

Review

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Mass screening is a key component to fight against SARS-CoV-2 and return to normalcy

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Abstract: The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) had highly transmissible and pathogenic, which caused serious economic loss and hazard to public health. Different countries have developed strategies to deal with the COVID-19 pandemic that fit their epidemiological situations, capacities, and values. Mass screening combined with control measures rapidly reduced the transmission of the SARS-CoV-2 infection. The COVID-19 pandemic has dramatically highlighted the essential role of diagnostics capacity in the control of communicable diseases. Mass screening has been increasingly used to detect suspected COVID-19 cases and their close contacts, asymptomatic case, patients attending fever clinics, high-risk populations, employees, even all population to identify infectious individuals. Mass screening is a key component to fight against SARS-CoV-2 and return to normalcy. Here we describe the history of mass screening, define the scope of mass screening, describe its application scenarios, and discuss the impact and challenges of using this approach to control COVID-19. We conclude that through a comprehensive screening program and strong testing capabilities, mass screening could help us return to normalcy more quickly.

Keywords: close contacts; diagnostics capacity; mass screening; SARS-CoV-2.

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was first reported in December 2019, which had highly transmissible and pathogenic [1, 2]. On 30 January 2020, the World Health Organization (WHO)

declared the outbreak of novel coronavirus as a public health emergency of international concern (PHEIC), mainly based on that the number of infected people in China has increased and the outbreak has occurred in several countries [3]. On 11 February 2020, the WHO named coronavirus disease 2019 (COVID-19), which seriously threatens human health and public safety [4]. On 11 March 2020, the WHO officially characterized COVID-19 as a global pandemic [5]. To date, the novel coronavirus disease COVID-19 has rapidly spread to over 200 countries, areas and territories. Globally, there have been more than 214 million confirmed cases of COVID-19 and more than four million death cases.

At present, different countries have developed strategies to deal with the COVID-19 pandemic that fit their epidemiological situations, capacities, and values. Global COVID-19 prevention and control strategies mainly include containment, mitigation and suppression strategies. China mainly taken containment and suppression strategies, which effectively stopped the transmission of SARS-CoV-2 [6]. Nonpharmaceutical interventions (NPIs) have been widely used to control the transmission of COVID-19 all over the world, which were essential components of the government response to outbreaks [7–9]. These have included patient isolation, contact tracing, contact isolation, travel restrictions, wearing face masks, cancellation of mass gatherings, closure of schools and expanding social distancing [10–12]. All the measures were aimed at reducing COVID-19 transmission and returning to normal life. Testing of reported symptomatic cases and tracing their contacts aims to provide a more targeted measure, but on various occasions, it was hard to effectively control the transmission of COVID-19 [13, 14].

Fast and widespread identification of infectious people is crucial to fighting against the outbreak of COVID-19. Over the past year, the ongoing pandemic has overwhelmed testing capacity and facilities. When the COVID-19 outbreak occurs, we can identify close contacts of confirmed cases through epidemiological investigation. However, mass screening has been increasingly used to detect suspected COVID-19 cases and their close contacts, asymptomatic case, patients attending fever clinics, high-risk populations, employees, even all population to

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identify infectious individuals. Although, screening on its own will not stop the spread of SARS-CoV-2, as part of a strategy, screening integrated with following isolation of those with positive test results in Beijing and Qingdao city have managed to control the widespread transmission of SARS-CoV-2 virus. Here we describe the history of mass screening, define the scope of mass screening, describe its application scenarios, and discuss the impact and challenges of using this approach to control COVID-19. We conclude that through a comprehension screening program and strong testing capabilities, mass screening could help us return to normalcy more quickly.

The development of screening

The commission on chronic illness conference on preventive aspects of chronic disease held in 1951 [15], defined screening as “the presumptive identification of unrecognized disease or defect by the application of tests, examinations, or other procedures which can be applied rapidly. Screening tests sort out apparently well persons who probably have a disease from those who probably do not. A screening test is not intended to be diagnostic. Persons with positive or suspicious findings must be referred to their physicians for diagnosis and necessary treatment”. In general, it had taken the definition to imply a relatively simple (though not necessarily unsophisticated) method of case-finding. According to the form of the screening, the screening can be divided into mass screening, selective screening, and multiple screening.

Mass screening is a term used to indicate the large-scale screening of whole population groups [15]. It referred to screening where no selection of population groups is made. Selective screening is used for the screening of selected high-risk groups in the population. It may still be large-scale, and can be considered as one form of population screening. Multiple screening has evolved by combining single screening tests. Where much time and effort has been spent by a population in attending for a single test. It is natural to consider the economy of offering other tests at the same time. Multiple screening has been defined as “the application of two or more screening tests in combination to large groups of people” [16]. In 1967, two comprehensive multiple screenings were carried out in South East London involving 7,000 adults aged 40–65 followed-up for nine years [17]. Finally, the cost of multiple screening was too high. This was relatively ineffective and uneconomic. Hence, in 1976, a Canadian Task Force on the Periodic Examination was commissioned, leading to some reports and recommendations concerning

screening [17]. In 1984, the United States Preventive Services Task Force was formed, which in 1989 published the recommendations on screening [17]. Different organizations have different definitions of screening (Table 1).

Table 1: Definitions of screening.

Source	Year	Definition
US Commission on Chronic Illness [22]	1957	Screening is the presumptive identification of unrecognized disease or defect by the application of tests, examinations or other procedures, which can be applied rapidly. Screening tests sort out apparently well persons who apparently have a disease from those who probably do not.
Mckeown [23]	1968	Screening is medical investigation which does not arise from a patient’s request for advice for a specific complaint.
Wilson and Jungner [24]	1968	Mass screening is the large-scale screening of whole population groups. Selective screening is screening of certain high-risk groups in the population. Multiphasic screening is the administration of two or more screening tests to large groups of people. Surveillance is long-term observation of individual populations. Case-finding is screening of patients already in contact with the health services to detect disease and start treatment. Early disease detection refers to all types of screening.
NSC-First Report [25]	1998	Screening is the systematic application of test or inquiry to identify individuals at sufficient risk of a specific disorder to warrant further investigation or direct preventive action among persons who have not sought medical attention on account of symptoms of that disorder.
NSC-Second Report [26]	2000	Screening is a public health service in which members of a defined population, who do not necessarily perceive that they are at risk of, or are already affected by, a disease or its complications, are asked a question or offered a test to identify those individuals who are more likely to be helped than harmed by further tests or treatment to reduce the risk of disease or its complications.

Much screening practice evolved in the USA during the 1950s in the form of multiple screening programmes [15–19]. In 1961, Thorner and Remein of the US Public Health Service published the first comprehensive review of the principles of screening [20]. In 1968, Wilson and Jungner produced their *Principles and Practice of Screening for Disease*, which was published as a World Health Organization monograph [15]. Screening is the examination of entire population or group within population to determine their risk factors of disease and the disease at its asymptomatic stage. Successfully identifying human immunodeficiency virus (HIV) infection during pregnancy through screening tests reduced vertical transmission of HIV to less than 1% when effective combination antiretroviral therapy [21]. During the A (H1N1) 2009 pandemic, screening has the potential to detect travellers with clinical signs or symptoms of infectious disease, exit population, and targeted entry population [22]. In August 2014, the Ebola virus disease (EVD) outbreak in West Africa a public health emergency of international concern. Entry screening at airports, ports, and land crossings is the key measure for Ebola virus disease preparedness. Entry screening is a public health intervention used to identify persons with possible symptoms of, or risk of exposure to, Ebola virus disease (EVD), to prevent them from further travel. Screening measures are based upon risk and can be adapted for airports, land border crossings and sea ports. Objectives of entry screening is to identify ill travellers with signs and symptoms consistent with EVD, and their potential contacts, to identify appropriate public health measures, such as treatment and isolation, that are commensurate with the risks and do not unduly interfere with international travel, to protect the health of travellers, their families, and the population of the destination country [23]. Entry screening for Ebola prevention and control measures can also be applied to SARS-CoV-2 control. In 1999, Crossroads proposed comprehensive principles for evaluating a screening programme from an ethical, legal and social behavioural perspective. These principles should be fully considered when screening.

- (1) The burden of screening diseases is an important factor.
- (2) The target population for screening can be defined.
- (3) The disease is in the latent period or preclinical stage at a high level.
- (4) The natural history of the condition, including development from latent to declared disease, should be adequately understood.
- (5) There should be a suitable test or examination.
- (6) There is a comprehensive screening program.

- (7) The components of the screening program are continuous and well-developed.
- (8) Screening subjects can benefit from physical, psychological and social life.
- (9) Screening should be equipped with disease monitoring, prevention, treatment, education and social support.
- (10) A consensus must be reached beforehand on the measures to be taken against those who have a positive screening result.
- (11) The costs of screening should be weighed in terms of economics, psychology, sociology, medicine and health services.
- (12) Screening programmes should be accepted by the target population but should not negatively affect non-participants.
- (13) Confidentiality measures shall be taken for those screened and those not attending.

The basic steps in a screening programme for reducing disease risk in whole population (Figure 1). Figure 1 is referred to the textbook *Screening: Evidence and practice* [15]. Our goal is to screen the target population from the whole population. The initial screening process was named as “sieve”. Due to the accuracy of test results, this process will produce false negatives and uncertain test result. The subsequent testing and investigation were named as “sort”. This diagnostic phase also will produce diagnostic results negative and uncertain results. We carried out a diagnostic test for the positive population of screening test. Some interventions will be implemented for people with positive diagnoses, such as quarantine of infectious diseases. Assessment must also be an integral part of any screening process. In 1971, Cochrane and Holland came up with seven criteria included simplicity, acceptability, accuracy, cost, repeatability, sensitivity, and specificity for evaluation, which are still in effect today [24] (Table 2).

Scope of mass screening for SARS-CoV-2

Mass screening for SARS-CoV-2 is the most practical way to solve the current crisis [25, 26]. Screening test to identify SARS-CoV-2 infections is critical to controlling the global COVID-19 pandemic starting in late 2019. In some countries, the large-scale use of screening tests have become a cornerstone of successful containment strategies [27]. In 2020, Singapore used a comprehensive surveillance system to detect as many cases as possible, and to contain

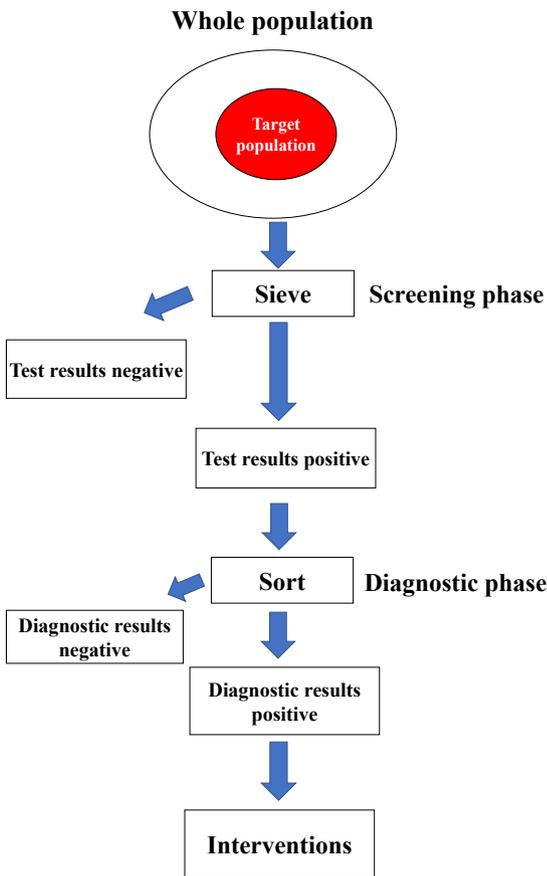


Figure 1: The flow diagram of screening. The targeted population were screened from the whole population. The first phase was “sieve”, which could test the positive population. The second phase was “sort”, which could diagnostic the positive population. This process removes some false positives. We can take the interventions to the targeted population.

Table 2: Summary of the screening evaluation.

Index	Criteria
Simplicity	The test should be simple to perform, easy to interpret and, where possible, capable of use by paramedics and other personnel.
Acceptability	Since participation in screening is voluntary, the test must be acceptable to those undergoing it.
Accuracy	The test must give a true measurement of the condition or symptom under investigation.
Cost	The expense of the test must be considered in relation to the benefits of early detection of the disease.
Repeatability	The test should give consistent results in repeated trials.
Sensitivity	The test should be capable of giving a positive finding when the individual being screened has the condition being sought.
Specificity	The test should be capable of giving a negative finding when the individual being screened does not have the condition being sought.

them at the individual level [27]. At the same time, it coupled with community-based measures proportionate to the transmission risk. This strategy has been effective in containing spread. Singapore’s surveillance for COVID-19 aimed to identify as many cases as possible using complementary detection methods. The detection methods of mass screening included real-time reverse transcription polymerase chain reaction (RT-PCR), antigen tests, and serological tests. In Singapore, PCR testing for COVID-19 is available at all public hospital laboratories to increase diagnostic capacity. Serological tests were used to investigate linkages between cases and clusters. RT-PCR detection of respiratory tract specimens in the laboratory is the reference standard for the diagnosis of COVID-19. Besides, point-of-care techniques and serological immunoassays are emerging rapidly. There are more and more screening methods. But there are still many problems in different countries. Many wealthy countries have faced challenges in providing testing and specimen collection that have inhibited rapid growth in testing capacity [28]. In a low-resource environment, these challenges may be even greater [28]. Urging clinical and public health needs are now driving an unprecedented global effort to improve the detection capacity for SARS-CoV-2 infection.

Diagnostic testing was implemented at different time among persons with proven or suspected COVID-19 (Figure 2). The mass screening could be used to test the asymptomatic infection at incubation period of SARS-CoV-2 infection. Test design must consider several parameters, such as assay sensitivity, specificity testing, the limit of detection, turnaround time, throughput, batch processing and cost. It is important to recognize the acceptable diagnostic accuracy of the test [29]. Although real-time reverse transcription polymerase chain reaction (RT-PCR) analyzed for respiratory specimens in the laboratories is the cornerstone of COVID-19 diagnostic tests, several novel or adjuvant diagnostic methods are developed and evaluated [30, 31]. Accurate rapid screening tests for SARS-CoV-2 infection could contribute to clinical and public health strategies to manage the COVID-19 pandemic. So, we have summarized some commonly used screening methods for COVID-19 detection, included molecular tests, antigen tests, and serologic tests (Table 3). The advantages and disadvantages of diagnostic testing for SARS-CoV-2 were showed in Table 3.

Molecular tests

Molecular tests are widely used to screen for COVID-19. The current gold standard for virus detection is nucleic acid

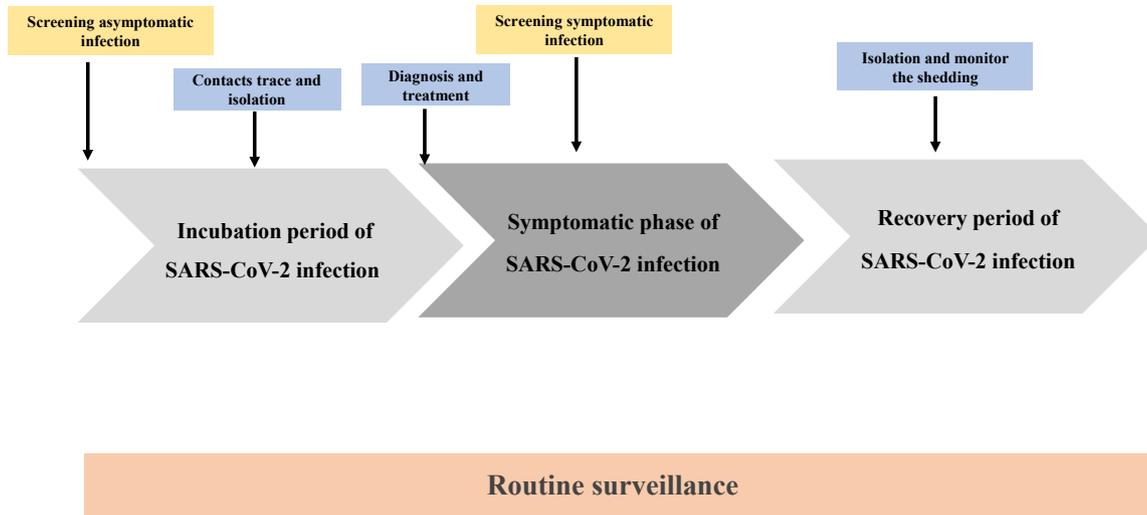


Figure 2: The diagnostic testing among persons was used to screened the patients of COVID-19. The screening could conduct from the incubation period to the symptomatic phase. At different phase, different measures were implemented.

Table 3: The advantages and disadvantages of diagnostic testing for SARS-CoV-2.

Test methods	Advantages	Disadvantages
Molecular tests	Wide dynamic detection range, wide application range, high specificity and sensitivity	Requires higher laboratory conditions, amplification efficiency is easily affected, takes at least several hours
Antigen tests	Rapid test to results and low-cost detection	The sensitivity is relatively low
Serologic tests	Assess individual and population immunity	Likely false native in early disease

amplification test, which is identified with nasopharyngeal swab by quantitative RT-PCR. In acute respiratory infection, RT-PCR assay is the most effective method for detection causative viruses from respiratory specimens. The study found that RT-PCR assays were highly sensitive, with best results obtained for E gene and RdRp genes of SARS-CoV-2 assays (5.2 and 3.8 copies per reaction at 95% detection probability, respectively) [32]. The limit of detection from tests were 3.9 and 3.6 copies per reaction for E and RdRp genes, respectively [32]. In China, the detection kits of SARS-CoV-2 target highly conserved ORF1ab and N genes for RT-PCR. Another study targeted the ORF1ab and N gene regions of SARS-CoV-2, they developed the RT-PCR assays to detect SARS-CoV-2 in human clinical specimens [33]. The assays have a dynamic range of at least seven orders of magnitude and the limits of detection were ≥ 10 copies per reaction of the positive control plasmid [33]. The oropharyngeal and nasopharyngeal swab should be transferred

into universal transport medium, which were used to detect for RT-PCR. By monitoring SARS-CoV-2 viral loads in upper respiratory specimens obtained from patients, higher viral loads (inversely related to Ct value) were detected soon after symptom onset, with higher viral loads detected in the nose than in the throat by RT-PCR assays [34]. The mean viral load of severe cases was around 60 times higher than that of mild cases, suggesting that higher viral loads might be associated with severe clinical outcomes, compared the viral dynamics in mild and severe cases of COVID-19 [35].

All kinds of clinical specimens, such as bronchoalveolar lavage, sputum, saliva, nasopharyngeal swabs, oropharyngeal swabs, feces, and blood for detection of SARS-CoV-2 virus were evaluated. Nasopharyngeal swabs and oropharyngeal swabs are currently standard respiratory specimens of the upper respiratory tract recommended for diagnostic testing for COVID-19. However, the collection of nasopharyngeal swabs presents challenges, such as exposure to health care workers, supply chain constraints related to swabs and personal protective equipment, and difficulties with self-collection.

Saliva samples are very easy to collect, which could solve the problem of nasopharyngeal swabs collected. The study evaluated the nasopharyngeal swabs and saliva samples collected from individuals in either a healthcare or a community setting [36]. There are two protocols for nucleic acid extraction from saliva samples, one is direct nucleic acid extraction, just like nucleic acid extraction from nasopharyngeal swabs [36]. Another protocol was that the saliva samples were processed with homogenization step using a bead mill homogenizer before RNA extraction [36]. The first protocol showed that lower

sensitivity (50%) of saliva samples compared with nasopharyngeal swabs samples (89.7%). The concordance for positive results between the two tests was only 39.7%. The detection rate in saliva samples was significantly lower compared with that in nasopharyngeal swabs samples. Besides, the cycle threshold values of N and ORF1ab genes were significantly higher in saliva samples compared with nasopharyngeal swabs. The second protocol demonstrated that the detection rate for SARS-CoV-2 was significantly higher in saliva compared with nasopharyngeal swabs (97.8 vs. 78.9%). The concordance for positive results between two tests was 76.8%. Besides, the cycle threshold values of N and ORF1ab genes were significantly lower in saliva samples compared with nasopharyngeal swabs. These results suggested that the sensitivity of saliva samples observed in the first protocol was lower due to inadequate sample handling compared to the second protocol. A five-sample pooling strategy with saliva was successful validation. The pooled testing results demonstrated that the positive percentage agreement was 95%. The study showed that the saliva samples are more sensitive than nasopharyngeal swabs samples if properly collected and treated prior to extraction and PCR. However, it must be emphasized that homogenization is an additional step in the workflow that will increase processing time compared with nasopharyngeal swabs samples. In addition, laboratories intending to perform saliva testing in different regions of the world should evaluate similar homogenizing methods, because homogenizers may not be available in these regions. A study was implemented to examine the use of weekly saliva RT-PCR testing for SARS-CoV-2 detection among nursing home workers as a strategy to control disease transmission within nursing home in Belgium. Weekly saliva RT-PCR testing for SARS-CoV-2 demonstrated large-scale feasibility and efficacy in disrupting the chain of transmission [37].

The method of RT-PCR poses wide dynamic detection range, wide application range, high specificity and sensitivity. To assess the diagnostic accuracy of molecular tests for SARS-CoV-2 infection, a systematic review showed that the sensitivity and specificity of molecular tests were 95.1 and 98.8%, respectively [38]. But RT-PCR detection method requires higher laboratory conditions, and the amplification efficiency is easily affected by PCR inhibitors [39, 40]. Besides, the method takes at least several hours.

In 1943, Dorfman used the pooling strategy to detect the affected members of large population [41]. Dorfman's strategy was to test all groups, and then to test every number of every infected group. A pool was considered positive if the viral gene was amplified, and individual

samples within the pool were re-tested individually. To identify individuals infected with SARS-CoV-2, the naive approach is to test everyone separately. However, it is far more efficient to pool samples and test these sample pools together at low prevalence. Cost reductions can be achieved by pooling subsamples and testing them in groups. At present, the pooling strategy was used for SARS-CoV-2 tests [42]. A balance must be considered between increasing the group size and retaining test sensitivity, as sample dilution increases the likelihood of false negative test result for individuals with a low viral load in the sampled region at the time of the test [43]. The use of group testing by RT-PCR on a massive scale to monitor infection rates closely and continually in a population, along with the rapid and effective isolation of people with SARS-CoV-2 infections, provides a promising pathway towards the long-term control of coronavirus disease 2019 (COVID-19) [43]. By used and monitored a large-scale, adaptive 8- and 5-samples pooling of nasopharyngeal sample lysates were used to detect the SARS-CoV-2 [44]. Data analysis of nearly 135,000 pooled samples revealed high empirical efficiency of sample pooling, outweighing a minor, clinically insignificant loss of sensitivity [45]. The pooled testing strategy spared 76% of RNA extraction and RT-PCR tests, even in the setting of a changing prevalence rate (<1%–6%) [44]. In UK, pooled testing for SARS-CoV-2 could provide the solution to testing strategy [45]. Pooling of up to 30 samples per pool can increase test capacity with existing equipment and test kits and detects positive samples with sufficient diagnostic accuracy [46]. Another study evaluated the pooled testing of nasopharyngeal swabs for SARS-CoV-2 by comparing the sensitivity of individual sample testing with 4- and 8-pool sample testing [47]. The results showed that the sensitivity becomes considerably lower when the samples are pooled, which is high percentage of false negative reports with larger sample pool size and when the patient viral load is low or weak positive samples [47]. In our country, the pooling strategy was widely used for the mass screening of SARS-CoV-2 in Beijing and Qingdao. The pooling detection strategy in China generally adopts 5 samples or 10 samples for testing (Figure 3). This measure saves the test time and cost, which effectively detects the nucleic acid of SARS-CoV-2.

Antigen tests

Overall, the preferred screening test is simple, inexpensive, acceptable to the participants, and has a high sensitivity and specificity. Although RT-PCR is accurate and reliable,

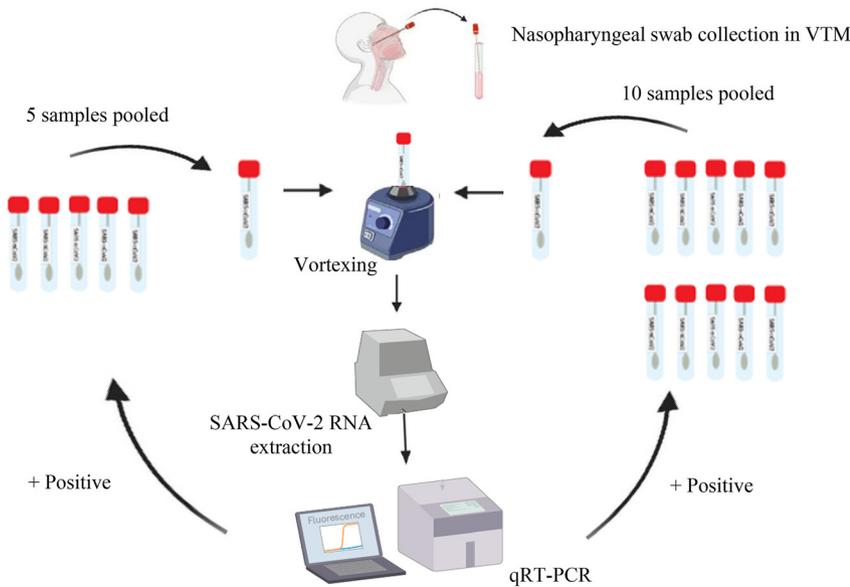


Figure 3: The schematic overview of sample pooling strategy of SARS-CoV-2 detection for mass screening. We can collect the nasopharyngeal swabs samples. Pooled testing of 5 and 10 samples pooled for SARS-CoV-2 virus was implemented when mass screening for population. A pool was considered positive if the viral gene was amplified, and individual samples within the pool were re-tested individually.

it takes a long time as a screening test. A rapid alternative is serological detection based on antibody, but these methods cannot be used for early diagnosis of infection. Theoretically, rapid antigen testing can provide the advantages of rapid test to results and low-cost detection of SARS-CoV-2. Tests for detecting respiratory syncytial virus or influenza virus antigens by immunoassay directly from clinical specimens have been commercially available for decades with low complexity and can provide results within minutes at the point of care [48]. Current tools for influenza and respiratory syncytial virus lack optimal sensitivity to exclude disease, and the same challenge is likely to be present for SARS-CoV-2, where testing needs to be conducted under clear guidance of correct interpretation [49, 50]. Rapid antigen testing method has been widely used for detection the human coronavirus included severe acute respiratory syndrome (SARS), NL63, 229E [51–53]. In 2016, the antigen test of middle east respiratory syndrome coronavirus (MERS-CoV) was developed, which based a monoclonal antibody to detect N protein for MERS-CoV. Despite its high specificity, the rapid MERS-CoV antigen test has a moderate sensitivity [54]. Indirect immunofluorescence with virus specific monoclonal antibody for the detection of human coronaviruses strains 229E and OC43 could detected coronaviruses in cell suspensions with infectious viral titers of 4.25 log TCID₅₀/mL for 229E and 2.0 log TCID₅₀/mL for OC43, but this method had low sensitivity compared with RT-PCR [55]. A fluorescence immunochromatographic assay for detecting nucleocapsid protein of SARS-CoV-2 in nasopharyngeal swab sample

and urine within 10 min was developed, the results were consistent with nucleic acid test for the same samples [56]. The nucleocapsid protein assay is an accurate, rapid, early and simple method for diagnosis of COVID-19, but the sensitivity is relatively low. The high viral titers may improve the diagnostic sensitivity of rapid antigen tests for human coronaviruses [57].

Quantitative RT-PCR detection of SARS-CoV-2 from nasopharyngeal swab specimens requires medical personnel with protective equipment and takes a long time. By eliminating these problems, self-collected saliva tests have a substantial logical advantage over non-self-collected nasopharyngeal swab specimens. In June 2020, a chemiluminescence enzyme immunoassay was developed to detect SARS-CoV-2 nucleoproteins in nasopharyngeal swab samples or saliva samples within 35 min in Japan [58]. A chemiluminescence enzyme immunoassay was used monoclonal antibodies against SARS-CoV-2-N protein. At the same time, the collected saliva specimens were extracted RNA, which were detected by RT-PCR. The results showed that the antigen concentration detected by chemiluminescence enzyme immunoassay was highly correlated with RNA viral load detected by RT-PCR, indicating that chemiluminescence enzyme immunoassay is a reliable and accurate detection method. Previous study on the SARS-CoV-2 antigen in nasopharyngeal swab specimens showed the detection results were consistent with the results of RT-PCR in 91.4% of 313 samples and 98.2% of 548 samples [59, 60]. Kobayashi reported showed the concordance was lower with 66% of 100 nasopharyngeal swab

specimens [61]. The limitations of the study included the lack of longitudinal clinical follow-up and the low number of positive cases in the airport isolation cohort [58]. They were unable to verify that whether the participants who tested negative did not subsequently infected COVID-19 [58]. A systematic review showed that the sensitivity in symptomatic, symptomatic (up to seven days from onset of symptoms), and asymptomatic infection were 72, 78.3 and 58.1%, respectively [38]. Average specificities were high in symptomatic and asymptomatic participants, and for most brands the overall summary specificity was 99.6% [38]. Simple antigen-based tests, if sufficiently sensitive, may be useful in low-resource settings and home settings to provide isolation and spatial isolation measures for patients without serious disease and their contacts.

Serologic tests

Serological assays are particularly important for antibody testing in the diagnosis of novel and emerging human coronaviruses included SARS-CoV and MERS-CoV [62, 63]. Antibodies are formed by the body's immune system in response to infections, and can be detected in whole blood, plasma or serum for laboratory. In these cases, the affected patient may not test positive for viral RNA, especially in the early stages of disease, but an immune response can be retrospectively shown to have occurred.

Serology tests to detect the presence of antibodies to SARS-CoV-2 could identify previous SARS-CoV-2 infection, and may help to confirm the presence of current infection. Serology can be used as an adjuvant diagnostic tool when SARS-CoV-2 is detected, especially rapid antigen and/or molecular assays are neither available nor stable. Serological tests used to identify IgA, IgM, and IgG antibodies against SARS-CoV-2 from clinical blood or saliva specimens, such as enzyme linked immunosorbent assay (ELISA), may be less complex than molecular tests and have the potential to be used for diagnosis in some cases [64]. A patient infected with SARS-CoV antibody response can take from days to weeks to reliably detect [64]. Negative results do not rule out SARS-CoV-2 infection, especially in those who have had recent exposure to the virus. Antibody cross-reactivity to non-SARS-CoV-2 coronavirus proteins is also a potential problem, so the positive results could be the result of past or present infection with other human coronavirus [65]. Serological tests may be more relevant when patients receive medical treatment for late-stage complications of the disease, when RT-PCR may be false negative because the virus declines over time [66].

The development of serological tests that can accurately assess prior infection and immunity to SARS-CoV-2 is critical for epidemiological studies, ongoing surveillance, vaccine research, and potential risk assessment for health care workers. Immunoassays have been marketed in some countries, but their diagnostic accuracy and optimal use remain unclear. In China, many serology devices have received urgent approval from the National Medical Products Administration (NMPA), such as Wongfo Biotech, Innovita Biological technology, and Wantai Kairui Biotechnology.

Antibody tests are being considered and evaluated for both identification of past SARS-CoV-2 infection and current infection. The antibody test sensitivity was strongly related to time since onset of symptoms, with low sensitivity (IgG 66.5% and IgM 58.4%) between 8 and 14 days [67]. The sensitivity for IgG and IgM were 88.2 and 75.4%, respectively between 15 and 21 days. Between 22 and 35 days, the sensitivity for IgG and IgM were 80.3 and 68.1%, respectively. The specificity was high (>98%) for IgG and IgM antibody. The sensitivity of antibody screening test is too low in the first week since symptom onset to have a primary role for the diagnosis of COVID-19, but they may still have a role complementing other testing in individuals presenting later. Antibody tests are likely to have a useful role for detecting previous SARS-CoV-2 infection if used 15 or more days after the onset of symptoms.

New technologies, such as clustered regularly interspersed short palindromic repeats (CRISPR) based diagnostics, are being used to develop fast, simple, low-cost, portable, and temperature-stable detection methods for deployment in non-traditional and resource-limited environments, such as airports and border crossings. The COVID-19 pandemic has greatly highlighted the important role of diagnostic technology in controlling infectious diseases.

The COVID-19 pandemic has dramatically highlighted the essential role of diagnostics in the control of communicable diseases. Generally, RT-PCR for nuclear acid test is used to detect the current infectious individuals, while serum antibody test is mostly used to investigate the COVID-19 prevalence. Mass nucleic acid screening has been used in various forms and it has important differences from testing that aims to reach a diagnosis when someone has sought help for a problem, it usually carries out among people with no signs or symptoms, with the aim of early quarantine, and rapid finding and testing of close contacts, and interrupt spread. At present, most countries performed mass screening by using highly sensitive and specific nucleic acid testing assays to detect SARS-CoV-2 infection. However, nucleic acid testing has some limitations, including not only cost- and infrastructure-related issues

but also possible false negative results, as suggested by the discrepancy observed among viral RNA testing, antigen test, serology test, and clinical symptoms in some cases. A study showed that the combination of viral RNA and antibody detection significantly improved the sensitivity of COVID-19 diagnosis, even in the early phase of one week after the onset of symptoms [68]. Intensive deployment of diagnostic technology may help a small number of countries succeed in controlling transmission [69]. Urgent clinical and public health needs are now drive an unprecedented global effort to improve SARS-CoV-2 detection capabilities.

Application of mass screening for SARS-CoV-2

The outbreak of SARS-CoV-2 has placed unprecedented strain on worldwide. Isolation of cases and contact tracing is used to control outbreaks of infectious diseases, and has been used for the COVID-19 [70]. A test, trace, and isolate system can effectively tackle the COVID-19 outbreak all over the world [71]. Under this premise, we should first conduct diagnostic test to identify SARS-CoV-2 infections. Up to now, the transmission risk of SARS-CoV-2 was from asymptomatic carriers. Previous studies have shown that asymptomatic individuals infected with SARS-CoV-2 virus were infectious, and might subsequently become symptomatic [72]. It was obvious that asymptomatic carriers can transmit the virus even if they were asymptomatic. Based on estimated sequential intervals and incubation periods, modeling suggested that up to 44% of transmission occurred before symptoms appeared [73]. The possibility of transmission by asymptomatic carriers was considered a possible driver of the pandemic. Appropriate measures were recommended, such as mass screening in pre-symptomatic and asymptomatic individuals in high-risk communities. However, whether asymptomatic carriers play an essential role in population-based transmission has remained an essential question.

Mass screening has been suggested as a systematic means of identifying positive carriers including asymptomatic individuals. It was used early in the pandemic in China and South Korea [74]. Asymptomatic individuals are similarly infectious as symptomatic patients, meaning that population wide containment strategies must rely on mass screening to ensure an early breaking the chains of infection if these can be delivered effectively. Containment of future outbreaks will largely depend on early detecting in work departments and geographic areas.

Mass screening combined with other control measures was highly successful in rapidly reducing SARS-CoV-2 transmission in some countries [75]. Mass screening could be able to maintain the health and welfare of our staff, rapid identification and isolation of infected health-care workers so as to protect patients and the wider community, and more rapid return to work of staff during this challenging period [76]. Mass screening has been used in various forms and it has important differences from testing that aims to reach a diagnosis when someone has sought help for a problem, it usually carries out among people with no signs or symptoms, with the aim of early quarantine, and rapid finding and testing of close contacts, and interrupt spread. Mass screening has played important role in controlling the outbreak of COVID-19.

Mass screening for SARS-CoV-2 in China

The mass screening in outbreak response mainly focused on early detection of infectious individuals among screened population. The strict lockdown measures were imposed in Wuhan from January 23 to April 8, 2020. During the first two months after Wuhan reopened, sporadic cases still appeared. However, there were still concerns that the risk of COVID-19 outbreak will seriously affect industrial production and social services, and restrict the normal life of residents. In order to determine the COVID-19 epidemic of Wuhan, the government of Wuhan performed a citywide nucleic acid screening for SARS-CoV-2 between May 14 and June 1, 2020. All city residents aged six years or older were eligible and 9,989,828 (92.9%) participated [77]. No new symptomatic cases and 300 asymptomatic cases were identified. The detection rate was 0.303/10,000 and the prevalence of SARS-CoV-2 infection in Wuhan was very low [77]. All asymptomatic positive cases, positive cases, and their close contacts were isolated for at least two weeks until the results of nucleic acid testing were negative. In the screening programme, 76.7 and 23.3% of the samples underwent single and mixed testing, respectively. The screening of COVID-19 infection in Wuhan is a large-scale programme in post-lockdown settings, which offered the valuable lessons of international significance as more countries and cities around the world entering the post-lockdown phase. This study provides the organization process, detailed technical methods used, the results of mass nucleic acid screening, and has important reference significance for other countries. In order to resume industrial production, social services, and the normal lives of residents, the city government of Wuhan

carried out a comprehensive citywide nucleic acid screening of SARS-CoV-2 infection. The impacts and cost-effectiveness of the citywide screening of SARS-CoV-2 infections on population's health, health behaviours, economy, and society are required to fully evaluate.

On 11 June 2020, a 52-year-old man suffering from fever and cough was diagnosed with COVID-19 in Beijing, after a 56-day zero new case interval. By RT-PCR tests, two environmental samples from Xinfadi Market (XFDM) were positive for SARS-CoV-2. To evaluate the extent of infection spreading, a screening campaign of SARS-CoV-2 infection was implemented over the city by Beijing Center for Disease Prevention and Control. Between 15 June and 10 July, a total of more than 10 million citizens, and 5,342 environmental samples were screened [25]. Eventually 368 positive cases were confirmed [25]. On October 11, 2020, three cases of SARS-CoV-2 were reported in Qingdao. The local authorities launched aggressive contact tracing and isolation of close contacts of confirmed infection. The government of Qingdao launched a city-wide, grid-based mass screening by RT-PCR. In order to minimize processing time and save resources, a pooled test method was used. Every pool contained from 3 to 10 people. If a pooled sample was positive, individual sample in the pool was launched RT-PCR testing [26]. Until October 16, a total of 10.9 million people were tested. Nine positive cases related to the original cluster were tested using pooled testing [26]. All testing were identified 12 cases. Testing millions of people in a short period is challenging and requires effective coordination and implementation, as well as the cooperation of residents.

In July 2021, the B.1.617.2 variant of SARS-CoV-2 was widely spread in Nanjing. In addition, the B.1.617.2 variant spread other cities from Nanjing. Mass screening was carried out to identified the infected individuals and curb the epidemic in Nanjing and other cities.

Mass screening for SARS-CoV-2 in New Zealand

An aggressive approach has enabled New Zealand to end community transmission of SARS-CoV-2. In New Zealand, the full lockdown measure allowed the country to effectively manage borders, and do contact tracing, testing, and surveillance. New Zealand has done a large amount of testing as a measure of COVID-19 eliminating, the testing included large scale testing of symptomatic people in early outbreak detection and testing of specific communities who are at higher risk of acquiring the virus [78].

Mass screening for SARS-CoV-2 in Slovakia

With the development of rapid antigen tests, population screening for SARS-CoV-2 infection has become feasible and could reduce transmission. Mass screening combined with other control measures was conducted in Slovakia. In October 2020, Slovakia became the first country in the world to use rapid antigen tests in a campaign targeting the whole population in order to identify infections. The effectiveness of population-wide, rapid antigen test-based screening and other control measures in reducing SARS-CoV-2 infection prevalence is reported [79]. The study evaluated the impacted of repeated mass antigen testing coupled with quarantine measures on the spread of the COVID-19 pandemic using epidemiology data from Slovakia, which could be an effective tool in mitigating pandemic [80]. The most important limitation of this observational study is that we were unable to clearly distinguish the effect of the mass testing campaigns from that of the other nonpharmaceutical interventions introduced at a similar time, that have led to a reduction in contacts and mobility.

Mass screening for SARS-CoV-2 in Luxembourg

The strict lockdown measures were imposed by Luxembourg government in mid-March 2020 [81]. This was followed by a gradual easing of restrictions, which aimed at launching population-wide SARS-CoV-2 screening programmes to pre-emptive break the infection chains. RT-PCR was performed using SARS-CoV-2 single well dual-target (ORF1ab and N gene) assay for nucleic acids extracted from oropharyngeal swabs [81]. To save time and cost, a collection strategy was implemented involved mixing samples from four different individuals prior to RNA extraction and re-analyzing samples a second time separately from the positive sample pool. The mass screening identified 850 cases and an additional 249 cases resulting from contact tracing [81]. To assess the impact of mass screening on the epidemic dynamics, the SEIR model was used to evaluate the situation in Luxembourg in terms of implemented measures and identified case by mass screening and contact tracing. The analysis of the amplitude of active cases showed 29% would have increased in the peak of active cases without mass screening. The positive effect of mass screening was also highlighted by the total number of cases, which would have been 42.9% higher if the testing strategy had not been implemented.

The synergistic effect of mass screening and contact tracing was investigated to close tracing of mass screening indicator cases in the simulation. The effect of mass screening could be reduced by more than half without contact tracing.

Mass screening for SARS-CoV-2 in South Korea

South Korea experienced the COVID-19 outbreak and performed the rapidly screening testing. In the early epidemic phase, South Korea carried out the aggressive testing policy, which reduced the transmission and reproduction number, and the availability of medical interventions and resources for severe patients was related to a reduction in case fatality rate [75]. This study showed that the epidemiological spread patterns of COVID-19 in Korea and suggested the effectiveness of intervention policies based on aggressive testing and policies for severe patient care.

Mass screening for SARS-CoV-2 in Liverpool, UK

Liverpool was the first city in England to roll out mass testing of its population for COVID-19. All people living or working in Liverpool were be offered COVID-19 tests from Friday 6 November, regardless of whether they have symptoms [82]. It is urgent the capacity of weekly viral detection. The diagnosis capacity, together with intensive contact tracing, could enable the country to resume normal life immediately. Within the tested population anyone infected would be detected within about a week of becoming infectious. This emergency measure would only be needed for about two months but could be rapidly reintroduced to control any future epidemic caused by the COVID-19 [74]. Measures to prevent household transmission and to support those in isolation will be vital and might be a core part of a government's policy. It is time that UK government carried out the measure of "test and trace" to "test, trace, isolate, and support" [83]. However, mass testing for COVID-19 in the UK caused an unevaluated, under designed, and costly mess. According to the UK's National Screening Committee for appraisal of a programme's viability, effectiveness, and appropriateness, it does not do well and has been already roundly criticized [84–86].

When mass screening is conducted, all potential high-risk populations were tested for SARS-CoV-2 by diagnosis methods. This includes not only the close contacts, but

also persons that exposed to polluted environment, and community-population that living in the community where test positive case was detected. Mass screening combined with other control measures can rapidly and effectively control the epidemic of COVID-19.

Pros and Cons of mass screening for SARS-CoV-2

Screening is a strategy to detect an unrecognized disease or symptoms in individuals who are asymptomatic. In the case of screening for SARS-CoV-2 infections, the goal of screening is to detect infected individuals to reduce onward transmission. Improved testing capacity is the core of the mass screening program. Study showed if large scale testing was not available in Wuhan, the turning point of suspected SARS-CoV-2 case number will comes six days later, and the cost of isolating the suspected cases will 124% increased [87]. As we have known that asymptomatic persons account for approximately 40%–45% of SARS-CoV-2 infections [88], the RT-PCR in early outbreaks allow the early detection and isolation of cases who has not shown symptoms, consequently, reduce the risk of transmission by asymptomatic and pre-symptomatic cases [89]. The same goal can be reached by mass testing of community population, and the large scaled community testing allow a clear picture of the epidemiological situation on the ground. The contribution to work resumption is also meaningful, as work resumption is an important step of returning to normal life [90].

However, there is opposite view toward mass screening. The most controversial thing is the cost [91]. Undeniable, mass testing is financial, material and human source consuming. Mass screening was a cross-sectional screening programme, and it was hard to assess the test results with change over time. A systematic review with meta-analysis was performed to evaluate the sensitivity and specificity of diagnostic tests for COVID-19. According to the type of sample, the sensitivity of nasopharyngeal aspirate/swab and throat swab were 73.3% higher than the rectal stool, urine, and plasma [92]. While the testing capacity and materials storage are still very limited in some countries [77], it is unseemly to test the general population whose positive rate may very low. Another concern is the risk of transmission caused by mass screening, as tests are usually conducted for a large number of peoples at the same time, which poses an inherent risk of transmission of infectious disease. Widespread COVID-19 testing is essential to suppress the viral transmission. A big concern is

availability, but the accuracy of detection is a long-term consideration. Although RT-PCR is relatively accurate, false positives and false negatives will accumulate as mass screening strategies are employed [93]. On one hand, misdiagnoses could have major implications on the ability to detect the infections individuals [94]. On the other hand, false alarms may cause unnecessary isolation to those innocent people and their close contacts [95]. Although a positive nucleic test can indicate the presence of viral RNA, some false negative results may be due to the relatively low viral load in asymptomatic infected people, and the accuracy of the diagnostic method cannot be accurately detected [93]. We found only modelling studies on the effectiveness question with unrealistically positive assumption the effectiveness question with unrealistically positive assumptions [96]. Regarding effectiveness of screening for SARS-CoV-2 infection, there wasn't study addressing this question [96].

The need to mobilize sufficient medical personnel to conduct the nasopharyngeal swabs could be a major obstacle to countries. Nasopharyngeal swabs can be self-administered and therefore reduce demand on trained personnel and transmission risk in the process of sample collection or even may enable testing at home. Mass screening was used rapid antigen tests for the whole population in Slovakia. Rapid antigen tests were less sensitive in detecting infections with low viral load but have been found to detect the vast majority of infectious infections. So, it easily caused false negatives. Mass screening in UK was used the lateral flow test, with a high false negative rate. This is also a huge problem in a low prevalence setting. Besides, in the mass screening process, data protection and privacy laws were ignored [97]. It is potentially in breach of the *Declaration of Helsinki* [97]. Ethical and law problem is also being concerned in terms of refusal testing and privacy protection [98]. Some countries proactively open the data of confirmed cases to the public or share it with medical institutions to find close contacts more efficiently [99]. This resulted in too much information leakage. Based on some of the above reasons, mass screening programme has been blamed as unevaluated, under designed, and costly mess [84].

Essential condition of mass screening for SARS-CoV-2

There are certain conditions that must be met for a successful mass testing program. First, the mass screening

program must have clear aim. The aim of testing determines the target population and the methods of test, and clear aim helps the stakeholders to make a rational plan. Second, the workers should have a reasonable plan and overall arrangement. Mass testing consumes a lot of manpower, material and financial resources, and involves many parts, so a comprehensive programme plan is critical to achieve the goal. The programme should not only define the processes of sample collection and testing but also have to involve the interpreting of test results and disposal of test-positive and -negative cases. Importantly, mass screening should be used combined with followed isolation and contact tracing measures. Third, the laboratory possesses reliable reagent and unified methods. Fast point-of-care tests were used in some places, however, the accuracy of the methods is unsure. Fourth, sufficient facilities and manpower that able to test the vast quantities of samples. Mass testing means that testing must be widely available, large university and commercial laboratories will contribute to the roll-out, supplementing the capacity of the lighthouse labs. Fifth, the sampling site requires proper field organization. To curb the chances of infection in crowds waiting for test, sample collections is suggested to be done by physicians in outdoors or in a separate facility outside the main building complexes. In addition, temperature monitor, mandatory mask wearing and >1 m social distance are required to be complied by the waiting crowds. When going to the sampling site, the participants need wear mask and follow the command and guidance of the field staff.

Prospects of mass screening for SARS-CoV-2

Mass screening is an intervention strategy for COVID-19 control in the general population regardless of the presentation of symptoms (Figure 4). Mass screening was considered to be a viable strategy to control the SARS-CoV-2 epidemic. The strategy can identify the isolate asymptomatic cases in the early stages of infection and reduce the risk of virus transmission [100]. China has conducted mass screening in many cities, including Beijing, Qingdao, Shijiazhuang, Dalian, Kashi, and Nanjing. Luxembourg and Slovakia also implemented similar mass screening at the national level to allow partial lockdown measures to be eased. The large-scale testing conducted after the reopening of Wuhan city was mainly aimed at ensuring that Wuhan was not affected by COVID-19 and

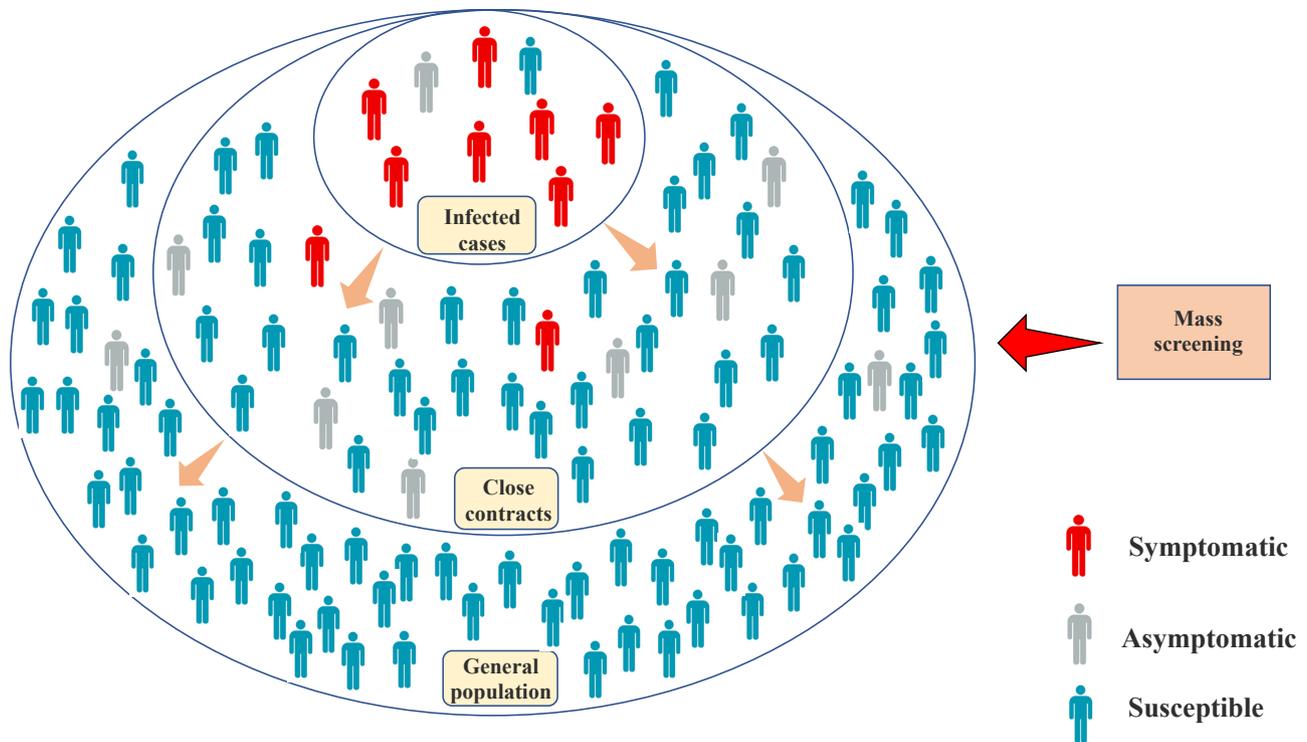


Figure 4: Diagram of controlling the transmission of COVID-19 with mass screening and contact tracing. The mass screening is an intervention strategy for COVID-19 control in the general population. We can identify the isolate asymptomatic cases with SARS-CoV-2, isolate the cases, and trace the close contacts.

rebuild the public's confidence in normal economic activities [100]. In contrast, the purpose of mass screening in other Chinese cities was to rule out the risk of a possible pandemic. Mass screening may provide a potential solution to curb the epidemic by detecting most infected individuals in a short time and may be particularly helpful in identifying asymptomatic cases to curb further transmission [101, 102]. This will be depended on the capacity of the healthcare system and the feasibility implementation of programme.

Mass screening is a fundamental aspect of COVID-19 control, and a core component of a combination approach that able to detect the case as early as possible and avoid secondary cases. Through mass screening, we can identify the infections with SARS-CoV-2, isolate the cases, and trace the close contacts (Figure 4). These measures can help us contain the transmission of COVID-19. Additionally, mass screening helps to understand the prevalence of infection and promote work resumption. A successful mass screening programme with comprehensive plan might restore normal life many months earlier than mass vaccination [103]. Mass screening will be critical when pursuing an exit strategy from strict lockdown measures that curb spread of the virus.

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