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SURVIVAL OF CORONAVIRUSES IN DIFFERENT INTERMEDIATE HOST: SPIKE PROTEIN, NUCLEOTIDE SIMILARITY, AMINO ACID SUBSTITUTION, AMINO ACID SIMILARITY AND RECEPTOR CHANGE

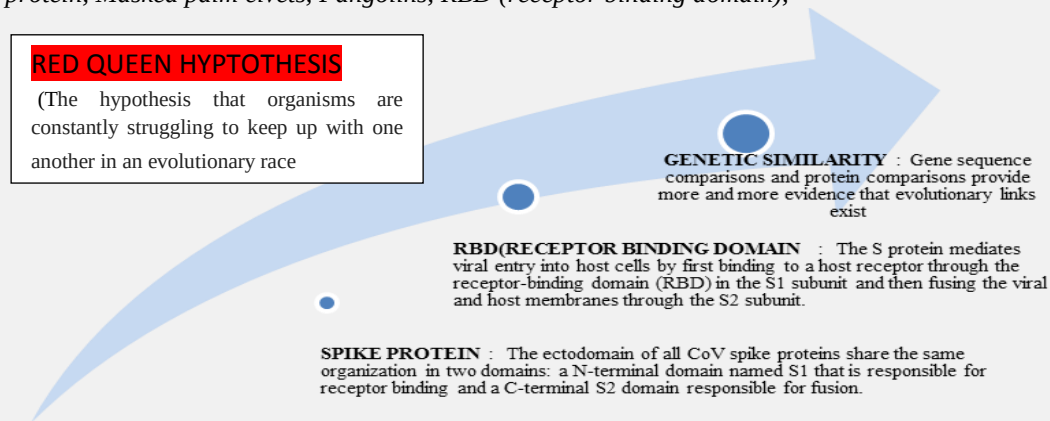
Ashwini A Mankar[1], Vijay S Harode[2]

Assistant Professor, Department of Biotechnology, Kamla Nehru Mahavidyalaya, Nagpur (INDIA)[1]
Assistant Professor, Department of Biotechnology, Kamla Nehru Mahavidyalaya, Nagpur (INDIA)[2]
mankarashwini3@gmail.com, vijayharode29@gmail.com

ABSTRACT

The New Year began with a new threat to the human health, Severe Acute Respiratory Syndrome (SARS)-CoronaVirus (CoV)-2. SARS-CoV-2 is a novel virus (identified as the cause of coronavirus disease 2019 (COVID-19) that began in Wuhan, China in late 2019) belonging to the Coronavirus family, the 7th recognized as a human pathogen and the 3rd causing a severe clinical syndrome after SARS-CoV (identified in 2002 as the cause of an outbreak of severe acute respiratory syndrome (SARS) and Middle East Respiratory Syndrome (MERS)-CoV1(was identified in 2012 as the cause of Middle East respiratory syndrome (MERS).[Lai CC et al; 2020]. Intermediate host played an important role in transmission of coronaviruses from them to humans such as in case of COVID 19 (pangolins or bats yet to be confirmed), SARS-CoV (masked palm civets), MERS (camel) which arises many questions that why there is need of changing the intermediate host for its survival. In this review we will see the modifications done in respect for its survival which includes S protein , nucleotide, amino acid substitution, genetic similarity

Keywords: *S protein, Masked palm civets, Pangolins, RBD (receptor binding domain),*



I INTRODUCTION

In December 2019, there was an outbreak of pneumonia with an unknown cause in Wuhan, Hubei province in China, with an epidemiological link to the Huana Seafood Wholesale Market, which is alive animal and seafood market. Clinical presentations of this disease greatly resembled viral pneumonia. Through deep sequencing on the lower respiratory tract samples of patients, a novel coronavirus named the 2019novel coronavirus (2019nCoV) was identified **[**SongZ. et al; 2019**].** As of 16 April 2020, more than 2.06 million cases [CSSE 2020] of

COVID-19 have been reported in 210 countries and territories**,[**Worldometer 2019**]** resulting in more than 1,37,000 deaths. More than 5,12,000 people have recovered **[**SongZ. et al; 2019**]** although there may be a possibility of relapse or reinfection**.[** Politi , Daniel 2020 **;** Feng, Emily 2020**]** The deaths per diagnosed cases varies significantly between countries**.[** Centre for Evidence-Based Medicine., 2020**]** SARS-CoV first emerged in 2002–2003 in Guangdong, China as an atypical pneumonia marked by fever, headache and subsequent onset of respiratory symptoms such as cough and pneumonia, which may later develop into life-threatening respiratory failure and acute respiratory distress syndrome **[**Graham. et al., 2013**]** . Being highly transmissible among humans, it quickly spread across 29 countries, infecting more than 8000 individuals with a mortality rate of about 10% **[**Frieman 2008**;** Peiris. et al., 2004**].** Originally, palm civets were thought to be the natural reservoir for the virus **[**Wang et al 2005**].** However, subsequent phylogenetic studies pointed to the bat origin of SARS-CoV based on sequences of SARS-like virus found in bats [Hu. et al., 2005**].** The MERS-CoV epidemic surfaced in Saudi Arabia in 2012 with similar clinical symptoms as SARS-CoV but with a much higher mortality rate of about 35% **[**Kim. et al., 2016**]** MERS-CoV specific antibodies were only found in dromedary camels **[**Reusken.et al., 2013**]**. Another evidence to link MERS-CoV to dromedaries was found after two human cases of MERS-CoV infection, diagnosed in October of 2013, and were linked to a farm in Qatar **[**Haagmans BL.et al., 2013 **]**. In response, all the 14 dromedary camels on that farm were tested with RT-PCR. Eleven dromedary camels had positive nasal swabs for MERS-CoV which suggest that dromedary camels were responsible for transmission and considered as host.**[** Haagmans BL.et al., 2013**]** Unlike SARS-CoV, which exhibits super-spreader events, transmission of MERS-CoV is geographically limited **[**Graham. et al., 2013**].** In fact, reported cases of MERS-CoV often stem from outbreaks within the Middle Eastern countries or recent travel to the region **[**Oboho. et al., 2015**]**

Why there is a need to keep changing a pathogen host for the sake of survival: Insights to some modifications!!!

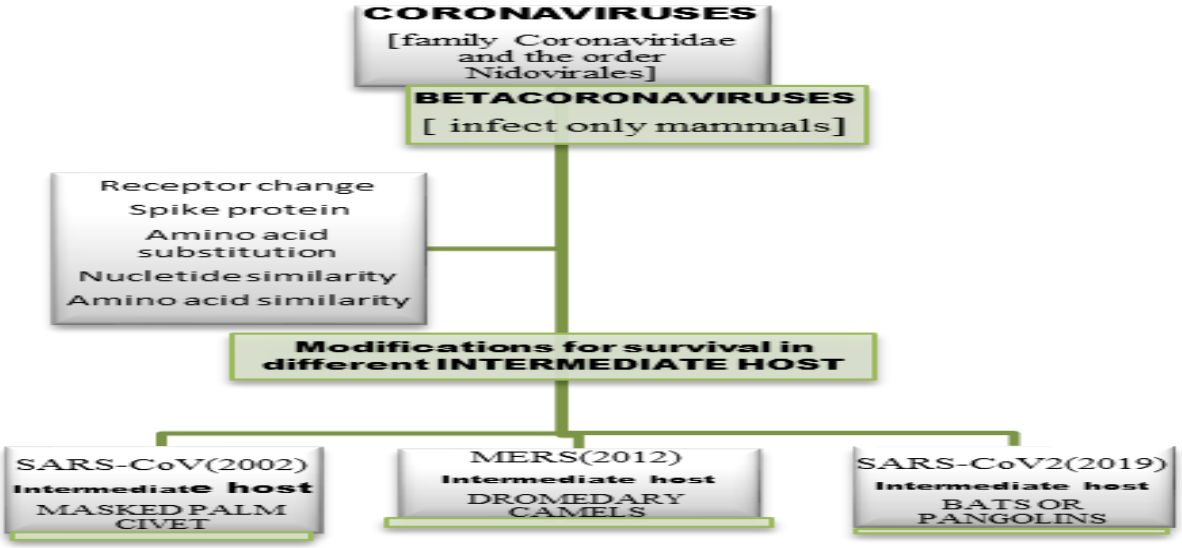
The pathogen's evolutionary survival depends on infecting new hosts - and jumping to other species is one way to do this. The new host's immune systems try to kill off pathogens, meaning the two are locked in an eternal evolutionary game of trying to find new ways to vanquish each other. For example, about 10% of infected people died during the 2003 SARS epidemic, compared with under 0.1% for a "typical" flu epidemic. Environmental and climate change are removing and altering animals' habitat, changing how they live, where they live and who eats whom. While there might be pathogens that can infect other hosts and cause disease, the inability to pervade, or spread, throughout the infected host species indicates that the pathogen is not adapted to that host species. In this case, the ability or lack thereof a pathogen to adapt to its host environment is an indicator of the pathogen's fitness or virulence. If a pathogen has high fitness in the host environment, or is virulent, it will be able to grow and spread quickly within its host. For instance, in what's called the **red queen hypothesis,** hosts are constantly shifting genetically via sexual reproduction in order to continue changing so pathogens have less of a chance to be well adjusted to the host. If the host keeps changing via gene shuffling in the form of reproduction, then hosts will have to continuously evolve with the host to keep up with its changes. This sets up a moving target for co-evolving pathogens**. [**Den Bakker et al., 2011**]** This would be the reason why the pathogens continuously change their host in order to sustain itself by doing some or the other modification. As we know the new coronavirus continues to spread around the globe, researchers say the virus is changing its genetic makeup slightly like all viruses — change small parts of their genetic code all the time. They proofread their own genomes when they copy themselves, cutting out things that don't seem right. “They maintain this ability to keep their genome pretty much intact by doing some or the other modification," says Vineet Menachery, a virologist at the University of Texas Medical Branch.”

So, according to the research it seems that the modifications are done in respect to genetic makeup, receptor binding domain, receptors and some nucleotide substitution in order to survive in different intermediate host so that it could percolate and result in spillover to humans.

Comparison of the changes done while changing the [host for the sake of spillover to humans (SARS-CoV1 , SARS-CoV2 , MERS) …

In case of **SARS-CoV** the host were considered to be Masked palm civet [Wang.et al., 2005] and when the genome sequences of masked palm civets were compared with 92 human SARS-CoVs (Song et al., 2005), a total of 212 single-nucleotide variation (SNV) sites with multiple

occurrences were identified, among which 209 sites were in the protein coding regions . At least one of the mutations, aa 487 in the spike protein, was shown to be important in the adaptation of SARS-CoV to human cells (Li et al., 2005c; Qu et al., 2005). Analysis of the structures of the spike protein and its cellular receptor angiotensin converting enzyme 2 (ACE2) (Li et al., 2003) indicated that aa 487 of spike is important for spike-ACE2 interaction and a mutation from **Ser487 (civet) to Thr487** appeared to enhance human-to-human transmission of the SARS-CoV .Residues 344 and 360 of the spike protein are distant from the binding interface in the spike-ACE2 complex (Li et al., 2005a), and these two mutations do not affect binding affinity or infectivity of the SARS-CoV (Li et al., 2005c; Qu et al., 2005). Whether the rest of the amino acids residues play important roles during cross species transmission or human-to-human transmission shall need further investigations. The genome sequences of SL-CoVs isolated from horseshoe bat shared a sequence identity of 89–90%. Similar sequence identity, 87–92%, exists between bat and human or civet isolates. (Wu et al., 2005; Kan et al., 2005).



ACCORDING TO THE DATA PUBLISHED AND THE INFORMATION IT SUGGEST THAT SARS-CoV2 MAY BE A MUTATED FORM OF SARS-CoV1 AS IT SEEKS MANY SIMILARITIES WITH THE NOVEL CORONAVIRUS 2019 AND THE ALSO THAT PANGOLINS SHOW MORE RESEMBLANCE TO THE SARS-CoV2 AND COULD BE CONSIDERED AS AN INTERMEDIATE HOST

RECEPTOR	ACE2	DPP4	ACE2
SPIKE PROTEIN		>	97.5%
NUCLEOTIDE SIMILARITY	99.94%	>99.5%	93.2%
AMINO ACID SIMILARITY	93%	-	94.1%
AMINO ACID SUBSTITUTION	SER 487 [HOST] ⬇ THR487 [HUMANS]	-	PRO38 [HOST] GLU268 ⬇ ⬇ SER 38 [HUMANS] ALA268

In similar way in case of **SARS-CoV2** it is not confirmed whether the spread was from Pangolins or Bats the evidences and information shows that Pangolin-CoV showed high overall genome sequence identity to SARS-CoV-2 (91.02%) throughout the genome also preprint studies found that CoVs in pangolins shared 90.3% [K. Xiao.et.al;2020] and 92.4% [T.T.-Y. Lam.et.al;2020] DNA identity with SARS-CoV-2, approximating the 91.02% identity to SARS-CoV-2.Pangolin-CoV genes shared high average 93.2% nucleotide identity , higher amino acid sequence identity to SARS-CoV-2 genes including the spike (S) protein (97.5%) which suggest that high S protein amino acid identity implies functional similarity between Pangolin-CoV & SARS-CoV-2, [P. Liu, W. Chen, J.-P. Chen 2019] also the two amino acid sites (38P and 268Q) are shared by Pangolin-CoV, and SARS-CoVs, which are mutated to 38S and 268A in SARS-CoV-2 which[Tao Zhang,et.al;2020] indicate that Pangolin-CoV might be the common origin of SARS-CoV-2 and also the coronavirus isolated from pangolin is similar at 99% in a specific region of the S protein, which corresponds to the 74 amino acids involved in the ACE (Angiotensin Converting Enzyme 2) receptor binding domain, the one that allows the virus to enter human cells to infect them. [Alexandre Hassanin 2020**]** Dipeptidyl peptidase 4 (DPP4, also known as CD26) was identified as a functional receptor for **MERS-CoV** [Wang. Et al., 2004] and it is relatively conserved among mammalian species, DPP4- recognizing RBD is localized to the S1 C-terminal portion of S protein of MERS-CoV [Lu g.at al.,2013; Wang N.et al.,2013; Chen Y. et al.,2013]. The core subdomain of MERS-CoV RBD is structurally similar to that of the SARS-CoV RBD, but the external subdomain (also named as RBM) is different to that of the SARS-CoV.

II CONCLUSION

The relationship between a virus and its host is a complicated affair: a myriad of factors from the virus and the host are involved in viral infection and consequential pathogenesis. As intracellular obligate parasites, viruses have also evolved various strategies to hijack the host machineries. In this review, we saw the modification in various factors responsible for the survival of Coronaviruses in different intermediate host as it keeps changing its host in order to sustain itself. For years, HCoVs have been identified as mild respiratory pathogens that affect the human population. However, it was the emergence of SARS-CoV that thrust these human viruses into the spotlight of the research field. Therefore, most of the HCoV research today is pertained towards SARS-CoV. While the recent MERS-CoV outbreak has been mostly limited to the Middle East region, it is more likely that more emerging of remerging HCoVs might surface to threaten the global public health as seen from the high mortality rates in past two outbreaks :SARS-CoV(10%) and MERS-CoV(35%). Therefore, study of the pathogenesis of all HCoVs would gain more insights for the development of antiviral therapeutics and vaccines

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AUTHOR'S INFORMATION:

1 Ashwini .A. Mankar (Assistant Professor)

mankarashwini3@gmail.com

Department of Biotechnology

Kamla Nehru Mahvidyalaya , Nagpur

CSIR NET , ARS NET, ICAR JRF AIEEA

2 Vijay.S. Harode (Assistant Professor)

vijayharode29@gmail.com

Department of Biotechnology

Kamla Nehru Mahvidyalaya , Nagpur

CSIR NET , ARS NET, GATE, SET