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The D614G substitution in the S gene and clinical information for patients with COVID-19 detected in Hong Kong

Gannon C.K. Mak⁎, Angela W.L. Lau, Andy M.Y. Chan, Desmond Y.W. Chan, Dominic N.C. Tsang

All from Centre for Health Protection, Department of Health, Hong Kong Special Administrative Region, China.

The SARS-COV-2 virus is a new type of coronavirus causing the pandemic in 2020, the spike (S) protein is a key target for vaccine development [1], virus entry [2,3] and infectivity [4–6]. Recent epidemiological and experimental studies showed that the D614G substitution in the SARS-COV-2 S protein might increase mortality and infectivity [7–11]. However, there is scanty data to correlate severity for COVID-19 patients having this substitution. We focused our study on SARS-COV-2 S gene in order to characterize viral mutations.

In an attempt to understand the relevance of D614G substitution among COVID-19 patients in Hong Kong, full length S gene sequences from severe and non-severe cases were examined. COVID-19 patients were confirmed by RT-PCR as described [12]. The severe cases were classified as described previously [13]. For this analysis, the original specimens of the respiratory samples from COVID-19 patients were sequenced using Sanger method. Only one specimen from each patient was included. The PCR amplification and DNA sequencing of the full length of S gene were performed using eight pairs of in-house designed primers (available on request). The SARS-COV-2 virus reference sequence, NC_045512 (GenBank accession number), was used in this analysis.

From 21 Jan 2020 to 12 Jun 2020, a total of 113 cases were sequenced. Among them, 11 and 102 were severe and non-severe cases respectively. Of 11 severe cases, 4 (36.4 %) showed D614G substitution while 39 (38.2 %) non-severe cases showed D614G substitution. There is no association of D614G with severe illness (p = 1.000, Fisher’s exact test, doubled one-sided). Of the 49 cases (6 severe cases and 43 non-severe cases) sequenced between January and February, none of them showed D614G. The first case of D614G appeared on March, however, D614G were found coincidentally in both severe and non-severe cases of the same month. The S gene sequences generated in this study have been deposited in the Global Initiative for Sharing All Influenza Data (GISAID) database. The corresponding patient information was also available in the Appendix A.

For the 113 cases sequenced, the S protein sequence of the 44 (38.9 %) were identical to the reference sequence. Reviewing the non-synonymous substitutions for the remaining 69 sequences, none of them were located within receptor binding domain between positions 400 – 600. Besides position 614, non-synonymous substitutions were found in other 13 positions. However, none of them were associated with severity (Appendix A).

SARS-COV-2 is an RNA virus which evolves rapidly. It is interesting to see that the dominance of 61 G virus is increasing over the 614D virus [14–16]. Although functional characteristics are unknown, numerous S gene mutations are reported regularly in GISAID [17]. It is thus important that laboratory surveillance continues to monitor the mutations of the S gene for SARS-COV-2 viruses. Concurrent genetic surveillance would facilitate early detection of sites that can increase mortality and infectivity as well as sites that are selected for the virus to escape immunological restraint especially when the vaccine is available.

Declaration of Competing Interest

None.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.jcv.2020.104550.

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