CC BY

Evaluating a SARS-CoV-2 screening strategy based on serological tests

Valutazione di una strategia di screening per l'infezione da SARS-CoV-2 basata su test sierologici

Michela Baccini,[1,2] Alessandra Mattei,[1,2] Emilia Rocco,[1,2] Giulia Vannucci,[1,2] Fabrizia Mealli[1,2]

[1] Department of Statistics, Computer Science, Applications (DISIA), University of Florence, Florence (Italy)
[2] Florence Center for Data Science, Florence (Italy)

Corresponding author: Michela Baccini; michela.baccini@unifi.it

WHAT IS ALREADY KNOWN

- **More intensive testing of suspected SARS-CoV-2 cases are required to identify people with ongoing infection to be early quarantined.**
- **Serological tests, while useful to investigate the extent of the contagion in the community by detecting individuals who have developed antibodies, may lead to high numbers of false negative and false positive results if used with the intent of identifying subjects with ongoing infection, for reasons related to the SARS-CoV-2 antibody dynamics.**
- **Despite this drawback, different countries and regions have proposed to use this kind of test in heterogeneous screening strategies aimed to detect subjects with ongoing infection.**

WHAT THIS STUDY ADDS

- **According to probabilistic laws, the probability of a positive serological test in subjects with ongoing SARS-CoV-2 infection likely ranges from 0.65 to 0.77. The probability of a negative serological test in subjects without an ongoing infection is not lower than 0.98.**
- **Screening procedures which use serological tests at a first step and then send positive subjects to RT-PCR tests should be avoided unless the prevalence and the level of contagion, as expressed by the infection reproduction number, are very low.**
- **Cost-effectiveness evaluations should be performed before any implementation of new screening strategies on the general population or on specific subgroups.**

ABSTRACT

BACKGROUND: facing the SARS-CoV-2 epidemic requires intensive testing on the population to early identify and isolate infected subjects. Although RT-PCR is the most reliable technique to detect ongoing infections, serological tests are frequently proposed as tools in heterogeneous screening strategies.

OBJECTIVES: to analyse the performance of a screening strategy proposed by the local government of Tuscany (Central Italy), which first uses qualitative rapid tests for antibody detection, and then RT-PCR tests on the positive subjects.

METHODS: a simulation study is conducted to investigate the number of RT-PCR tests required by the screening strategy and the undetected ongoing infections in a pseudo-population of 500,000 subjects, under different prevalence scenarios and assuming a sensitivity of the serological test ranging from 0.50 to 0.80 (specificity 0.98). A compartmental model is used to predict the number of new infections generated by the false negatives two months after the screening, under different values of the infection reproduction number.

RESULTS: assuming a sensitivity equal to 0.80 and a prevalence of 0.3%, the screening procedure would require on average 11,167 RT-PCR tests and would produce 300 false negatives, responsible after two months of a number of contagions ranging from 526 to 1,132, under the optimistic scenario of a reproduction number between 0.5 to 1. Resources and false negatives increase with the prevalence.

CONCLUSIONS: the analysed screening procedure should be avoided unless the prevalence and the rate of contagion are very low. The cost and effectiveness of the screening strategies should be evaluated in the actual context of the epidemic, accounting for the fact that it may change over time.

Keywords: serological test, sensitivity, specificity, compartmental model, false negative

RIASSUNTO

INTRODUZIONE: per affrontare l'epidemia di SARS-CoV-2 sono necessarie strategie di screening sulla popolazione per identificare e isolare precocemente i soggetti infetti. Sebbene il test basato su RT-PCR sia la tecnica al momento più affidabile per rilevare le infezioni in corso, i test sierologici sono spesso proposti in strategie di screening eterogenee.

OBIETTIVI: valutare le prestazioni e i costi in termini di numero di test effettuati per una strategia di screening implementata dalla Regione Toscana, che utilizza dapprima test rapidi qualitativi per il rilevamento di anticorpi, quindi test RT-PCR su soggetti positivi.

METODI: attraverso uno studio di simulazione, sono stati stimati il numero di test RT-PCR richiesti dalla strategia di screening e il numero delle infezioni in corso non rilevate in una pseudo-popolazione di 500.000 soggetti, sotto diversi scenari di prevalenza e sensibilità del test sierologico. È stato, infine, usato un modello compartimentale per prevedere il numero di nuove infezioni generate dai falsi negativi due mesi dopo lo screening, ipotizzando valori differenti del numero di riproduzione dell'infezione (R_0).

RISULTATI: assumendo una sensibilità pari a 0,80 e una prevalenza dello 0,3%, la procedura di screening richiederebbe in media 11.167 test RT-PCR e produrrebbe 300 falsi negativi, responsabili dopo due mesi di un numero di contagi compreso tra 526 e 1.132, sotto lo scenario ottimistico di un valore R_0 tra 0,5 e 1. La quantità di risorse richieste e il numero di falsi negativi aumentano all'aumentare della prevalenza.

CONCLUSIONI: la procedura di screening analizzata dovrebbe essere evitata, a meno che la prevalenza e il tasso di contagio non siano molto bassi. Il suggerimento generale è che il costo e l'efficacia delle strategie di screening siano sempre valutati tenendo conto del contesto reale dell'epidemia e dei suoi possibili cambiamenti nel corso del tempo.

Parole chiave: test sierologico, sensibilità, specificità, modello compartimentale, falsi negativi

INTRODUCTION

The severe acute respiratory syndrome Coronavirus 2 (SARS-CoV-2) epidemic has rapidly spread around the world. Most European countries have implemented progressive measures of physical distancing. In Italy, from March 9th to May 4th, citizens were prohibited from leaving home except in cases of proven need or urgency. Then, the restrictions have been progressively lifted on. During the post lockdown period, scientists have advocated the need of implementing 3T-strategies: «testing» the subjects in order to detect ongoing infections, «tracing» the contacts of the infected, and appropriately «treating» the cases.[1] The first two steps of the 3T-strategies are aimed to contain contagion, minimizing the chance of uncontrolled disease spreading. These steps motivate the need to introduce screening strategies able to early detect subjects with ongoing infection. The WHO has stressed that more intensive testing of suspected cases is required to identify and early quarantined infected people.[2]

Knowledge of diagnostic tests for SARS-CoV-2 is still evolving, and a clear understanding of the nature of the tests and interpretation of their findings is not yet there. The most reliable diagnostic test for SARS-CoV-2 is the infections-reverse transcriptase-polymerase chain reaction (RT-PCR) test, although evidence arose that its accuracy could be not maximum.[3-6]

A wide range of serology immunoassays (IAs) have been developed as well.[7,8] These include automated chemiluminescent IA (CLIA), manual ELISA, and rapid lateral flow IA (LFIA), which detect the immunoglobulin M (IgM) and immunoglobulin G (IgG) produced in persons in response to SARS-CoV-2 infection.[9,10]

Due to the limited availability of reagents for RT-PCR tests and the relative low cost of serological test, different countries and regions have proposed the use of IAs in combination with RT-PCR in heterogeneous screening strategies to detect subjects with ongoing infection, despite serological tests are not appropriate to reveal the presence of viral material during the infection. In fact, while they are useful to investigate the extent of the contagion in the community by detecting individuals who have developed antibodies, IAs may lead to high false negative and false positive rates if used with the intent of identifying subjects with ongoing infection, for reasons related to the SARS-CoV-2 antibody dynamics.[6,7,10-12]

In this paper, one screening strategy that has been proposed by the governor of Tuscany Region (Central Italy) with decree No. 54 of May 6th 2020 is analysed.[13] This strategy, similar to others implemented elsewhere, first uses qualitative serological rapid tests, and then RT-PCR tests in case of positive immune response. The strategy is going to be applied to a large portion of the regional population: half a million people, approximately 1/8 of the whole population. Under different scenarios of prevalence of infection, we assess the performance of this screening strategy in terms of expected number of RT-PCR tests to be used on subjects with ongoing infection (cases) and on subject who are not currently infected, as well as in terms of number of cases that the procedure will not be able to detect. In order to contextualize the danger derived from the infections left undetected, we quantify the potential contagion deriving from these false negatives under different hypotheses on the infection reproduction number, R_0, that is the average number of contagions deriving from one infected individual.[14-15]

METHODS

ACCURACY OF THE SEROLOGICAL TEST IF USED TO DETECT ONGOING INFECTION

Rapid point-of-care tests for detection of antibodies have been widely developed and marketed and are of variable quality. These tests are purely qualitative in nature and indicate the presence or absence of antibodies of SARS-CoV-2. A positive result, intended as detection of IgG and/or IgM antibodies,[10] may arise in case of 1) previous infection; 2) ongoing infection (with antibodies already developed); 3) false detection of antibodies. A negative test may arise in case of 1) no previous infection; 2) early stage of an ongoing infection (with antibodies not yet developed); 3) false detection of absence of antibodies.[16]

Serological tests generally have a relatively high sensitivity as tests for detecting the presence of antibodies.[8-10] However, due to reasons related to antibodies kinetics, infected individuals need some time to develop antibodies, and thus, serological tests may wrongly report a negative result on cases at an early stage of infection. Therefore, serological tests are powerful diagnostic tools for patients with no symptoms or mild to moderate illness who undergo the test after two or more weeks from the infection onset, but may perform poorly if used as screening tests for detecting ongoing infections. [6,8-10]

According to a recent Cochrane collaboration meta-analysis and a report of the European network for health technology assessment (EUNetHTA), the sensitivity of the serological tests increases with the time from the infection.[9-10] In particular, the Cochrane review reports the following estimates of the serological tests sensitivity: 0.301 in the first week since people first noticed symptoms, 0.722 in the second week, 0.914 in the third, and 0.960 after the third week. For sake of simplicity, let us initially assume that time since the first symptoms approximates time from the infection (i.e. time from infection to first symptoms is negligible). According to the estimates reported in the Cochrane review, the sensitivity of the serological tests when used to detect ongoing infections can be decomposed as follows:

$$Pr(+|O) = Pr(+|O, W_1) \times Pr(W_1|O) + Pr(+|O, W_2) \times Pr(W_2|O) + \\ + Pr(+|O, W_3) \times Pr(W_3|O) + Pr(+|O, 3W) \times Pr(3W|O) \quad (1)$$

where + = the serological test is positive, O = the subject has ongoing infection, W_i = the test is performed during the i^{th} week from infection onset (i=1,2,3), $3W$ = the test is performed after the third week.

Let us assume that the average time to recovery for an infected is 4-6 weeks.[17] Considering that $Pr(W_i|O)$ = $Pr(3W|O)$ = 1/4 if the time from infection to recovery is 4 weeks, and $Pr(W_i|O)$ = 1/6 and $Pr(3W|O)$ = 0.5 if it is 6 weeks (i=1,2,3), the sensitivity $Pr(+|O)$ ranges from 0.729 to 0.813.

If, more realistically, time from infection to first symptoms is assumed to be not negligible, the sensitivity $Pr(+|O)$ is expected to range between even lower values. For example, if the average time from infection to first symptoms is one week and during this week the sensitivity is 0.301, reasoning as in equation 1, we find that $Pr(+|O)$ ranges between 0.559 to 0.670.

Regarding specificity, let – denote the event that the serological test is negative, $\bar{I}$ = the subject has never been infected, $\bar{O}$ = the subject does not have an ongoing infection. Under the optimistic assumption that the serological test is perfectly able to detect antibodies, unless it is performed at an early stage of an ongoing infection, it is possible to demonstrate that the specificity of the serological test, if used to detect ongoing infections, is:

$$Pr(-|\bar{O}) = Pr(-|\bar{I}) \times \frac{Pr(\bar{I})}{Pr(\bar{O})'} \quad (2)$$

where $Pr(-|\bar{I})$ is the specificity of the serological test if used to detect antibodies (see the appendix for details). If the ratio $Pr(-|\bar{I})/Pr(-|\bar{O})$ is close to 1 – which is a reasonable approximation for the Tuscany region, where at the present stage of the epidemic the prevalence of previous infections in the population is around 1%[18] and the prevalence of ongoing infection is very low – then $Pr(-|\bar{O}) \approx Pr(-|\bar{I})$. Considering that $Pr(-|\bar{I})$ is approximately 1 for some serological tests and never lower than 98%,[9-10,19-20], we set $Pr(-|\bar{O})$ to 0.98, in the analysis. It is worth noting that this specificity value represents an optimistic scenario which could be not appropriate in situations where the ratio $Pr(\bar{I})/Pr(\bar{O})$ is not close to 1. This could be the case for some northern Italian regions, where the prevalence of previous infections has been estimated to be larger than 3%, with a maximum of 7.5% in the Lombardy Region.[18]

MONTE CARLO SIMULATION

The present study evaluates the performance of the screening strategy in a Monte Carlo (MC) simulation study on pseudo-populations of 500,000 subjects, characterized by different percentage of individuals with ongoing infection. We consider the following values of prevalence of ongoing infections: 0.003, 0.005, 0.010, 0.015, 0.02. The first three scenarios are in accordance with studies reporting proportions of infected people around 0.3% and never greater than 1% over the last four months of the coronavirus pandemic, the last two correspond to scenarios where the level of contagion is slightly worse.[14,21]

According to the results obtained in the previous section, this study first focuses on an optimistic scenario where the sensitivity of the serological test in detecting ongoing infections is 0.80 and the specificity is 0.98. Then, taking 0.80 as an upper bound of the sensitivity level, the study also performs simulations with sensitivity equal to 0.5, 0.6 and 0.7.

For sake of simplicity, RT-PCR test specificity and sensitivity have been assumed as equal to 1, even if a certain percentage of false negatives is expected from this procedure as well.[4-5] Under this assumption, the sensitivity and specificity of the serological test can be interpreted as relative to the RT-PCR test.

For each simulation setting, 500 Monte Carlo (MC) iterations are run and in each iteration various performance measures of the strategy are calculated: total number of RT-PCR tests, number of RT-PCR tests respectively performed on subjects with ongoing infection and on subjects who are not currently infected, number of false negatives, and negative predictive value. It is worth noting that, due to the fact that (false and true) positives from the serological test undergo a RT-PCR test, the screening protocol does not produce false positives (the specificity of the strategy is 1 by design), thus the positive predictive value is 1.

Starting from the estimated numbers of false negatives arising from the Monte Carlo simulations, the present study uses a Susceptible-infected-recovered (SIR) compartmental model to predict the number of infected originated by those that the screening left undetected. Assuming a time of infectivity equal to three weeks,[22] both the number of circulating subjects who can spread the contagion (assuming that all subjects in the population are screened at the same time) and the cumulative number of new infections two months after the screening are calculated, derived from the undetected infected subjects. Calculations are done under the different prevalence scenarios used in the MC simulations and different hypothetical values of the infection reproduction number, R_0: 0.5, 1, 1.5, 2.

RESULTS

In relative terms, the strategy appears to perform well: the negative predictive values are very high, with a mean always close to 1 and a very low MC variability (see supplementary figure S1). However, the actual impact can be better understood by looking at the absolute values of the total number of RT-PCR tests performed at the second step of the screening procedure and the number of false negatives. Figure 1 shows the number of RT-PCR tests that have to be performed (on individuals with ongoing infection and on individuals who are not currently infected) and the expected number of false negatives (currently infected indi-

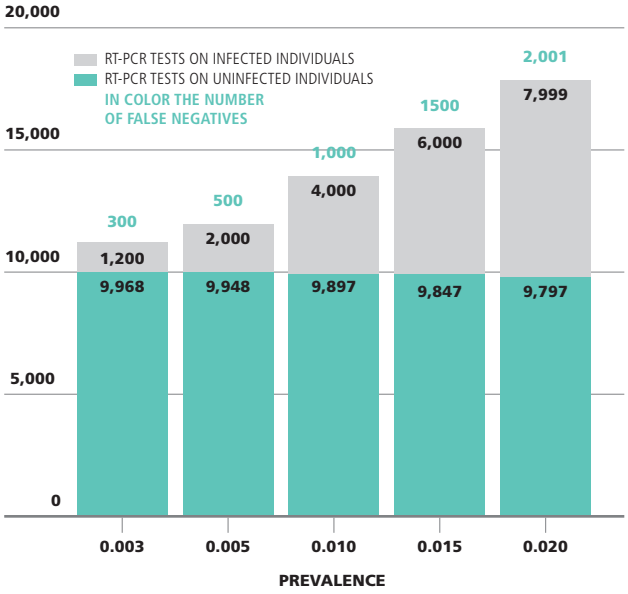

Figure 1. Number of RT-PCR tests (on currently infected subjects and on subjects who are not currently infected) and number of false negatives (currently infected individuals who are not detected) arising from the screening procedure, by prevalence of ongoing infections in the population (sensitivity of the serological test=0.80; specificity of the serological test=0.98).
Figura 1. Numero di test RT-PCR (su soggetti che hanno l'infezione in corso e su soggetti che non hanno l'infezione in corso) e numero di falsi negativi (individui con infezione in corso che non vengono rilevati) generati dalla procedura di screening, per prevalenza di infezioni in corso nella popolazione (sensibilità del test sierologico: 0,80; specificità del test sierologico: 0,98).

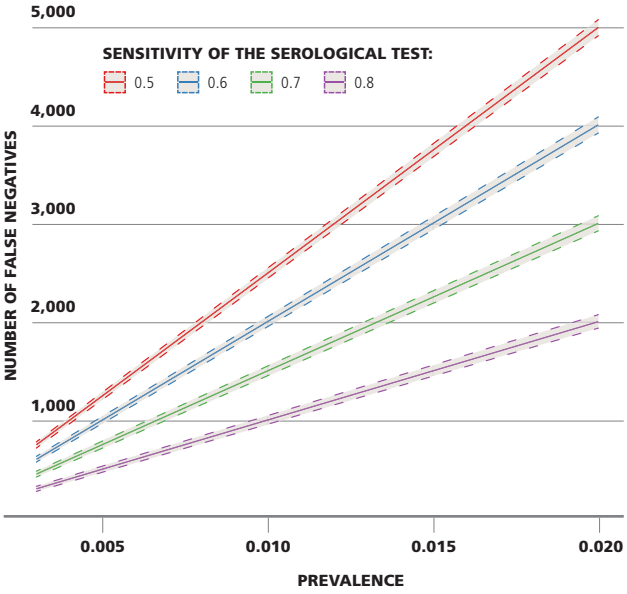

Figure 2. Number of false negatives (currently infected individuals who are not detected) arising from the screening procedure, by sensitivity of the serological test and prevalence of ongoing infections in the population (specificity of the serological test=0.98).
Figura 2. Numero di falsi negativi (individui con infezione in corso che non vengono rilevati) che emergono dalla procedura di screening, per sensibilità del test sierologico e prevalenza di infezioni in corso nella popolazione (specificità del test sierologico: 0,98).

PREVALENCE (%)	FALSE NEGATIVES	NEW INFECTED GENERATED FROM THE FALSE NEGATIVES			
		R=0.5	R=1	R=1.5	R=2
0.3	300	526	1,132	2,945	8,665
0.5	500	876	1,884	4,891	14,263
1	1,000	1,752	3,762	9,698	27,670
1.5	1,500	2,627	5,632	14,422	40,297
2	2,001	3,502	7,495	19,066	52,213

Table 1. Cumulative number of new infected induced by the false negatives after two months from the screening, by prevalence of ongoing infections in the population and infection reproduction number (sensitivity of the serological test=0.80, specificity of the serological test=0.98).
Tabella 1. Numero cumulato di nuove infezioni indotte dai falsi negativi dopo due mesi dallo screening, per prevalenza di infezioni in corso nella popolazione e per numero di riproduzione dell'infezione (sensibilità del test sierologico: 0,80; specificità del test sierologico: 0,98).

viduals who are not detected by the serological test). In addition to the MC mean, table S1 shows the MC 5th and 95th percentiles of these quantities. For instance, consider a population where the prevalence is 0.3% and the specificity and sensitivity of the serological test are, respectively, 0.98 and 0.80 (first bar in figure 1 and first row in table S1). Three important findings are noted: first, the total number of RT-PCR tests required by the procedure increases with the prevalence of ongoing infections in the population (thus the gain in comparison to administering the RT-PCR test to all subjects decreases). Second, a large number of RT-PCR tests is performed on uninfected individuals: the screening strategy requires that, in addition to the 500,000 serological tests, about 11,168 RT-PCR tests are administered, 9,968 of which undergone on people who are not actually infected. Only the remaining 1,200 tests reveals the presence of the infection. Third, the screening strategy leads to 300 false negatives, i.e. the serological test is not able to detect the presence of antibodies in 300 infected individuals, possibly because it is done in the first weeks from the onset of the infection. No test will be performed on these infected individuals, who, therefore, will not be kept in quarantine.
As the prevalence of ongoing infections increases (figure 1) the number of false negatives increases, too. On the other hand, the number of RT-PCR tests performed on the individuals with positive serological test but not currently infected slightly decreases. Leaving the specificity of the serological test unchanged, figure 2 reports the expected

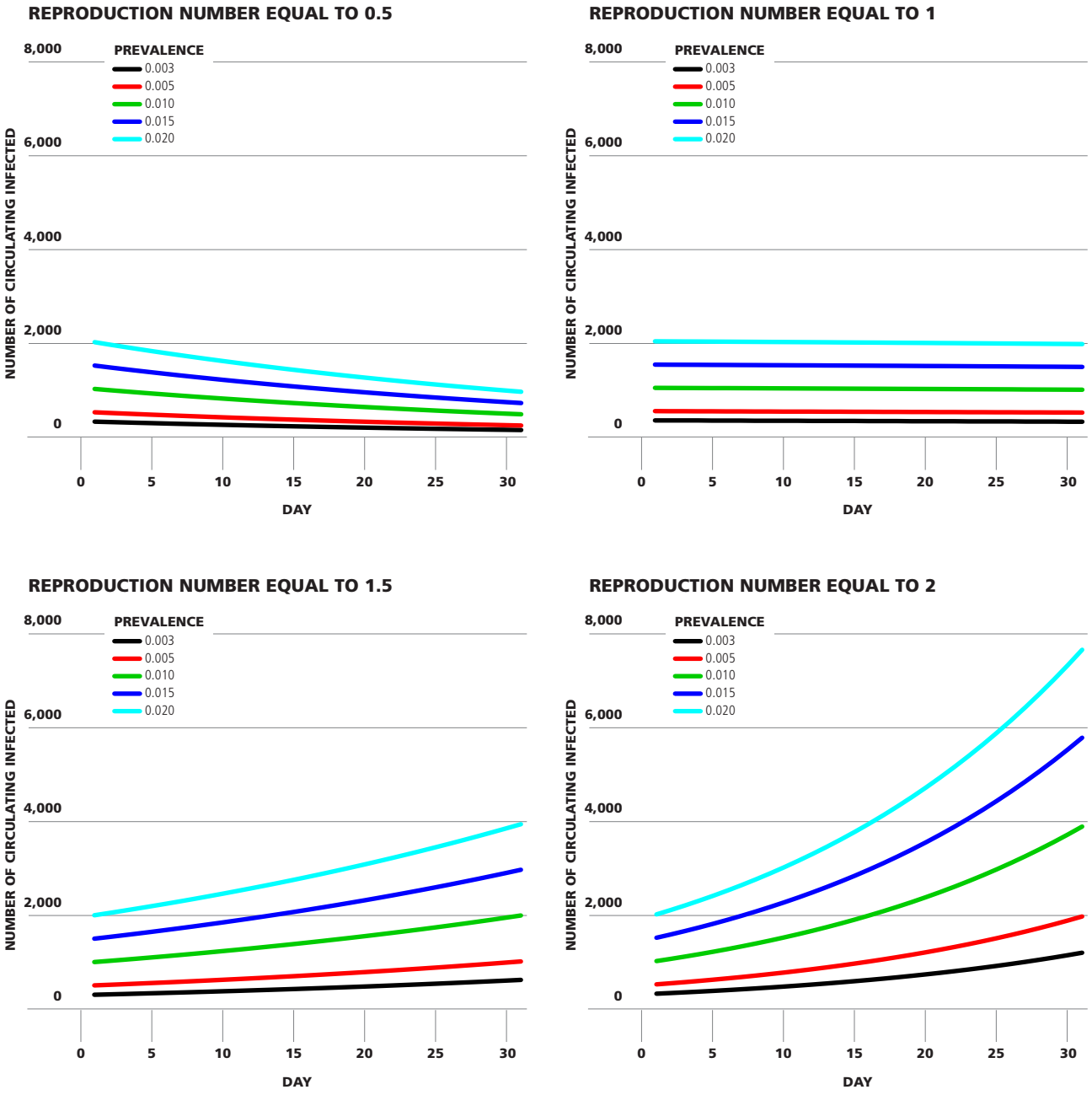

Figure 3. Number of infected subjects induced by the false negatives to the serological test, circulating in the region during the first 30 days after the screening, by initial prevalence of ongoing infections in the population and infection reproduction number.
Figura 3. Numero di individui infetti prodotti dal contatto con i falsi negativi al test sierologico, che circolano in regione durante i primi 30 giorni a partire dallo screening, per prevalneza iniziale delle infezioni in corso nella popolazione e numero di riproduzione dell'infezione.

number of false negatives as the sensitivity varies, for different values of prevalence.
The results of the SIRD model are reported in table 1 and figure 3. For this analysis the sensitivity of the test is assumed to be equal to 0.80 and a specificity equal to 0.98. In figure 3 we report the daily number of infected individuals derived from the initial false negatives, who can spread the contagion in the region during the first month after the screening. The number of infected subjects decreases over time if the reproduction number, R_0, is lower than 1; it is stable if R_0 is exactly 1 and it increases if R_0 is higher than 1. The induced epidemic strongly depends on the initial number of false negatives, and thus on the prevalence of infection at the time of the screening.
The total number of new infections derived from the initial false negatives after two months from the screening is reported in table 1. If R_0 is lower than 0.5 and the prevalence is lower than 1%, the new infected are less than 1,800. But a value of R_0 equal to 1 is sufficient to double the burden of infection originated from the unde-

tected infected individuals. If the reproduction number is equal to 2, the new infections are expected to be more than 8,000, even for very low values of the initial prevalence of ongoing infections.

DISCUSSION

The results of this study provide important insights on the cost-effectiveness of the analysed strategy. First of all, in a time of scarce availability of reagents for RT-PCR analysis, it should be carefully evaluated whether it is feasible to perform such a large number of RT-PCR tests on subjects who have resulted positive to the rapid serological test, by also considering that most of these positives are false positives, i.e. positives to the serological test but not currently infected. In this regard, it is important to remark that the specificity value used in the analysis is optimistically high and that, in situations where the probability for an individual to have been infected and subsequently recovered is higher, the number of positive serological tests on people who are not currently infected could be much more relevant. This consideration, which analytically derives from the formula in equation (2) and the underlying assumptions, is related to the fact that the sensitivity of the serological test in detecting the SARS-CoV-2 antibodies in case of past infections is very high.

The fact that, as described in the Decree,[13] serological tests will be carried out on a voluntary basis further undermines the performance of the strategy. Alternative less laborious and less costly screening procedures and strategies that improve the accuracy of the serological tests should be considered and evaluated. For instance, in order to increase the positive predictive value of the serological test, a questionnaire collecting information on SARS-CoV-2 symptoms could be firstly administered and individuals with SARS-CoV-2 symptoms encouraged to undergo the test. Additionally, with the aim to reduce the costs related to the RT-PCR tests performed at the second step of the screening procedure, individuals with a positive serological result might be preventively quarantined and undergone a second serological test after a pre-fixed time period, defined on the basis of SARS-CoV-2 antibodies kinetics. In this case, social and economic costs deriving from the preventive quarantine should be quantified as well.

From an efficacy perspective, the results of this study show that the main pitfall of the strategy is the number of undetected infections, which increases as the prevalence of subjects with ongoing infection in the population increases. False negatives could cause false reassurance, behavioral changes, and disease spread. It is also important that the number of false negatives is evaluated in the context of the epidemic. In fact, each undetected case can generate new infections and the transmission can happen with different strength, depending on many factors mainly related to the implementation of measures of physical distancing and plans aimed to early detect and isolate new cases. In the simplified SIR model used in this work R_0 was changed to define different scenarios of transmission, showing that the same number of initial undetected infections can be responsible of very different numbers of new infected individuals after two months from the screening. In particular, strategies like the one analysed in this paper might be particularly ineffective in the long-run, when, with the progressive time from the lockdown, an increase of R_0 is expected, or when applied to sub-populations where the number of potential at risk contacts is higher than among the general population, such as patients or workers of hospitals and nursing homes. In addition, it should be also considered that the relative gain in terms of resources of applying this two-step screening procedure in respect to testing all subjects through individual RT-PCR tests decreases with the prevalence, and that, as the prevalence increases, the number of false negatives increases as well. Therefore, this kind of procedure should be absolutely avoided unless the prevalence of current infections is very low. In this sense, even if in the analysis prevalence values higher than 2% are not considered, it is reasonable to assert that in case of infection outbreak or high-risk subpopulations, the serological test could show a very poor performance if the interest is in detecting ongoing infections.

Regarding the methods used, it is worth noting that, because the calculations of the quantities of interest are relatively simple, MC simulations are not strictly needed here, but they allow to perform cost-effectiveness evaluations in a simple way, sufficiently general to be extended, if necessary, to more complex problems involving composite and/or multi-stage screening procedures.

In the context of the SARS-CoV-2 pandemic, evaluation procedures such as that one used in this paper should be routinely performed before any implementation of new screening strategies on the general population or on specific subpopulations, particularly in the presence of resources constraints. Unlike in the case of non-communicable diseases, the danger deriving from cases that are not detected by the screening should be contextualized accounting for the strength of the epidemic spread, which depends not only on the social distancing measures adopted to slow the contagion, but also on the measures undertaken for early detection and isolation of the subjects with ongoing infection, thus ultimately on the effectiveness of the screening strategies themselves.

Conflict of interest: none declared.

Acknowledgements: the authors would like to thank the working group on Covid-19 at the Department of Statistics, Computer Science, Applications of the University of Florence; specifically: Carla Rampichini, Anna Gottard, Matteo Pedone, Fiammetta Menchetti, Silvia Noirjean, Cecilia Viscardi, Giulia Cereda.

Submission date: 14.07.2020
Accepted on: 07.10.2020

REFERENCES

1. Recom magazine. Vespignani: «Per sconfiggere il virus servono le 3T». Available from: https://www.reccom.org/2020/05/04/vespignani-per-sconfiggere-il-virus-servono-le-3t/
2. World health organisation. (2020) Strategies and planning. Geneva, WHO, 2020. Available from: https://www.who.int/emergencies/diseases/novel-coronavirus-2019/strategies-and-plans
3. Lippi G, Simundic AM, Plebani M. Potential preanalytical and analytical vulnerabilities in the laboratory diagnosis of coronavirus disease 2019 (Covid-19). Clin Chem Lab Med 2020;58(7):1070-76.
4. Abdalhamid B, Bilder CR, McCutchen EL, Hinrichs SH, Koepsell SA, Iwen PC. Assessment of specimen pooling to conserve SARS CoV-2 testing resources. Am J Clin Pathol.2020;153(6):715-18.
5. He JL, Luo L, Luo ZD, et al. Diagnostic performance between CT and initial real-time RT-PCR for clinically suspected 2019 coronavirus disease (Covid-19) patients outside Wuhan, China. Respir Med 2020;168:105980.
6. Sethuraman N, Jeremiah SS, Ryo A. Interpreting diagnostic tests for SARS-CoV-2. JAMA. 2020;323(22):2249-51.
7. Italian Ministry of health. Covid-19: test di screening e diagnostici. Rome, Italian Ministry fof health, 2020. Available from: http://www.normativasanitaria.it/jsp/dettaglio.jsp?id=74021
8. The Johns Hopkins center for health security. Serology-based tests for Covid-19. Baltimore, MD, USA, Center for health security, 2020. Available from: https://www.centerforhealthsecurity.org/resources/COVID-19/serology/Serology-based-tests-for-COVID-19.html
9. European network for health and technology assessment, RCRC01 Authoring team. Rapid collaborative review on the current role of antibody tests for novel coronavirus SARS-COV-2 in the management of the pandemic. Diemen, Netherlands, EUnetHTA, 2020. (last accessed: 23.06.2020). Available from: https://eunethta.eu/rcr-ot-01/
10. Deeks JJ, Dinnes J, Takwoingi Y, et al. Antibody tests for identification of current and past infection with SARS-CoV-2. Cochrane Database Syst Rev 2020;6(6)CD013652.
11. Watson J, Whiting PF, Brush JE. Interpreting a covid-19 test result. BMJ 2020;369:m1808.
12. Haugh T, Bedi S. Just because you test positive for antibodies doesn't mean you have them. Ney York Times, 2020. Available from: https://www.nytimes.com/2020/05/13/opinion/antibody-test-accuracy.html
13. Presidency of Tuscany region. Decree n. 54/2020. Ulteriori disposizioni in merito ai test seriologici rapidi connessi alla emergenza pandemia da Covid-19. Firenze, Tuscany, 2020. Available from: https://www.cicogninirodariprato.edu.it/wp-content/uploads/2020/05/ordinanza_del_presidente_n-54_del_06-05-2020.pdf
14. Baccini M, Cereda G, Viscardi C, et al. Oltre il picco: analisi e previsioni dell'epidemia da SARS-CoV-2 in Toscana. Epidemiol & Prev repository, 2020. Available from: https://repo.epiprev.it/index.php/2020/04/10/oltre-il-picco-analisi-e-previsioni-dellepidemia-da-sars-cov-2-in-toscana/
15. Murray JD. Mathematical Biology. I: An introduction. Berlin: Springer; 2002.
16. Li Z, Yi Y, Luo X, et al. Development and clinical application of a rapid IgM-IgG combined antibody test for SARS-CoV-2 infection diagnosis. J Med Virol 2020;10.1002/jmv.25727.
17. World Health Organization. Countries / Western Pacific / China. Available from: https://www.who.int/china
18. Istituto nazionale di statistica. Primi risultati dell'indagine di sieroprevalenza sul SARS-CoV-2. Roma: Istat; 2020. Available from: https://www.istat.it/it/files//2020/08/ReportPrimiRisultatiIndagineSiero.pdf
19. Gorlani P. Coronavirus a Brescia, test sierologici: il 62,2% degli isolati ha anticorpi, 1 su 5 è ancora infetto. Brescia, Corriere della sera, 2020. Available from: https://brescia.corriere.it/notizie/cronaca/20_maggio_23/test-sierologici-622per-centodegli-isolati-ha-anticorpiuno-5-ancora-infetto-cc3e316a-9cda-11ea-a31e-977f755d9d62.shtml
20. Marrone C. Coronavirus, quanto sono davvero accurati i test sierologici? Corriere della sera, 02.05.2020. Available from: https://www.corriere.it/salute/malattie_infettive/20_maggio_02/coronavirus-quanto-sono-davvero-accurati-test-sierologici-01ec7db0-8c52-11ea-9e0f-452c0463a855.shtml
21 Woodhill N. Coronavirus (Covid-19) infection survey pilot: England, 14 May 2020. London: Statistical bulletin of office for national statistics; 2020. Available from: https://www.ons.gov.uk/peoplepopulationandcommunity/healthandsocialcare/conditionsanddiseases/bulletins/coronaviruscovid19infectionsurveypilot/england14may2020
22. World health organization. Transmission of SARS-CoV-2: implications for infection prevention precautions. Geneva: WHO; 2020. Available from: https://www.who.int/publications/i/item/modes-of-transmission-of-virus-causing-covid-19-implications-for-ipc-precaution-recommendations