

Detection and characterisation capability and capacity for SARS-CoV-2 variants within the EU/EEA

16 February 2021

Key messages

- In most Member States, the sequencing capacity for identification of SARS-CoV-2 variants is below the recommendation set by the European Commission to sequence 5–10% of SARS-CoV-2 positive specimens.
- Although the majority of the EU/EEA countries are actively investigating the emergence of SARS-CoV-2 variants, three countries are not doing so.
- Many countries are increasing or planning to increase their sequencing capacity but have indicated the need for support from ECDC.
- Specific needs include support with sequencing capacities and protocols and with bioinformatics in particular.
- For many countries, the turn-around time for PCR pre-screening results shared with public health authorities is longer than 48 hours.

Background and methods

ECDC's rapid risk assessment on the spread of new SARS-CoV-2 variants of concern in the EU/EEA, published on 21 January 2021, concludes that the risk associated with the introduction and community spread of variants of concern has increased to high/very high. Member States are asked to increase their efforts to detect introductions of known variants and the emergence of new variants by increasing the level of surveillance and sequencing for a representative sample of community COVID-19 cases [1].

ECDC has produced guidelines on establishing sequencing capacities and capabilities, to help when taking decisions on which technologies to use and/or deciding on the role of sequencing for SARS-CoV-2 outbreak investigations and surveillance [2].

A European Commission Recommendation dated 19 January 2021 states that all EU Member States should reach a capacity of sequencing at least 5% - and preferably 10% - of positive test results [3]. To reach this target, there needs to be a significant increase in sequencing capacity in Member States.

ECDC has mapped the detection and characterisation capability and capacity for SARS-CoV-2 variants across the EU/EEA. This is the fifth laboratory capacity survey since the beginning of the COVID-19 pandemic in December 2019. The latest ECDC rapid assessment of laboratory practices and needs related to COVID-19 was published on 18 January 2021 [4].

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On 13 January 2021, a questionnaire (see Annex 1) was sent out to 30 EU/EEA Member States using the EU Survey Tool. The recipients included ECDC Operational Contact Points for COVID-19 for Microbiology, ECDC National Focal Points for Viral Respiratory Diseases, ECDC National Coordinators, EWRS Contact Point and Health Security Committee (HSC) Members.

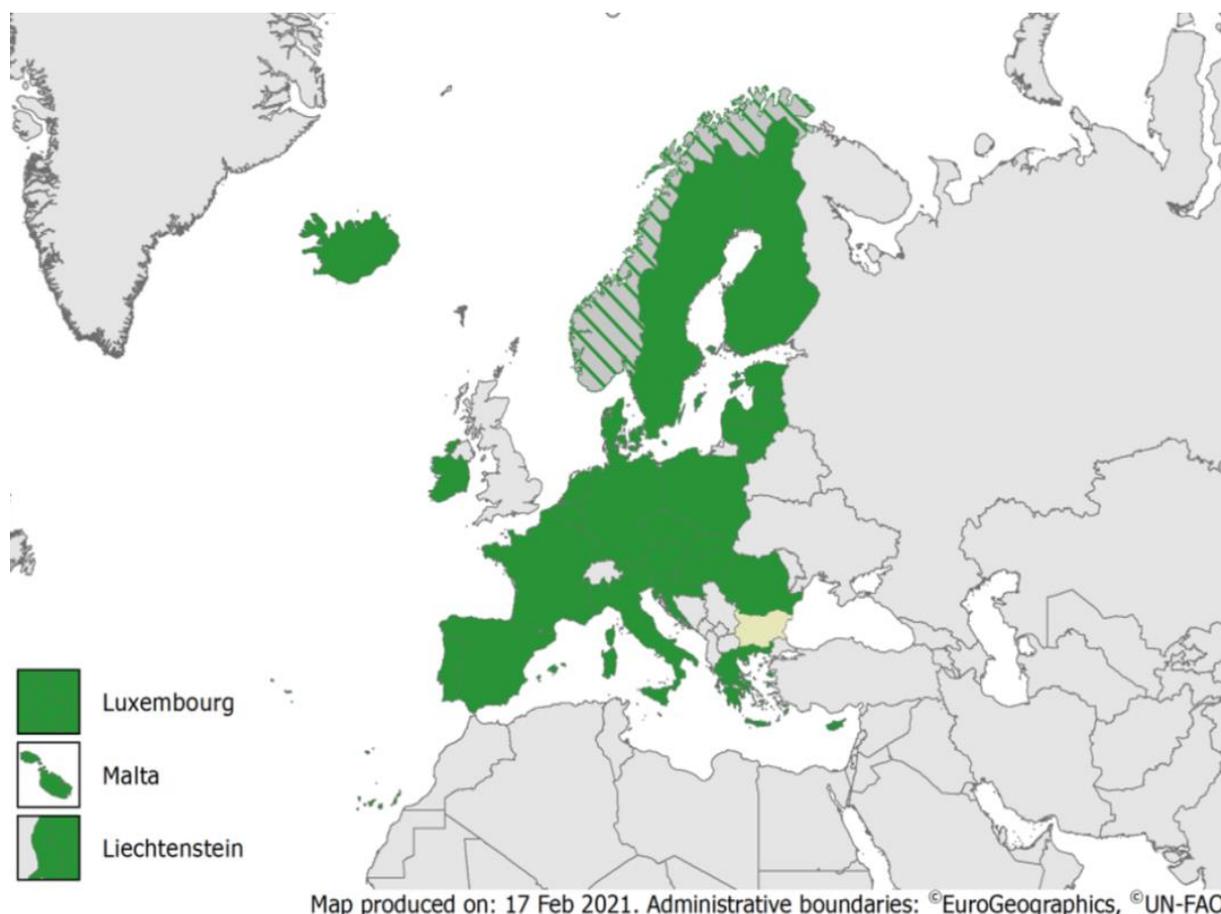
Due to technical issues, seventeen of the 29 responding countries did not have access to the entire set of questions. Those countries were contacted individually by email on 30 January 2021 and asked to send their responses to the missing questions by email.

Results

On 22 January 2021, 29 EU/EEA Member States had replied to the survey (Figure 1). Sixteen out of the seventeen countries that had not been able to submit answers to all questions, completed the survey by 4 February 2021. In total, twenty-eight of the twenty-nine responding countries submitted complete responses to the survey. For one country, responses to nine questions (Questions 11 to 19) were missing.

Among those responding from the 29 countries, 10 were ECDC National Focal Points (NFP) for Viral Respiratory Diseases, and 17 were ECDC Operational Focal Points (OCP) for COVID-19 Microbiology. Six responding institutions reported being Contact Points of the EU Early Warning and Response System (EWRS), one registered as a Member of the Health Security Committee (HSC) and two reported being ECDC National Coordinators. One institution also reported being the ECDC National Microbiology Focal Point and National COVID-19 Laboratory Network Coordinator. Other institutions also registered as NFPs for Threat Detection, NFPs for Viral Respiratory Diseases, OCPs for COVID-19 Epidemiology and NFPs for Surveillance.

Figure 1. Countries (n=29) that took part in the survey (green), EU/EEA, January 2021



Countries that were invited but did not provide responses are depicted in light green. Countries with incomplete responses are depicted with stripes. Countries outside the EU/EEA are shown in grey.

Investigating the emergence of new SARS-CoV-2 virus variants

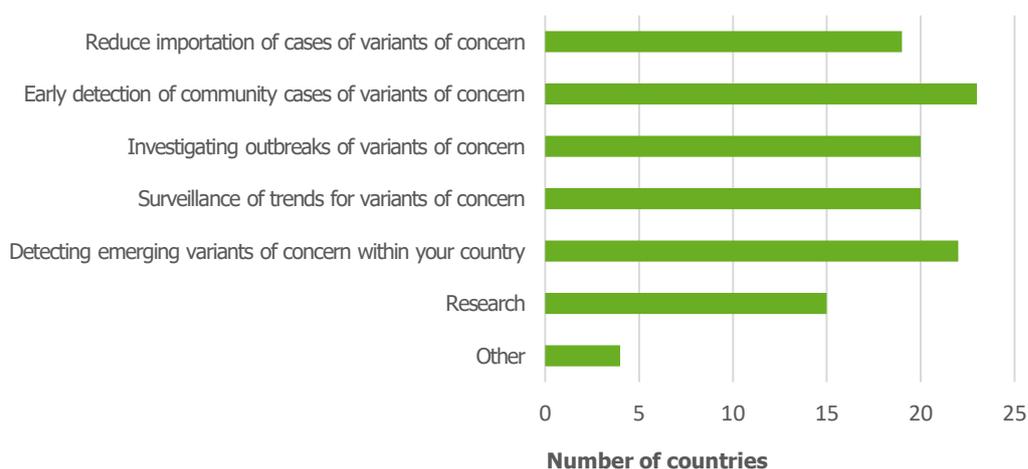
We asked the countries if they were actively investigating the emergence of new SARS-CoV-2 virus variants. Twenty-six out of 29 countries replied 'yes', and three countries replied 'no' to this question. One of these three countries commented that they were not actively investigating the emergence of SARS-CoV-2 virus variants, however they had investigated an unspecified number of SARS-CoV-2-positive specimens by sequence analysis and could show the presence of virus variants in the country. The selection of positive specimens was not further explained. One of the three countries added a comment stating that an unspecified number of specimens has been sequenced for research purposes.

Purpose of pre-screening for SARS-CoV-2 variant viruses

Pre-screening for known variant SARS-CoV-2 viruses is done by RT-PCR using a primer set that can indicate presence of the variant.

The 26 countries that actively investigate the emergence of SARS-CoV-2 virus variants could clarify the purpose of pre-screening by selecting predefined options. The answers are presented in Figure 2. Most countries (n=19), perform pre-screening to reduce importation of variants of concern (VOC) – i.e. to reduce the spread of imported VOC, for early detection of community cases of VOC (n=23) or to investigate outbreaks of VOC (n=20). In addition, monitoring trends of VOC and detecting emerging VOC were commonly reported as purposes (n=20). A total of 15 countries reported research questions as being the purpose for the pre-screening of virus variants.

Figure 2. Purpose of pre-screening for SARS-CoV-2 variant viruses among countries of the EU/EEA, January 2021



NB - Multiple choices could be made.

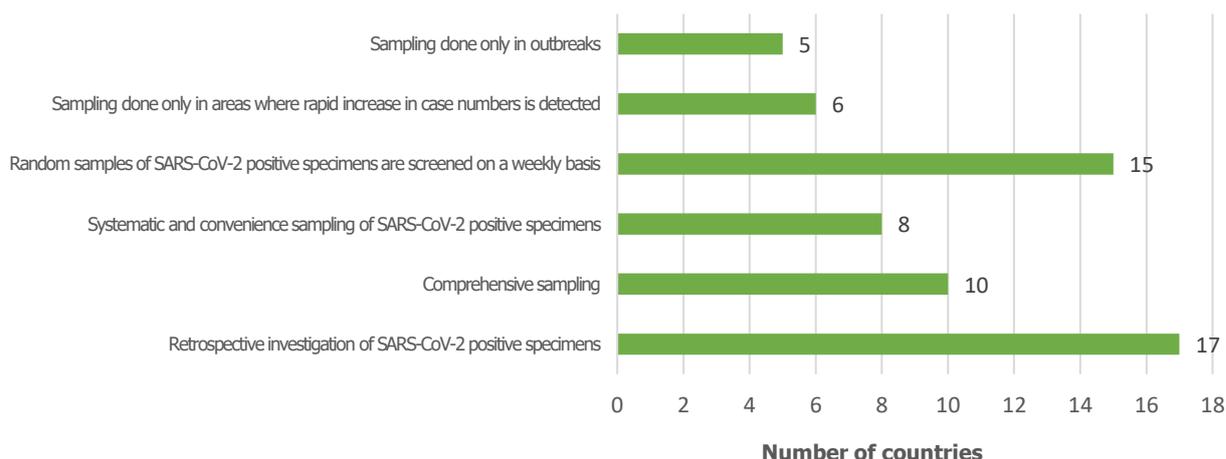
Sampling strategies for SARS-CoV-2 variant viruses

Countries were asked about sampling strategies for SARS-CoV-2 variants (Figure 3.)

Weekly screening of random positive SARS-CoV-2 specimens and the retrospective investigation of SARS-CoV-2-positive specimens were the most common approach. Ten countries are performing comprehensive testing – i.e. all SARS-CoV-2-positive specimens are pre-screened by PCR for variants and those found to be positive are subsequently sequenced. Eight countries reported systematic and convenience sampling of SARS-CoV-2-positive specimens. Five countries are only sampling in outbreaks and six are only sampling in areas where a rapid increase in case numbers is detected.

An additional sampling strategy which was not listed but mentioned by the countries was to sample target groups considered at a very high risk of being infected with a SARS-CoV-2 virus variant. Here, sequencing of selected samples was performed on specimens from i) people in geographical areas where a variant was widespread ii) outbreaks with high secondary attack rate iii) cases of reinfection iv) cases involving infection after vaccination v) cases with an unexpected adverse clinical evolution and vi) situations involving an unexpectedly high rate of hospitalisation/death. Furthermore, one country was sequencing selected samples to monitor laboratory diagnostic efficiency.

Figure 3. Sampling strategies for variant viruses in EU/EEA Member States, January 2021



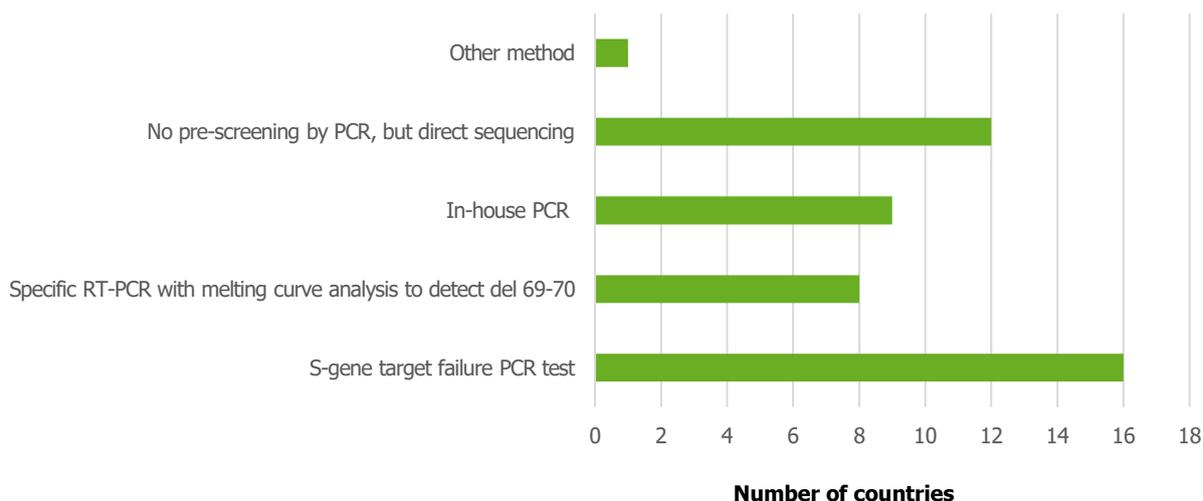
Multiple choices could be made.

Methods for pre-screening for SARS-CoV-2 variant viruses

Countries reported on the methods used for pre-screening SARS-CoV-2 virus variants (Figure 4). Sixteen countries used S-gene target failure RT-PCR tests, such as Thermo Fisher’s TaqPath [5] or the protocol developed by Yale University [6]. Twelve countries reported that they do not pre-screen using RT-PCR but sequence the samples directly. It is important to note that different laboratories in a country may not have the same strategy or capabilities. For example, direct sequencing of samples coming from individuals returning from abroad may still be possible even if pre-screening is not possible, while another laboratory in the same country would pre-screen any SARS-CoV-2 positive specimen to identify virus variants. In addition, some countries undertake direct sequencing of SARS-CoV-2-positive specimens from individuals with a travel history and individuals linked to verified cases infected with a virus variant without previous pre-screening of the specimens.

Specific RT-PCR, with melting curve analysis to identify the deletion of amino acid positions 69 and 70 were employed by eight countries and in-house RT-PCR tests by nine countries. An N501Y mutation-specific PCR, developed by the company TibMolBiol, is in use in one country, while three other countries are still evaluating this method.

Figure 4. Methods for pre-screening of SARS-CoV-2 variant viruses in EU/EEA Member States, January 2021



NB - Multiple choices could be made

Pre-screening capacity of SARS-CoV-2 variant viruses

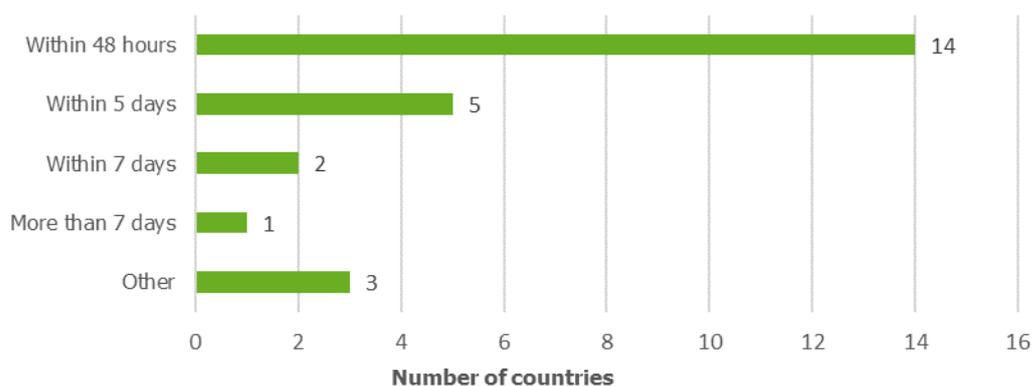
Among the 29 countries that responded, one explicitly stated that it does not pre-screen SARS-CoV-2 positive specimens to identify virus variants. The three countries which do not actively investigate the emergence of SARS-CoV-2 variants did not respond to this question. Twelve countries either did not provide an estimate of their pre-screening capacity or did not know the current capacity for pre-screening of SARS-CoV-2 positive specimens countrywide. Three countries reported a weekly capacity of more than 25 000 specimens. Six countries pre-screened up to 1 000 samples per week and seven countries reported having capacity to pre-screen 1 000 to 25 000 specimen per week.

Seven countries announced that their pre-screening capacity will increase in the coming weeks.

Turn-around time for pre-screening

Countries were asked about the turn-around time for PCR pre-screening results shared with public health authorities. (Figure 5). Fourteen countries reported a turn-around time of 48 hours or less. Five countries reported that results are shared within five days, two countries required seven days and one country reported that results were shared after more than seven days. The remaining seven countries, including three countries that indicated 'Other' in their response, did not state a specific turn-around time. This is because they either do not pre-screen SARS-CoV-2 positive specimens at all, do not do so regularly enough to be able to define a representative turn-around time (n=3) or did not provide an answer to this question (n=4).

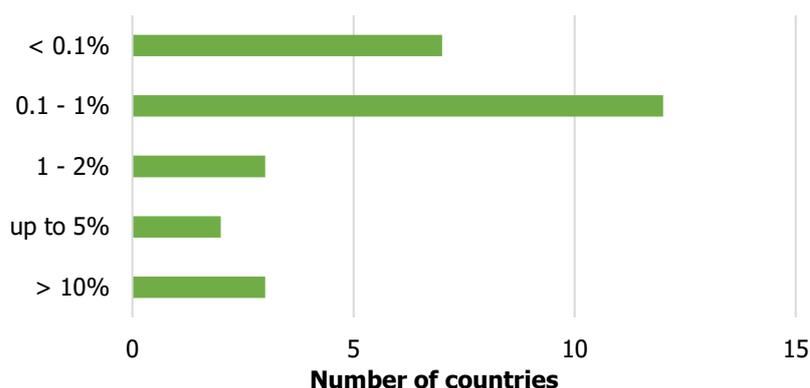
Figure 5. Turn-around time of PCR pre-screening results to be shared with public health authorities in EU/EEA countries, January 2021



Proportion of SARS-CoV-2 positive specimens characterised by sequencing

Countries were asked to report what proportion of SARS-CoV-2-positive specimens they characterised by sequencing (Figure 6). The majority, nineteen countries, sequenced less than 1% of positive specimens. Of these, seven countries did not even achieve a proportion of 0.1%, and 12 countries achieved between 0.1% and 1% of specimens sequenced. Three countries stated that they achieve a proportion of 1–2% and two countries achieve up to 5%. Three countries characterise more than 10% of positive specimens. Notably, one country reported that they sequence all SARS-CoV-2-positive specimens with an RT-PCR cycle threshold value below 32.

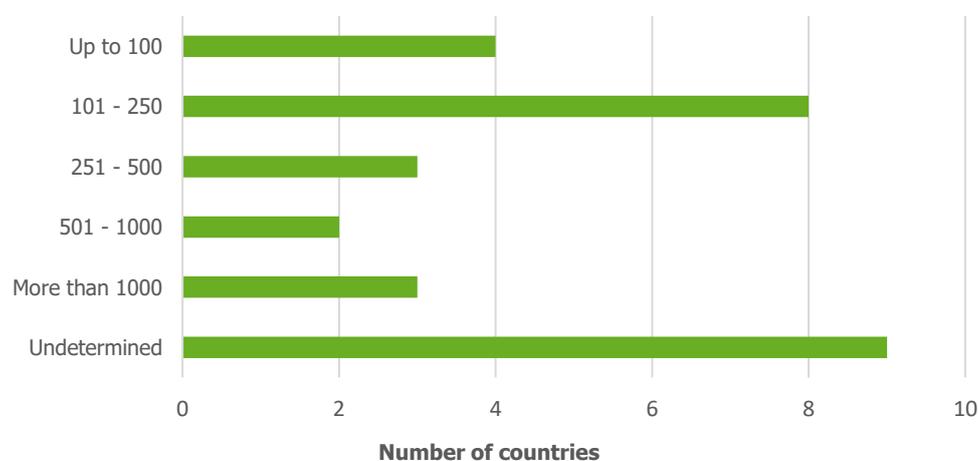
Figure 6. Current proportion of SARS-CoV-2 positive specimens characterised by sequencing in EU/EEA countries, January 2021



Sequencing capacity

The countries were asked to define their current sequencing capacity per week by the number of processed specimens (Figure 7). Four countries could sequence up to 100 specimens per week and eight countries reported a sequencing capacity of 101–250 specimens per week. Three countries could sequence between 251 and 500 specimens, while only two countries reported a capacity of 501 to 1 000 specimens per week. A sequencing capacity of more than 1 000 specimens per week was achieved by three countries. Nine countries did not submit any estimate on their weekly sequencing capacity. As of 22 January 2021, only five countries could meet the sequencing volume of a minimum of 500 specimens processed per week, as per ECDC's statement [1].

Figure 7. Number of countries in the EU/EEA with the capacity to sequence the indicated number of specimens per week, as of 22 January 2021



Antigenic characterisation of SARS-CoV-2 positive specimens

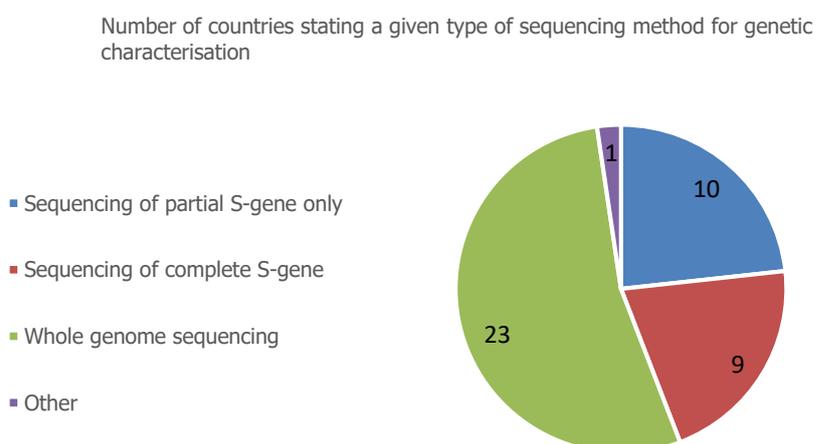
Countries were asked about the proportion of SARS-CoV-2-positive specimens selected for antigenic characterisation (i.e. virus neutralisation assays). Seven countries reported they performed antigenic characterisation of SARS-CoV-2 positive specimens. However, none of the countries performed this on a routine basis and countries were either in the process of implementing the method or had characterised only a few specimens. Seventeen countries stated that they did not perform antigenic characterisation, while four countries did not provide an answer to this question.

Sequencing methods for genetic characterisation

Countries were asked which sequencing approach they apply to SARS-CoV-2 positive specimens to identify variants. The results are shown in Figure 8.

Ten countries reported that they sequenced parts of the S-gene and nine countries reported that they sequenced the complete S-gene. Whole genome sequencing was used by twenty-three countries. One country was using metagenomic sequencing based on an Illumina sequencing protocol. It is important to note that multiple choices could be made in this question and several countries used two or more methods.

Figure 8. Methods used for antigenic characterisation of SARS-CoV-2 positive specimens among EU/EEA countries, January 2021



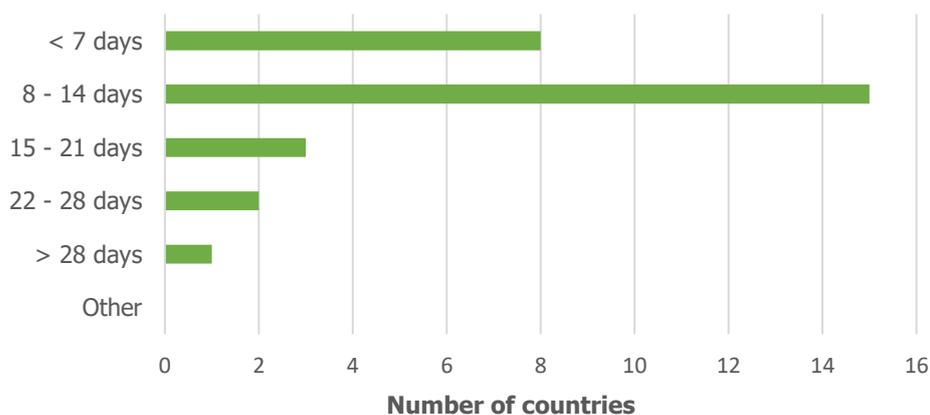
NB - Multiple choices could be made.

Turn-around time from sample collection to processed sequences

Those countries that responded reported the average turn-around times from sample collection to processed sequences being made available to national public health institutes (Figure 9). Most countries reported an average turn-around time with a maximum of two weeks. Eight countries reported their results within a week and fifteen countries within eight to 14 days. Three countries reported a turn-around time of 15 to 21 days and two countries reported times of 22 to 28 days or more (one of these two countries reported that the turn-around time was longer than 28 days).

It is important to note that some countries differentiated their answers further and reported different turn-around times for different methods. Results from Sanger sequencing may be available within seven days, while whole genome sequencing (WGS) usually takes longer.

Figure 9. Average turn-around times from sample collection to processed sequence results being made available to national public health institutes in EU/EEA countries, January 2021

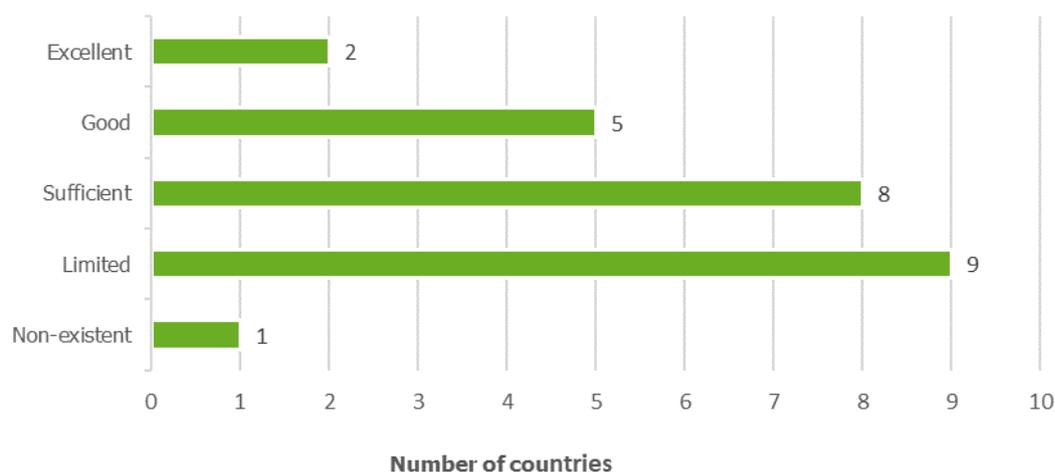


Capability to detect SARS-CoV-2 variant viruses

Countries were asked to assess their capability to detect variants in a timely manner in order to provide results for public health action (Figure 10.)

At the time of the survey, only two countries described their capability as excellent, five countries assessed their capability as good. Eight described their capability as sufficient and nine countries described their capability as limited. One country assessed its capacity to detect SARS-CoV-2 variant viruses as non-existent.

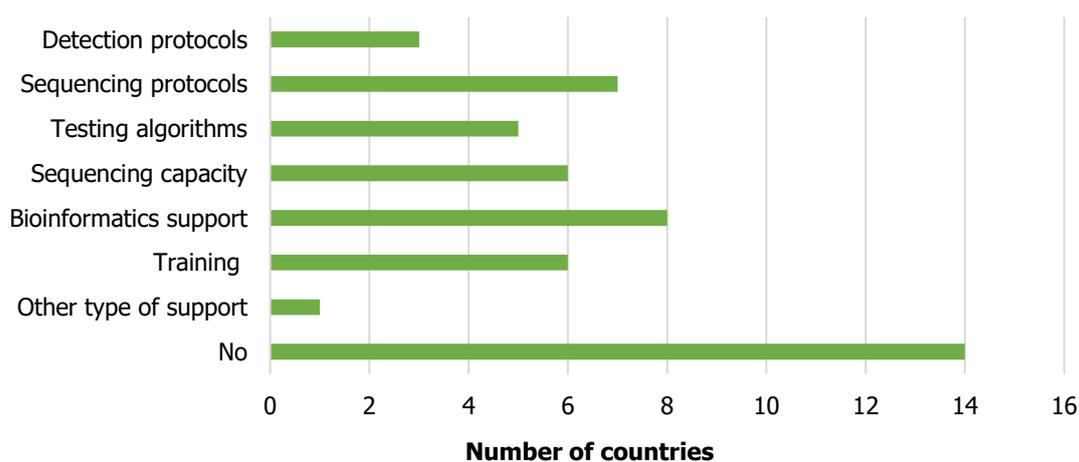
Figure 10. Assessment of detection capabilities to identify SARS-CoV-2 variants in countries of the EU/EEA, January 2021



Technical support for implementation of variant virus surveillance

Countries were asked whether they needed technical support for the implementation of variant virus surveillance. Fourteen countries responded that they did not require support. Fourteen countries reported a need for support as specified in Figure 11. Detection and sequencing protocols, testing algorithms, bioinformatic support, training and sequencing capacity were listed as areas where support is needed. One country specified the sharing of sampling strategy as an area requiring support and one country did not provide a response to this question.

Figure 11. Type of support needed for implementation of variant virus surveillance in EU/EEA countries, January 2021

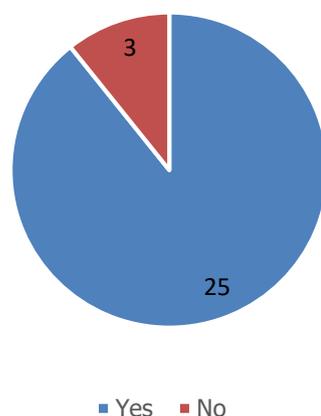


NB - Multiple choices could be made.

Reporting SARS-CoV-2 variant viruses in TESSy

Countries were asked if they plan to report variant virus results to the European Surveillance System (TESSy) (Figure 12). While most countries (25) are planning to report their results on variant viruses to TESSy, three are not planning to do so. It is important to note that not all countries plan to report on a weekly basis and one country specifically stated that they will not.

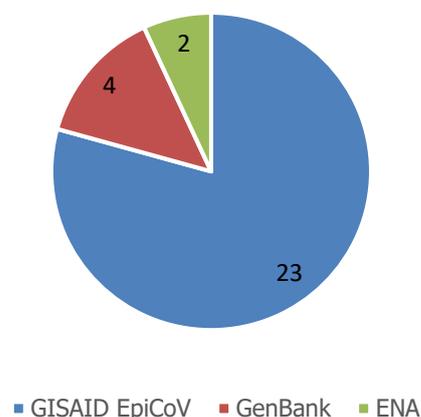
Figure 12. Number of EU/EEA countries planning to report variant virus results to TESSy, January 2021



Sequence data

When asked if countries were submitting sequence data to any international databases, all but one responded that they did. More specifically, twenty-three countries were reporting to GISAID EpiCoV, four countries were reporting to GenBank and two countries were reporting to European Nucleotide Archive (ENA) (see Figure 13). It should be noted that countries were able to make multiple choices in their answer to this question and four countries stated that they report to more than one database.

Figure 13. Countries of the EU/EEA reporting data to different platforms providing open access to genomic data collections, as of January 2021



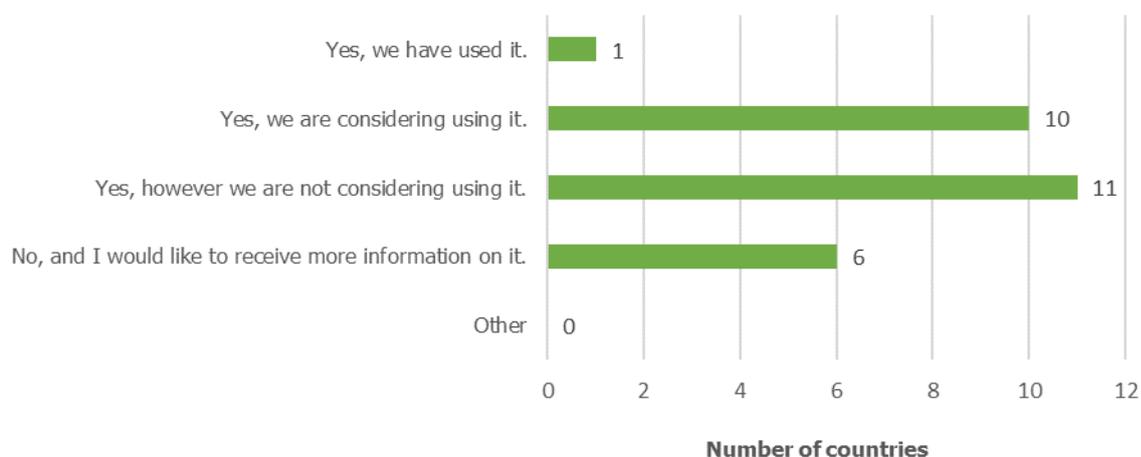
NB - Multiple choices could be made.

Sequencing support from ECDC

Countries were asked if they were aware of ECDC's offer of sequencing support. The results are presented in Figure 14. In total, twenty-two countries confirmed that they were aware of the offer, however, only one reported that they had made use of this offer. Ten countries were considering the offer, while eleven countries were not considering the offer of receiving support from ECDC. Six countries stated that they were not aware of the support ECDC can offer in terms of sequencing capacity, and that they wished to receive more information.

ECDC will be scheduling individual follow-up calls, in particular with the countries that had indicated a need for support.

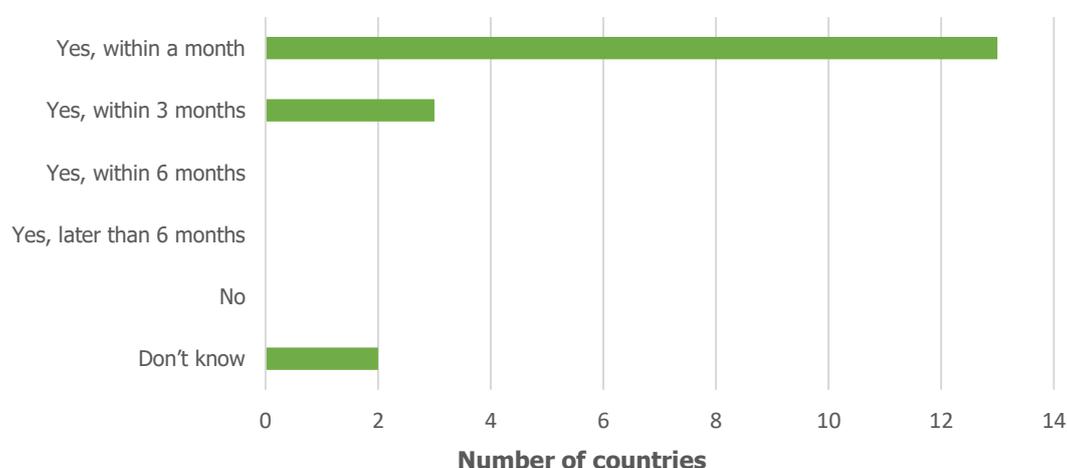
Figure 14. Awareness of ECDC's offer for sequencing support among EU/EEA countries in January 2021



Surveillance of SARS-CoV-2 variant viruses

When asked about their plans to do surveillance of variant viruses, ten countries stated that they had already started surveillance of variant viruses. Thirteen countries stated that they planned to commence surveillance within a month (Figure 15). Among those countries which will soon be starting variant surveillance activity is one of the three countries that were not actively investigating the emergence of SARS-CoV-2 variants by the time the survey was sent out. The other two countries did not report any plans to start surveillance of SARS-CoV-2 variants.

Figure 15. Countries within the EU/EEA are planning to start surveillance of variant viruses, as of January 2021



Countries were also asked to describe how they were going to implement surveillance of variant viruses. Nine countries reported that they are in the process of developing a surveillance strategy, without going into further detail. Three countries described their plan briefly as consisting of pre-screening followed by sequencing. One country referred to the National Genomic Programme which covers the surveillance of variant viruses, and two countries confirm that all SARS-CoV-2-positive specimens are being sequenced. Two countries stated that they are in the process of increasing their sequencing capacity. Five countries described their surveillance strategy by defining the sampling strategy, which included the selection of representative samples from across countries (n=5) and from outbreaks (n=4), sampling of returning travellers (n=2), and selecting of samples from individuals with unusual manifestation – e.g. young age, lack of comorbidities (n=3) and vaccinated individuals (n=3). One country is extending its sentinel surveillance and two countries are collecting the sequencing results at the national public health authorities where the sequences are being analysed.

Conclusions

This survey is the first to assess the current detection and characterisation capability and capacity for SARS-CoV-2 variants in the EU/EEA countries. All EU/EEA Member States were asked to complete the 20 questions to describe a situation which was representative of their entire country (see questionnaire in Annex 1).

As of 22 January 2021, the vast majority of the responding countries (26 of 29) stated that they were actively investigating the emergence of SARS-CoV-2 variants. However, three countries reported that they are not actively investigating the emergence of variants. All three of these countries did, however, indicate that they had detection capability for SARS-CoV-2 variants.

The European Commission set the recommended target for the proportion of positive SARS-CoV-2 specimens to be sequenced at 5–10% [3]. This survey indicates that, as of 22 January 2021, only three countries could meet this target and report a sequencing rate of over 10%. Two other countries achieved the lower limit of the recommended sequencing rate (i.e. 5%). None of the remaining countries achieved the recommended proportion of sequenced positive SARS-CoV-2 specimens.

In the rapid risk assessment published on 21 January 2021, ECDC stated that, as a minimum, 500 randomly selected samples per week should be sequenced by each country [1]. Based on the survey result, five countries have the necessary sequencing capacity as of 22 January 2021.

As it is of key importance to understand the bottlenecks and challenges faced when investigating variants, ECDC has invited countries to individual meetings to discuss this further. ECDC initiated these discussions on 7 February 2021, starting with the countries with no or limited capacity to detect VOCs.

To help countries with low or limited sequencing capacities to be able to detect and monitor capacity of VOCs, ECDC and the European Commission are planning support activities. As a first step, ECDC will offer sequencing services for Member States and these services will be tailored to meet the operational needs in countries. This includes offering additional services besides the actual sequencing (e.g. shipping services, cDNA synthesis and basic bioinformatics analysis). ECDC and the European Commission are also discussing how to support Member States in their capacity strengthening efforts in the mid- to long-term perspective.

In response to the VOCs, many countries are currently increasing both their sequencing capacity and capability. In recent months, some EU/EEA Member States have increased their sequencing capacity significantly – for example by reprioritising existing capacity within their country. However, such reprioritisation is only possible in countries where the technology has already been implemented. Besides sequencing capacity, many countries requested additional support with sequencing protocols and bioinformatics.

In order to facilitate the exchange of scientific knowledge and best practices, ECDC established a virology characterisation working group in January 2021 and is continuing with the regular ECOVID-LabNet information sharing. ECOVID-LabNet has been proven to be a valuable interaction platform and activities within this network will be continued and expanded. Together with the World Health Organization's Regional Office for Europe, ECDC has set up an EZCollab platform, a space where protocols and other documents can easily be shared within the network.

ECDC is in progress of launching new molecular External Quality Assessments (EQAs), including variant viruses and setting up support for virus neutralisation assay.

Limitations of the study

Due to a technical issue with the survey, not all those countries that responded could access the complete set of questions before the closing of the deadline for the online questionnaire. All these seventeen countries were contacted individually and all but one completed the survey belatedly. It is important to note that the responses from those sixteen countries were submitted up to eight days after the original deadline on 22 January 2021.

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Annex 1. Questionnaire

Question 1. Is your country actively investigating the emergence of new SARS-CoV-2 virus variants?

- Yes
- No
- Don't know

Question 2. What is the purpose of pre-screening for variants in your country? Multiple answers possible.

Definition of pre-screening for variant SARS-CoV-2 viruses: Pre-screening for variant SARS-CoV-2 viruses by using PCR to detect suspected variant virus cases in the population, e.g. screening for infection with viruses with a deletion in position 69-70 (del69-70) of spike protein.

- Reduce importation of cases of variants of concern
- Early detection of community cases of variants of concern
- Investigating outbreaks of variants of concern
- Surveillance of trends for variants of concern
- Detecting emerging variants of concern within your country
- Research
- Other.

Question 3. What is your country's sampling strategy for variant viruses? Multiple answers possible.

- Sampling done only in outbreaks
- Sampling done only in areas where rapid increase in case numbers is detected
- Random samples of SARS-CoV-2 positive specimens are screened for variants on a weekly basis
- Systematic and convenience sampling of SARS-CoV-2 positive specimens, e.g. first 10 specimens of the week are screened for variants
- Comprehensive sampling (all SARS-CoV-2 positive specimens are pre-screened by PCR for variants, e.g. using S-gene drop-out, and positive ones subsequently sequenced)
- Retrospective investigation of SARS-CoV-2 positive specimens.

Question 4. Please describe your sampling strategy in short or upload your file.

Question 5. What methods do you use for SARS-CoV-2 variant virus pre-screening? Multiple answers possible.

- S-gene target failure PCR test (e.g. Thermo Fisher TaqPath or Yale university)
- Specific RT-PCR with melting curve analysis to detect del 69-70 (TibMolBio – VirSNiP SARS-CoV-2 Spike N501Y)
- In-house PCR (please comment if you have a protocol available for sharing within the ECOVID-LabNet)
- No pre-screening by PCR, but direct sequencing (please comment if you have a protocol available for sharing within the ECOVID-LabNet)
- Other method, please describe below.

Question 6. What is your current pre-screening capacity for variant viruses (e.g. del 69-70 viruses), per week by number of processed specimens? Please comment also on any possible plans for changing your capacity.

Question 7. What is the turn-around time of PCR pre-screening results shared with public health authorities?

- Within 48 hours
- Within 5 days
- Within 7 days
- More than 7 days
- Other, please provide comments.

Question 8. What is the current proportion of SARS-CoV-2 positive specimens characterised by sequencing in your country? (Number of sequenced specimens / number of total positive specimens.)

- < 0.1%
- 0.1-0.5%
- 0.5-1%
- 1-2%
- 2-5%
- 5-10%
- > 10%.

Question 9. What is your current sequencing capacity per week by number of processed specimens?

Question 10. What proportion of your SARS-CoV-2 positive specimens are you characterising by antigenic characterisation i.e. neutralisation assay? (Number of antigenically characterised / number of positive specimens)

- None
- We characterise the following number of our positive specimens antigenically (please give the number of characterised / number of positive specimens) in the comment field.

Question 11. Which type of sequencing do you use for genetic characterisation? Multiple answers allowed.

- Sequencing of partial S-gene only
- Sequencing of complete S-gene
- Whole genome sequencing
- Other.

Question 12. What is the average turn-around time from sample collection to processed sequences being available for the national public health institute in your country?

- < 7 days
- 8 - 14 days
- 15 - 21 days
- 22 - 28 days
- > 28 days
- Other, comments.

Question 13. How would you describe your capability to detect variants in a timely manner to act upon the results for public health response?

- Non-existent
- Limited
- Sufficient
- Good
- Excellent.

Question 14. Do you need technical support for implementation of variant virus surveillance in your country and if yes, what type of support?

- Multiple answers allowed.
- Detection protocols
- Sequencing protocols
- Testing algorithms
- Sequencing capacity
- Bioinformatics support, e.g. phylogenetic analysis
- Training (please specify in comments)
- Other type of support (please describe in comments below).
- No.

Question 15. Does your country plan to report variant virus results to TESSy?

- Yes
- No.

Question 16. Does your country submit sequence data to any international database?

- Yes
- No.

Question 17. Where does your country submit sequence data? Multiple answers possible.

- GISAID EpiCoV
- GenBank
- ENA
- Other sequence database.

Question 18. Are you aware of ECDC's offer for sequencing support?

- Yes, we have used it.
- Yes, we are considering using it.
- Yes, however, we are not considering using it
- No, and I would like to receive more information on it.
- Other.

Question 19. Do you plan to start surveillance of variant viruses?

- Yes, within a month
- Yes, within three months
- Yes, within six months
- Yes, later than six months
- No
- Don't know.

Question 20. How do you plan to implement surveillance of variant viruses?