Analysis of Delta (Indian) Variant of SARS-CoV-2 Infectivity using Resonant Recognition Model

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Running Title: SARS-CoV-2 Delta Variant

Abstract: This manuscript is continuation of our previous work, where we have analyzed different variants of SARS-CoV-2 virus (UK, South African, Brazilian, and Indian (Kappa)) using Resonant Recognition Model (RRM), which is biophysical model capable to analyze protein function and interaction. We have previously identified correlation between infectivity of these SARS-CoV-2 virus variants with strength of signal at RRM characteristic frequencies for each variant. Here, we have extended this analysis for Delta (Indian) SARS-CoV-2 virus variant, which is extremely infectious and is rapidly spreading around the World. Our results with Delta (Indian) variant are in complete agreement with our previous RRM proposition that viral infectivity is proportional to strength of signal at RRM characteristic frequency. These results can explain why Delta (Indian) variant is more infectious. With strong correlation obtained in all these examples, we can propose here that RRM model can be used as general tool to analyze infectivity of mutated virus variants.

Keywords: Delta Variant of SARS-CoV-2, Prediction of Infectivity, Resonant Recognition Model, COVID-19

Introduction

With COVID-19 pandemic still raging all around the World, which is fueled with more infectious new mutant variants of SARS-CoV-2 virus, like Delta (Indian) variant, it is important to have a tool to analyze and predict infectivity of any new variant even before it has spread across the community. We have already applied the Resonant Recognition Model (RRM), which is biophysical model capable to analyze protein function and interaction [1-3], to analyze UK, South African, Brazilian, and Indian (Kappa) variants of SARS-CoV-2 [4-5]. The RRM model has identified parameters (frequencies) that characterize SARS-CoV-2 spike protein, as well as its S1 fragment critical for viral interaction with host cell receptor [3-4]. Comparing the strength of signal using S/N – signal to noise ratio, at these characteristic RRM frequencies, we have identified that higher S/N at these frequencies relates to more infectious variants of SARS-CoV-2 [4-5]. Here, we have extended this analysis to new extremely infectious and worrisome Delta (Indian) variant, lineage B.1.617.2. This Delta variant is rapidly spreading around the World and is characterized by the changes within the spike protein and attributes [6], as presented in Figure 1.

Figure 1. Changes and attributes within the spike protein of Delta (Indian) variant.

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Here, we have analyzed infectivity of Delta (Indian) variant of SARS-CoV-2, using the RRM model in the same way and as continuation of our previous work presented in Reference 4.

Methods
Resonant Recognition Model
The Resonant Recognition Model (RRM) is theoretical, biophysical model that can analyze interaction and recognition between proteins and their targets, which could be other proteins, DNA, RNA, or small molecules, has been published previously in details [1-2,7-11]. The RRM model is based on the findings that certain periodicities (frequencies) within the distribution of energy of delocalized electrons along protein are critical for protein biological function and/or interaction with their targets. The distribution of these energies is calculated by assigning each amino acid physical parameter representing the energy of delocalized electrons of each amino acid. Next step was to calculate spectral characteristics of such energy distribution (signal) using Fourier Transform, where the linear numerical signal is transformed into the frequency domain and is characterized by number of different frequencies making up the original signal. Comparing such spectra using cross-spectral function for proteins sharing the same biological function/interaction, it has been shown that they share the same frequency within the spectrum of energy distribution along the protein [1-2,7-10]. Peak frequencies in such multiple cross-spectral function present common frequency components for all sequences analyzed. The comprehensive analysis done so far confirms that all protein sequences with the common biological function and/or interaction have common frequency component, which is specific feature for the observed function/interaction [1-2,9-11]. Thus, each specific biological function/interaction within protein is characterized by its specific frequency. The strength of the signal either in spectrum or cross-spectrum, can be measured by signal-to-noise ratio (S/N). The S/N is calculated as the ratio between the signal intensity at the specific frequency and the mean value over the whole spectrum. Higher S/N value means stronger signal at the specific frequency and possibly the stronger biological function/interaction related to this frequency.

It is extremely important to understand that the RRM model is particularly efficient when it is applied to viruses. In general, viruses are mutating very quickly, making extremely hard to make vaccine or cure with current approaches based on homology. However, even when viruses are mutating so often and so quickly, they are still keeping their specific functionality. The RRM is not looking at all into homology of mutated viruses, but it is looking for characteristic parameter(s) identifying virus protein's biological function/interaction [1-2]. Thus, the RRM analysis can identify the common characteristic frequency of viral activity, which does not depend on virus mutations as long as they keep their functionality. Based on this common characteristic frequency, the RRM can analyze the strength of viral activity by calculating S/N at this characteristic frequency for different mutants. This RRM approach has been experimentally tested on the example of HIV virus, which is highly mutated, but all isolates keep the same RRM characteristic frequency [12-14].

Based on our previous work [4-5], where we have found strong correlation between different SARS-CoV-2 variant's infectivity and S/N ratio for spike proteins and their S1 fragments at previously identified RRM characteristic frequencies, we have analyzed here Delta (Indian) variant of SARS-CoV-2 virus, which has been found to be extremely more infectious than previously analyzed variants, with the aim to explore if this extreme infectibility can also be identified using the RRM model.

Results
Previous RRM Results with SARS-CoV-2 Spike Proteins
The spike proteins are on the surface of coronaviruses and are the first to approach and recognize receptors on host cells. We have previously analyzed spike proteins, using the RRM model, from different coronaviruses with the aim to find out if there is any common component that can characterize spike's recognition and interaction with host cells [4,11]. The result of this analysis is the most prominent common RRM frequency at $f1=0.2827\pm0.0009$, as presented in Figure 2 [4,11].
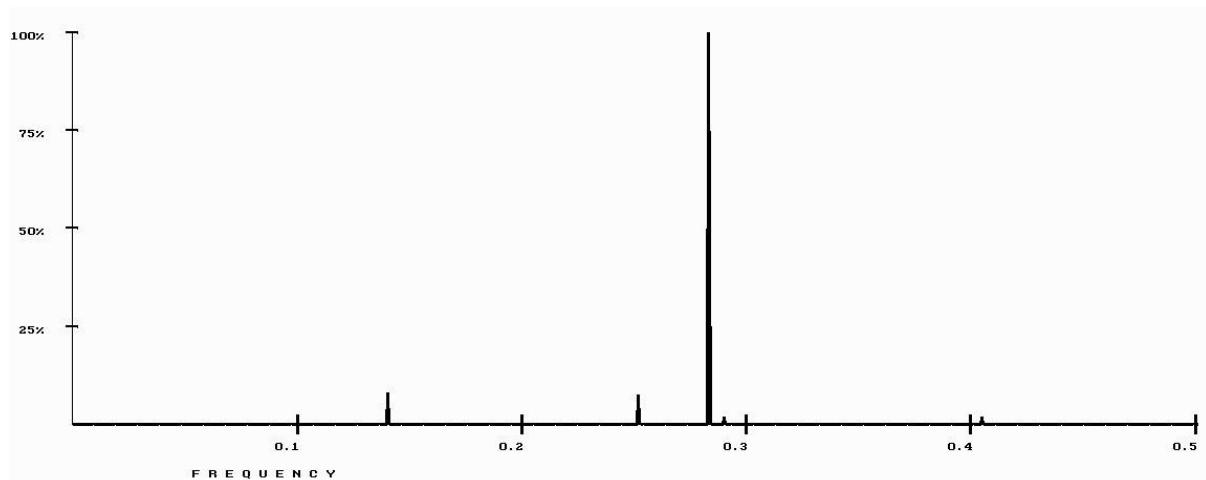
Figure 2. RRM cross-spectrum of spike proteins with common characteristic frequency at f1=0.2827±0.0009 [4,11].

The one unique common RRM characteristic frequency f1 for all analyzed spike proteins from different coronaviruses points out that all coronaviruses have one and the same RRM characteristic, crucial for viral recognition and interaction with host cells.

The SARS-CoV-2 viral infection is through interaction between spike S1 fragments and receptors on the surface of host cells. These host cells receptors for some coronaviruses, like HCoV-NL63, SARS-CoV (the virus that causes SARS) [15] and SARS-CoV-2 (the virus that causes COVID-19) [16] is angiotensin-converting enzyme 2 (ACE2). The ACE2 receptor has been found to be on the surface of various cell membranes, including cells in the lungs, heart, arteries, kidney, and intestines. The binding of the S1 spike protein fragments of SARS-CoV and SARS-CoV-2 viruses to the ACE2 receptor on the surface of cells leads to endocytosis and translocation of the virus into endosomes within the cells [17].

We have previously analyzed S1 fragments from coronaviruses that affect humans, using the RRM model, and the most prominent common RRM frequency has been found at f2=0.3145±0.0019, as presented in Figure 3 [3-4,11].
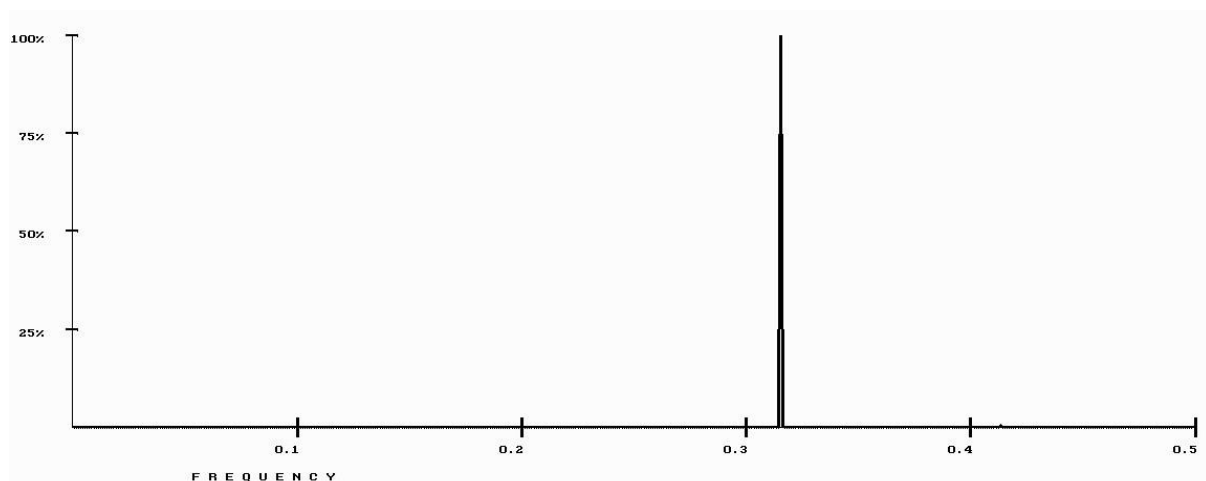

Figure 3. RRM cross-spectrum of spike S1 fragments from coronaviruses affecting humans with common characteristic frequency at f2=0.3145±0.0019 [3].

To analyze interaction between S1 spike fragments and ACE2 receptors, we have compared these two groups of proteins, using the RRM cross-spectral function and the most prominent common RRM frequency was also found to be at f2=0.3145±0.0019, same as presented in Figure 3 [3]. Thus, according to RRM principles, the frequency f2 is characterizing the interaction between S1 spike fragments and ACE2 receptors.

RRM Analysis of Different SARS-CoV-2 Variants
Once when we have identified the characteristic RRM frequency f1 for spike proteins, as well as the characteristic RRM frequency f2 for interaction of S1 fragments of spike proteins with ACE2 receptors, we are able to identify how much

certain mutations can affect strength of the signal within mutated proteins at these specific RRM frequencies. We have calculated previously [4-5] the strength of the signal, as signal-to-noise ratio (S/N), at RRM characteristic frequencies f1 and f2 within the original strain of SARS-CoV-2 virus (P0DTC2 from UniProt database). For this strain S/N for frequency f1 within the whole spike protein was found to be at 0.90 and S/N for frequency f2 within the S1 fragment of spike protein was found to be at 1.71, as presented in Table 1 and Figure 4.

For all other variants (UK, South African, Brazilian, Indian-Kappa, Indian-Delta), we have applied relevant mutations as listed in Table 1. Then we have calculated S/N ratio at frequencies f1 and f2 for each of mutated variants separately and results are presented in Table 1 and Figure 4.

SARS-CoV-2 Variant	S/N at f1=0.2827	S/N at f2=0.3145	Mutations
Brazilian variant	0.81		K417T, E484K, N501Y
Original variant P0DTC2	0.90		-
South African variant	0.94		N501Y, K417N, E484K
Indian variant (Kappa)	1.13		L452R, E484Q
UK variant	1.36		N501Y, 69-70del, P681H, D614G, N439K, Y453F
Delta (Indian) variant	0.64		T19R, 156-157del, R158G, L452R, T478K, D614G, P681R, D950N
Brazilian variant S1		1.42	K417T, E484K, N501Y
Original S1 fragment		1.71	-
South African variant S1		1.80	N501Y, K417N, E484K
Indian variant S1 (Kappa)		1.84	L452R, E484Q
UK variant S1		2.65	N501Y, 69-70del, P681H, D614G, N439K, Y453F
Delta (Indian) variant S1		**4.08**	**T19R, 156-157del, R158G, L452R, T478K, D614G, P681R**

Table 1. S/N ratio at RRM characteristic frequency f1 for whole spike protein and frequency f2 for S1 spike fragment calculated for following SARS-CoV-2 variants: original variant (P0DTC2), Brazilian variant, South African variant, Indian (Kappa) variant, UK variant and Delta (Indian) variant highlighted in red, all presented with related mutations in regard to original variant.
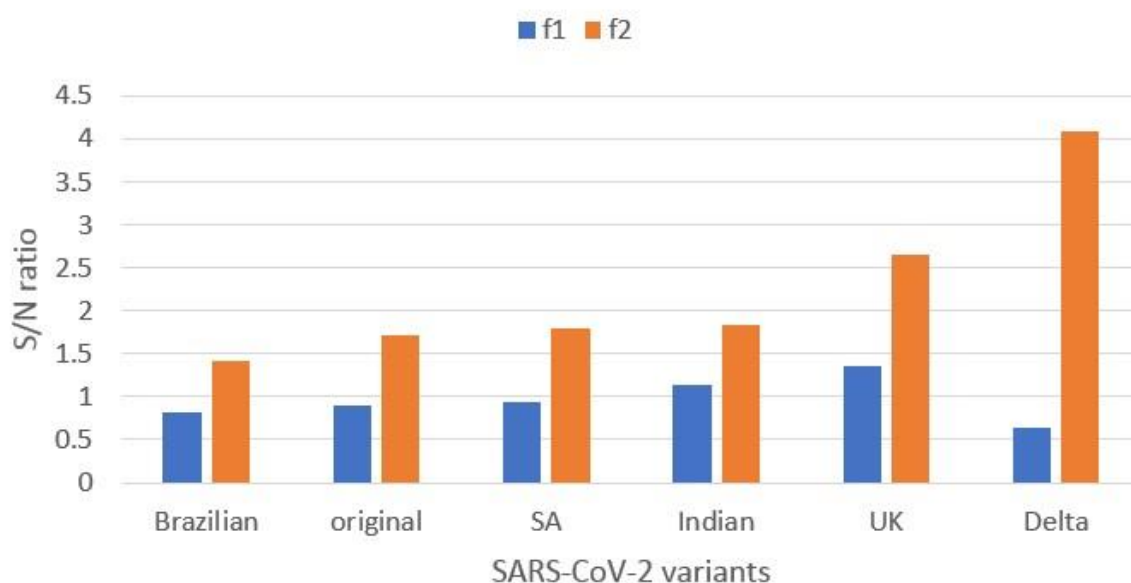

Figure 4. S/N ratio at RRM characteristic frequency f1 in blue for whole spike protein and frequency f2 in orange for S1 spike fragment for: original variant (P0DTC2), Brazilian variant, South African variant, Indian (Kappa) variant, UK variant and Delta (Indian) variant.

It can be observed from Table 1 and Figure 4, that South African, Indian (Kappa) and UK variants have increased S/N ratio in comparison to original virus at both characteristic frequencies f1 and f2, while Brazilian variant has decreased S/N ratio in comparison to original virus at both characteristic frequencies f1 and f2, as previously presented [4-5]. In this previous work, we have found that there is significant correlation between S/N ratio at both frequencies f1 and f2, with infectivity of these variants [4-5]. Such correlation is expected as higher S/N ratio at characteristic frequencies critical for SARS-CoV-2 spike activity, according to RRM model, show that mutated variants with higher S/N ratio are more active and consequently more infectious. When we have analyzed even more infectious Delta (Indian) variant, we have found extremely higher S/N ratio than at any other variants of concern at RRM frequency f2, as presented in Table 1 and Figure 4. However, S/N ratio for Delta (Indian) variant is much lower than at any other variants of concern at RRM frequency f1, as presented in Table 1 and Figure 4. Having in mind that Delta (Indian) variant is extremely infectious, more than other variants of concern, and that there is much higher S/N ratio at frequency f2, but lower at frequency f1, we can propose here that frequency f2 is critical for infectivity of mutated SARS-CoV-2 virus variants. Such result is expected as frequency f2 is related to recognition/interaction between S1 fragments of spike proteins and ACE2 receptors. Our results are in complete agreement with RRM proposition that SARS-CoV-2 viral activity and infectivity is proportional to S/N ratio especially at frequency f2 and thus can explain why Delta (Indian) variant has much higher prevalence for interaction with ACE2 receptor and consequently is more infectious.

3D Presentation of Delta (Indian) Variant Mutations

It is interesting to point out that although majority of mutations of Delta (Indian) variant are within S1 fragment of SARS-CoV-2 spike protein but are not within the 3D structure of SARS-CoV-2 spike receptor binding domain bound with ACE2 (PDBe>6m0j [18]). Only mutations: L452R and T478K are within the 3D structure of SARS-CoV-2 spike receptor binding domain bound with ACE2 and are presented in Figure 5 with CPKs in red.
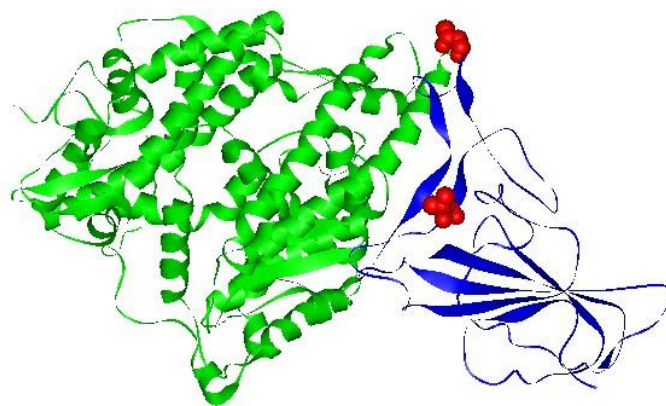

Figure 5. 3D structure of SARS-CoV-2 spike receptor binding domain bound with ACE2 (PDBe>6m0j [18]) with Delta (Indian) variant mutations highlighted with CPKs in red.

The fact that most of mutations characterizing Delta (Indian) variant of SARS-CoV-2, which was identified as the most infectious variant so far, are not within 3D spike receptor binding domain is highlighting the idea that not only the amino acids within binding domain are important for binding properties, but also amino acids which are structurally further from binding domain are important as well. This is the advantage of RRM model, as it does not look into structural characteristics of binding domain, but it looks into the whole protein's biophysical parameters that are important for its binding activity.

Discussion and Conclusion

With recently appearing new mutants of SARS-CoV-2 virus, where some of these variants are more infectious, there is a need to establish theoretical approach that can predict infectivity of mutated viruses, just from known mutations, sometimes even before these mutations appear in nature. We have already employed the RRM model, which is biophysical theoretical model, capable to analyse protein's interactions/functions to examples of SARS-CoV-2 variants: Brazilian, South African, Indian (Kappa) and UK [4-5]. By identifying characteristic parameters (frequencies) for spike activity and its interaction with ACE2 receptor, we found that amplitude's S/N at these frequencies are directly correlated with infectivity of these previously analysed variants of SARS-CoV-2 virus [4-5], as presented in Table 1 and Figure 4. We have extended here this approach to new, even more infectious Delta (Indian) variant of SARS-CoV-2 virus. However, with this analysis we have found that only S/N ratio for Delta (Indian) variant at frequency f2=0.3145±0.0019, responsible for interaction between S1 spike fragment and ACE2 receptor, is much higher than for other variants, while S/N ratio at frequency f1=0.2827±0.0009 for Delta (Indian) variant is much lower than for other variants, as presented in Table 1 and Figure 4.

Thus, we can propose here that frequency f2 is critical for infectivity of mutated SARS-CoV-2 virus variants. Such result is expected as frequency f2 is related to recognition/interaction between S1 fragments of spike proteins and ACE2 receptors. Our results are in complete agreement with RRM proposition that SARS-CoV-2 viral activity and infectivity is proportional to S/N ratio especially at frequency f2 and thus can explain why Delta (Indian) variant has much higher prevalence for interaction with ACE2 receptor and thus is more infectious. These results are establishing the RRM model as theoretical model capable of analysing and predicting strength of mutated viruses, just from known mutations.

Although majority of mutations of Delta (Indian) variant are within S1 fragment of SARS-CoV-2 spike protein, they are not within the 3D structure of SARS-CoV-2 spike receptor binding domain bound with ACE2 (PDBe>6m0j [18]). This indicates that not only the amino acids within binding domain are important for binding properties between protein and its receptor, but also amino acids which are structurally further from binding domain are important as well. This is the advantage of RRM model, as it does not look into structural characteristics of binding domain, but it looks into the whole protein's biophysical parameters that are important for its binding activity.

Baring all above in mind, we can propose here that RRM model, can be used as general tool for analysis of infectivity and strength of mutated virus variants, both existing and possible new once.

Contributions
Conceptualization, I.C.; Methodology, I.C. and D.C.; Software, D.C.; Resources I.L.; Writing—Original Draft Preparation—Review and Editing, I.C., D.C. and I.L.

Competing Interests
Authors declare they have no competing interests.

Human/Animal Involvement
Authors declare that there were no human participants nor any animal involvement in this study.

Funding

Acknowledgement

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