On January 30, 2020, the World Health Organization (WHO) declared a public health emergency of international concern due to the dizzying increase in the number of people affected by a new coronavirus called SARS CoV-2. On March 11, 2020, after its global spread, it was established as a pandemic. Subsequently, the emergence of new variants was reported at the international level, implying complex epidemiological scenarios for which it is necessary to maintain operational, active and robust genomic surveillance systems to monitor the evolution of the pandemic worldwide and of the epidemic in each country 1,2.

Global surveillance of SARS-CoV-2 genetic sequences supports the response to the COVID-19 pandemic. It also facilitates the monitoring of the geographical and temporal spread of SARS-CoV-2, the early detection and evaluation of mutations that may influence the pathogenic capacity, virus transmission or response measures taken (non-pharmacological measures, vaccines, treatment and diagnostic tests) 3,4.
In Ecuador, on February 29, 2020, INSPI as National Reference Laboratory in coordination with the National Liaison Center (CNE) of the MSP notified the first case of COVID-19 in the country and the second to officially announce in South America the introduction of the virus in the region, by which time it was the first laboratory in South America to implement the technique for detection thanks to the support provided by PAHO/WHO to the National Influenza Center in Ecuador. From that date on, the virus spread rapidly in Ecuadorian territory and the National Authorities implemented a series of sanitary, social, economic and security measures to control and mitigate the presence of the virus. Subsequently, studies of the complete genome of the virus were initiated in the country by academic institutions and public and private institutions that reported the presence of variants of concern and importance in public health. In this context, the country has made efforts in different areas to continue with the genomic surveillance process to better understand the evolution, transmission dynamics and behavior of the virus over time, information necessary to determine mitigation strategies for the spread of the virus by national authorities. Until May 24, 2021, according to WHO classification, the variants of concern (VOC) and variants of interest (VOI) are the following:

Note: It is worth mentioning that the variants described here are subject to changes and modifications according to the virological behavior and the designation made by the WHO expert committee.

Based on the presence of variants of concern and interest that are constantly evidenced, as well as the rate of evolution and adaptation of the virus, the following operational plan for the Systematic and Routine Genomic Surveillance of SARS-CoV-2, named by the authors of this publication, is proposed for the first time in Ecuador, considering the need to implement a national genomic surveillance system that performs and reports on a frequent basis the monitoring of genetic changes, mutations presented by the virus and is carried out as another microbiological technique within the portfolio of services provided by reference laboratories, public health laboratories or public or private institutions. This information will be vital for decision making by National Health Authorities, as well as to increase the number of sequences available for national authorities and the international scientific community through the publication of 90 to 100% of the sequences obtained in the GISAID platform. The present plan presents the following objectives:

To characterize the circulation patterns at the national level for the detection of SARS-CoV-2 variants of public health importance and to generate essential knowledge to complement epidemiological and virological surveillance.

To generate virological data to understand the patterns of evolution and dispersion of SARS-CoV-2 in the country, as well as the impact on the epidemiological behavior of COVID-19 and on the public health measures adopted to control the disease.

**VARIANTS OF CONCERN (VOC)**

- B.1.1.7
- B.1.351
- P.1
- B.1.617

**VARIANTS OF INTEREST (VOI)**

- B.1.525
- B.1.427/B.1.429
- B.1.1.28.2, alias P.2
- B.1.1.28.3, alias P.3
- B.1.526 con E484K o S477N
- B.1.616
Coordinate and integrate national data from various laboratories with sequencing capacity, providing a public health approach. To take advantage of the existing infrastructure of laboratories that perform sequencing in coordination with public health and academia, in order to expand the response capacity of the SARS CoV2 Genomic Surveillance System.

The methodology to be used is divided into phases: first, the Operational Plan for Routine Systematic Genomic Surveillance of SARS-CoV-2 will be socialized; institutions in the country with massive sequencing capacity for SARS-CoV-2 will be identified by sending contact information to INSPI; technical meetings, virtual or face-to-face working groups and constant reporting of variants detected through official channels.

The members of the SARS-CoV-2 National Routine and Systematic Genomic Surveillance Network are: health facilities without the capacity to perform real-time RT-PCR tests for the detection of SARS-CoV-2; health facilities with the capacity to perform real-time RT-PCR tests for the detection of SARS-CoV-2, but without sequencing capacity; laboratories that have the capacity to perform genomic sequencing (public, private or academic) from the different sectors involved and that have expressed their willingness to participate in the SARS-CoV-2 National Routine and Systematic Genomic Surveillance Network; laboratories that have the capacity to perform genomic sequencing; laboratories that have the capacity to perform real-time RT-PCR tests for the detection of SARS-CoV-2, but without sequencing capacity; laboratories with sequencing capacity, providing a public health approach. To take advantage of the existing infrastructure of laboratories that perform sequencing in coordination with public health and academia, in order to expand the response capacity of the SARS CoV2 Genomic Surveillance System.

Sample selection criteria are established based on convenience sampling strategies and random residual diagnostic sampling for selection of SARS CoV-2 samples considering: different age groups; health units with 50-100% increase in notification; other provinces or geographic locations within the country, related to an exponential growth of cases that has not been explained by the relaxation of measures; periods of time with irregular reporting of exponential increase of cases; non-specific severity: severe and fatal cases (consider hospital indicators); clusters of severe cases in persons < 60 years old and without underlying conditions; increased incidence of pediatric cases; cases in with suspected reinfection with SARS-CoV-2; cases in fully immunized persons; persons from countries where SARS-CoV-2 variants of concern (VOC) and of public health importance (VOI) are circulating; human-animal interface (sites where animal cases have been detected) or potential cases of zoonotic origin); unusual behavior of covid-19 diagnostic tests determined by laboratories that are part of the comprehensive public health network 5,6. For this reason, it should be considered to analyze the severity caused by the virus in the population by means of: percentage of hospitalization, percentage of admission to ICU, as well as, percentages of positivity, effective reproductive index, demography by provinces to complement the process of sample selection.

Regarding the type of sample and appropriate conditions, the sample of choice will be aopharyngeal swab, however, samples of respiratory origin may be taken ensuring a good quality of sampling (tracheal aspirate, bronchoalveolar lavage). In the case of deceased patients, lung or respiratory tract tissue samples may also be considered useful, provided that appropriate autopsy conditions, particularly respiratory protection, are in place. These samples should be placed in viral transport medium ensuring the cold chain or in DNA/RNA preservers that allow stabilization of their genetic material. The samples must maintain a constant cold chain for a certain period of time; if they cannot be sent during this period, it is recommended to freeze them until they are sent. For this type of study, it will be avoided to submit the sample to multiple freeze-thaw cycles, since it would cause the degradation of the viral RNA and would affect its quality. The sample should be accompanied by its respective epidemiological record with a request for determination of variants validated according to the criteria described above. It is recommended to obtain a number of 100 SARS-CoV-2 samples per month at the national level according to the laboratory capacities and the criteria mentioned above. The turnaround time for sequencing
results is approximately 315 working days. Viruses possess the biological characteristic of changing or evolving over time, which is considered an expected process. So far, hundreds of SARS CoV-2 variations have been identified worldwide. Since the initial genomic characterization of SARS-CoV-2, the virus has been divided into different genetic groups or clades. The occurrence of silent or non-silent mutations is an event that must be analyzed using appropriate bioinformatics tools and properly trained experienced personnel. In fact, some specific mutations define the viral genetic groups currently circulating worldwide and could impact their level of transmissibility, epidemiology and even lethality.

The purpose of the systematic and routine national genomic surveillance of SARS CoV-2 is to strengthen the strategies for the management of genomic data in the integrated health system, through a notification flow for public health purposes, following the protocols of the International Health Regulations, ethics committees and other legal regulations in force established for this purpose; and under no circumstances is it intended to have an impact on the clinical treatment or diagnosis of the patient.

It is worth mentioning the inter-institutional participation of various actors from public and private laboratories, academia, other institutions and organizations that have the capacity to perform massive sequencing, generating more information that will allow a better understanding of the evolution of the virus in the country; to date, academic institutions together with INSPI have coordinated efforts with the MSP and other public institutions contributing to the identification of existing SARS-CoV-2 variants and those that will continue to appear over time.

Another product of this genomic surveillance system is to take advantage of the existing infrastructure of the various public and private laboratories in order to integrate and analyze national genetic data on the circulation of SARS CoV-2 variants in a systematic and timely manner with a focus on information that allows the implementation of public health measures.

In this context, based on the importance of articulating efforts for the benefit of public health in the country, INSPI invited public and private institutions and academia with sequencing capacity that are carrying out variant identification through research projects, agreements or other national and international collaborations, to be part of the National Systematic and Routine Genomic Surveillance Network of SARS-CoV-2, in order to link the information generated in a single comprehensive genomic surveillance system. In order to better understand the dynamics of virus transmission, phylodynamics and its molecular evolution, becoming a very useful epidemiological tool in monitoring the infection during the health emergency in the Ecuadorian territory and in the global context. Consequently, it contributes with robust data of vital importance to strengthen the epidemiological surveillance of the virus, at the country level and to the necessary public health measures, which will allow the National Authorities to make timely decisions in the management of the COVID-19 pandemic. Ultimately, laboratories will be able to perform the sequencing of samples according to the criteria established in the operational plan for the National Routine and Systematic Genomic Surveillance of SARS-CoV-2 and this information will be reported as soon as possible to the National Health Authorities, as well as, to the GISAID platform so that the national and international scientific community can have access to the genetic behavior of the virus in the country, in the region and according to the procedures described in the International Health Regulations, considering that Ecuador is part of the PAHO/WHO COVID-19 Regional Genomic Surveillance Network.

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8. GISAID initiative. Available at: https://www.gisaid.org/