%; specificity: 92.4–98.2%), and culture (highest diagnostic accuracy).[27]

BAL can help rule out opportunistic infections in immunocompromised hosts,[1] with a sensitivity up to 98% for *Pneumocistis jiroveci*. Sensitivity of smear microscopy for TB disease in HIV-positives ranges from 10 to 30%, increasing to 85.7% and 52–95% when nucleic acid amplification techniques and culture are adopted, respectively.[27–29] In immunocompromised hosts with invasive aspergillosis, BAL can help detect fungal hyphae (34–64% of the cases) and galactomannan antigen (sensitivity and specificity of respectively 79–90% and 84–94%), and can increase the rate of culture positivity (23–85%).[1,30,31]

BAL shows a low accuracy in the diagnosis of peripheral lung malignancies (mean sensitivity 43%), whereas lymphangitic carcinomatosis and pulmonary lymphoma may be diagnosed using BAL samples.[6,32,34]

Bronchial washing

Bronchial washing (BW) consists of instillation and subsequent aspiration of saline mixed up with bronchial secretions, into a specific bronchial trap. It may be useful to assess the microbiology of central airways secretions.[6] In the diagnosis of TB, BW smear microscopy and Xpert MTB/RIF show a sensitivity of 25–41% and 80–92.3% and a specificity of 87.7–95.8% and 81.6–98.6%, respectively.[27,33–35]

A limited diagnostic support was found for endobronchial lung cancers (mean sensitivity: 47%). (Table 1).

The diagnostic yield of bronchoscopy when bioptic techniques (*i.e.*, endobronchial needle aspiration and forceps biopsy) are used is not affected by BW.[36]

Needle aspiration

Needle aspiration, which is the most versatile bronchoscopic sampling technique, is recommended for the diagnosis of endobronchial and peripheral lesions and in case of hilar/mediastinal lymph adenopathies (Table 1).[37,38]

A thin (25–19 gauge), retractable needle attached to the distal tip of a flexible catheter is inserted into the

Table 2 Factors which can influence the diagnostic yield of bronchoscopic sampling techniques in the diagnosis of peripheral pulmonary lesions.

Lesion size
CT bronchus sign presence
Navigational modalities employment
ROSE presence
Malignant nature of the lesion

working channel of the endoscope and is pushed into the target lesion, while the catheter is moved back and forth for few seconds at its proximal end. The vacuum inside the syringe causes tissue to be suctioned into the needle.[37,38] The needle may be inserted in an endobronchial lesion under direct endoscopic vision and into a hilar/mediastinal lymph node, through the tracheobronchial wall, with or without endoscopic ultrasound guidance. Fluoroscopy and/or other navigational techniques are necessary to reach peripheral lung abnormalities.[38]

The collected specimen may be smeared on a glass slide or directly placed in formalin solution (technique named formalin-fixed, paraffin-embedded cell-block). Rapid on-site evaluation (ROSE) of the aspirates may be performed, allowing bronchoscopists to stop sampling when sufficient material has been harvested for diagnosis and molecular analysis, thus potentially avoiding useless samplings and reducing the complications of bronchoscopy.[36,37]

Endobronchial lesions

Endobronchial needle aspiration (EBNA) is a useful and safe technique adopted for the diagnosis of endobronchial lesions (mainly lung neoplasms).

It has a mean sensitivity of 56%, with a rate of complications (mostly minor bleedings) <1%.[36,39,40] It significantly increases the accuracy of bronchoscopy in the diagnosis of central lung cancers when combined with endobronchial forceps biopsy. EBNA is particularly helpful in sampling submucosal/peribronchial (*i.e.*, growing in deeper layers of the airways) and necrotic lesions. Needle can penetrate the mucosa and can sample neoplasms spreading in the deeper layers.[36]

In the diagnosis of endobronchial tuberculosis, Altin et al. reported a lower sensitivity of EBNA than forceps biopsy in the detection of granulomas (19% *vs* 84%, respectively).[27,41]

Peripheral pulmonary lesions

Transbronchial needle aspiration with the guidance of fluoroscopy has been adopted to sample peripheral lung lesions (both nodules and masses) since 1984 (Fig. 1B).[42] A recent systematic review and meta-analysis showed a diagnostic yield of 53% and a rate of complications <9%, with pneumothorax and bleeding being the most frequent events.[43] Several clinical and procedural variables may affect its accuracy: CT bronchus sign, an underlying malignant process, diameter of the lesions >3 cm, and ROSE employment are the most important predictive factors of a positive aspirate (Table 2). Notably, data on comparison between TBNA and transbronchial forceps biopsy (TBB) in studies where both procedures were performed in the same patients showed a significant diagnostic advantage when TBNA is performed (diagnostic yield: 60% *vs*. 45%, respectively), although studies have shown that TBNA is still a underused sampling technique.[43,44]

Recently, new navigational methods, which may be coupled with fluoroscopy, have been adopted to sample peripheral lesions. TBNA guided by electromagnetic navigation bronchoscopy (EMN) showed a diagnostic yield of 46.3%, while needle aspiration guided by radial probes endobronchial ultrasounds (rEBUS) of 49.5—62.5%.[44,45] When added to TBB, rEBUS-TBNA significantly increases the accuracy of bronchoscopy in the diagnosis of peripheral lesions.[45]

Hilar and mediastinal lymph adenopathies

Conventional transbronchial needle aspiration (*i.e.*, not guided by ultrasounds) was introduced by Wang in 1984.[42]

American College of Chest Physician (ACCP) guidelines showed a sensitivity of 78% in the diagnosis and staging of non-small cell lung cancer, with a complication rate of 0.3%.[46,47] It was also used for the diagnosis of sarcoidosis stage I and II and mediastinal tuberculosis (sensitivity of 72–79% and 65–100%, respectively %).[27,48–50]

Currently, cTBNA has been replaced by endobronchial ultrasound (EBUS)-guided TBNA: it includes an echobronchoscope (*i.e.*, a bronchoscope with a convex probe at the distal end) and allows a real-time visualization of the lymph nodes and mediastinal vessels. Unlike cTBNA, EBUS-TBNA can diagnose lymph adenopathies sized <1 cm and lymph node stations without endobronchial landmarks.[6,47] EBUS-TBNA shows a higher sensitivity and negative predictive value (89% and 91%, respectively) than conventional technique in the diagnosis and staging of NSCLC.[39,47] Its sensitivity in the diagnosis of sarcoidosis and tuberculous lymph adenopathies is 79–84%[51,52] and 87%, respectively. Both conventional and ultrasound-guided techniques increase the diagnostic accuracy of bronchoscopy when combined with other sampling techniques (bronchial and transbronchial forceps biopsy and BAL).[51,52]

Recently, a new needle aspiration technique, named endoscopic ultrasound (with bronchoscope) fine needle aspiration (EUS-B-FNA) has been proved to be effective[53]: an ultrasound guided needle aspiration of mediastinal lymph adenopathies is performed with an echobronchoscope introduced in the esophagu,.[53] Transbronchial and transesophageal needle sampling can be performed with the same instrument, in the same endoscopic session, and by the same operator (*i.e.*, a trained pulmonologist), thus maximizing time and reducing costs. The transesophageal approach can be also used to sample nodes within reach of EBUS, when the clinical conditions contraindicate the transbronchial route (*e.g.*, respiratory failure, cough, *etc.*).[54,55]

The combined approach increases the accuracy of endosonography and is now recommended by international guidelines.[56]

EUS-B-FNA may safely diagnose extra-thoracic targets, such as abdominal lymph nodes, liver and left adrenal glands metastatic lesions.[57] Both EBUS-TBNA and EUS-B-FNA may diagnose lung parenchymal lesions adjacent to the central airways and the esophagus.[58,59]

Needle size does not significantly affects the diagnostic yield.[60]

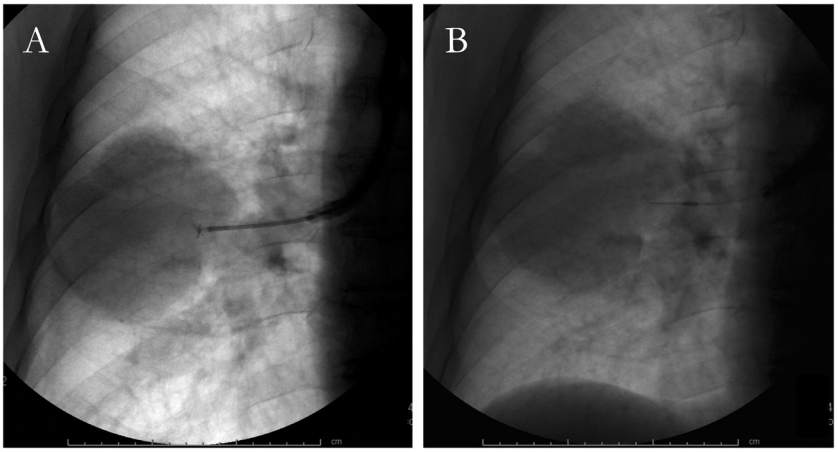

Figure 1 Conventional fluoroscopy-guided transbronchial forceps biopsy (A) and needle aspiration (B) of a right pulmonary mass.

Several studies demonstrated the suitability of ultrasound-guided needle aspiration samples for molecular analysis in advanced NSCLC, on both cytology smears and cell-block preparations.[61–63]

Complications of endosonographic needle aspiration procedures are rare (serious adverse events rate of 0.14%).[64]

Forceps biopsy

Forceps has been adopted to collect lung tissue samples through the bronchoscope since the initial implementation of bronchial endoscopy (Table 1).[65]

Endobronchial lesions

Endobronchial biopsy (EBB) is recommended for the diagnosis of visible endobronchial lesions[39]: forceps should be opened outside the distal end of the operating channel and pushed against the lesion providing the right orientation to the instrument, according to the localization of the target site. The tip of the forceps is then closed, pulled out of the operating channel of the bronchoscope and the specimen is then placed in formalin solution.[66] The different characteristics of the forceps (serrated or smooth edge, fenestrated or unfenestrated cups, needle between the cups) make it potentially suitable for specific settings/lesions. However, the diagnostic yield of various forceps biopsy types was not statistically different.[38]

EBB is usually employed for suspected bronchogenic cancer with a sensitivity of 74%[39]: ≥3 biopsies are recommended for diagnosis, although at least 6 biopsies can provide sufficient tissue for immunohistochemical and molecular testing.[39,67] Several studies[36,68,69] demonstrated that the combination of EBB and endobronchial needle aspiration can achieve the best diagnostic performance.

EBB, when combined with transbronchial biopsy, can increase the sensitivity of bronchoscopy by 10–20% in the diagnosis of sarcoidosis: sampling should be performed where the mucosa is abnormal and in the first and second carina if the mucosa seems normal (4–6 endobronchial biopsies); 30% with normal mucosa may have positive EBB.[70–72]

Forceps biopsy showed a sensitivity of 72.2–100.0% in the detection of TB granulomas (endobronchial TB), and may be useful in ruling out malignancies.[27,41,73]

The most frequent complication is minor bleeding, which can resolve spontaneously in the majority of the cases or can be treated with ice-cold saline or vasoconstrictive agents (*e.g.*, epinephrine). On this basis, caution may be warranted when sampling is performed for vascularized lesions (*i.e.*, carcinoids).[74,75]

Peripheral lung lesions

A transbronchial biopsy is performed when the lesion cannot be directly assessed with the bronchoscope: it is wedged in the bronchus pertaining to the anatomical site of the lesion, and the closed forceps are pushed in the peripheral area of the lung, opened at 5—6 mm from the lesion and then closed to collect sample (Fig. 1A).[6] Fluoroscopy guidance can improve the sensitivity in case of peripheral focal and diffuse cancer lesions.[6,76,77] Observational studies have demonstrated that navigational methods (*i.e.*, electromagnetic navigation bronchoscopy, radial probes ultrasounds, virtual bronchoscopy) and/or ultrathin instruments may increase the diagnostic yield of conventional, fluoroscopy-guided technique (77–84%).[44,78–80]

The diameter of the lesion affects the accuracy of the technique: the sensitivity is <35% in case of nodules sized <2 cm.[39] Moreover, sensitivity is 24% performing only a single biopsy and 70% when six biopsies are collected.[81,82] The presence of the CT-bronchus sign is associated with a higher yield (Table 2).[83,84]

TBB may increase the sensitivity of BAL for the diagnosis of *Pneumocystis jirovecii* pneumonia, including non–HIV patients.[85] In sputum smear negative or sputum scarce TB patients with peripheral lung lesions, TBB[86–88] may help detect cytological and histological TB findings (*i.e.*, necrotizing granulomatous inflammation), ruling out malignancies.[89]

Finally, TBB is a safe and repeatable procedure monitoring early signs of graft rejection in lung transplant recipients.[90]

Mild bleeding and pneumothorax are the most frequent complications. Pneumothorax can occur in 1–5% of the cases; its variability can depend on the use of mechani-

cal ventilation, surrounding emphysema, lesion near to the pleura, and poor expertise of the healthcare worker.[6]

Bleeding can be a risk in patients with coagulopathy disorders and/or taking anticoagulant and anti-platelet drugs.[6]

Interstitial lung diseases

Sensitivity of TBB in diffuse lung disease varies widely.[91] The main limitations are the small size of the sample and the difficult preservation of the tissue integrity, for which a surgical biopsy or a cryobiopsy may be more suitable.[92] Exceptions are conditions involving the centrilobular region (both at the terminal and respiratory bronchioles or along the lymphatic distribution, such as sarcoidosis, hypersensitivity pneumonitis, organizing pneumonia, eosinophilic pneumonias, and lymphangitic spread of malignancy). Sensitivity ranges from 55% in stage I to 80% in stage III[93] in sarcoidosis. Higher yield is found when biopsies are performed in >1 lobe and in the area of the most affected tissue in stage II/III disease.[94]

Guidelines on Idiopathic Pulmonary Fibrosis (IPF) do not recommend for or against TBB when the HRCT suggest a probable UIP pattern. In this context, TBB could be only clinically helpful to exclude mimickers.[15,95]

Brushing

Endobronchial lesions and peripheral lung abnormalities

Brushing consists of a rigid central wire surrounded by brushes of various sizes and shapes. A brush inserted through the operating channel performs both a back and forth and a spinning movement on the surface of the mucosa. Cytological material may be smeared on glass slides or placed in formalin solutions. The diameter or the length of the brush does not affect the diagnostic yield[6] (mean sensitivity in the diagnosis of endobronchial malignancy: 61%).[39] Addition of bronchial brushing to forceps biopsy and needle aspiration does not increase the sensitivity of bronchoscopy.[36,40,69]

It showed a diagnostic yield of 47–54% in the diagnosis of peripheral lesions,[39] which is usually lower than that reported for TBB and TBNA, even if guided by novel methods of navigation (Table 1).[39,44,80]

Quantitative cultures of protected brushing (*i.e.*, a double-lumen catheter brush system with a distal occluding plug to prevent secretions from entering the catheter during passage through the bronchoscope channel) can be performed to diagnose pneumonia in critically ill patients, (mean sensitivity: 89%).[96,97]

Minor bleeding is the most likely incidental complication.

Cryobiopsy

Cryoprobe is a therapeutic and diagnostic tool traditionally adopted for endobronchial tumour ablation and airway recanalization or by removal of blood clots and foreign bodies.[98–100]

Only recently several studies proved its accuracy as endobronchial and transbronchial biopsy technique (Table 2).[101]

Its activity is based on the principle of the Joule-Thomson effect, wherein the adiabatic expansion of a compressed gas leads to a rapid cooling. The cooled tip of the cryoprobe, inserted in the working channel of the bronchoscope, adheres to the tissue due to crystallization of water molecules at the interface. After a few seconds of cooling, the probe is extracted with a specimen, which is placed in formalin.

Cryobiopsy may be used with flexible bronchoscopes with local anaesthesia, deep sedation and/or general anaesthesia, with laryngeal mask or in patient intubated with orotracheal tube or rigid tracheoscope (during spontaneous breathing or mechanical ventilation). Intubation with deep sedation or general anaesthesia and administration of a bronchial blocker are recommended to prevent severe bleedings in the diagnostic work-up of ILD.[102]

Endobronchial lesions

Hetzel et al. demonstrated the higher efficacy of cryobiopsy in the diagnosis of endobronchial malignant lesions when compared with conventional forceps: it can collect larger specimens without disrupting the morphological structure. Cryoprobes increase the diagnostic yield of bronchoscopy (up to 95%), without a higher rate of bleeding.[98] Several studies have confirmed these findings,[103] including the safety in the diagnosis of carcinoid tumours.[104]

Peripheral lung lesions

Schumann et al. evaluated the accuracy in the diagnosis of peripheral lesions with the guidance of rEBUS: the diagnostic yield of 74.2% was not significantly higher in comparison with that of EBUS-guided forceps biopsy (61.3%); however, samples were significantly larger than those collected by TBB.[105]

Other studies showed a diagnostic yield of 69–85% when guided by ultrasounds or EMN, and confirmed the advantage of larger samples and a better preserved architecture, thus improving the specimen quality for the molecular diagnosis.[106–110]

Mild bleeding and pneumothorax were the most common incidental adverse events.

Interstitial lung diseases

Transbronchial lung cryobiopsy (TBLC) is a minimally invasive alternative to surgical lung biopsy (SLB), which is the gold standard in the histopathological diagnosis of many ILD. Conventional forceps biopsies are inadequate in diseases characterized by a heterogeneous histological pattern and in those with histological abnormalities located at the periphery of the secondary lobule (*e.g.*, usual interstitial pneumonia, UIP).[102]

Larger biopsy size and lack of crush artifact make cryobiopsy more suitable for the diagnosis of diffuse lung diseases if compared with conventional forceps biopsy.[111]

IPF Guidelines recommend cryobiopsy only in experienced centers, when HRCT pattern is probable UIP, indeterminate for UIP or suggesting an alternative diagnosis.[15]

Tomassetti et al., who recruited 117 patients with fibrotic ILD needing a pathological diagnosis, demonstrated that the addition of TBLC was associated to an increased diagnostic confidence in the multidisciplinary diagnosis of idiopathic pulmonary fibrosis, similar to that provided by SLB.[112]

Samples should be taken under fluoroscopic guidance in the distal part of the lung parenchyma, avoiding high density fibrotic areas. Biopsy should be performed at a dis-

tance >1 cm from the pleura to reduce the occurrence of pneumothorax.[113]

As suggested by Ravaglia et al., collection of ≥2 samples from two different segments in the same lobe or from different lobes in case of inter-lobar radiographic heterogeneity is recommended to increase the diagnostic yield.[102,113–115]

A systematic review and meta-analysis showed a pooled diagnostic yield of 79%.[116]

Two studies evaluated the accuracy of cryobiopsy in comparison with surgical biopsy: Romagnoli et al. found a poor concordance between TBLC and SLB (concordant coefficient (k): 0.22, percentage agreement: 38%),[117] whereas a multicentre, prospective study , found a histopathological agreement of 70.8% (weighted k: 0.70) and a final diagnostic agreement of 76.9% (k: 0.62).[118]

The frequency of pneumothorax and moderate/severe bleeding is 9.5% and 1.1–8.7%, respectively.[113]

Guidelines suggest the use of fluoroscopy, of a bronchial blocker, and a small (*i.e.*, 1.9 mm) cryoprobe to reduce the complication rate.[113]

One prospective study reported on the utility of a radial EBUS miniprobe to avoid injuries of pulmonary vessels during biopsy.[119] Confocal laser microscopy (CLM) is a minimally invasive endoscopic technique that provides real time *in vivo* microscopic imaging of the distal lung through a thin probe advanced through the working channel of the bronchoscope until the alveolar area. Preliminary data demonstrated that CLM may be a useful guidance tool for transbronchial cryobiopsies. It helps to distinguish fibrotic *vs*. not fibrotic areas and to avoid the pleura thereby reducing the risk of pneumothorax.[120]

Likewise, Cone beam CT-guided TBLC, which evaluates the probe-to-pleura relationship based on 3D CT scans, has a safe profile, with low risk of pneumothorax and moderate/severe bleeding.[121]

The main contraindications are bleeding diathesis, use of anticoagulants, thienopyridines, antiplatelet drugs, and thrombocytopenia ($<50 \times 10^9$/L), pulmonary hypertension, and severe respiratory functional impairment.[102]

New tools

New flexible needles of different size have recently been introducedonto the market for endosonographic sampling of hilar/mediastinal lymph nodes. New needles may provide more visibility on ultrasound images while the needle penetrates the lymph node, more flexibility to target paratracheal and hilar stations, and a larger amount of tissue for histopathological analysis.[122]

New tools for diagnosis of endobronchial and peripheral lesions can be directly inserted into the working channel of the endoscope or passed into a guide sheath to reach peripheral lung abnormalities. Triple cytology needle brush may trap larger tissue samples. A new core biopsy system (*i.e.* GenCut core biopsy system) consists of a flexible tool with a rounded, blunt tip, a port and blade along the distal and lateral sides with a hollow core: suction is applied follow by rotation and agitation to collect intact tissue.[44,80]

Cautery-assisted transbronchial forceps biopsy (ca-TBFB) is a new sampling technique which collects larger amount of tissue from mediastinal lymph nodes: a target lymph node is identified with EBUS while an electrocautery knife is advanced through the working channel of the endoscope toward the airway wall. Then, cautery is applied and the knife inserted through the tracheal/bronchial wall defect (under EBUS real-time guidance), created by the cautery edge. After the penetration, the knife is withdrawn and a spiked forceps advanced into the lymph node to collect the sample.[123,124]

Two observational studies showed a higher sensitivity in comparison with that of EBUS-TBNA in the diagnosis of sarcoidosis and lymphoma.[123,124]

Another study proved an increased sensitivity of EBUS forceps biopsy in patients with mediastinal lymph nodes in whom ROSE of EBUS-TBNA failed to show positive findings.[125]

References

1. Du Rand IA, Blaikley J, Booton R, Chaudhuri N, Gupta V, Khalid S, et al. British Thoracic Society guideline for diagnostic flexible bronchoscopy in adults. Thorax. 2013;68 SUPPL. 1, http://dx.doi.org/10.1136/thoraxjnl-2013-203618.
2. Krasnik M, Vilmann P, Larsen SS, Jacobsen GK. Preliminary experience with a new method of endoscopic transbronchial real time ultrasound guided biopsy for diagnosis of mediastinal and hilar lesions. Thorax. 2003;58(12):1083–6, http://dx.doi.org/10.1136/thorax.58.12.1083.
3. Ninan N, Wahidi MM. Basic bronchoscopy: Technology, techniques, and professional fees. Chest. 2019;155(5):1067–74, http://dx.doi.org/10.1016/j.chest.2019.02.009.
4. Yamamoto S, Ueno K, Imamura F, Matsuoka H, Nagatomo I, Omiya Y, et al. Usefulness of ultrathin bronchoscopy in diagnosis of lung cancer. Lung Cancer. 2004;46(1):43–8, http://dx.doi.org/10.1016/j.lungcan.2004.03.005.
5. Sferrazza Papa GF, Mondoni M, Volpicelli G, Carlucci P, Di Marco F, Parazzini EM, et al. Point-of-care lung sonography: An audit of 1150 examinations. J Ultrasound Med. 2017;36(8):1687–92, http://dx.doi.org/10.7863/ultra.16.09007.
6. Miller RJ, Casal RF, Lazarus DR, Ost DE, Eapen GA. Flexible bronchoscopy. Clin Chest Med. 2018;39(1):1–16, http://dx.doi.org/10.1016/j.ccm.2017.09.002.
7. Gasparini S. Indications for diagnostic bronchoscopy in adults. Monaldi Arch Chest Dis - Pulm Ser. 2011;75(1):24–31, http://dx.doi.org/10.4081/monaldi.2011.236.
8. Mondoni M, Carlucci P, Cipolla G, Fois A, Gasparini S, Marani S, et al. Bronchoscopy in patients with hemoptysis and negative imaging tests. Chest. 2018;153(6):1510–1, http://dx.doi.org/10.1016/j.chest.2017.12.028.
9. Mondoni M, Carlucci P, Cipolla G, Fois A, Gasparini S, Marani S, et al. Bronchoscopy to assess patients with hemoptysis: Which is the optimal timing? BMC Pulm Med. 2019;19(1):1–6, http://dx.doi.org/10.1186/s12890-019-0795-9.
10. Mondoni M, Carlucci P, Job S, Parazzini EM, Cipolla G, Pagani M, et al. Observational, multicentre study on the epidemiology of haemoptysis. Eur Respir J. 2018;51(1):10–3, http://dx.doi.org/10.1183/13993003.01813-2017.
11. Mondoni M, Papa GFS, Sotgiu G, Carlucci P, Pellegrino GM, Centanni S. Haemoptysis: A frequent diagnostic challenge. Eur Respir J. 2016;47(1):348–50, http://dx.doi.org/10.1183/13993003.01344-2015.
12. Mondoni M, Radovanovic D, Sotgiu G, Di Marco F, Carlucci P, Centanni S, et al. Interventional pulmonology techniques in elderly patients with comorbidities. Eur J Intern Med. 2019;59(September 2018):14–20, http://dx.doi.org/10.1016/j.ejim.2018.09.015.
13. Facciolongo N, Patelli M, Gasparini S, Agli LL, Salio M, Simonassi C, et al. Incidence of complications in bronchoscopy.

Multicentre prospective study of 20,986 bronchoscopies. Monaldi Arch Chest Dis - Pulm Ser. 2009;71(1):8–14, http://dx.doi.org/10.4081/monaldi.2009.370.
14. Raghu G, Rochwerg B, Zhang Y, Garcia CAC, Azuma A, Behr J, et al. An official ATS/ERS/JRS/ALAT clinical practice guideline: Treatment of idiopathic pulmonary fibrosis: An update of the 2011 clinical practice guideline. Am J Respir Crit Care Med. 2015;192(2):e3–19, http://dx.doi.org/10.1164/rccm.201506-1063ST.
15. Raghu G, Remy-Jardin M, Myers JL, Richeldi L, Ryerson CJ, Lederer DJ, et al. Diagnosis of idiopathic pulmonary fibrosis. An official ATS/ERS/JRS/ALAT clinical practice guideline. Am J Respir Crit Care Med. 2018;198(5):e44–68, http://dx.doi.org/10.1164/rccm.201807-1255ST.
16. Mondoni M, Sotgiu G. Bronchoscopic management of peripheral pulmonary lesions: Robotic approach paves the way to the future. BMC Pulm Med. 2019;19(1):8–10, http://dx.doi.org/10.1186/s12890-019-0927-2.
17. Meyer KC, Raghu G, Baughman RP, Brown KK, Costabel U, Du Bois RM, et al. An official American Thoracic Society clinical practice guideline: The clinical utility of bronchoalveolar lavage cellular analysis in interstitial lung disease. Am J Respir Crit Care Med. 2012;185(9):1004–14, http://dx.doi.org/10.1164/rccm.201202-0320ST.
18. Hiwatari N, Shimura S, Takishima T, Shirato K. Bronchoalveolar lavage as a possible cause of acute exacerbation in idiopathic pulmonary fibrosis patients. Tohoku J Exp Med. 1994;174(4):379–86, http://dx.doi.org/10.1620/tjem.174.379.
19. Kim DS, Park JH, Park BK, Lee JS, Nicholson AG, Colby T. Acute exacerbation of idiopathic pulmonary fibrosis: Frequency and clinical features. Eur Respir J. 2006;27(1):143–50, http://dx.doi.org/10.1183/09031936.06.00114004.
20. Baughman RP. Technical aspects of bronchoalveolar lavage: Recommendations for a standard procedure. Semin Respir Crit Care Med. 2007;28(5):475–85, http://dx.doi.org/10.1055/s-2007-991520.
21. Wells AU. The clinical utility of bronchoalveolar lavage in diffuse parenchymal lung disease. Eur Respir Rev. 2010;19(117):237–41, http://dx.doi.org/10.1183/09059180.00005510.
22. De Lassence A, Fleury-Feith J, Escudier E, Beaune J, Bernaudin JF, Cordonnier C. Alveolar hemorrhage. Diagnostic criteria and results in 194 immunocompromised hosts. Am J Respir Crit Care Med. 1995;151(1):157–63, http://dx.doi.org/10.1164/ajrccm.151.1.7812547.
23. Baqir M, Vassallo R, Maldonado F, Yi ES, Ryu JH. Utility of bronchoscopy in pulmonary Langerhans cell histiocytosis. J Bronchology Interv Pulmonol. 2013;20(4):309–12, http://dx.doi.org/10.1097/LBR.0000000000000021.
24. Ohshimo S, Bonella F, Cui A, Beume M, Kohno N, Guzman J, et al. Significance of bronchoalveolar lavage for the diagnosis of idiopathic pulmonary fibrosis. Am J Respir Crit Care Med. 2009;179(11):1043–7, http://dx.doi.org/10.1164/rccm.200808-1313OC.
25. Arcadu A, Moua T. Bronchoscopy assessment of acute respiratory failure in interstitial lung disease. Respirology. 2017;22(2):352–9, http://dx.doi.org/10.1111/resp.12909.
26. Nieto JMS, Alcaraz AC. The role of bronchoalveolar lavage in the diagnosis of bacterial pneumonia. Eur J Clin Microbiol Infect Dis. 1995;14(10):839–50, http://dx.doi.org/10.1007/BF01691489.
27. Mondoni M, Repossi A, Carlucci P, Centanni S, Sotgiu G. Bronchoscopic techniques in the management of patients with tuberculosis. Int J Infect Dis. 2017;64:27–37, http://dx.doi.org/10.1016/j.ijid.2017.08.008.
28. Miro AM, Gibilara E, Powell S, Kamholz SL. The role of fiberoptic bronchoscopy for diagnosis of pulmonary tuberculosis in patients at risk for AIDS. Chest. 1992;101(5):1211–4, http://dx.doi.org/10.1378/chest.101.5.1211.
29. Kennedy DJ, Lewis WP, Barnes PF. Yield of bronchoscopy for the diagnosis of tuberculosis in patients with human immunodeficiency virus infection. Chest. 1992;102(4):1040–4, http://dx.doi.org/10.1378/chest.102.4.1040.
30. Hage CA, Carmona EM, Epelbaum O, Evans SE, Gabe LM, Haydour Q, et al. Microbiological laboratory testing in the diagnosis of fungal infections in pulmonary and critical care practice. An Official American Thoracic Society Clinical Practice Guideline. Am J Respir Crit Care Med. 2019;200(5):535–50, http://dx.doi.org/10.1164/rccm.201906-1185ST.
31. Zou M, Tang L, Zhao S, Zhao Z, Chen L, Chen P, et al. Systematic review and meta-analysis of detecting galactomannan in bronchoalveolar lavage fluid for diagnosing invasive aspergillosis. PLoS One. 2012;7(8):e43347, http://dx.doi.org/10.1371/journal.pone.0043347.
32. Rennard SI. Bronchoalveolar lavage in the diagnosis of cancer. Lung. 1990;168 Suppl:1035–40, http://dx.doi.org/10.1007/bf02718241.
33. Jo YS, Park JH, Lee JK, Heo EY, Chung HS, Kim DK. Discordance between MTB/RIF and real-time tuberculosis-specific polymerase chain reaction assay in bronchial washing specimen and its clinical implications. PLoS One. 2016;11(10):1–12, http://dx.doi.org/10.1371/journal.pone.0164923.
34. Barnard DA, Irusen EM, Bruwer JW, Plekker D, Whitelaw AC, Deetlefs JD, et al. The utility of Xpert MTB/RIF performed on bronchial washings obtained in patients with suspected pulmonary tuberculosis in a high prevalence setting. BMC Pulm Med. 2015;15(1):4–8, http://dx.doi.org/10.1186/s12890-015-0086-z.
35. Le Palud P, Cattoir V, Malbruny B, Magnier R, Campbell K, Oulkhouir Y, et al. Retrospective observational study of diagnostic accuracy of the Xpert® MTB/RIF assay on fiberoptic bronchoscopy sampling for early diagnosis of smear-negative or sputum-scarce patients with suspected tuberculosis. BMC Pulm Med. 2014;14(1):1–7, http://dx.doi.org/10.1186/1471-2466-14-137.
36. Mondoni M, Carlucci P, Di Marco F, Rossi S, Santus P, D'Adda A, et al. Rapid on-site evaluation improves needle aspiration sensitivity in the diagnosis of central lung cancers: A randomized trial. Respiration. 2013;86(1):52–8, http://dx.doi.org/10.1159/000346998.
37. Trisolini R, Patelli M, Ceron L, Gasparini S. Transbronchial needle aspiration. Monaldi Arch Chest Dis - Pulm Ser. 2011;75(1):44–9, http://dx.doi.org/10.1017/9781316084182.011.
38. Gasparini S. Bronchoscopic biopsy techniques in the diagnosis and staging of lung cancer. Monaldi Arch Chest Dis - Pulm Ser. 1997;52(4):392–8.
39. Rivera MP, Mehta AC, Wahidi MM. Establishing the diagnosis of lung cancer: Diagnosis and management of lung cancer, 3rd ed: American college of chest physicians evidence-based clinical practice guidelines. Chest. 2013;143 5 SUPPL:e142S–65S, http://dx.doi.org/10.1378/chest.12-2353.
40. Kaçar N, Tuksavul F, Edipoğlu Ö, Ermete S, Güčlü SZ. Effectiveness of transbronchial needle aspiration in the diagnosis of exophytic endobronchial lesions and submucosal/peribronchial diseases of the lung. Lung Cancer. 2005;50(2):221–6, http://dx.doi.org/10.1016/j.lungcan.2005.05.018.
41. Altin S, Çikrikçioğlu S, Morgül M, Koşar F, Özyurt H. 50 endobronchial tuberculosis cases based on bronchoscopic diagnosis. Respiration. 1997;64(2):162–4, http://dx.doi.org/10.1159/000196662.
42. Wang K-P, Haponik EF, Britt EJ, Khouri N, Erozan Y. Transbronchial needle aspiration of peripheral pulmonary nodules. Chest. 1984;86(6):819–23, http://dx.doi.org/10.1378/chest.86.6.819.

43. Mondoni M, Sotgiu G, Bonifazi M, Dore S, Parazzini EM, Carlucci P, et al. Transbronchial needle aspiration in peripheral pulmonary lesions: A systematic review and meta-analysis. Eur Respir J. 2016, http://dx.doi.org/10.1183/13993003.00051-2016. ERJ-00051-2016.
44. Khandhar SJ, Bowling MR, Flandes J, Gildea TR, Hood KL, Krimsky WS, et al. Electromagnetic navigation bronchoscopy to access lung lesions in 1,000 subjects: First results of the prospective, multicenter NAVIGATE study. BMC Pulm Med. 2017;17(1):59, http://dx.doi.org/10.1186/s12890-017-0403-9.
45. Chao T-Y, Chien M-T, Lie C-H, Chung Y-H, Wang J-L, Lin M-C. Endobronchial ultrasonography-guided transbronchial needle aspiration increases the diagnostic yield of peripheral pulmonary lesions: A randomized trial. Chest. 2009;136(1):229–36, http://dx.doi.org/10.1378/chest.08-0577.
46. Holty JEC, Kuschner WG, Gould MK. Accuracy of transbronchial needle aspiration for mediastinal staging of non-small cell lung cancer: A meta-analysis. Thorax. 2005;60(11):949–55, http://dx.doi.org/10.1136/thx.2005.041525.
47. Silvestri GA, Gonzalez AV, Jantz MA, Margolis ML, Gould MK, Tanoue LT, et al. Methods for staging non-small cell lung cancer: Diagnosis and management of lung cancer, 3rd ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. Chest. 2013;143(5):e211S–50S, http://dx.doi.org/10.1378/chest.12-2355.
48. Bilaçeroğlu S, Günel O, Eris N, Çağırıcı U, Mehta AC. Transbronchial needle aspiration in diagnosing intrathoracic tuberculous lymphadenitis. Chest. 2004;126(1):259–67, http://dx.doi.org/10.1378/chest.126.1.259.
49. Trisolini R, Agli LL, Cancellieri A, Poletti V, Tinelli C, Baruzzi G, et al. The value of flexible transbronchial needle aspiration in the diagnosis of stage I sarcoidosis. Chest. 2003;124(6):2126–30, http://dx.doi.org/10.1378/chest.124.6.2126.
50. Trisolini R, Tinelli C, Cancellieri A, Paioli D, Alifano M, Boaron M, et al. Transbronchial needle aspiration in sarcoidosis: Yield and predictors of a positive aspirate. J Thorac Cardiovasc Surg. 2008;135(4):837–42, http://dx.doi.org/10.1016/j.jtcvs.2007.11.011.
51. Agarwal R, Srinivasan A, Aggarwal AN, Gupta D. Efficacy and safety of convex probe EBUS-TBNA in sarcoidosis: A systematic review and meta-analysis. Respir Med. 2012;106(6):883–92, http://dx.doi.org/10.1016/j.rmed.2012.02.014.
52. Trisolini R, Lazzari Agli L, Tinelli C, De Silvestri A, Scotti V, Patelli M. Endobronchial ultrasound-guided transbronchial needle aspiration for diagnosis of sarcoidosis in clinically unselected study populations. Respirology. 2015;20(2):226–34, http://dx.doi.org/10.1111/resp.12449.
53. Hwangbo B, Lee HS, Lee GK, Lim KY, Lee SH, Kim HY, et al. Transoesophageal needle aspiration using a convex probe ultrasonic bronchoscope. Respirology. 2009;14(6):843–9, http://dx.doi.org/10.1111/j.1440-1843.2009.01590.x.
54. Mondoni M, D'Adda A, Terraneo S, Carlucci P, Radovanovic D, DI Marco F, et al. Choose the best route: Ultrasound-guided transbronchial and transesophageal needle aspiration with echobronchoscope in the diagnosis of mediastinal and pulmonary lesions. Minerva Med. 2015;106(5):13–9.
55. Oki M, Saka H, Ando M, Tsuboi R, Nakahata M, Oka S, et al. Transbronchial vs transesophageal needle aspiration using an ultrasound bronchoscope for the diagnosis of mediastinal lesions: A randomized study. Chest. 2015;147(5):1259–66, http://dx.doi.org/10.1378/chest.14-1283.
56. Vilmann P, Clementsen PF, Colella S, Siemsen M, De Leyn P, Dumonceau JM, et al. Combined endobronchial and oesophageal endosonography for the diagnosis and staging of lung cancer: European society of gastrointestinal endoscopy (ESGE) Guideline, in cooperation with the European respiratory society (ERS) and the European society of tho. Eur Respir J. 2015;46(1):40–60, http://dx.doi.org/10.1183/09031936.00064515.
57. Meena N, Hulett C, Patolia S, Bartter T. Exploration under the dome: Esophageal ultrasound with the ultrasound bronchoscope is indispensible. Endosc Ultrasound. 2016;5(4):254–7, http://dx.doi.org/10.4103/2303-9027.187886.
58. Argento AC, Puchalski J. Convex probe EBUS for centrally located parenchymal lesions without a bronchus sign. Respir Med. 2016;116:55–8, http://dx.doi.org/10.1016/j.rmed.2016.04.012.
59. Skovgaard Christiansen I, Kuijvenhoven JC, Bodtger U, Naur TMH, Ahmad K, Singh Sidhu J, et al. Endoscopic ultrasound with bronchoscope-guided fine needle aspiration for the diagnosis of paraesophageally located lung lesions. Respiration. 2019;97(4):277–83, http://dx.doi.org/10.1159/000492578.
60. Wahidi MM, Herth F, Yasufuku K, Shepherd RW, Yarmus L, Chawla M, et al. Technical aspects of endobronchial ultrasound-guided transbronchial needle aspiration: CHEST guideline and expert panel report. Chest. 2016;149(3):816–35, http://dx.doi.org/10.1378/chest.15-1216.
61. Chaddha U, Hogarth DK, Murgu S. The role of endobronchial ultrasound transbronchial needle aspiration for programmed death ligand 1 testing and next generation sequencing in advanced non-small cell lung cancer. Ann Transl Med. 2019;7 (*August 2019) Ann Transl Med (Focus ''Diagnostic Ther Adv Manag Lung Cancer'')* http://atm.amegroups.com/article/view/24632
62. Biswas A, Leon ME, Drew P, Fernandez-Bussy S, Furtado LV, Jantz MA, et al. Clinical performance of endobronchial ultrasound-guided transbronchial needle aspiration for assessing programmed death ligand-1 expression in nonsmall cell lung cancer. Diagn Cytopathol. 2018;46(5):378–83, http://dx.doi.org/10.1002/dc.23900.
63. Labarca G, Folch E, Jantz M, Mehta HJ, Majid A, Fernandez-Bussy S. Adequacy of samples obtained by endobronchial ultrasound with transbronchial needle aspiration for molecular analysis in patients with non–small cell lung cancer. Systematic review and meta-analysis. Ann Am Thorac Soc. 2018;15(10):1205–16, http://dx.doi.org/10.1513/AnnalsATS.201801-045OC.
64. von Bartheld MB, van Breda A, Annema JT. Complication rate of endosonography (endobronchial and endoscopic ultrasound): A systematic review. Respiration. 2014;87(4):343–51, http://dx.doi.org/10.1159/000357066.
65. Andersen H, Fontana R, Harrison E. Transbronchoscopic lung biopsy in diffuse pulmonary disease. Dis Chest. 1965;48(2):187–92.
66. Dionísio J. Diagnostic flexible bronchoscopy and accessory techniques. Rev Port Pneumol. 2012;18(2):99–106, http://dx.doi.org/10.1016/j.rppneu.2012.01.003.
67. Ofiara LM, Navasakulpong A, Ezer N, Gonzalez AV. The importance of a satisfactory biopsy for the diagnosis of lung cancer in the era of personalized treatment. Curr Oncol. 2012;19(0):16–23, http://dx.doi.org/10.3747/co.19.1062.
68. Govert JA, Dodd LG, Kussin PS, Samuelson WM. A prospective comparison of fiberoptic transbronchial needle aspiration and bronchial biopsy for bronchoscopically visible lung carcinoma. Cancer Cytopathol. 1999;87(3):129–34, http://dx.doi.org/10.1002/(SICI)1097-0142(19990625)87:3<129::AID-CNCR5>3.0.CO;2-G.
69. Dasgupta A, Jain P, Minai OA, Sandur S, Meli Y, Arroliga AC, et al. Utility of transbronchial needle aspiration in the diagnosis of endobronchial lesions. Chest. 1999;115(5):1237–41, http://dx.doi.org/10.1378/chest.115.5.1237.

70. Shorr AF, Torrington KG, Hnatiuk OW. Endobronchial biopsy for sarcoidosis: A prospective study. Chest. 2001;120(1):109–14, http://dx.doi.org/10.1378/chest.120.1.109.
71. Bjermer L, Thunell M, Rosenhall L, Stjernberg N. Endobronchial biopsy positive sarcoidosis: Relation to bronchoalveolar lavage and course of disease. Respir Med. 1991;85(3):229–34, http://dx.doi.org/10.1016/S0954-6111(06)80085-5.
72. Mondoni M, Radovanovic D, Valenti V, Patella V, Santus P. Bronchoscopy in sarcoidosis: Union is strength. Minerva Med. 2015;106 2 Suppl 2:1–7.
73. Ozkaya S, Bilgin S, Findik S, Kök HÇ, Yuksel C, Atıcı AG. Endobronchial tuberculosis: Histopathological subsets and microbiological results. Multidiscip Respir Med. 2012;7(1):34, http://dx.doi.org/10.1186/2049-6958-7-34.
74. Pue CA, Pacht ER. Complications of fiberoptic bronchoscopy at a university hospital. Chest. 1995;107(2):430–2, http://dx.doi.org/10.1378/chest.107.2.430.
75. Bernasconi M, Koegelenberg CFN, Koutsokera A, Ogna A, Casutt A, Nicod L, et al. Iatrogenic bleeding during flexible bronchoscopy: Risk factors, prophylactic measures and management. ERS Monogr. 2017;3(2), http://dx.doi.org/10.1183/23120541.00084-2016.
76. de Fenoyl O, Capron F, Lebeau B, Rochemaure J. Transbronchial biopsy without fluoroscopy: a five year experience in outpatients. Thorax. 1989;44(11), http://dx.doi.org/10.1136/thx.44.11.956, 956 LP - 959.
77. Anders CGT, Johnson MJE, Bush MBA, Matthews CJI. Transbronchial biopsy without fluoroscopy: A seven-year perspective. Chest. 1988;94(3):557–60, http://dx.doi.org/10.1378/chest.94.3.557.
78. Chen A, Chenna P, Loiselle A, Massoni J, Mayse M, Misselhorn D. Radial probe endobronchial ultrasound for peripheral pulmonary lesions. A 5-year institutional experience. Ann Am Thorac Soc. 2014;11(4):578–82, http://dx.doi.org/10.1513/AnnalsATS.201311-384OC.
79. Kato A, Yasuo M, Tokoro Y, Kobayashi T, Ichiyama T, Tateishi K, et al. Virtual bronchoscopic navigation as an aid to CT-guided transbronchial biopsy improves the diagnostic yield for small peripheral pulmonary lesions. Respirology. 2018;23(11):1049–54, http://dx.doi.org/10.1111/resp.13377.
80. Pritchett MA, Schampaert S, de Groot JAH, Schirmer CC, van der Bom I. Cone-Beam CT with augmented fluoroscopy combined with electromagnetic navigation bronchoscopy for biopsy of pulmonary nodules. J Bronchology Interv Pulmonol. 2018;25(4):274–82, http://dx.doi.org/10.1097/LBR.0000000000000536.
81. Eberhardt R, Anantham D, Ernst A, Feller-Kopman D, Herth F. Multimodality bronchoscopic diagnosis of peripheral lung lesions. Am J Respir Crit Care Med. 2007;176(1):36–41, http://dx.doi.org/10.1164/rccm.200612-1866OC.
82. Popovich J, Kvale PA, Eichenhorn MS, Radke JR, Ohorodnik JM, Fine G. Diagnostic accuracy of multiple biopsies from flexible fiberoptic bronchoscopy. Am Rev Respir Dis. 1982;125(5):521–3, http://dx.doi.org/10.1164/arrd.1982.125.5.521.
83. Gaeta M, Pandolfo I, Volta S, Russi EG, Bartiromo G, Girone G, et al. Bronchus sign on CT in peripheral carcinoma of the lung: Value in predicting results of transbronchial biopsy. Am J Roentgenol. 1991;157(6):1181–5, http://dx.doi.org/10.2214/ajr.157.6.1950861.
84. Naidich DP, Sussman R, Kutcher WL, Aranda CP, Garay SM, Ettenger NA. Solitary pulmonary nodules: CT-bronchoscopic correlation. Chest. 1988;93(3):595–8, http://dx.doi.org/10.1378/chest.93.3.595.
85. Broaddus C, Dake MD, Stulbarg MS, Blumenfeld W, Hadley WK, Golden JA, et al. Bronchoalveolar lavage and transbronchial biopsy for the diagnosis of pulmonary infections in the acquired immunodeficiency syndrome. Ann Intern Med. 1985;102(6):747–52, http://dx.doi.org/10.7326/0003-4819-102-6-747.
86. Jacomelli M, Silva PRAA, Rodrigues AJ, Demarzo SE, Seicento M, Figueiredo VR. Bronchoscopy for the diagnosis of pulmonary tuberculosis in patients with negative sputum smear microscopy results. J Bras Pneumol. 2012;38:167–73.
87. Chan T-C, Pai P-L, Shaw S-L, Fan I-C. Spatiotemporal evolution of market towns in the jiangnan area during the ming-qing dynasties of China. Hist Methods A J Quant Interdiscip Hist. 2015;48(2):90–102, http://dx.doi.org/10.1080/01615440.2014.995783.
88. Theron G, Zijenah L, Chanda D, Clowes P, Rachow A, Lesosky M, et al. Feasibility, accuracy, and clinical effect of point-of-care Xpert MTB/RIF testing for tuberculosis in primary-care settings in Africa: A multicentre, randomised, controlled trial. Lancet. 2014;383(9915):424–35, http://dx.doi.org/10.1016/S0140-6736(13)62073-5.
89. Mondoni M, Fois A, Centanni S, Sotgiu G. Could BAL Xpert MTB/RIF replace transbronchial lung biopsy everywhere for suspected pulmonary TB patients? Int J Tubercolosis Lung Dis. 2016;20(8):2016.
90. Higenbottam T, Stewart S, Penketh A, Wallwork J. Transbronchial lung biopsy for the diagnosis of rejection in heart-lung transplants patients. Transplantation. 1988;46(4):532–9.
91. Kebbe J, Abdo T. Interstitial lung disease: The diagnostic role of bronchoscopy. J Thorac Dis. 2017;9 Suppl 10:S996–1010, http://dx.doi.org/10.21037/jtd.2017.06.39.
92. Leslie KO, Gruden JF, Parish JM, Scholand MB. Transbronchial biopsy interpretation in the patient with diffuse parenchymal lung disease. Arch Pathol Lab Med. 2007;131(3):407–23, http://dx.doi.org/10.1043/1543-2165(2007)131[407:TBIITP]2.0.CO;2.
93. Rohatgi PK, Kuzmowych TV, Delaney MD. Indications for transbronchial lung biopsy in the diagnosis of intrathoracic sarcoidosis. Respiration. 1981;42(3):155–60, http://dx.doi.org/10.1159/000194422.
94. Chapman JT, Mehta AC. Bronchoscopy in sarcoidosis: Diagnostic and therapeutic interventions. Curr Opin Pulm Med. 2003;9(5):402–7, http://dx.doi.org/10.1097/00063198-200309000-00011.
95. Ensminger SA, Prakash UBS. Is bronchoscopic lung biopsy helpful in the management of patients with diffuse lung disease? Eur Respir J. 2006;28(6):1081–4, http://dx.doi.org/10.1183/09031936.06.00013106.
96. de Jaeger A, Litalien C, Lacroix J, Guertin M-C, Infante-Rivard C. Protected specimen brush or bronchoalveolar lavage to diagnose bacterial nosocomial pneumonia in ventilated adults: A meta-analysis. Crit Care Med. 1999;27(11):2548–60.
97. Chastre J, Combes A, Luyt C-E. The invasive (quantitative) diagnosis of ventilator-associated pneumonia. Respir Care. 2005;50(6), 797 LP - 812 http://rc.rcjournal.com/content/50/6/797.abstract
98. Hetzel J, Eberhardt R, Herth FJF, Petermann C, Reichle G, Freitag L, et al. Cryobiopsy increases the diagnostic yield of endobronchial biopsy: A multicentre trial. Eur Respir J. 2012;39(3), http://dx.doi.org/10.1183/09031936.00033011, 685 LP - 690.
99. Sehgal IS, Dhooria S, Behera D, Agarwal R. Use of cryoprobe for removal of a large tracheobronchial foreign body during flexible bronchoscopy. Lung India. 2016;33(5):543–5, http://dx.doi.org/10.4103/0970-2113.188978.
100. Fruchter O, Kramer MR. Retrieval of various aspirated foreign bodies by flexible cryoprobe: In vitro feasibility study. Clin Respir J. 2015;9(2):176–9, http://dx.doi.org/10.1111/crj.12120.
101. Babiak A, Hetzel J, Krishna G, Fritz P, Moeller P, Balli T, et al. Transbronchial cryobiopsy: A new tool

for lung biopsies. Respiration. 2009;78(2):203–8, http://dx.doi.org/10.1159/000203987.
102. Hetzel J, Maldonado F, Ravaglia C, Wells AU, Colby TV, Tomassetti S, et al. Transbronchial cryobiopsies for the diagnosis of diffuse parenchymal lung diseases: Expert statement from the cryobiopsy working group on safety and utility and a call for standardization of the procedure. Respiration. 2018;95(3):188–200, http://dx.doi.org/10.1159/000484055.
103. Rubio ER, Le SR, Whatley RE, Boyd MB. Cryobiopsy: Should this be used in place of endobronchial forceps biopsies? Biomed Res Int. 2013;2013, http://dx.doi.org/10.1155/2013/730574.
104. Boyd M, Sahebazamani M, Le S, Rubio E. The safety of cryobiopsy in diagnosing carcinoid tumors. J Bronchol Interv Pulmonol. 2014;21(3):234–6, http://dx.doi.org/10.1097/LBR.0000000000000079.
105. Schuhmann M, Bostanci K, Bugalho A, Warth A, Schnabel PA, Herth FJF, et al. Endobronchial ultrasound-guided cryobiopsies in peripheral pulmonary lesions: A feasibility study. Eur Respir J. 2014;43(1), http://dx.doi.org/10.1183/09031936.00011313, 233 LP - 239.
106. Taton O, Bondue B, Gevenois PA, Remmelink M, Leduc D. Diagnostic yield of combined pulmonary cryobiopsies and electromagnetic navigation in small pulmonary nodules. Pulm Med. 2018;2018, http://dx.doi.org/10.1155/2018/6032974.
107. Nasu S, Okamoto N, Suzuki H, Shiroyama T, Tanaka A, Samejima Y, et al. Comparison of the utilities of cryobiopsy and forceps biopsy for peripheral lung cancer. Anticancer Res. 2019;39(10):5683–8, http://dx.doi.org/10.21873/anticanres.13766.
108. Herath S, Yap E. Novel hybrid cryo-radial method: An emerging alternative to CT-guided biopsy in suspected lung cancer. A prospective case series and description of technique. Respirol Case Reports. 2018;6(2):e00287, http://dx.doi.org/10.1002/rcr2.287.
109. Arimura K, Tagaya E, Akagawa H, Nagashima Y, Shimizu S, Atsumi Y, et al. Cryobiopsy with endobronchial ultrasonography using a guide sheath for peripheral pulmonary lesions and DNA analysis by next generation sequencing and rapid on-site evaluation. Respir Investig. 2019;57(2):150–6, http://dx.doi.org/10.1016/j.resinv.2018.10.006.
110. Arimura K, Kondo M, Nagashima Y, Kanzaki M, Kobayashi F, Takeyama K, et al. Comparison of tumor cell numbers and 22C3 PD-L1 expression between cryobiopsy and transbronchial biopsy with endobronchial ultrasonography-guide sheath for lung cancer. Respir Res. 2019;20(1):185, http://dx.doi.org/10.1186/s12931-019-1162-3.
111. Colby TV, Tomassetti S, Cavazza A, Dubini A, Poletti V. Transbronchial cryobiopsy in diffuse lung disease: Update for the pathologist. Arch Pathol Lab Med. 2017;141(7):891–900, http://dx.doi.org/10.5858/arpa.2016-0233-RA.
112. Tomassetti S, Wells AU, Costabel U, Cavazza A, Colby TV, Rossi G, et al. Bronchoscopic lung cryobiopsy increases diagnostic confidence in the multidisciplinary diagnosis of idiopathic pulmonary fibrosis. Am J Respir Crit Care Med. 2016;193(7):745–52, http://dx.doi.org/10.1164/rccm.201504-0711OC.
113. Maldonado F, Danoff SK, Wells AU, Colby TV, Ryu JH, Liberman M, et al. Transbronchial cryobiopsy for the diagnosis of interstitial lung diseases: CHEST guideline and expert panel report. Chest. 2020;157(4):1030–42, http://dx.doi.org/10.1016/j.chest.2019.10.048.
114. Ravaglia C, Wells AU, Tomassetti S, Gurioli C, Gurioli C, Dubini A, et al. Diagnostic yield and risk/benefit analysis of trans-bronchial lung cryobiopsy in diffuse parenchymal lung diseases: A large cohort of 699 patients. BMC Pulm Med. 2019;19(1):16, http://dx.doi.org/10.1186/s12890-019-0780-3.
115. Ravaglia C, Wells AU, Tomassetti S, Dubini A, Cavazza A, Piciucchi S, et al. Transbronchial lung cryobiopsy in diffuse parenchymal lung disease: Comparison between biopsy from 1 segment and biopsy from 2 segments - diagnostic yield and complications. Respiration. 2017;93(4):285–92, http://dx.doi.org/10.1159/000456671.
116. Johannson KA, Marcoux VS, Ronksley PE, Ryerson CJ. Diagnostic yield and complications of transbronchial lung cryobiopsy for interstitial lung disease. A systematic review and metaanalysis. Ann Am Thorac Soc. 2016;13(10):1828–38, http://dx.doi.org/10.1513/AnnalsATS.201606-461SR.
117. Romagnoli M, Colby TV, Berthet J-P, Gamez AS, Mallet J-P, Serre I, et al. Poor concordance between sequential transbronchial lung cryobiopsy and surgical lung biopsy in the diagnosis of diffuse interstitial lung diseases. Am J Respir Crit Care Med. 2019;199(10):1249–56, http://dx.doi.org/10.1164/rccm.201810-1947OC.
118. Troy LK, Grainge C, Corte TJ, Williamson JP, Vallely MP, Cooper WA, et al. Diagnostic accuracy of transbronchial lung cryobiopsy for interstitial lung disease diagnosis (COLDICE): A prospective, comparative study. Lancet Respir Med. 2019;(November), http://dx.doi.org/10.1016/S2213-2600(19)30342-X.
119. Gnass M, Filarecka A, Pankowski J, Soja J, Bugalho A, Szlubowski A. Transbronchial lung cryobiopsy guided by endobronchial ultrasound radial miniprobe in interstitial lung diseases: Preliminary results of a prospective study. Polish Arch Intern Med. 2018;128(4):259–62, http://dx.doi.org/10.20452/pamw.4253.
120. Wijmans L, Bonta PI, Rocha-Pinto R, de Bruin DM, Brinkman P, Jonkers RE, et al. Confocal laser endomicroscopy as a guidance tool for transbronchial lung cryobiopsies in interstitial lung disorder. Respiration. 2019;97(3):259–63, http://dx.doi.org/10.1159/000493271.
121. Zhou G, Ren Y, Li J, Yang T, Su N, Zhao L, et al. Safety and diagnostic efficacy of cone beam computed tomography-guided transbronchial cryobiopsy for interstitial lung disease: A cohort study. Eur Respir J. 2020;(March), http://dx.doi.org/10.1183/13993003.00724-2020.
122. Krimsky WS, Pritchett MA, Lau KKW. Towards an optimization of bronchoscopic approaches to the diagnosis and treatment of the pulmonary nodules: A review. J Thorac Dis. 2018;10 Suppl 14:S1637–44, http://dx.doi.org/10.21037/jtd.2018.04.38.
123. Ray AS, Li C, Murphy TE, Cai G, Araujo KLB, Bramley K, et al. Improved diagnostic yield and specimen quality with ebus-guided forceps biopsies: A retrospective analysis. Ann Thorac Surg. 2019;(November), http://dx.doi.org/10.1016/j.athoracsur.2019.08.106.
124. Bramley K, Pisani MA, Murphy TE, Araujo KL, Homer RJ, Puchalski JT. Endobronchial ultrasound-guided cautery-assisted transbronchial forceps biopsies: Safety and sensitivity relative to transbronchial needle aspiration. Ann Thorac Surg. 2016;101(5):1870–6, http://dx.doi.org/10.1016/j.athoracsur.2015.11.051.
125. Mehta RM, Aurangabadbadwalla R, Singla A, Loknath C, Munavvar M. Endobronchial ultrasound-guided mediastinal lymph node forceps biopsy in patients with negative rapid-on-site-evaluation: A new step in the diagnostic algorithm. Clin Respir J. 2020;14(4):314–9, http://dx.doi.org/10.1111/crj.13133.

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LETTER TO THE EDITOR

Clinical and radiological improvement of protracted COVID-19 and Good syndrome secondary to advanced thymoma

Good's syndrome (GS) is a rare clinical entity that affects both sexes over 40 years of age. GS is characterized by the presence of thymoma associated with hypogammaglobulinemia with loss of B cells in most cases. In addition, altered T cell function in the presence of normal or elevated T cell counts has been reported. This disease produces severe immunodeficiency, and an increased incidence of opportunistic and viral infections may complicate the clinical course of affected patients. Few cases of SARS-CoV-2 infection and GS have been reported with different outcomes.[1,2] Although immunocompromised patients are prone to prolonged COVID-19 courses, however, at the same time they might be protected from severe disease mediated by an overstimulation of the immune system.[3] We report a prolonged COVID-19 infection with SARS-CoV-2 viremia in a GS patient who recovered after standard treatment concomitant with convalescent plasma.

The index patient was a 44-year-old male diagnosed with advanced thymoma (type B1) who in the past 6 years had undergone surgical excision and adjuvant mediastinal radiotherapy. After 2 years of stable results, he developed recurrent metastatic disease in multiple sites, for which he underwent multiple surgeries and radiotherapy. On presentation, he was on systemic treatment with gemcitabine and capecitabine for 5 months. The patient was admitted to our institution due to worsening dyspnea, non-productive cough and nasopharyngeal swab indicating positive for SARS-CoV-2 by polymerase chain reaction (PCR). He also reported a SARS-CoV-2 infection 8 months before, with no residual symptoms. Mild hypoxemia (PaO_2 56 mmHg) with hypocapnia ($PaCO_2$ 32 mmHg) and respiratory alkalosis (pH 7.48) were present. While a moderate increase in CRP (3.37 mg/dl; normal values $<$ 0.3 mg/dl) was noted, D-dimer levels (289 ng/mL; normal values $<$500), LDH, CK, renal and liver function were within normal range. Low flow O_2 with nasal cannula (FiO_2 24%) was started for normalization of hypoxemia and hypocapnia.

Chest computed tomography (CT) showed chronic actinic alterations in the mediastinum and left lung, with architectural distortion (unchanged in comparison to previous exams), and ground-glass opacities distributed in the medullary and peripheral regions of the lungs. Compared to the CT performed 8 months before (during the first COVID-19 diagnosis), the pulmonary changes were present in distinct lung fields and with greater extension (Fig. 1). A diagnostic workup was started to rule out infectious causes. Drug pneumonitis was deemed unlikely as the lung parenchymal changes preceded acute onset.

A bronchoscopy with bronchoalveolar lavage (BAL) was performed with positive SARS-CoV2 PCR. BAL cellularity demonstrated 5% macrophages, 92% neutrophils, 3% lymphocytes, 0% eosinophils. Multiplex PCR for other respiratory viruses, Pneumocystis *jiroveci* PCR, bacteria culture, acid-fast bacilli smear and galactomannan on BAL were negative. Additionally, cytomegalovirus (CMV) PCR was positive (viral load of 2350 IU/mL). Gancyclovir was initiated for suspected superimposed CMV pneumonitis and an immunodeficiency screening was ordered. Serologic test for SARS-Cov-2 antibodies was negative. The first immunological evaluation showed a reduction of the gamma-globulin peak in serum electrophoresis with reduced immunoglobulin serum levels. The evaluation of peripheral lymphocyte subsets showed inversion of the CD4/CD8 ratio with no peripheral B cells. Immunoglobulin serum levels were extremely low for all classes. The immunological work-up is summarized in Table 1. With these immunological findings associated with thymoma, the patient was diagnosed with GS and was put on facilitated subcutaneous immunoglobulin replacement treatment. Additionally, dexamethasone 6 mg every 24 h was initiated due to hypoxemia and accompanying COVID-19 infection.

The patient's symptoms and hypoxemia improved with this referred treatment. However, after seven days, the symptoms worsened, with CT showing an increase in the ground glass and reticular opacifications and consolidations (Fig. 2A). A new BAL was performed, which revealed cellularity of 4% macrophages, 92% neutrophils, 4% lymphocytes, 0% eosinophils, besides persistence of SARS-CoV-2 PCR with low cycle threshold value (CT=13), suggesting high viral load. CMV BAL PCR remained positive (11600 IU/mL). Unexpectedly, *Pneumocystis jiroveci* PCR was positive (334,000 copies/mL). Sulfamethoxazole/trimethoprim was initiated to treat Pneumocystis pneumonia (PCP) and was later replaced by primaquine plus clindamycin due to hepatotoxicity. Corticosteroid therapy was maintained throughout the treatment.

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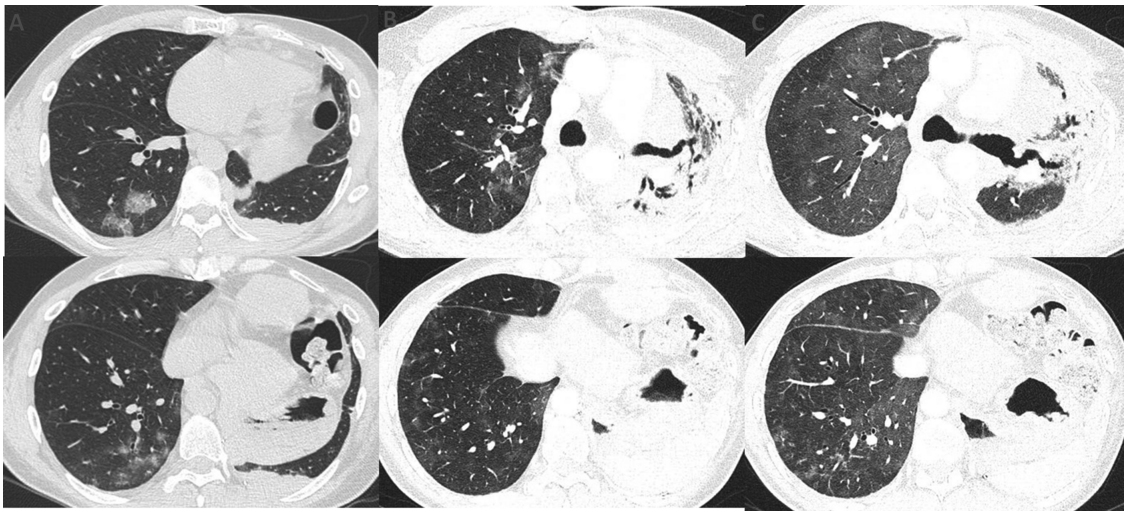

Fig. 1 CT axial slices during the evolution of the case. **(A *first column*)** CT image from the first diagnosis of SARS-CoV-2 infection 8 months before showing ground glass opacities in the right lower lobe. **(B *middle column*)** CT image two months later, before the chemotherapy started, showing ground glass opacities in distinct lung fields. **(C *third column*)** CT image revealing persistent ground glass opacities with greater extension on patient's presentation to emergency care.

After 21 days of treatment for PCP and CMV, the patient's symptoms improved and O_2 saturation returned to 94% in ambient air. However, a repeat CT revealed new ground-glass opacities in the right lower lobe. A third BAL was performed, demonstrating cellularity of 53% macrophages, 31% neutrophils, 16% lymphocytes, 0% eosinophils, negative CMV and Pneumocystis PCR; COVID-19 PCR remained positive, with a persistent low cycle threshold. Based on clinical stability, the patient was followed up on conservative management with a gradual taper of steroids.

Table 1 Patient's immunological evaluation for diagnosis of GS.

	Index patient	Normal range for age
WBC (cells/mm3)	9540 (cells/mm3)	4500-10800 (cells/mm3)
ANC (cells/mm3)	5370 (cells/mm3)	1500-8000 (cells/mm3)
ALC (cells/mm3)	3590 (cells/mm3)	1000-5200 (cells/mm3)
Hb (g/dL)	11.2 (g/dL)	12.0-16.0 (g/dL)
PLTs (cells/mm3)	360000 (cells/mm3)	100000-400000 (cells/mm3)
B cells (CD20+)	0%	8-18%
T cells (CD4+)	22.9%	31-56%
T cells (CD8+)	72.8%	17-41%
CD4+/CD8+	0.3	0.9-2.6
T cells (CD3+)	97%	59-81%
IgA	5 mg/dL	50-400 mg/dL
IgG	151 mg/dL	600-1500 mg/dL
IgM	4 mg/dL	50-300 mg/dL
SARS-CoV-2 IgG antibody test	0.1	$<$ 0.6 = negative
SARS-CoV-2 IgM antibody test	0.1	$<$ 0.6 = negative

Five days after the last bronchoscopy, the patient presented with new-onset hypoxemia, PaO_2/FiO_2 was 170, requiring oxygen through a high-flow nasal catheter. Repeat chest CT angiography showed small subsegmental thromboembolism and revealed new ground-glass opacities and consolidation (Fig. 2B). A diagnosis of protracted COVID-19 was considered. The patient was treated with four units of convalescent COVID-19 donor plasma over 48 h. Corticosteroid tapering was reversed, and its dosage increased. Additionally, anticoagulation therapy was started due to pulmonary embolism. Over the next 24 h, the patient showed substantial clinical improvement with gradual resolution of the hypoxemia, with no requirement for oxygen support four days after the first plasma infusion. A new CT demonstrated a significant reduction of pulmonary compromise (Fig. 2C).

Although not universally, most recent studies define protracted COVID as a disease course with a duration of symptoms beyond the usual natural history (i.e. more than 4 weeks) accompanied by positive respiratory sample PCR as evidence of persistent viral shedding.[1—4] Protracted COVID-19 with viral RNA shedding over 15 days has been described in elderly or immunocompromised patients and severe cases.[4—7] Convalescent plasma therapy (CPT) is a passive polyclonal antibody administration to provide immediate immunity in an attempt to improve the survival rates in severe acute respiratory syndromes of viral etiology (including COVID-19).[4,5] According to some reports, CPT with anti—SARS-CoV-2 antibodies could be a promising approach in the context of protracted COVID-19 in individuals unable to mount a specific humoral response to SARS-CoV-2.[5,6] On the contrary, a randomized trial demonstrated that CPT in hospitalized patients with COVID-19 pneumonia overall did not reduce the progression to severe respiratory failure or death within 30 days.[8]

The present case constitutes the second case report in literature showing prolonged SARS-CoV-2 infection due to GS, and the first one demonstrating clinical and radiological response following a treatment plan that also included CPT infusion. London et al.[9] presented a 41-year-old woman with thymoma and GS, and reported improved COVID-19

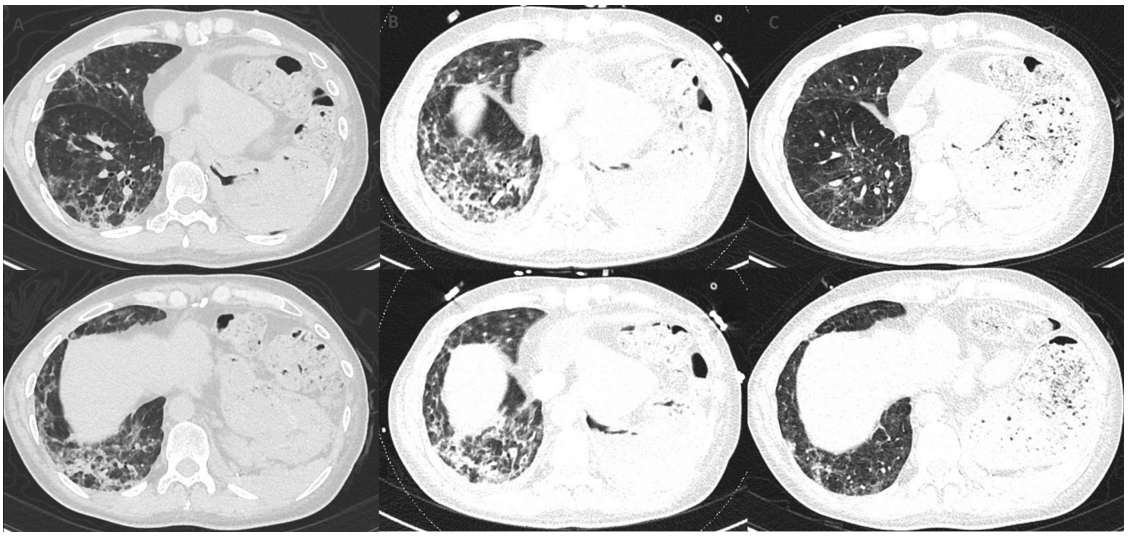

Fig. 2 CT axial slices during the evolution of the case. **(A *first column*)** CT after treatment for CMV and intravenous immunoglobulin demonstrating new ground glass opacities along with consolidations and reticular opacities. **(B *middle column*)** Despite treatment for both CMV and PCP, CT image reveals worsening in pulmonary compromise. **(C *third column*)** CT demonstrating significant radiological improvement after CPT followed by substantial clinical improvement in a short period of time.

symptoms after 4 days of therapy and negative nasopharyngeal PCR following CPT. Our case demonstrates the imaging response in serial CT scans, with significant reduction of ground-glass opacities with standard treatment concomitant with CPT reinforcing the role of ineffective humoral response in protracted forms of COVID-19.

Authors contributions

Milena Tenorio Cerezoli and Felipe Marques da Costa were the pulmonologists conducting the clinical case. João Antônio Gonçalves Garreta Prats was the collaborator infectologist. William Nassib William Junior and Diego Vinícius Gonçalves Santana were the consultant oncologists. Augusto Kreling Medeiros and Ulysses S. Torres were the clinical radiologists accompanying the case. This very complex case demanded a multidisciplinary approach. All the authors, in conjunction, after a meeting of the multidisciplinary team, opted for conceiving this clinical case study and submitting it to Pulmonology. The preliminary draft was written by Augusto Kreling Medeiros, followed by revision and proofreading by all authors, who added insights/ corrections regarding their area of expertise. A final revision was performed by Ulysses S. Torres, with approvation by all authors.

Conflicts of interest

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References

1. Pozzi MR, Baronio M, Janetti MB, Gazzurelli L, Moratto D, Chiarini M, et al. Fatal SARS-CoV-2 infection in a male patient with Good's syndrome. Clin Immunol. 2021;223:108644.
2. Cos Esquius ML, Montesinos IL, Martinez RG, Nuñez JE, Rabasco AC, González BS, et al. Severe COVID-19 pneumonia in Good syndrome with a favorable outcome. Clin Immunol. 2022;235:108789.
3. Guevara-Hoyer K, Antrás JF, Gastardi JC, Sánchez-Ramón S. Immunodeficiency and thymoma in Good syndrome: two sides of the same coin. Immunol Lett. 2021;231:11–7.
4. Tiberghien P, de Lamballerie X, Morel P, Gallian P, Lacombe K, Yazdanpanah Y. Collecting and evaluating convalescent plasma for COVID-19 treatment: why and how? Vox Sang. 2020;115: 488–94.
5. Clark E, Guilpain P, Filip IL, Pansu N, Le Bihan C, Cartron G, et al. Convalescent plasma for persisting COVID-19 following therapeutic lymphocyte depletion: a report of rapid recovery. Br J Haematol. 2020;190(3):e154–6.
6. Moutinho-Pereira S, Calisto R, Sabio F, Guerreiro L. High-titre convalescent plasma therapy for an immunocompromised patient with systemic lupus erythematosus with protracted SARS-CoV-2 infection. BMJ Case Rep. 2021;14(8):e244853.
7. Betrains A, Godinas L, Woei-A-Jin FJSH, Rosseels W, Van Herck Y, Lorent N, et al. Convalescent plasma treatment of persistent severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) infection in patients with lymphoma with impaired humoral immunity and lack of neutralising antibodies. Br J Haematol. 2021;192(6):1100–5.
8. Menichetti F, Popoli P, Puopolo M, Alegiani SS, Tiseo G, Bartoloni A, et al. Effect of high-titer convalescent plasma on progression to severe respiratory failure or death in hospitalized patients with COVID-19 pneumonia: a randomized clinical trial. JAMA Netw Open. 2021;4(11):e2136246.
9. London J, Boutboul D, Lacombe K, Pirenne F, Heym B, Zeller V, et al. Severe COVID-19 in patients with B cell alymphocytosis and response to convalescent plasma therapy. J Clin Immunol. 2021;41(2):356–61.

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LETTER TO THE EDITOR

COVID-19 vaccine booster in healthcare workers - reasons for refusing

World Health Organization declared COVID-19, a disease caused by SARS-CoV-2, a pandemic on March 11, 2020.[1] For this reason, in order to combat the pandemic, vaccines against COVID-19 were developed and administered at an unprecedented speed.[2]

Although a third of the population of the European Union was still not vaccinated at the beginning of October 2021, the need to administer booster doses of vaccines against COVID-19 was already under public discussion.[2]

In Portugal, the booster dose started in November 2021; the first phase included health professionals, the population over 80 years old and the population between 50 and 79 years old with an increased risk pathology.[3]

There is currently some preliminary evidence about the increased effectiveness of the booster dose, concerning protection against severe COVID-19 and the decrease in the risk of transmissibility.[4] However, the social uncertainty regarding the efficacy and safety of the vaccine, may be associated with a reduction in the 'population's motivation to take the booster dose.

In the first stage of vaccination of the booster dose against COVID-19, a study was conducted at IPO Porto, where healthcare workers (HCW) who refused the booster dose were identified. Then, an inquiry was carried out by telephone to record the reasons for the refusal.

A database was created to keep records of all HCW

The Institution's Ethics Committee approved the study and informed consent was obtained from all participants.

This is a retrospective observational and the data was colleted from HCW from November 25 to December 3, 2021.

In this first stage of vaccination, 1376 booster doses of the BNT162b2 vaccine were administered, which 57 HCW refused. Professionals who had SARS-CoV-2 infection or complete vaccination schedule for less than five months were excluded. In Table 1, the reasons for refusal are identified.

Of the 57 HCW included in the study, about 93% were female and 7% male. Regarding the professional category, it was mostly nurses and operational assistants who refused the vaccine administration (54.4% and 28.1%, respectively); however, these are also the most prevalent professional categories in the institution. 15 HCW reported more than one reason for hesitation. The 31 nurses who refused correspond to 3.3% of all nurses called to get the booster dose. Of the 318 operational assistants called, 16 refused, corresponding to 5%.

Table 1 — Reasons for refusing the booster dose of the COVID-19 vaccine.

	n (%)
Acute disease	17 (29,8)
Hesitation	15 (26,3)
- Adverse effects in the previous vaccination scheme	10 (31,3)
- Considering the administration of the booster dose premature	5 (15,6)
- Doubts about the increased effectiveness of the booster dose	4 (12,5)
- Fear of short or long term applications to health	4 (12,5)
- Not considering being in a risk group	4 (12,5)
- Doubts about the safety of a new administration	3 (9,4)
- Adverse effects of booster dose on co-workers, friends or family	2 (6,3)
Temporary incapacity due to chronic disease	10 (17,5)
Pregnancy	6 (10,5)
Risk pregnancy	3 (5,2)
Absence due to vacation	2 (3,5)
Recent rubella vaccination	1 (1,8)
Autoimmune disease, therefore under immunosuppressants	1 (1,8)
Recent abortion	1 (1,8)
Breastfeeding period	1 (1,8)

Thus, there were a small number of HCW who refused the booster dose. Of the total of 1468 professionals called, 57 refused, corresponding to about 3.9%.

Public trust is a fundamental element of vaccination interventions and policies that achieve high coverage and the most effective way to increase COVID-19 vaccine uptake is to make vaccination straightforward, so that it acts on existing intentions to vaccinate. [5,6]

Pfizer has reported that a third dose of its vaccine provided levels of neutralizing antibodies against omicron that were similar to those seen after two doses of the vaccine against the original virus (wildtype).[7]

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These results can be explained by the fact that in Portugal, a campaign is being carried out in the media to inform the population about the efficacy and safety of the vaccine. The significant investment in the construction and adaptation of spaces for vaccination centres in all regions of the country is notable, thus contributing to excellent results in the vaccination coverage of the Portuguese population. In addition, the Occupational Medicine service clarified the HCW and monitored the vaccination process in the institution, as had already happened in the previous vaccination process (with 92% of healthcare workers being vaccinated in the institution), ensuring information and safety.

According to data from the general health department in Portugal, it is known that by 7 January 2022, more than 3.2 million people have had the booster dose against COVID-19. There are still insufficient data to assess the prevalence of refusal of this dose in the general population.

This study shows recent data on the booster dose, allowing us to conclude that we are all on the right track to fight the pandemic. However, the sample is small and, therefore, further studies will be needed to obtain more information about the `population's behavior regarding booster dose.

References

1. The WHO Just Declared Coronavirus COVID-19 a Pandemic Time. Available online: https://time.com/5791661/who-coronavirus-pandemic-declaration/ (accessed 8 January 2022).
2. Rzymski P, Camargo CA, Fal A, et al. COVID-19 vaccine boosters: the good, the bad, and the ugly. Vaccines. 2021;9:1299.. https://doi.org/10.3390/vaccines9111299 (accessed 8 January 2022).
3. Direção Geral da Saúde (DGS). Norma n° 002/2021- Campanha de Vacinação Contra a COVID-19. (accessed 8 January 2022).
4. Bar-On YM, Goldberg Y, Mandel M, et al. Protection of BNT162b2 vaccine booster against Covid-19 in Israel. N Engl J Med. 2021;385:1393—400. 10.1056/NEJMoa2114255 (accessed 8 January 2022).
5. Larson H, Heymann D. Public health response to influenza A (H1N1) as an opportunity to build public trust. JAMA. 2010;303 (3):271—2. 10.1001/jama.2009.2023 (accessed 8 January 2022).
6. Brewer N, Chapman G, Rothman A, et al. Increasing vaccination: putting psychological science into action. Psychol Sci Public Interest. 2017;18(3):149—207. 10.1177/1529100618760521 (accessed 8 January 2022).
7. Mahase E. Covid-19: omicron and the need for boosters. BMJ. 2021;375:n3079.. http://dx.doi.org/10.1136/bmj.n3079 (accessed 8 January 2022).

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LETTER TO THE EDITOR

Laboratory activity testing the lung function during 16 months of the Covid-19 pandemic

Dear Editor

Problems related to the clinical and organizational management of Covid-19 by pulmonologists have been vast.[1] Since March 2020, because of the SARS CoV2 correlated pandemic,[2] all lung function testing laboratories (LFTLs) have been reconfigured[3] and examinations reduced or suspended. This reorganization has forced to changes in tasks, roles, protocols and scheduling of the LFTLs, starting from the initial proposed indications and suggestions.[4,5]

This descriptive report aims to explore and measure, retrospectively, the impact of the relocation of an LFTL service, giving details on routine measures collected by an Integrated Health Care Record database.

We retrospectively (study approved by Ethics Committee CEC 2440; March 12th, 2020) undertook a revision of the number and quality of the examinations (Forced Expiratory Volume at first second-FEV_1, Residual Volume-RV, arterial blood gases, Lung carbon monoxyde diffusion-DLCO) over 16 months (27th May 2020—11th October 2021, i.e. pandemic period) with respect to data from a corresponding (1st November 2018 - 29th February 2020) pre-pandemic period. Our hospital is a referral institution for pulmonary rehabilitation, performing in and outpatient diagnosis for post-acute, chronic subjects [6,7] and post-Covid-19 population[8] located in the Lombardy region, with high specialization and a multidisciplinary team. The particular mission of the hospital is linked to a) outpatient diagnosis for chronic respiratory diseases, preoperative needs and post-Covid-19 follow-up; b) care of patients with complex disabilities and needs, suffering from severe chronic and exacerbated respiratory failure including pneumonia post-Covid-19 sequelae.

Patients attended our outpatient LFTL service having been referred by their general practitioner or by an external pulmonologist for lung and disability assessment. Inpatients were transferred to our hospital directly or within 30 days following an acute illness, that had previously required either acute hospital care or home management by the general practitioner.

Our LFTL has undergone a profound transformation as imposed by the Italian Health Authorities and was interrupted from March to May 2020 before re-opening; there was no reduction or increase in human resources (doctors and technicians). Specific instructions and suggestions were introduced during the pandemic, following Scientific Societies,[4,5] for organization, waiting areas, re-organizing testing, tests procedure, increase of testing times, need for more consumables, use of UV sanitization, chemical sanitization of the plethysmography box between patients, use of antimicrobial filters during maneuvers, protection of healthcare workers during lung function testing, training in use of medical masks such as FFP2,[9] frequent hand hygiene, interpersonal distancing and re-indications for lung function tests. More space in waiting rooms, dedicated slots for exams, differentiation of spirometers, increased ventilation times to face the risk of contagion and requests for a negative swab for patients before accessing the LFTL have also been provided.

Table 1 shows LFTL volume activities. During the pandemic time, we performed 3663 LFTL tests. This is 2.5% more than pre-pandemic, resulting in a decrease in inpatient tests of 16.4% and an increase of outpatient tests by 23.8%. In pre-Covid time, DLCO was 7.5% of total examinations, rising during the pandemic time to 19.3% (17.2% of outpatient and 22.0% of inpatient tests). During the pandemic time 19.5% of total examinations referred to Covid-19 patients, DLCO being the most commonly requested test. The important decrease in inpatient requests was due to the reduction of availability of rehabilitative beds and came along with a significant increase in outpatient testing to determine and quantify long Covid-19.

During the pandemic period, 277 post Covid-19 patients, both inpatient and outpatients, attended our LFTL. One year after infection, these patients presented pathological values (<80% of predicted values) in 22%, 20%, and 63% of the cases for FEV_1, RV, and DLCO respectively.

To reduce the spread of the severe acute respiratory Coronavirus 2 syndrome, many LFTLs have been closed or significantly reduced their testing capacity,[3] while respiratory physicians, researchers, and administrators have begun to consider how different options to conventional pulmonary function testing could be integrated into the patients'care.[3] Since the pandemic beginning, scientific Societies have had a justified position of extreme caution in the execution of LFT.[4,5] The same Societies have provided a set of information about the risks and recommendations for LFT, updating

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Table 1 Lung function testing laboratory (LFTL) volume activities.

	Pre-pandemic time			Pandemic time					
	LFTL tests			LFTL tests			LFTL tests on COVID-19 patients		
	Total	Outpatient	Inpatient	Total	Outpatient	Inpatient	Total	Outpatient	Inpatient
FEV_1, n	808	322	486	609 [#]	346	263	153	92	61
% of the total examination	22.6			16.6			21.4		
RV, n	1373	772	601	1370 [*]	836	534	178	120	58
% of the total examination	38.4			37.4			25.0		
DLCO, n	267	98	169	705 [@]	359	346	240	143	97
% of the total examination	7.5			19.3			33.7		
PaO_2/FiO_2, n	1124	495	629	979 [+]	547	432	142	92	50
% of the total examination	31.5			26.7			19.9		
Total Examinations, n	3572	1687	1885	3663	2088	1575	713	447	266

Legend: Data shown as number and (%). Abbreviations: LFTL: lung function testing laboratory; FEV_1 = forced expiratory volume at first second; RV = residual volume; DLCO = lung carbon monoxyde diffusion; PaO_2 = arterial oxygen tension; FiO_2 = inspiratory fraction of oxygen;

[*] p= 0.0178

[#] p= 0.0001

[@] p< 0.0001

[+] p = 0.1376 for comparison between total values of each evaluation collected during pandemic and pre-pandemic.

them continuously during the pandemic time,[4,5] and giving the possibility for a rapid reopening with maximum safety.[5]

To the best of our knowledge, this is the first Italian study showing what has changed in terms of quantity and quality since the reopening of a lung function testing laboratory after the deep Covid-19 crisis. Despite the strong perplexities, fear and resistance for a full resumption of activity, our service was able to restart its mission in maximal safety; the substantial restoration of the volumes of activity, even in a progressive way during the months of reopening, has allowed us to give significant answers especially to the strong demand for outpatient diagnostics. It is interesting to note how the requests received at our service have been deeply influenced by the post Covid-19 problems as demonstrated by the strong increase in testing for lung volume diffusion, one of the more significant and useful tests for post Covid-19 sequelae.[10-12] DLCO acquired in fact relevance due to its specific capacity to evaluate interstitial lung disease impairment and due to easy and non-invasive execution of the maneuver requested.[10-12]

Our data have demonstrated that the maximum pre-caution/safety and the essential need to measure the lung function may coexist, independently from the kind of respiratory disease. Furthermore, our data may be of interest for doctors and health organizers to develop a policy that provides strategies of re-opening and re-organisation of lung function laboratories; at the same time, it may be of interest to focus on which kind of performance (inpatient vs outpatient, types of respiratory function tests) is more requested during a pandemic time.

Our results have highlighted the impact of the SARS- CoV-2 pandemic on a lung function testing laboratory facility in a typical Italian rehabilitative hospital, showing a significant change in terms of quantity, modality and quality activities. Lung function testing laboratory should be considered a key component in the health follow up, planning a response to a respiratory pandemic crisis.

Author contributions

Conceptualization, writing original paper, review and supervision: Michele Vitacca; Data collection, revision of the paper: Michela Mineni, Gundi Steinhilber; Data curation, investigation, revision of the paper: Beatrice Salvi, Laura Comini; Investigation, Formal analysis, revision of the paper: Mara Paneroni. All Authors approved the current version of the manuscript.

Conflicts of interest

The authors have no conflicts of interest to declare.

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References

1. Winck JC, Ambrosino N. COVID-19 pandemic and non invasive respiratory management: every goliath needs a David. an evidence based evaluation of problems. Pulmonology. 2020;26(4):213—20. https://doi.org/10.1016/j.pulmoe.2020.04.013.
2. Worldometer [Internet]. COVID-19-19 coronavirus pandemic. https://www.worldometers.info/coronavirus/WHO. (Accessed on: 12 November 2021).
3. Kouri A, Gupta S, Yadollahi A, Ryan CM, Gershon AS, To T, et al. Addressing reduced laboratory-based pulmonary function testing during a pandemic. Chest. 2020;158(6):2502—10. https://doi.org/10.1016/j.chest.2020.06.065.
4. Recommendation from ERS Group 9.1 (Respiratory function technologists /Scientists) lung function testing during COVID-19 pandemic and beyond. https://panafricanthoracic.org/images/pdf/African_Paeds_LF/COVID-19/ERS_91_Statement_on_lung_function_during_COVID-19_Final_with_Contributors.pdf (Accessed on: 12 November 2021).
5. Milanese M, Corsico AG, Bellofiore S, Carrozzi L, Di Marco F, Iovene B, et al. SIP (Italian Respiratory Society). Suggestions for lung function testing in the context of COVID-19. Respir Med. 2020;177:106292. https://doi.org/10.1016/j.rmed.2020.106292.
6. Ceriana P, Nava S, Vitacca M, Carlucci A, Paneroni M, Schreiber A, et al. Noninvasive ventilation during weaning from prolonged mechanical ventilation. Pulmonology. 2019;25(6):328—33. https://doi.org/10.1016/j.pulmoe.2019.07.006.
7. Maestri R, Bruschi C, Fracchia C, Pinna GD, Fanfulla F, Ambrosino N. Physiological and clinical characteristics of patients with COPD admitted to an inpatient pulmonary rehabilitation program: a real-life study. Pulmonology. 2019;25(2):71—8. https://doi.org/10.1016/j.pulmoe.2018.07.001.
8. Zampogna E, Paneroni M, Belli S, Aliani M, Gandolfo A, Visca D, et al. Pulmonary rehabilitation in patients recovering from COVID-19. Respir Med. 2021;184:106465. https://doi.org/10.1016/j.rmed.2021.106465.
9. Ippolito M, Vitale F, Accurso G, Iozzo P, Gregoretti C, Giarratano A, et al. Medical masks and respirators for the protection of healthcare workers from SARS-CoV-2 and other viruses. Pulmonology. 2020;26(4):204—12. https://doi.org/10.1016/j.pulmoe.2020.04.009.
10. Bellan M, Soddu D, Balbo PE, Baricich A, Zeppegno P, Avanzi GC, et al. Respiratory and psychophysical sequelae among patients with COVID-19 four months after hospital discharge. JAMA Netw Open. 2021;4(1):e2036142. https://doi.org/10.1001/jamanetworkopen.2020.36142.
11. Anastasio F, Barbuto S, Scarnecchia E, Cosma P, Fugagnoli A, Rossi G, et al. Medium-term impact of COVID-19 on pulmonary function, functional capacity and quality of life. Eur Respir J. 2021;58(3):2004015. https://doi.org/10.1183/13993003.04015-2020.
12. Torres-Castro R, Vasconcello-Castillo L, Alsina-Restoy X, Solis-Navarro L, Burgos F, Puppo H, et al. Respiratory function in patients post-infection by COVID-19: a systematic review and meta-analysis. Pulmonology. 2021;27(4):328—37. https://doi.org/10.1016/j.pulmoe.2020.10.013.

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LETTER TO THE EDITOR

Impact of the COVID-19 pandemic on in-hospital diagnosis of tuberculosis in non-HIV patients

Dear editor,

The coronavirus disease 2019 (COVID-19) pandemic brought unprecedented consequences for everyday activities, damaging economies and severely affecting healthcare systems.[1] A modelling analysis commissioned by the STOP TB Partnership has indicated that the COVID-19 pandemic has deeply affected the efforts on tuberculosis (TB) prevention, case detection and management. Reductions in TB diagnoses during the pandemic have been reported worldwide which has been attributed to a reduction in admissions due to lockdown, as well as to the abundance of public health measures directed against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) at the expense of other respiratory infectious diseases.[2]

In Portugal, the first COVID-19 case was recorded on the 2nd March 2020. We retrospectively reviewed adult hospitalised patients with TB, not associated with human immunodeficiency virus (HIV) infection or acquired immunodeficiency syndrome (AIDS) disease, in our tertiary hospital, and compared those admitted between March 2018 and June 2019 (pre-pandemic) with those admitted from March 2020 to June 2021 (pandemic). Chi-square test was used to compare categorical variables. Independent-samples t-test was used to evaluate differences in continuous variables with normal distribution and Mann-Whitney U tests were applied to evaluate differences in continuous variables with skewed distribution. Patient data were entirely anonymized and authorized by the Responsible for Access to Information (RAI) of Centro Hospitalar Universitário de São João. The registration protocol complies with the ethical guidelines of the Declaration of Helsinki and it was approved by the Ethics and Health Committee of Centro Hospitalar Universitário de São João.

We analysed a total of 100 cases (58 from pre-pandemic period and 42 from pandemic period) and the 42 cases linked with the time of pandemic reflected 72.4% of the admissions comparing to the homologous pre-pandemic one. There were fewer admissions in the first three months of the pandemic (18 in the pre-pandemic vs 7 in the pandemic, $p = 0.101$), coinciding with a mandatory household lockdown period and in line with reports disclosing partial disruptions in TB case detection and treatment during the same three months.[3]

Patients were significantly older in the pandemic group (54.5 years vs 63.0 years, $p = 0.015$), (Table 1). In both groups, most were male (75.9% in the pre-pandemic vs 78.6% in the pandemic sets, $p = 0.250$) and had smoking history (62.1% vs 61.9%, $p = 0.987$). Pre-existing lung disease was present in 32.8% ($n = 19$) and 38.1% ($n = 16$) of the patients ($p = 0.992$), with chronic obstructive lung disease (COPD) being the most frequent among those - 36.8% and 43.8%. Previous TB diagnosis was found in 6.9%, $n = 4$ and 9.5%, $n = 4$ of the cases in each group ($p = 0.805$). A variety of immunosuppression status was present in 46.6% ($n = 27$) of the patients in the pre-pandemic group vs 16.7% ($n = 7$) in the pandemic, a difference that was statistically significant ($p = 0.002$).

Disseminated tuberculosis was identified in 20.7% ($n = 12$; pre-pandemic group) and 31.0% ($n = 13$; pandemic group) of cases ($p = 0.324$). However, a significant increase in disseminated tuberculosis in immunocompetent patients was seen in the pandemic group - $n = 11$, 84.6% vs $n = 4$, 33% ($p = 0.009$) - affecting mainly bone, liver and genitourinary system. Exclusive pulmonary tuberculosis was found in 51.7% ($n = 30$) and 47.6% ($n = 20$) of patients ($p = 0.421$), respectively. Despite no difference in cavitating pulmonary disease ($p = 0.239$), bilateral lesions were more frequent in the pandemic period - $n = 12$, 60% vs $n = 9$, 30% ($p = 0.035$) - as was previously reported.[4] Concerning patients with negative sputum smears at presentation ($n = 32$ vs $n = 17$), and patients with numerous (>50/field) bacilli ($n = 7$ vs $n = 12$), the differences found across the two populations did not however achieve a statistically significant difference ($p = 0.055$). The length of stay was longer (median, 19.5 days [IQR 9.75-51.00] vs. 40.5 days [IQR 13.25-67.25]) in the pandemic period.

As previously stated by Visca et al., COVID-19 may occur at any time during a patient's TB journey and may cause a spectrum of host immunological responses. Nevertheless, more evidence is needed to understand the potential of COVID-19 to modify TB severity or to promote reactivation of TB infection.[5] We detected 3 cases of COVID-19 and tuberculosis co-infection, all male with no previous TB diagnosis. One had exclusive pulmonary tuberculosis and was immunocompromised due to chemotherapy, the other two patients had pleural and disseminated tuberculosis and no co-morbidities. No deaths were recorded.

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Table 1 Patients' characteristics and clinical presentations. Data are presented as frequencies and percentages for categorical variables and as medians and interquartile ranges (IQR) for continuous variables.

	March 2018-June 2019 (*n* = 58)	March 2020-June 2021(*n* = 42)	*p*-value
Age (years)	54.5 (41.75-67.50)	63.0 (47.75-77.00)	0.015
Male, *n* (%)	44 (75.9)	33 (78.6)	0.250
Smoking habits, *n* (%)			
Previous or active	36 (62.1)	26 (61.9)	0.987
Non-smokers	20 (34.5)	16 (38.1)	0.847
Passive	2 (3.4)	0	0.224
Previous or active drug abuse, *n* (%)	5 (8.6)	4 (9.5)	0.367
Previous pulmonary tuberculosis, *n* (%)	4 (6.9)	4 (9.5)	0.805
Pre-existing lung disease, *n* (%)	19 (32.8)	16 (38.1)	0.992
Chronic obstructive lung disease	7 (36.8)	7 (43.8)	
Non-cystic fibrosis bronchiectasis	3 (15.8)	4 (25.0)	
Silicosis	3 (15.8)	3 (18.7)	
Asthma	2 (10.5)	2 (12.5)	
Hypersensitivity pneumonitis	2 (10.5)	0	
Alpha-1 antitrypsin deficiency	1 (5.3)	0	
Cystic lung disease	1 (5.3)	0	
Immunocompromised status, *n* (%)	27 (46.6)	7 (16.7)	0.002
Chronic alcoholism	12 (44.5)	3 (42.9)	
High dose steroids	6 (22.2)	2 (28.5)	
Hematologic malignancy	5 (18.5)	2 (28.5)	
Monoclonal antibody treatment	4 (14.8)	0	
Pulmonary tuberculosis, *n* (%)	49 (84.5)	38 (90.5)	0.379
Restricted to lungs	30 (51.7)	20 (47.6)	0.421
Disseminated	12 (20.7)	13 (31.0)	0.324
Pleuropulmonary	7 (12.1)	5 (11.9)	0.601
Extrapulmonary tuberculosis, *n* (%)	9 (15.5)	4 (9.5)	0.379

For McQuaid et al., two of the main concerns of the COVID-19 pandemic on TB would be a greater impact on patients with drug-resistant TB and a net increase in deaths in all scenarios with some level of health service disruption.[6] According to our data, during the pandemic period, 95.2% (*n* = 40) of the cases were drug-sensitive and mortality was lower (12.1%, *n* = 7, vs. 4.8%, *n* = 2) but no statistical difference was found (*p* = 0.208).

In our study, less immunocompromised patients hospitalized with TB were seen in the pandemic period. Particular care with social distancing, self-quarantine and the thorough use of facial masks by these patients might have had an impact in this finding. In the pandemic setting there was: (i) a clear increase in extended pulmonary forms, (ii) a significant rise of disseminated tuberculosis cases in immunocompetent patients and (iii) a tendency to find a greater proportion of numerous bacilli on smears, indicating that people might have tolerated longer symptomatic periods before seeking medical aid, as they were reluctant to go to the hospital, leading to diagnostic delay and to an increased risk of TB transmission in households and communities. Rodrigues et al. reached the same conclusion, finding delays in the diagnosis of tuberculosis in the outpatient tuberculosis centres (OTBC), even though they had been open during the pandemic.[7] In our population, COVID-19 did not impact in a rise of drug-resistant cases or higher mortality, but more data are needed in the following years.

References

1. Migliori GB, Tiberi S, García-Basteiro AL, Duarte R. Tuberculosis and its future in the COVID-19 era: the Pulmonology series 2021. Pulmonology. 2021;27(2):94—6. https://doi.org/10.1016/j.pulmoe.2020.10.005.
2. Wee LE, Goh KCM, Conceicao EP, Tan JBX, Sng LH, Venkatachalam I. Increased detection of pulmonary tuberculosis amongst hospitalised inpatients during the COVID-19 pandemic. Eur Respir J. 2021;57(5):2100616. https://doi.org/10.1183/13993003.00616-2021.
3. Comella-del-Barrio P, De Souza-Galvão ML, Prat-Aymerich C, Domínguez J. Impact of COVID-19 on Tuberculosis Control. Arch Bronconeumol. 2021;57:5—6. https://doi.org/10.1016/j.arbres.2020.11.016.
4. Aznar ML, Espinosa-Pereiro J, Saborit N, et al. Impact of the COVID-19 pandemic on tuberculosis management in Spain. Int J Infect Dis. 2021;108:300—5. https://doi.org/10.1016/j.ijid.2021.04.075.
5. Visca D, Ong CWM, Tiberi S, et al. Tuberculosis and COVID-19 interaction: a review of biological, clinical and public health effects. Pulmonology. 2021;27(2):151—65. https://doi.org/10.1016/j.pulmoe.2020.12.012.
6. McQuaid CF, McCreesh N, Read JM, et al. The potential impact of COVID-19-related disruption on tuberculosis burden. Eur Respir J. 2020;56(2):2001718. https://doi.org/10.1183/13993003.01718-2020.
7. Rodrigues I, Aguiar A, Migliori GB, Duarte R. Impact of the COVID-19 pandemic on tuberculosis services. Pulmonology. 2022;28(3):210—9. https://doi.org/10.1016/j.pulmoe.2022.01.015.

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LETTER TO THE EDITOR

The unfriendly side of "happy hypoxaemia": Sudden cardiac death

A 31 year-old female patient was admitted to our dedicated COVID-19 Intensive Care Unit with hypoxaemic respiratory insufficiency due to COVID-19 pneumonitis. Her past medical history was unremarkable, except for extreme obesity (BMI 70·3). She was not taking any medications, and was not under any regular medical care. Her chest X-ray at admission showed bilateral interstitial pulmonary consolidations (Fig. 1A). At admission, our patient was tachypneic, with a respiratory rate of 30-40/min, with SpO_2 85% to 90% while on a non-rebreather mask. We started our patient on dexamethasone, tocilizumab and enoxaparin in a therapeutic dose. Oxygenation improved markedly upon initiation of non-invasive ventilation (NIV) through a mask interface (IPAP 12 cmH_2O, EPAP 10 cmH_2O, FiO_2 started at 100% and titrated to SpO_2). Initially, while on NIV, she maintained $SpO_2 > 90\%$, with FiO_2 of 60-80%, whereas saturations would drop within minutes while on a non-rebreather mask (15L / min), during attempted breaks from NIV. We did not offer high-flow nasal cannula (HFNC) oxygen therapy, partially because of its excessively high oxygen consumption, which presented logistical barriers in our resource-limited setting during a pandemic. Furthermore, we expected "true" positive pressure ventilation to have more beneficial effects on obesity-related, position-dependent atelectasis, and on a potentially present obstructive sleep apnea. Over the first week of admission, she developed progressive hypoxaemia, while denying dyspnoea. Our patient repeatedly refused intubation and invasive ventilation, citing a lack of symptoms, and a fear for a worse outcome with mechanical ventilation. On day three, the patient desaturated to S_pO_2 of about 50% (despite $FiO_2$100%), while denying dyspnoea, and without exhibiting tachypnea. Following prone positioning, saturations recovered to above 90%. Intermittent awake proning was, from that moment on, continued three times daily. On day seven, S_pO_2 again dropped to 30-55%, from then on never exceeding 60%, and at times reaching levels as low as 21%, with no further response to prone positioning. Changes in ventilatory settings (EPAP as low as 6 and up to 14cmH_2O; as CPAP, or with pressure support of up to 6 cmH_2O; FiO_2 from here on never below 100%) yielded no improvement in respiratory parameters. An arterial line was placed in the left brachial artery, to allow for regular blood sampling. Arterial blood was extremely dark (Fig. 1B), with a P_aO_2 of 28mmHg. Accidental venous sampling was excluded with an adequate arterial blood pressure waveform. There was no methemoglobinaemia. Meanwhile, our patient was able to speak coherent, full sentences, and denied having any sensation of dyspnoea, although she was at times tachypneic. She remained firm in her view, and persistently refused endotracheal intubation. After enduring extreme hypoxaemia for about two days in a seemingly stable condition, she suddenly developed ventricular fibrillation, rapidly followed by asystole and death. Resuscitative efforts were not successful.

Silent, or "happy" hypoxaemia is a well-known phenomenon in COVID-19.[1,2] Several theories have been proposed for its existence, most revolving around intrapulmonary shunting as the primary driver of hypoxaemia, with relative preservation of lung compliance in the early stages of the disease,[3] and resultant normocarbia or even hypocarbia,[4] although a neural factor has also been proposed.[5] In our case, we think a certain level of hypoxaemia at baseline associated with previously undiagnosed, but potentially present obesity hypoventilation syndrome may have contributed to our patient's tolerance of extremely low arterial oxygen levels.

The optimal timing of intubation in severe COVID-19 remains controversial. Early on in the pandemic, prominent authors urged clinicians to intubate early, in order to prevent patient self-induced lung injury (P-SILI), which was hypothesised to result from excessive respiratory effort.[6] Furthermore, non-invasive respiratory support was feared to lead to droplet formation, putting health-care workers at risk.[7] Conversely, it has been argued that early, potentially unnecessary intubation may increase mortality by exposing patients to the risks of sedation and invasive ventilation.[8] As the pandemic progressed, many clinicians adopted a "wait-and-see" strategy, where patients are initially managed with non-invasive ventilatory support (including NIV and HFNC), and are intubated only upon failure of such therapies.[9] More recent studies yield conflicting results, and although non-invasive respiratory support appears safe, and has been shown to reduce the need for invasive ventilation, the optimum timing of intubation in COVID-19 is as of yet still unknown.[10] It may be reasonable to use a step-up approach, starting HFNC in patients who fail conventional oxygen therapy, failure of which could be followed by a trial of CPAP, and eventually, if indicated, intubation.[11]

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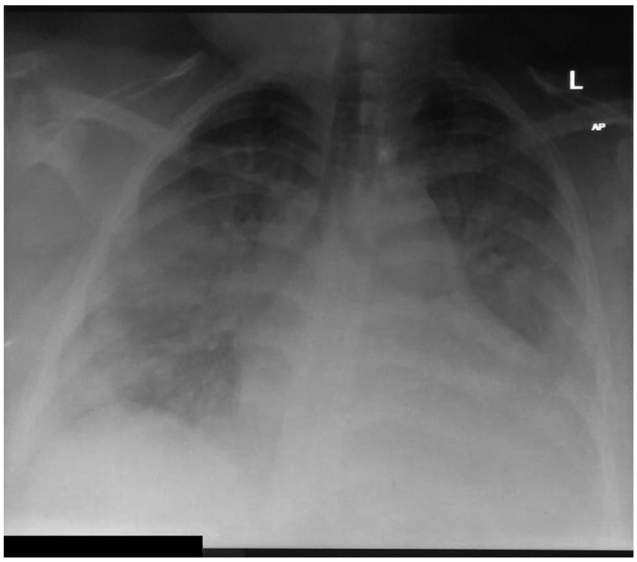
a



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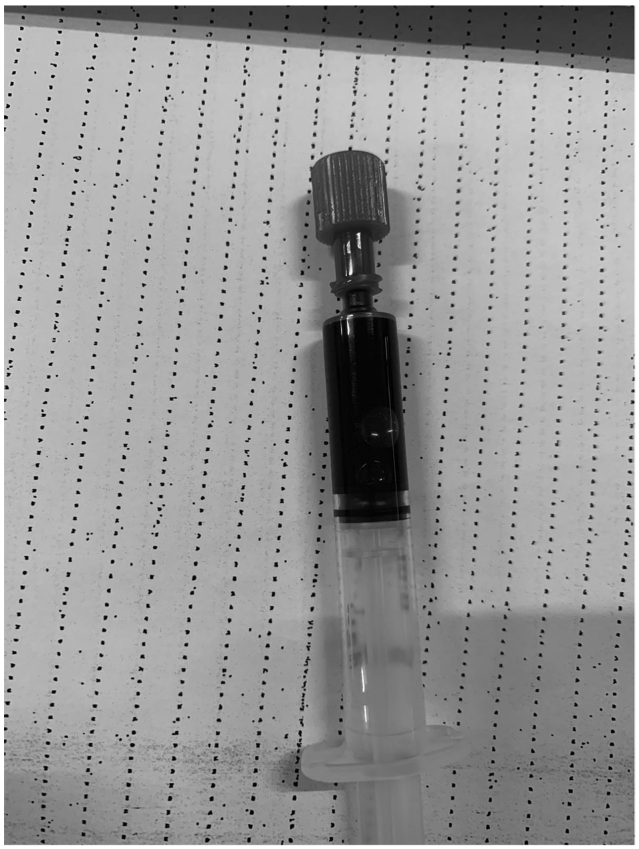

Fig. 1 (A) Chest X-ray taken in a patient with severe COVID-19. showing extensive bilateral opacities, with a slight predominance for the peripheral and basal lung fields. (B) Arterial blood sample, drawn from a patient with COVID-19 receiving non-invasive ventilation (EPAP 12; Pressure Support 4; FiO_2 100%). Blood gas analysis revealed a P_aO_2 of 28mmHg (P/F ratio 28mmHg). Despite oxygen saturations below 40%, the patient denied dyspnoea and did not have elevated work of breathing.

To aid in deciding whom to intubate, authors have proposed using the ROX index,[12] which, in several retrospective series, has been reported to predict failure of non-invasive respiratory support in COVID-19.[12,13] While most studies were performed in cohorts treated with HFNC, the ROX index has been reported to correlate with outcomes after CPAP as well.[14] It should be noted that different studies report different cut-offs (such as <3.85[12] and <5.99[13]). Our patient had ROX-indexes as low as 1.12, clearly indicating a high risk of treatment failure.

The phenomenon of seemingly well-tolerated hypoxaemia in COVID-19 has led to further controversy, as it seems counter-intuitive to intubate a patient who, despite having low oxygen saturations, is feeling well. This discrepancy between subjective and objective findings has led some authors to argue that such patients should not be intubated, as long as they remain asymptomatic, and do not exhibit increased work of breathing.[15] Even though we are inclined to agree with this concept in general, as long as hypoxaemia is mild to moderate, we believe our case demonstrates the dangers when such an approach is taken to the extreme. There is a point where hypoxaemia can lead to rapid cardiovascular decompensation,[2] where "happy" hypoxaemia can show its unfriendly side: sudden cardiac death.

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Conflicts of interest

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References

1. Tobin MJ, Laghi F, Jubran A. Why COVID-19 silent hypoxemia is baffling to physicians. Am J Respir Crit Care Med. 2020;202:356—60.
2. Tobin MJ. Basing respiratory management of COVID-19 on physiological principles. Am J Respir Crit Care Med. 2020;201:1319—20.
3. Haryalchi K, Heidarzadeh A, Abedinzade M, Olangian-Tehrani S, Ghazanfar Tehran S. The importance of happy Hypoxemia in COVID-19. Anesth Pain Med. 2021;11:e111872.
4. Bickler PE, Feiner JR, Lipnick MS, McKleroy W. "Silent" presentation of hypoxemia and cardiorespiratory compensation in COVID-19. Anesthesiology. 2021;134:262—9.
5. Rahman A, Tabassum T, Araf Y, Al Nahid A, Ullah MA, Hosen MJ. Silent hypoxia in COVID-19: pathomechanism and possible management strategy. Mol Biol Rep. 2021;48:3863—9.
6. Marini JJ, Gattinoni L. Management of COVID-19 respiratory distress. JAMA. 2020;323:2329—30.
7. Crimi C, Noto A, Cortegiani A, et al. Noninvasive respiratory support in acute hypoxemic respiratory failure associated with COVID-19 and other viral infections. Minerva Anestesiol. 2020;86:1190—204.
8. Tobin MJ, Laghi F, Jubran A. Caution about early intubation and mechanical ventilation in COVID-19. Ann Intensive Care. 2020;10. 78-020-00692-6.

9. Papoutsi E, Giannakoulis VG, Xourgia E, Routsi C, Kotanidou A, Siempos II. Effect of timing of intubation on clinical outcomes of critically ill patients with COVID-19: a systematic review and meta-analysis of non-randomized cohort studies. Crit Care. 2021;25. 121-021-03540-6.
10. Weerakkody S, Arina P, Glenister J, et al. Non-invasive respiratory support in the management of acute COVID-19 pneumonia: considerations for clinical practice and priorities for research. Lancet Respir Med. 2022;10:199—213.
11. Winck JC, Scala R. Non-invasive respiratory support paths in hospitalized patients with COVID-19: proposal of an algorithm. Pulmonology. 2021;27:305—12.
12. Myers LC, Mark D, Ley B, et al. Validation of respiratory rate-oxygenation index in patients with COVID-19-related respiratory failure. Crit Care Med. 2022.
13. Vega ML, Dongilli R, Olaizola G, et al. COVID-19 Pneumonia and ROX index: Time to set a new threshold for patients admitted outside the ICU. Pulmonology. 2022;28:13—7.
14. De Vita N, Scotti L, Cammarota G, et al. Predictors of intubation in COVID-19 patients treated with out-of-ICU continuous positive airway pressure. Pulmonology. 2022;28(3):173—80.
15. Voshaar T, Stais P, Köhler D, Dellweg D. Conservative management of COVID-19 associated hypoxaemia. ERJ Open Res. 2021;7:00026—2021. eCollection 2021 Jan.

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LETTER TO THE EDITOR

Multiple system atrophy: Inspiratory sighs as a key polysomnographic sign to early diagnosis?

Dear editor,

Multiple system atrophy (MSA) is a rapidly progressive adult neurodegenerative disease associated with sleep disorders, such as rapid eye movement (REM) sleep behavior disorder (RBD), obstructive sleep apnea syndrome (OSAS), or stridor.[1,2]

A 63-year-old hypertensive woman with sleep onset insomnia complained of harmful motor activity and vivid dreams. The patient had no history of snoring, or other night symptoms, or complaints related to excessive daytime sleepiness. Epworth sleepiness scale score was of 6. She underwent polysomnography which revealed micro-arousal index of 42.4%, respiratory disturbance index of 18.9/h, periodic limb movements (PLM) of 67.5/h, frequent

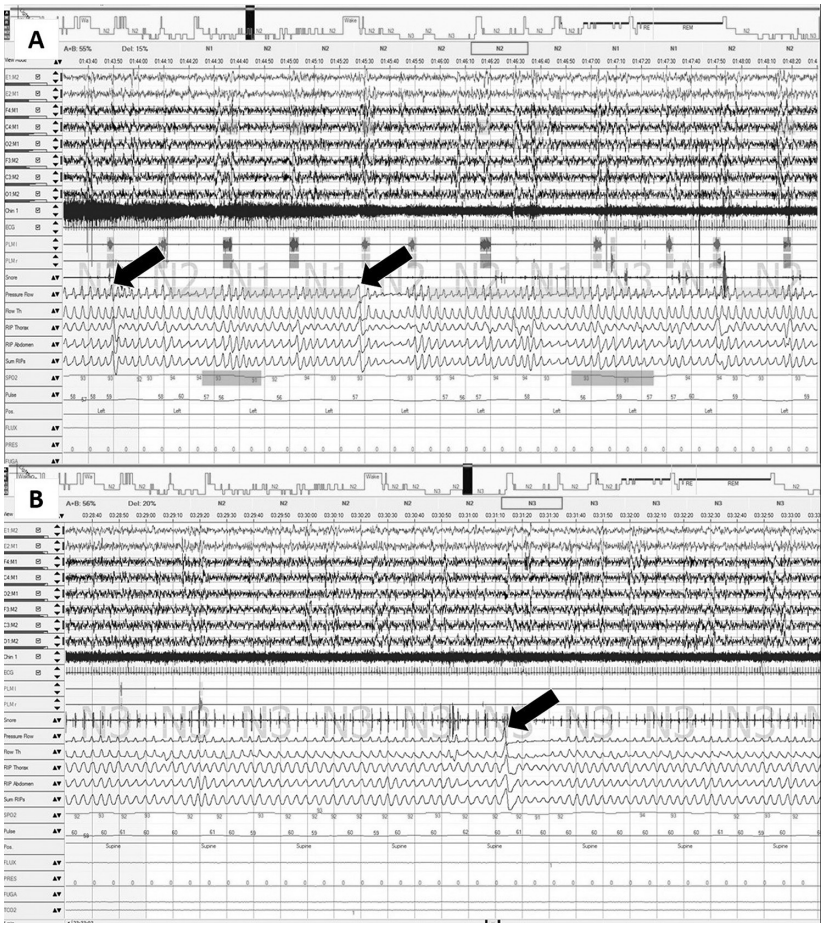

Figure 1 Examples of inspiratory sighs (black arrows), during N2 (A) and N3 (B) sleep. None of the sighs are associated to respiratory events.

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gesticulation and somniloquism and REM sleep without atonia. OSAS plus PLM syndrome and RBD were diagnosed, and she started continuous positive airway pressure treatment and clonazepam 0.5 mg/id. A year later she complained of multiple falls and orthostatic hypotension. Neurological examination revealed scanning dysarthria, distal polymini-myoclonus, limb dysmetria and ataxia A cerebellar-type probable MSA was diagnosed and the patient started levodopa/carbidopa 25/100 mg 3id partially improving symptoms. The polysomnography was reassessed and concluded the existence of multiple inspiratory sighs in different phases of NREM sleep (Fig. 1), a year prior the onset of neurological symptoms.

This typical but barely known polysomnographic respiratory finding in MSA — inspiratory sighs — can be present in short disease duration and could help the differential diagnosis. Specially in the presence of stridor, sighs may increase the likelihood of MSA.[3] Besides inspiratory sighs, our patient's polysomnography helps the diagnosis of OSAS, PLM syndrome, and RBD. These are common both in MSA and in Parkinson's disease, and usually start before neurological complaints.[2]

In conclusion, pulmonologists in sleep medicine should be aware of this polysomnographic picture (inspiratory sighs, OSAS, PLM syndrome, and RBD) since it could point towards the presence of an extrapyramidal disease, including MSA. Careful evaluation of polysomnography may help to identify early signs of these neurodegenerative diseases and lead to early referral for a neurological consultation.

Declaration of Competing Interest

The authors have no conflicts of interest to declare.

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Supplementary materials

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References

1. Cochen De Cock V. Sleep abnormalities in multiple system atrophy. Curr Treat Options Neurol. 2018;20(16). https://doi.org/10.1007/s11940-018-0503-8.
2. Fanciulli A, Wenning GK. Multiple-system atrophy. N Engl J Med. 2015;372(3):249—63. https://doi.org/10.1056/NEJMra1311488.
3. Parreira S, Antunes F, Coelho M, Bentes C, Peralta R. Sighs during sleep in multiple system atrophy. Sleep Med. 2021;78:75—80. https://doi.org/10.1016/j.sleep.2020.12.013.

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LETTER TO THE EDITOR

High-flow oxygen therapy in palliative care: A reality in a near future?

Dear Editor,

Duarte, et al. in the article review "High flow oxygen therapy in palliative care: A reality in a near future?" argue that High Flow Oxygen (HFO) is a reasonable palliative treatment in end-of-life patients.[1] This is a fact and I also share their opinion, since it makes it possible for people to communicate and feed themselves without an increased physical effort.

The authors also mention that this type of oxygen therapy has the benefit of producing fewer skin lesions. From my experience, it is true that it does not cause pressure ulcers, especially on the nasal bridge (even with the application of protective padding) and the whole feeling of claustrophobia,[2] as is often the case with non-invasive ventilation (NIV). However, HFO causes internal injuries in the nasal septum and a tamponing sensation with continued and prolonged use. There are visible effects reported by patients undergoing HFO in the pneumology service where I work. I emphasize that when this happens, patients remove the nasal cannula for a moment and try to clean it to diminish this nasal tamponade sensation, which eventually results in epistaxis. I should add that we always prefer a nasal catheter that is silicone-coated and as rigid as possible, in order to provide the best comfort for the patient.

I think it's important to analyze the benefits and drawbacks of this type of oxygen therapy recently used because, despite all the benefits it has, it is translates into discomfort. In the authors opinion, HFO is a reasonable palliative treatment in end-of-life patients, however this ends up being counterproductive since it cause injury and, consecutively, suffering in patients.

For end of life patients I do not believe, in most cases, it is the best option, and there is also no evidence that it has advantages over opioids and anxiolytics. As the authors says, dyspnea is the most prevalent symptom, and can be quite debilitating at all levels. The most important thing is the symptomatic relief of dyspnea, emphasizing that the use of oxygen therapy does not represent an improvement in survival in people with advanced disease.[3]

There is still an urgent need to look at the person suffering from an incurable disease in advanced and/or progressive stages, in order to promote well-being and quality of life.[4] Prevention and relief of pain are indispensable, not the removal of one type of pain in order to offer another. Symptomatic treatment continues to make more sense than inappropriate and excessive use of oxygen therapy.[5]

References

1. Duarte JC, Santos O, Lousada C, Reis-Pina P. High-flow oxygen therapy in palliative care: a reality in a near future? Pulmonology. 2021;27(6):479—80. https://doi.org/10.1016/j.pulmoe.2021.08.003.
2. Gay P. Complications of noninvasive ventilation in acute care. Respir Care. 2009;54(2):246—57. Available at http://rc.rcjournal.com/content/54/2/246/tab-pdf.
3. Martins M, Campôa E, Ferreira M, Reis-Pina P. Autonomy and dyspnea in palliative care: a case report. Pulmonology. 2020;26(2):105—7. https://doi.org/10.1016/j.pulmoe.2019.05.005.
4. Neto IG. Cuidados paliativos - conheça-os melhor. Lisboa: Fundação Francisco Manuel dos Santos; 2020. p. 13—40.
5. Frade L, Carreira N, Tosatto V, Marote S, Neto IG. Oxigenoterapia na doença avançada: conhece a evidência? Med Intern. 2019;26(4):320—5. [Internet]. Available at https://www.spmi.pt/revista/vol26/vol26_n4_2019_320_325.pdf.

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PHOTO

Late onset pulmonary vein stump thrombus seven years after left upper lobectomy

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The four authors have made substantial contributions to, not only the conception and design of the manuscript, but also the drafting and critical revision of the article.

There have been very few reported cases of pulmonary vein stump thrombus (PVST) after lobectomy, and the majority of them involved resection of the left upper lobe with subsequent thrombus formation in the left upper pulmonary vein stump.[1—3] The true incidence of PVST following lobectomy is unknown and likely underdiagnosed, especially in asymptomatic patients who do not undergo postoperative imaging with contrast-enhanced computed tomography (CT) or transesophageal echocardiogram. PVST after lobectomy is not a negligible finding because thromboembolic complications, including transient ischemic attack and stroke, have been reported.

A 75-year-old patient who had undergone a left upper lobectomy (LUL) seven years earlier for a lung adenocarcinoma was incidentally found to have a left superior PVST during a routine follow-up contrast-enhanced thoracic CT study. An 18F-fluorodeoxyglucose (FDG) positron emission tomography/CT (PET/CT) was very useful for excluding a tumor thrombus or any other form of malignancy. Three months after the patient was started with anticoagulation therapy, a new contrast-enhanced thoracic CT showed resolution of the PVST (Fig. 1). No other known causes for thrombus formation were identified.

PVST is very rare and has been associated with systemic embolism to vital organs (such as the brain, kidneys or bowel) if the thrombus enters the systemic arterial circulation. PVST is much more common in patients undergoing a LUL or a left pneumonectomy. In a study with 151 patients who underwent lobectomy, PVST was only detected in patients undergoing LUL, (17.9% of the LULs)[1]; in a more recent study with 1040 patients, PVST was also found in right-sided pulmonary resections, but less frequently than in left-sided interventions.[3] All previous reported cases of PVST following LUL were detected within 7 months postoperatively.[2] Length of the pulmonary vein stump has also been implicated in the risk of thrombosis.[1]

In conclusion, PVST is usually an early postoperative event that is more frequently observed in left-sided pulmonary resections (particularly LUL). PVST is likely an underdiagnosed condition, and more studies are needed to address if contrast-enhanced CT should be performed in high risk LUL patients. We believe that our case is interesting since the PVST developed 7 years after the LUL and because

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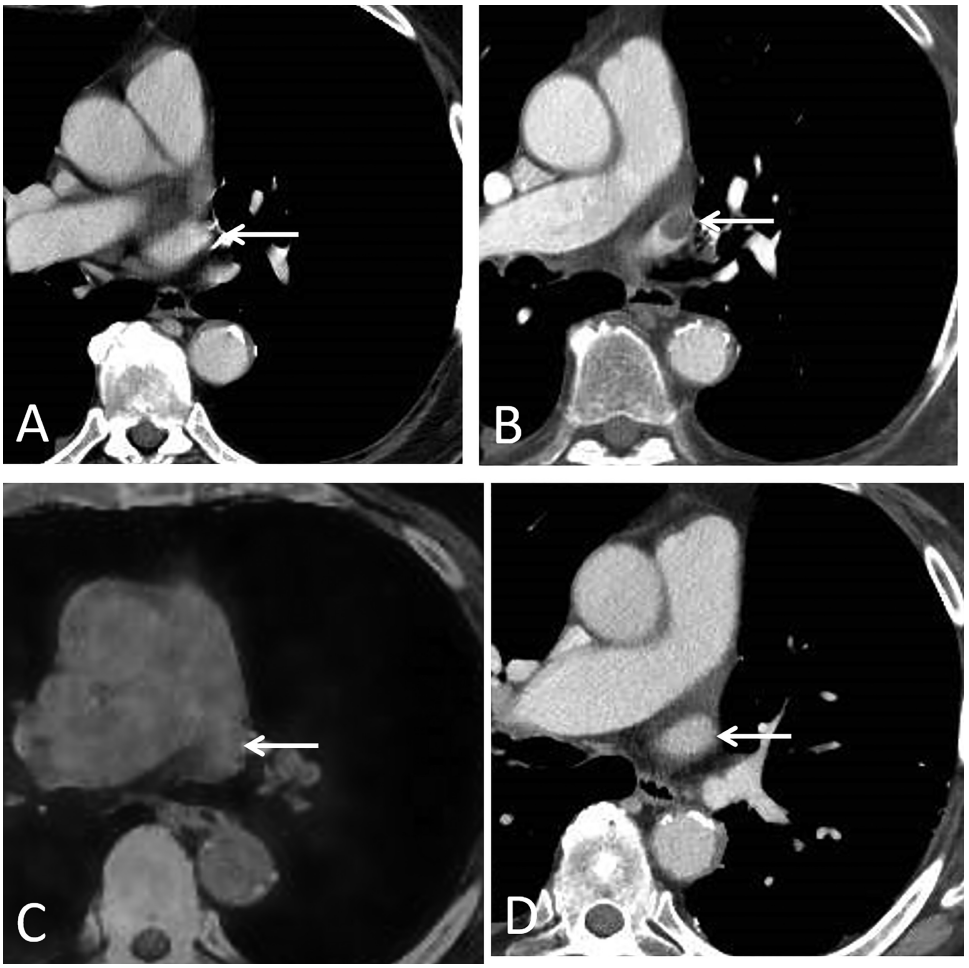

Fig. 1 A, Axial contrast-enhanced CT image (mediastinal window), obtained 5 years after the left upper lobectomy, shows a patent left superior pulmonary vein stump (arrow). B, Axial contrast-enhanced CT image (mediastinal window), obtained 7 years after the left upper lobectomy, shows a filling defect within the left superior pulmonary vein stump (arrow). C, Axial fused PET/CT image (obtained 3 days after B) demonstrates lack of FDG uptake at the level of the left superior pulmonary vein stump (arrow), suggesting a non-tumor thrombus. D, Axial contrast-enhanced CT image (mediastinal window), obtained 3 months after B, shows resolution of the left superior pulmonary vein stump thrombus (arrow).

FDG-PET/CT was useful for excluding a delayed tumor thrombus.

Conflicts of interest

The authors declare no conflict of interest.

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References

1. Ohtaka K, Hida Y, Kaga K, Takahashi Y, Kawase H, Hayama S, et al. Left upper lobectomy can be a risk factor for thrombosis in the pulmonary vein stump. J Cardiothorac Surg. 2014;9:5.
2. Chaaya G, Vishnubhotla P. Pulmonary vein thrombosis: a recent systematic review. Cureus. 2017;9(1):e993.
3. Hattori A, Takamochi K, Shiono S, Tsuboi M, Maniwa Y, Suzuki K, et al. Multicentre prospective observational study for pulmonary vein stump thrombus after anatomical lung resections. Eur J Cardiothorac Surg. 2021;61(1):92—9.

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PHOTO

Spontaneous elimination of a bronchial mucoid pseudotumor — A curious but pleasant surprise

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Dear Editor:

Tracheobronchial luminal filling defects are caused by several entities, comprising inflammatory or infectious etiologies, neoplasms (benignant and malignant), foreign bodies, and the abnormal accumulation of endogenous material (e.g., mucous plugs).[1] Mucoid pseudotumors simulating tracheobronchial tumors have been reported infrequently.[2] A typical CT characteristic of these pseudotumors is the presence of focal low-attenuation tissue mixed with air content, conferring a "bubbly" appearance, most frequently in dependent airway walls.[1] Hyperattenuating mucous plugging may also be observed, and is considered to be a relevant CT sign for the diagnosis of allergic bronchopulmonary aspergillosis.[3] In patients in the intensive care unit, mucous plugging may also be an important cause of bronchial obstruction leading to the acute onset of atelectasis and hypoxemia.[4]

Awareness of the CT characteristics of mucous plugging is important because this condition is observed frequently on chest imaging and is part of the differential diagnosis with parietal soft-tissue lesions, and because the failure to recognize it may lead to unnecessary bronchoscopic procedures. Asking the patient to cough and repeating the CT acquisition may be a useful technique, once mucous plugging may be cleared.[1] Unfortunately, CT does not always enable the safe ruling out of a parietal soft-tissue lesion; endoscopic procedures are mandatory to locally access the tracheobronchial tree and sample tissue for histopathological analysis.[1]

We would like to report the case of a 44-year-old man who presented complaining of cough and wheeze for the previous 3 days. His previous medical records were unremarkable, except for allergic rhinitis and depression. Spirometric evaluation showed a nonspecific ventilatory pattern, and diffusion and fractional exhaled nitric oxide test results were normal. Unilateral right wheezing was noted on lung auscultation. Initially, the clinical hypothesis of acute tracheobronchitis associated with the possibility of right-sided bronchial obstruction was formulated. The patient was referred for contrast-enhanced computed tomography (CT) examination, which showed a low-attenuating soft-tissue lesion completely obstructing the intermedius bronchi, raising the possibility of a tracheobronchial tumor (Fig. 1A). On the same day, after a coughing episode, the patient eliminated an elongated red and yellowish polypoid structure (Fig. 1B), identified on subsequent histopathological analysis as a plug containing fibrin, neutrophils, and cellular rests with no tissular organization. Follow-up unenhanced CT examination performed 1 day later revealed the complete disappearance of the previously detected bronchial soft-tissue lesion (Fig. 1C). On bronchoscopy, abundant mucoid airway fluid, but no parietal thickening or luminal foreign body, was observed. The patient's clinical symptoms resolved completely after steroid and antibiotic treatment.

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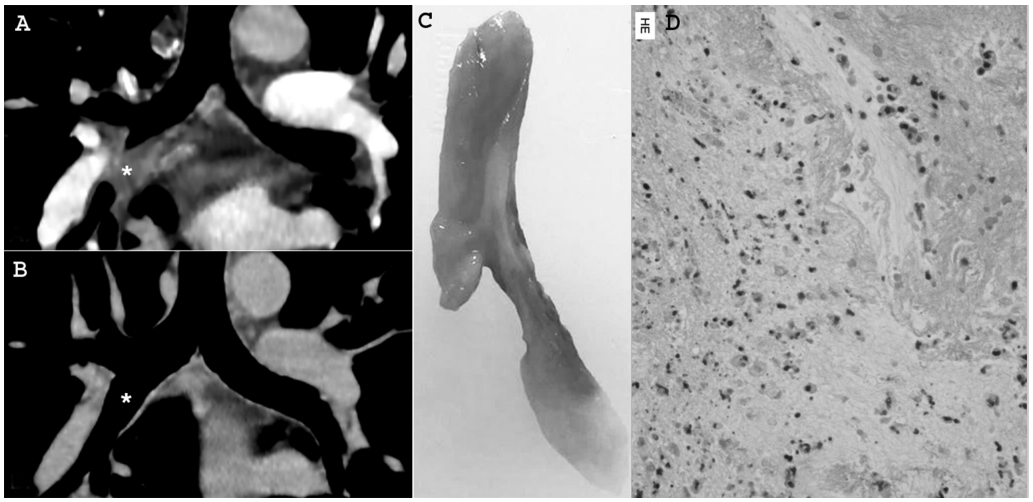

Fig. 1 In A, contrast-enhanced CT with coronal reconstruction revealed a soft-tissue mass obliterating the intermedius bronchi (asterisk). In B, follow-up unenhanced CT examination performed 1 day later showed the complete disappearance of the mass (asterisk). In C, the polypoid lesion (mucous plug) eliminated by the patient. In D, histological section of the specimen eliminated demonstrating that the plug contains fibrin, neutrophils, and cellular rests with no tissular organization (hematoxylin and eosin staining, x 100).

Authors' contributions

PPT, MFR and EM were responsible for the conception and design of the study, and wrote and edited the manuscript. PPT contributed to the drafting and revision of the manuscript. All authors read and approved the final manuscript.

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Conflicts of Interest

The author has no conflicts of interest to declare.

Supplementary materials

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References

1. Ngo AVH, Walker CM, Chung JH, Takasugi JE, Kanne JP, Reddy GP, et al. Tumors and tumorlike conditions of the large airways. AJR. 2013;201:301—13.
2. Itoi S, Ohara G, Kagohashi K, Kawaguchi M, Kurishima K, Satoh H. Cent Eur J Med. 2012;7:385—7.
3. Kouzaki Y, Kanoh S, Kawa A. High-attenuation mucus in allergic bronchopulmonary aspergillosis. Arch Bronconeumol. 2016;52:46—7.
4. Asrani A, Kaewlai R, Digumarthy S, Gilman M, Shepard JAO. Urgent findings on portable chest radiography: what the radiologist should know — review. AJR. 2009;196:S45—61.

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