# Review of: "Sequence evidence that the D614G clade of SARS-CoV-2 was already circulating in northern Italy in the fall of 2019"

Thomas Caspari[1]

1 Paracelsus Medical University Salzburg

Potential competing interests: No potential competing interests to declare.

On balance the article is well written and easy to read. It makes the case that the D614G variant (lineage TTTG) of SARS-CoV-2 may have arisen earlier than the Wuhan strain (lineage CCCA) that lacks this virulenz-enhancing mutation. The main piece of evidence are a series of genomic fragments that were detected in Italy roughly 3-4 months before the Wuhan outbreak. The author should please take note of the Hubei Index Case that reported the Wuhan strain also alread 1-2 month prior to the seafood market outbreak (Pekar et al., Science. 2021, 372(6540):412-417). Taken together, the observatios published here and the Science report support both a model such that the Wuhan wildtype (CCCA) and the D614G strain (TTTG) circulated both prior to the Wuhan event in December 2019. The question of whether they originated from the same zoonosis or from two animal-to-human transitions remains open.

Review Comments

Abstract: (Please keep in mind to write for an educated lay audience)

The Aspartate-614 to Glycine (D614G) mutation is a hallmark of all recent variants of SARS-CoV-2 as it increases the likelihood that two or even all three subunits of the viral Spike engage with the human Ace2 protein. The D614G clade….. (now continue with the abstract)

Page 1: please delete ..or lab-leak as this is not yet supported by strong evidence

Page 2 (end of first paragraph) say here at so fat the earliest detection of the D614G strain was in January 2020 independently in China and Germany (Yurkovetskiy, L.; Wang, X.; Pascal, K.E.; Tomkins-Tinch, C.; Nyalile, T.P.; Wang, Y.; Baum, A.; Diehl, W.E.; Dauphin, A.; Carbone,C.; et al. Structural and functional analysis of the D614G SARS-CoV-2 spike protein variant. Cell 2020, 183, 739–751.)

Page 2: please say from which sections of the SARS-CoV-2 genome the sections listed in Table 1 are (ie which gene or region)

Page 2: the relevance of the Brazilian sample from sewage needs further explanation and when were they collected?

Page 3: Around Feb 18th 2020 ?

Page 4: introduce lineage B and A to the reader in more detail and relate the CCCA and TTTG to these lineages

Page 4: I would delete the lab-leak section as it too speculative and ill supported

Page 4: say that the conclusion that the CCCA lineage might not be the source of the TTTG lineage is that your data might indicate the appearance of the TTTG in November 2019 prior to the Wuhan outbreak in December 2019. Please note the Hubei Index Case (Pekar J, Worobey M, Moshiri N, Scheffler K, Wertheim JO. Timing the SARS-CoV-2 index case in Hubei province. Science. 2021 Apr 23;372(6540):412-417)