Vol. 03, No. (2) 2021, Pg. 35-42

ENVIRO Dental Journal

Enviro Dental Journal

dentaljournal.org

Emerging Coronavirus Mutant – The Delta Plus: A Review

P RUJUTA* and C PURVA

Former Post graduate student, Department of Periodontology, The Oxford dental college, Bangalore, Karnataka, India.

Abstract

With emergence of COVID-19 pandemic, research on viruses has come into focus. Understanding the nature and lifecycle of a virus is extremely important in decoding its behaviour with evolution. Mutation in SARS-CoV-2 has helped virus-related adaptation and survival. Certain key mutations have paved way for dominance of virus over the host. These mutations contribute to change in the properties of the virus like enhanced viral entry inside the host cell, virulence, rate of replication, disease transmission, and reduced response to therapeutics and vaccines. Research shows the dominance of substitution mutations in spike protein of SARS-CoV-2. This has led to emergence of viral variants. Depending on their features they are further categorized into alternative of interest & alternative of concern. Classifying viral variants helps communicate globally and decide upon common measures to prevent and control the disease. Critically evaluating the key mutations and its effect on properties exhibited by virus will aid in understanding the viral pathogenicity and ultimately the rate of patient morbidity and mortality. There has been change in the signs and symptoms with the evolution of SARS-CoV-2 & its upcoming strains. Both systemic and oral manifestations faced a shift in clinical presentation to some extent. With emerging variant of SARS-CoV-2 R0/ rate of transmission of virus and its virulence are exponentially increasing. As dental surgeon are at high risk, precautionary measures should be taken and sterilization protocol has to be strictly followed.

Article History

Received: 24 October 2021

Accepted: 22 December 2021

Keywords

Covid-19;
Delta Plus;
Mutations;
Sars-Cov-2 Variants;
Spike Protein.

Introduction

COVID-19 (Coronavirus Disease – 2019) pandemic has rapidly spread worldwide & drastically brought down the economic status of many countries with human suffering.[1] The COVID-19 initiated by SARS-CoV-2 (Severe Respiratory Syndrome – Coronavirus – 2) undergoes constant changes for better adaptation and survival. These changes are nothing but mutations, which could help the virus dominate the host. Mutations in SARS-CoV-2

CONTACT Rujuta P drrujutapatil@gmail.com Former Post graduate student, Department of Periodontology, The Oxford dental college, Bangalore, Karnataka, India.

Doi: http://dx.doi.org/10.12944/EDJ.03.02.04

help them undergo structural changes over time. The key mutations contribute to change in the properties of the virus-like enhanced viral entry inside the host cell, virulence, rate of replication, disease transmission, and reduced response to therapeutics and vaccines.[2] This review emphases on SARS-CoV-2 mutations giving rise to different variants, emphasizing the Delta Plus variant, classification of emerging variants, and sensitivity to therapeutics in COVID-19 individuals.

Structural Changes of Spike Protein

The most important part of SARS-CoV-2 is Spike protein. It plays decisive part in a viral access inside host cell & rate of infectivity. The spike protein is 180 -200k Da and is present in a significantly inactive state. The activation takes place on interaction with the host cell, leading to dramatic structural changes in the viral spike protein.[3,4] The structure of spike protein is shown in Figure 1. Few critical structural modifications in spike protein take place in its Receptor Binding Domain (RBD), which include – a) Standing–up state; b) Lying–down state. The standing–up state of RBD helps in binding of viral spike protein to host cell receptor. The lying–down state hides the RBD making it non-accessible to the host immune surveillance. Along with this the higher affinity of RBD to hACE2 (human Angiotensin Converting Enzyme) receptor makes its entry inside host cells simpler and faster.[1]

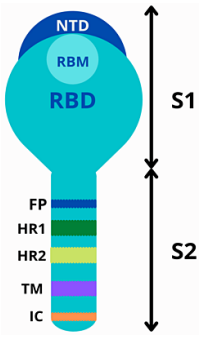

Fig. 1: Edifice of SARS-CoV-2 Spike Protein – Spike protein is divided into S1 & S2 subunits; S1 subunit comprises of - NTD – N-Terminal Domain, RBM – Receptor Binding Motif, RBD – Receptor Binding Domain; S2 subunit comprises of Fusion Peptide (FP), Heptad Repeat 1 (HR1), Heptad Repeat 2 (HR2), Transmembrane Domain (TM), & Intracellular Domain (IC).[5]

Mutations Leading to SARS-CoV-2 Variants

Mutations are a part of evolution. The new genetic variants as a result of mutation are passed on to future generations.[6] The genetic code of a virus changes naturally over time as and when it infects an animal or a person. The SARS-CoV-2 has also undergone mutations to adapt to the environment and increase the chances of its survival. These mutations have given upsurge to variants of SARS-CoV-2 (Table 1). Not all mutants are harmful, but the mutations in spike protein of SARS-CoV-2 have given rise to highly infective and antigenic variants. Genetic variants of SARS-CoV-2 are assessed using next generation sequencing.[7]

Table 1: SARS-CoV-2 Variants (8)

SARS-CoV-2 Variants	Mutation	Initial detection
Alpha	B.1.1.7	United Kingdom
Beta	B.1.351	South Africa in December 2020
Gamma	P.1	Travelers from Brazil, screened in Japan airport in early January.
Delta	B.1.617.2	India in December 2020.
Iota	B.1.526	United States (New York) – Nov -ember 2020
Epsilon	B.1.427	United States - (California)
Kappa	B.1.617.1	India – December 2020
Eta	B.1.525	United Kingdom/ Nigeria – Decem -ber 2020
Zeta	P.2	Rio de Janeiro, Brazil

Mutations in Alpha (B.1.1.7) & Beta (B.1.351) Variants

D614G Mutant SARS-CoV-2

D614G is mutation in carboxy (C)-terminal region of S1 subunit of spike protein. This region of the S1 subunit directly associates with S2. The mutation involves the substitution of aspartic acid at residue 614 (D614) with glycine (G614).[8,9] Evidence suggests that D614G does not escape the

immune-activated neutralizing antibodies from recognizing SARS-CoV-2. The phenomenon of immune escape is noticed in variants with mutation in RBD region of spike protein, as RBD is a target for neutralizing antibodies. RBD mediates a crucial step in viral entry by binding to cell-receptor protein ACE2 (Angiotensin Converting Enzyme – 2). The D614G mutation is not existing in RBD region of SARS-CoV-2 and yet the variant remains sensitive to host antibodies.[10]

Studies on animal models have shown increased viral transmission by D614G mutant SARS-CoV-2. The human studies have also evidenced enhanced viral imitation in human airway epithelial cells.[11,12,13,14] The SARS-CoV-2 spike protein variant D614G is one of only four particular nucleotide polymorphisms (SNPs). D614G enhances replication of SARS-CoV-2, thereby upsurges the chances of human-to-human transmission.[7] The molecular modeling and docking analysis identified an increase in viral S1 domain shedding in D614G mutant SARS-CoV-2, which could attribute to increased affinity of spike S1-S2 hinge region with TMPRSS2 (transmembrane serine protease 2) protease. The D614G correlated proportionally to viral load and infectivity.[15]

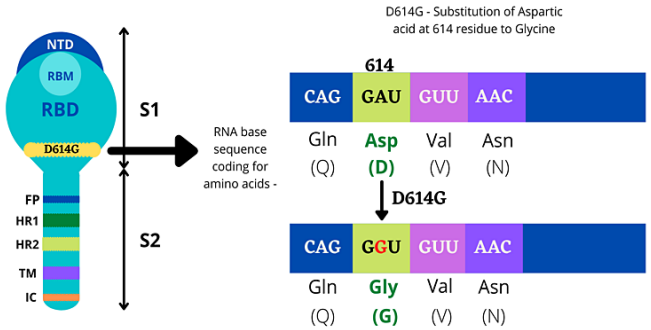

Fig. 2: D614G Mutation - D614G is a substitution mutation present in spike protein of SARS-CoV-2, wherein at 614th residue amino acid aspartic acid (D) is substituted by glycine (G). N501Y Mutant SARS-CoV-2

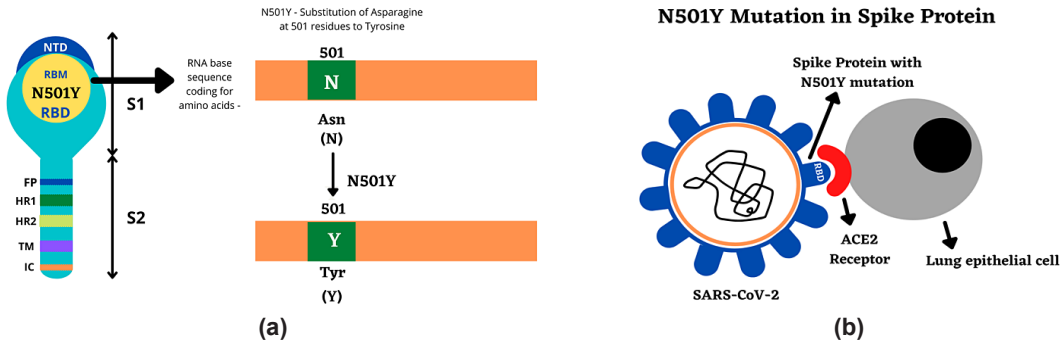

Fig. 3: N501Y mutation – a) N501Y is a substitution mutation present in Receptor Binding Domain of spike protein of SARS-CoV-2, wherein at 501st residue, amino acid asparagine (N) is substituted by tyrosine (Y). b) N501Y mutation leads to enhanced binding between spike protein & ACE2 receptor of host cell (eg: lung epithelial cell)

N501Y mutation is existing in receptor-binding domain (RBD) of spike protein at position 501, where amino acid asparagine (N) is supplanted with tyrosine (Y)[16] (Figure 3). Study on hamster model assessed 8 individual spike protein substitutions, out of which N501Y demonstrated enhanced fitness for repetition in upper airway. Enhanced fitness could be attributed to augmented replication & shedding of virus inside nasal cavity, responsible for the formation of efficient air-borne &/or fomite-mediated transmission. The N501Y substitution mutation contributes to phenotype of enhanced

viral transmission, suggesting N501Y to be the major cause for augmented transmission of this variant. Perfunctorily, N501Y substitution amended kinship of viral spike protein for cellular receptors & is foremost adaptive spike mutation of concern.[17] Host range, tissue tropism, transmission, & disease pathogenesis are greatly influenced by substitution mutations in spike protein, leading to enhanced viral-ACE2 receptor binding.[18]

This interaction promotes increased viral entry inside the host cell and further viral replication. All these contribute to increased viral virulence and pathogenicity ultimately, affecting the disease outcome.

Delta Variant and its Sub-lineage Delta Plus

The Delta variant (B.1.617.2) & its sub lineages (AY.1, AY.2, AY.3) have mutations in spike protein region (16). The substitution mutations in spike protein include T19R, V70F, T95I, G142D, E156-, F157-, R158G, A222V, W258L, K417N, L452R, T478K, D614G, P681R, D950N. The traits of delta variant & its sub lineages comprise of increased transmissibility with a potential lessening in neutralization by some EUA (Emergency Utilization Authority) like monoclonal antibody cures & by post-vaccination sera.[8] The most frequent spike protein mutation in delta sub-lineage AY.1/AY.2 is K417. The delta and its mutants are considered a variant of concern.[19] B.1.617.2.1 (AY.1) or generally acknowledged as Delta Plus variant implies Delta variant with additional mutation.[20]

Spike Protein Mutations Present in Delta Plus (Ay.1 – B.1.612.2.1) Variant – K417n, L452r and T478k

The Delta Plus variant caries K417N mutation in RBD of spike protein in addition to L452R and T478K mutations known to be present in parent Delta variant. The K417N mutation was previously reported to be present in Beta variant or lineage B.1.351 initially recognized in South Africa. As K417N, L452R, and T478K mutations are extant in spike protein of SARS-CoV-2, they have highest ability to impart pathogenic properties to the virus. The K417N amino acid substitution is present near the neutralizing antibody target domain, which made scientists conclude that the enhanced immune invasion property of SARS-CoV-2 might be contributed by K417N mutation. It might be synergistic with L452R, which is known to impart similar actions as that of K417N. The T478K transformation happens inside the interface between the RBD and the human receptor ACE2. The additional immune-escape-conferring mutation provides AY.1 lineage a slightly better fitness advantage than the Delta variant. Even so, more investigations are obligatory to comprehend mutual impression of these mutations into SARS-CoV-2 contagiousness, virulence, and capacity to evade host immune response activated on the administration of different COVID-19 vaccines.[21]

K417N - Key Mutation in Delta Plus Variant of SARS-CoV-2

Procurement of K417N mutation in Delta (B.1.617.2) ensued in progression of AY.1. Correspondingly, procurement of K417N & A222V mutations in Delta (B.1.617.2), ensued in the advancement of AY.2.[22] AY.1 variant known as delta plus carries the key spike mutation K417N, wherein lysine is substituted by asparagine at 417 residues. The spike conformation of SARS-CoV-2 variants is affected by amino acid substitutions, especially variant of concerns.[23] The evidence from free energy perturbation (FEP) calculations for interactions of RBD of spike protein with ACE2 receptor and antibody from COVID19 positive patient confirmed significantly increased S1 RBD-ACE2 interactions and drastically reduced STE90-C11 antibody.[24] These properties affect the rate of transmission, infectivity, and virulence of the virus.

Classification of Variants and its Significance

Variants are the consequences of mutation in the SASRS-CoV-2. Every variant affects human beings differently. Not all variants are harmful, but the effects of key pathogenic variants vary from trifling indications to severe forms of disease with hospitalization and sometimes death. Classifying these variants into different categories helps in defining the nature of the viral variant and consequential effects inside the host. Presently CDC (Centre for Disease Control) has classified them into four categories[8] -

- Variant of Interest (VOI)
- Variant of Concern (VOC)
- Variant of High Consequence (VOHC)
- Variant Being Monitored (VBM)

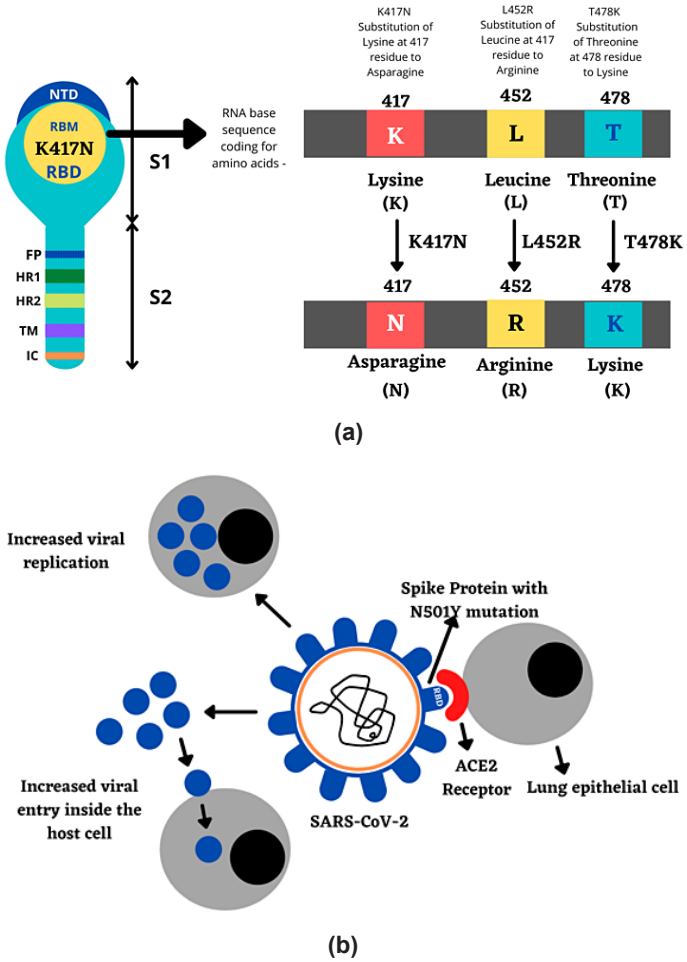

Fig. 4: K417N, L452R and T478K Mutation in Delta Plus – a) K417N, L452R and T478K are substitution mutations present in spike protein of SARS-CoV-2 Delta Plus variant. K417N involves substitution of amino acid Lysine at 417 residue to Asparagine. L452R involves substitution of Leucine at 417 residue to Arginine. T478K involves substitution of Threonine at 478 residue to Lysine; b) Consequences of mutations in spike protein of SARS-CoV-2 – Improved binding of RBD of spike protein to ACE Receptor

Variant of Interest

The virulence of any variant of SARS-CoV-2 depends on various factors. A variant is said to be of interest if the genetic mutations increase the rate of disease transmission and severity, lowers the response of patients to therapeutics, are capable of neutralizing the antibodies produced against previous infection or vaccination, & abridged sensitivity of variant to diagnostic techniques. Receptor binding region is critical for biding of SARS-CoV-2 to host ACE2 receptor. Mutations in the receptor-binding region are often seen in variants of interest, increasing their virulence. Such mutational changes are also concomitant with variant of interest. Increase in the SARS-CoV-2 positive cases or unique outbreak clusters is one of the attributes of the variant of interest. But the prevalence and spread need to be limited to be classified as a variant of interest. Appropriate public health actions against a variant of interest are required. These include enhanced sequence surveillance, increased laboratory characterization, and assessing the spread of the virus and its severity by epidemiological investigations. The monitoring of the effectiveness of therapeutic strategies and vaccination-mediated protection is required.[8]

Variant of Concern

A variant of concern is potentially more pathogenic than variant of interest. The attributes of variant of concern include evidence of a significantly increased disease transmissibility and severity with a higher rate of hospitalization or deaths. Characterized by a notable lessening in neutralization by antibodies engendered for the duration of erstwhile infection or vaccination along with a marked reduction in the effectiveness of therapeutic agents or vaccine-induced protection. The diagnostic aids nearly fail in detecting a variant of concern, as it interferes with the diagnostic test targets. All these attributes make a variant of concern dangerous with immediate attention and rigorous public health activities. These include statement to WHO (World Health Organization) under International Health Regulations, reporting to CDC, local or regional exertions to regulate spread, augmented testing, or research to fix efficacy of vaccines & cures in contradiction of variant. Additional considerations required include the development of newer diagnostic aids, modifications in the vaccines or therapeutics.[8]

Variant of Great Consequence

Variant of extraordinary significance has substantiation for reduced efficacy to preventive measures or medical countermeasures compared to previously circulating variants. It has all the traits of variant of concern, in accumulation to validated failure of diagnostic aids, evidence of significantly reduced effectiveness of vaccine with a higher figure of vaccine breakthrough cases, or lower vaccine-induced fortification in contradiction of severe disease. These variants also render patients markedly less responsive to multiple EUA or approved therapeutics. Appropriate actions include statement to WHO under International Health Regulations, reportage to CDC. Along with the proclamation of effective prevention stratagems or containment of conduction & commendations to apprise therapeutics & vaccines.[8]

Variant Being Monitored

Variants that continue to be a threat to the society are being monitored. Such variants are classified under variants being monitored. Evidence should indicate the negative impact of this variant on currently approved modalities to prevent or treat the disease. The variant should have the potential to increase the transmission of COVID-19 and cause server form of disease even at very low titres to be qualified as variant being monitored. Depending on the behaviour of variant and its negative impact on the society over time the classification of variant is changed.[8]

Discussion

The SARS-CoV-2 variants trigger immune response differently and signs and symptoms also vary accordingly. In India the first wave of COVID-19 which was dominated by the beta variant showed symptoms of loss of taste and smell sensation along with other oral manifestations like Oral dryness, vesiculobullous lesions, aphthous-like lesions, dysgeusia, & anosmia.[25] Triad xerostomia, taste dysfunction, and oral mucosal lesions are most commonly encountered oral manifestations of COVID-19 patients.[26] There has been change in signs and symptoms with the evolution of SARS-CoV-2 & its upcoming strains. Second wave in India dominated by Delta plus variant, created havoc and was predominated by severe form of pneumonia, shortness of breath, drop in oxygen saturation of blood, fever, cough etc. This shows that respiratory system was major target of virus and severely damaged the system. Oral symptoms developed within 2-14 days of virus were loss of taste, xerostomia, mucosal lesions, periodontal disease and COVID tongue. The COVID tongue is characterized by areas depapillation on dorsum. Literature also shows the possible association of increase in IL-6 cytokine with COVID tongue, which is the culprit cytokine for COVID-19 associated cytokine storm.[26,27] The delta plus variant is considered dangerous as it evades immunity developed post vaccination as well as natural immune response post COVID-19 recovery.[26] Both the systemic and oral manifestations faced a shift in clinical presentation to some extent. With emerging variant of SARS-CoV-2 R0/ rate of transmission of virus and its virulence are exponentially increasing. As dentists are at high risk, precautionary measures should be taken and sterilization protocol has to be strictly followed.

Conclusion

The type and location of mutations in SARS-CoV-2 are precarious in deciding characteristics

exhibited by the viral variant. Understanding the mutational intricacies guides in assessing the effect of viral infection on the population and also aids in formulating the appropriate and effective therapy and prophylactic measures. Variant of SARS-CoV-2, Delta is exceedingly virulent due to properties it possesses. Talk of hour is Delta plus, a sublineage of Delta variant containing mutations similar to that of the parent delta variant with few additional key mutations in spike protein that could contribute to SARS-CoV-2 pathogenicity and virulence, ultimately affecting the rate of patient morbidity and mortality. The structural and genetic changes in virus could affect the severity of disease and its clinical presentation. Yet more studies with larger sample sizes are required to confirm the virulent nature of Delta plus and its clinical implications.

Recommendations

- Key mutations in SARS-CoV-2
- Genetic composition, key mutations of Delta plus and its clinical implication
- Categorization of SARS-CoV-2 variants
- How & where SARS-CoV-2 mutations take place
- Alteration in Oral and systemic manifestations with emergence of delta plus variant

Acknowledgments

The author would like to thank Magadh University for granting the Ph.D. research work. The Department of Environmental Science, A.N. College, Patna of the Magadh University, is highly appreciated for allowed the GIS laboratory work. The author is also profoundly grateful to the National Remote Sensing Center (NRSC), Indian Space Research Organisation (ISRO), Govt. of India for their guidance during the Satellite data procurement.

Funding

The author(s) received no financial support for the research, authorship, and/or publication of this article.

Conflict of Interest

There is no funding or financial support for this research work.

References

1. Shang, J., Wan, Y., Luo, C., Ye, G., Geng, Q., Auerbach, A., Li, F. Cell entry mechanisms of SARS-CoV-2. *Proceedings of the National Academy of Sciences*. 2020; 117 (21):11727-11734.
2. https://www.who.int/en/activities/tracking-SARS-CoV-2-variants/.
3. Bosch, B. J., Van der Zee, R., De Haan, C. A., Rottier, P. J. The coronavirus spike protein is a class I virus fusion protein: structural and functional characterization of the fusion core complex. *Journal of virology.* 2003; 77 (16):8801-8811.
4. Huang, Y., Yang, C., Xu, X. F., Xu, W., Liu, S. W. Structural and functional properties of SARS-CoV-2 spike protein: potential antivirus drug development for COVID-19. *Acta Pharmacologica Sinica*. 2020; 41 (9):1141-1149.
5. Elaswad, A., Fawzy, M., Basiouni, S., Shehata, A. A. Mutational spectra of SARS-CoV-2 isolated from animals. *PeerJ*. 2020; 8: e10609.
6. Carlin, J. L. Mutations are the raw materials of evolution. *Nature Education Knowledge.* 2011; 3 (10):10.
7. Yurkovetskiy, L., Wang, X., Pascal, K. E., Tomkins-Tinch, C., Nyalile, T. P., Wang, Y., Luban, J. Structural and functional analysis of the D614G SARS-CoV-2 spike protein variant. *Cell*. 2020; 183 (3):739-751.
8. https://www.cdc.gov/coronavirus/2019-ncov/variants/variant-info.html
9. Zhang, L., Jackson, C. B., Mou, H., Ojha, A., Peng, H., Quinlan, B. D., Choe, H. (2020). SARS-CoV-2 spike-protein D614G mutation increases virion spike density and infectivity. *Nature communications.* 2020; 11 (1):1-9.
10. Callaway, E. Making sense of coronavirus mutations. *Nature.* 2020; 174-177.
11. Hou, Y. J., Chiba, S., Halfmann, P., Ehre, C., Kuroda, M., Dinnon, K. H., Baric, R. S. SARS-CoV-2 D614G variant exhibits efficient replication ex vivo and transmission in vivo. *Science*. 2020; 370 (6523): 1464-1468.
12. Plante, J. A., Liu, Y., Liu, J., Xia, H., Johnson, B. A., Lokugamage, K. G., Shi, P. Y. Spike mutation D614G alters SARS-CoV-2 fitness.

Nature. 2021; 592 (7852):116-121.

13. Chan, J. F. W., Zhang, A. J., Yuan, S., Poon, V. K. M., Chan, C. C. S., Lee, A. C. Y., ... & Yuen, K. Y. Simulation of the clinical and pathological manifestations of coronavirus disease 2019 (COVID-19) in a golden Syrian hamster model: implications for disease pathogenesis and transmissibility. *Clinical infectious diseases.* 2020; 71 (9):2428-2446.
14. Zhou, B. Thi Nhu Thao T, Hoffmann D, Taddeo A, Ebert N, Labroussaa F, Pohlmann A, King J, Steiner S, Kelly JN et al: SARS-CoV-2 spike D614G change enhances replication and transmission. *Nature.* 2021; 592 (7852):122-127.
15. Raghav, S., Ghosh, A., Turuk, J., Kumar, S., Jha, A., Madhulika, S., Odisha COVID-19 Study Group. Analysis of Indian SARS-CoV-2 genomes reveals prevalence of D614G mutation in spike protein predicting an increase in interaction with TMPRSS2 and virus infectivity. *Frontiers in Microbiology.* 2020; 11.
16. https://www.cdc.gov/coronavirus/2019-ncov/science/science-briefs/scientific-brief-emerging variants.html
17. Liu, Y., Liu, J., Plante, K. S., Plante, J. A., Xie, X., Zhang, X., Weaver, S. C. The N501Y spike substitution enhances SARS-CoV-2 transmission. *BioRxiv.* 2021.
18. Haddox, H. K., Dingens, A. S., Hilton, S. K., Overbaugh, J., Bloom, J. D. Mapping mutational effects along the evolutionary landscape of HIV envelope. *Elife*. 2018; 7: e34420.
19. https://dbtindia.gov.in/sites/default/files/INSACOG%20Bulletin%20July%2019.pdf
20. https://dbtindia.gov.in/pressrelease/qa-indian-sars-cov-2-genomics-consortium-insacog
21. https://www.proteogenix.science/product/sars-cov-2-rbd-of-spike-proteink417n-t478k-l452r-lineage-b-1-617-2-1-indian-delta-plus-variant/
22. https://dbtindia.gov.in/sites/default/files/INSACOG%20WEEKLY%20BULLETIN%20June%2030.pdf
23. Gobeil, S., Janowska, K., McDowell, S., Mansouri, K., Parks, R., Stalls, V., Acharya, P. Effect of natural mutations of SARS-CoV-2 on spike structure, conformation and antigenicity. *bioRxiv*. 2021.
24. Fratev F. The N501Y and K417N mutations in the spike protein of SARS-CoV-2 alter the interactions with both hACE2 and human derived antibody: A Free energy of perturbation study. *BioRxiv.* 2020.
25. Rodríguez MD, Romera AJ, Villarroel M. Oral manifestations associated with COVID-19. Oral diseases. 2020.
26. Amorim dos Santos J, Normando AG, Carvalho da Silva RL, Acevedo AC, De Luca Canto G, Sugaya N, Santos-Silva AR, Guerra EN. Oral manifestations in patients with COVID-19: A 6-month update. *Journal of dental research.* 2021 Nov;100 (12):1321-1329.
27. Iranmanesh B, Khalili M, Amiri R, Zartab H, Aflatoonian M. Oral manifestations of COVID-19 disease: A review article. Dermatologic therapy. 2021;34 (1): e14578.