DOI: 10.1111/1348-0421.12883

NOTE

Multiple clades of SARS-CoV-2 were introduced to Thailand during the first quarter of 2020

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Revised: 6 April 2021

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Funding information

National Research Council of Thailand, Grant/Award Number: NRCT 72/2563; King Chulalongkorn Memorial Hospital's Excellence Centre Program, Grant/Award Number: EC-63-30101-29; Biological Threat Reduction Program of the US Defense Threat Reduction Agency, Grant/Award Number: HDTRA 1-17-C-0004

Abstract

In early January 2020, Thailand became the first country where a coronavirus disease 2019 (COVID-19) patient was identified outside China. In this study, 23 whole genomes of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) from patients who were hospitalized from January to March 2020 were analyzed, along with their travel histories. Six lineages were identified including A, A.6, B, B.1, B.1.8, and B.58, among which lineage A.6 was dominant. Seven patients were from China who traveled to Thailand in January and early February. Five of them were infected with the B lineage virus, and the other two cases were infected with different lineages including A and A.6. These findings present clear evidence of the early introduction of diverse SARS-CoV-2 clades in Thailand.

KEYWORDS

clade, diverse, early, SARS-CoV-2, Thailand

Abbreviations: COVID-19, Corona Virus Disease 2019; GISAID, Global Initiative on Sharing All Influenza Data; Pangolin, Phylogenetic Assignment of Named Global Outbreak LINeages; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; WGS, whole-genome sequence.

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with the travel histories
he patients in this study along
SARS-CoV-2 isolates from t
ade and lineage identification of
TABLE 1 Cla

SARS-CoV-2 GISAID ID	SARS-CoV-2 GenBank Accession No.	Collection date	Nationality	Travel history ^å	GISAID clade	Nextstrain_clade	Pangolin_lineage	М
EP1_ISL_437620	MT447158.1	1/25/2020	Chinese	Wuhan, China	S	19B	A.6	icro
EPI_ISL_437616	MT447162.1	2/7/2020	Chinese	Wuhan, China	S	19B	Α	obio
EPI_ISL_437615	MT447163.1	2/22/2020	Thai	No travel history outside Thailand	S	19B	A.6	logy
EPI_ISL_437613	MT447165.1	2/25/2020	Thai	Hokkaido-Sapporo, Japan and Bangkok, Thailand (4 days before diagnosis)	S	19B	A.6	y and I
EP1_ISL_437612	MT447166.1	2/29/2020	Thai	No travel history outside Thailand	S	19B	A.6	mm
EP1_ISL_437609	MT447169.1	3/12/2020	Thai	(Poipet) Banteay Meanchey, Cambodia	S	19B	A.6	uno
EP1_ISL_437607	MT447171.1	3/16/2020	Thai	No travel history outside Thailand	S	19B	A.6	log
EP1_ISL_437605	MT447173.1	3/18/2020	Thai	No travel history outside Thailand	S	19B	A.6	у
EP1_ISL_437604	MT447174.1	3/19/2020	Thai	No travel history outside Thailand	S	19B	A.6	
EP1_ISL_437603	MT447175.1	3/19/2020	Thai	No travel history outside Thailand	S	19B	A.6	
EP1_ISL_437602	MT447176.1	3/20/2020	Thai	No travel history outside Thailand	S	19B	A.6	
EPI_ISL_437614	MT447164.1	2/25/2020	Thai	No travel history outside Thailand	0	19A	В	
EP1_ISL_437608	MT447170.1	3/13/2020	Thai	No travel history outside Thailand	0	19A	В	
EP1_ISL_437610	MT447168.1	3/12/2020	Thai	Chonburi, Thailand	IJ	20A	B.1	
EP1_ISL_437606	MT447172.1	3/15/2020	Belgian	Belgium	IJ	20A	B.1.8	
EP1_ISL_437623	MT447155.1	1/8/2020	Chinese	Wuhan, China	Γ	19A	В	
EP1_ISL_437621	MT447157.1	1/25/2020	Chinese	Zhejiang-Wuhan, China	Γ	19A	В	
EPI_ISL_437618	MT447160.1	1/29/2020	Chinese	Wuhan, China	Γ	19A	В	
EP1_ISL_437622	MT447156.1	1/23/2020	Chinese	Wuhan, China	0	19A	В	
EPI_ISL_437619	MT447159.1	1/26/2020	Chinese	Wuhan, China	0	19A	В	
EP1_ISL_437624	MT447154.1	2/1/2020	Thai	liyama, Japan	0	19A	В	
EPI_ISL_437617	MT447161.1 ^b	2/3/2020	Thai	No travel history outside Thailand	0	19A	В	
EPI_ISL_437611	MT447167.1	3/11/2020	British	London, UK	Λ	19A	B.58	BU
Abbreviation: Pangolin, Phylogenetic Assi ^a Travel history within the past 3 months. ^b Fatal case of COVID-19.	Abbreviation: Pangolin, Phylogenetic Assignment of Named Global Outbreak LINeages. ^{*1} ravel history within the past 3 months. ⁵ Patal case of COVID-19.	al Outbreak LINeages.						JATHONG E

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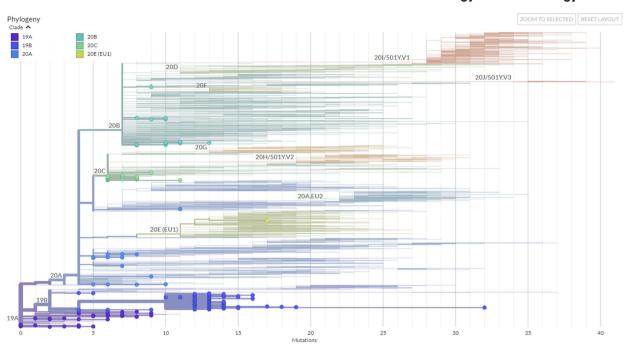


FIGURE 1 Sequencing data were uploaded to Nextclade (https://clades.nextstrain.org/, accessed on April 4, 2020) for phylogenetic analysis. Thai sequences (n = 243) from specimens sampled before March 31, 2020, were downloaded from the GISAID database on November 13, 2020, including the genomes from this study (n = 23). Colored circles show the sequencing data obtained in this analysis. Five Nextstrain clades were indicated; 19A (purple color), 19B (dark blue), 20A (blue), 20B (emerald green), and 20C (leaf green). The new GV GISAID clades (20E. EU1 Nextstrain clade) found in Thailand in October 2020 is in lime green circle

The coronavirus disease 2019 (COVID-19) pandemic has had devastating consequences globally, due in part to the greater infectivity of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as compared with other pathogens encountered over the past century. Thailand was the first country to report a confirmed COVID-19 case outside China, identified in a traveler who arrived at the Suvarnabhumi Airport in Bangkok from Wuhan, China on January 8, 2020.^{1,2} As of November 14, 2020, there were 3866 confirmed COVID-19 cases reported in Thailand with 60 deaths.³ Clade analysis from the Nextstrain database⁴ demonstrated that five clades of SARS-CoV-2 were found worldwide (as of November 13, 2020, from 184,320 full genomes), namely, clades 19A, 19B, 20A, 20B, and 20C, with more than 700 lineages characterized with Pangolin (Phylogenetic Assignment of Named Global Outbreak LINeages) software 5.0.

Whole-genome sequences (WGSs) of 23 COVID-19 cases found in Thailand between January and March 2020 were characterized using next-generation sequencing (MiSeq sequencer; Illumina, San Diego, CA, USA or PMseq; BGI Genomics, Hong Kong, China). All sequences were submitted to GenBank and Global Initiative on Sharing All Influenza Data (GISAID) databases. Accession numbers for both databases are presented in Table 1. The phylogenetic tree was constructed using Nextclade webapp (https://clades.nextstrain.org). All sequences were further identified for viral clade and lineage using the Nextstrain database⁴ and Pangolin software,⁵ respectively.

Six pangolin viral lineages were identified in our study including A, A.6, B, B.1, B.1.8, and B.58 (three Nextstrain viral clades including 19A, 19B, and 20A), as detailed in Table 1. Five Nextstrain viral clades were found in Thailand from the specimens collected between January and March 2020, as was publicized in GISAID on November 13, 2020^6 (n = 243; Figure 1 and Table S1). In another study from Thailand, five of eight GISAID viral clades including L, GH, GR, O, and S (T) were identified during the first wave of COVID-19 local outbreak in Thailand in March 2020.⁷

Travel histories of 23 patients were obtained from the outbreak investigation records from the Thai Ministry of Public Health (Table 1). All six patients in January 2020 traveled from China; of these, five were infected with the B lineage virus (including the first COVID-19 case in Thailand,^{1,2} MT447155.1), and one was infected with lineage A.6 (Table 1). Majority of the SARS-CoV-2 from the 9 Thai cases in this study belonged to A.6 lineage (9/23, 39.13%), of whom 7 had no history of travel outside of Thailand in the past 3 months. One had recently returned from Japan, while the other from Cambodia. A.6 lineage was most prevalent in Thailand during the first quarter of 2020 (125/243, 51.44%) (from GISAID database on November 13, 2020). By contrast, lineages B.1.8 and B.58 were identified from European travelers (Belgium and United Kingdom, respectively) who traveled to Thailand in March 2020 (Table 1).

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This study finds evidence of the early introduction of multiple SARS-CoV-2 viral clades into Thailand (Figure 1). All five current viral Nextstrain clades were identified among 243 viral sequences sampled between January and March 2020, reflecting considerable viral diversity since the early spread of the virus outside China. Three different viral lineages identified from seven Chinese travelers in January and early February 2020 revealed the evidence of early introduction of multiple SARS-CoV-2 viral lineages into Thailand (Table 1). The earliest lineage A virus, the root of the pandemic,⁵ was found in one patient from Wuhan who traveled to Thailand in early February 2020, while the viral lineage found in locally transmitted Thai cases was A.6, the one derived from lineage A (Figure 1 and Table 1).

The GISAID GV clade, the most recent clade identified on October 30, 2020, was first detected in the United Kingdom on March 22, 2020 (GISAID ID EPI ISL 420498, lineage B.1.153) but not found among the Thai entries in GISAID that were sampled between January and March 2020 (Figure 1). However, the first GV clade (lineage B.1.177) reported from Thailand was from a patient traveling from Belgium, whose specimen was collected on October 16, 2020. It may be that the GV clade of SARS-CoV-2 evolved in Europe in late March 2020, when Thailand went into a national lockdown with strict curfews beginning April 3, 2020. Thus, it is likely that Thai specimens during the early stages of the outbreak predated the evolution of GV. Alternatively, the number of WGS among the Thai entries in GISAID is inadequate (6.6%; 258 available WGS among 3866 confirmed cases to date as of November 13, 2020) as compared with Scotland where 49% of laboratory-confirmed cases before March 31, 2020, were sequenced for WGS.⁸ As there was increased inbound international air travel allowing specific groups to enter into Thailand in July 2020 according to the Emergency Decree, it is possible that new viral lineages may be evident among specimens collected subsequently in the state quarantine. Continued surveillance with WGS could help clarify the COVID-19 transmission dynamics worldwide and facilitate mutation identifications that will have an impact on the disease severity, transmissibility, development of diagnostic and therapeutic techniques, and certainly vaccine efficacy.^{9–11}

A more detailed study integrating clinical data, epidemiology including comprehensive travel history, and a larger set of complete viral genome isolates from the continued outbreak is needed to further inform the understanding of SARS-CoV-2 transmission.

ACKNOWLEDGMENTS

We gratefully acknowledge the authors of the originating and submitting laboratories of the genetic sequence and metadata shared through Global Initiative on Sharing All Influenza Data (GISAID), on which this study is based. We acknowledge Dr Paul Gaudio (Yale University) for his kind assistance in the critical editing of this manuscript. This study was supported by a research grant from the the National Research Council of Thailand (NRCT 72/2563), King Chulalongkorn Memorial Hospital's Excellence Centre Program (EC-63-30101-29), and the Biological Threat Reduction Program of the US Defense Threat Reduction Agency (Grant No. HDTRA 1-17-C-0004).

DISCLOSURE

The authors declare that there are no conflict of interests.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are openly available in [repository name] at https://www.gisaid.org [EPI_ISL437602-EPI_ISL437624] and https://www.ncbi. nlm.nih.gov/ [MT447154.1-MT447176.1].

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SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Buathong R, Chaifoo W, Iamsirithaworn S, et al. Multiple clades of SARS-CoV-2 were introduced to Thailand during the first quarter of 2020. Microbiol and Immunol. 2021;65: 405–409. https://doi.org/10.1111/1348-0421.12883