Conceptual model for NAb mRNA and PAC-MAN in-vivo delivery through rSARS-CoV-2 vector via ACE-2 receptors binding mechanism: Deep amalgamation approach to COVID-19 vaccine

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GRAPHICAL ABSTRACT

HIGHLIGHTS

- Development of a recombinant SARS-CoV-2 vector for in-vivo insertion of monoclonal SARS-CoV-2 neutralizing antibody gene and PAC-MAN via ACE-2 receptor binding mechanism
- Theoretical method for release of rSARS-CoV-2 to naturally infect humans through the nasal pathway and insert antibody genes which can develop immunity against pathogenic strains of the virus

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ABSTRACT

The paper presents a conceptual model for the development of recombinant SARS-CoV-2 as a vector for in-vivo delivery of viral neutralizing antibodies gene and PAC-MAN into human respiratory epithelial cells. The theoretical proposal suggests the sequence length, potential antibody candidates (as reported in the recent literature) and brief protocol which can be applied to produce rSARS-CoV-2 in the laboratory to test its effectiveness against most of the SARS-CoV-2 strains. Finally, a method for release of such functional vector is been discussed.

Keywords: *rSARS-CoV-2; virus vector vaccine; COVID-19 vaccine; SARS-CoV-2 neutralizing antibody; PAC-MAN in-vivo delivery*

INTRODUCTION

15th March 2020 (19:14 GMT), WHO announced the COVID-19 as a pandemic. This modern pandemic has constrained the whole world into a series of lockdowns, affected the global economy and changed the way humans interact.

Despite the most robust medical facilities and numerous advancements in medical sciences, scientists are still struggling to produce a safe and effective COVID -19 vaccine.

METHODS AND MATERIALS

Genomic composition analysis of this virus was the primary step to understand the virus-host interaction mechanism and disease propagation. The SARS-CoV-2 interact with ACE-2 enzyme on human cell and then gain entry inside the host cell (Zhang et al., 2020).

The original viral genome is available at NCBI (NC_045512) and we have performed its whole genome sequence analysis in our last preprint available at OSF preprint server, to better understand the potential role of each protein (Sharma et al., 2020).

Recently, a study published in Nature states the neutralizing potency of the monoclonal antibody S309 (**PDB: 6WS6**) (Pinto, et al., 2020).

Abbott et al., team reported a CRISPR-Cas13-based strategy, PAC-MAN (prophylactic antiviral CRISPR in human cells) and a group of six crRNAs for viral inhibition and efficient target RNA degradation in human lung epithelial cells that can target more than 90% of all coronaviruses (Abbott, et al., 2020).

We propose the use of a recombinant SARS-CoV-2 vector with S309 neutralizing antibody gene, CRISPR-Cas13d and its cognate crRNAs which could be delivered in RNA form for the in-vivo expression of PAC-MAN components. The process will involve the development of a recombinant virus system which can gain entry into the cell via the ACE-2 receptor binding mechanism and release the neutralizing antibody gene into the target cells.

The in-vivo expression of neutralizing antibody gene and PAC-MAN components by the cellular machinery will release the self-manufactured antibody into the system which will help humans to develop immunity against the pathogenic strains of the virus, as well as deploy PAC-MAN system into the cell to clean-up viral genome from already infected cells.

The rSARS-CoV-2 vector can be prepared with modern recombinant DNA technology. Major genome components required to create such rSARS-CoV-2 vector for in-vivo expression on neutralizing antibody are mentioned in table 1.

Table 1. *rSARS-CoV-2 whole-genome major components*

S.No.	Protein	Sequence length (aa)	NCBI Reference Sequence
1.	5' UTR	265	-
2.	Surface glycoprotein	1237	YP_009724390.1
3.	Envelope protein	75	YP_009724392.1
4.	Membrane glycoprotein	222	YP_009724393.1
5.	Nucleocapsid phosphoprotein	419	YP_009724397.2
6.	S309 Antibody gene	1332	6WS6
7.	3' UTR	229	-

Figure 1. *rSARS-Cov-2 vector design with both components.*

RESULTS AND DISCUSSION

The recombinant virus will follow the entry mechanism as the original pathogenic virus which will ensure the to-the-point delivery of the neutralizing antibody gene and PAC-MAN components. The antibody produced by this gene would be considered by the human body as a self-produced antibody hence, not produce an immune response against it. The neutralizing antibody will fight against any new virus in the system whereas the PAC-MAN will halt the viral gene expression by viral gene degradation. This deep amalgamation of both the techniques for neutralizing and viral genome degradation inside the infected cell can be beneficial for infected patients to fight off the virus as well as healthy individuals to build-up immunity against the virus. This portrays an approach to vector-based “Injection less” COVID-19 vaccine.

Deployment of the rSARS-CoV-2 vector

Finally, after the clinical testing and efficacy evaluation, the rSARS-CoV-2 vector may be deployed through the air pathway. The rSARS-CoV vector has NO disease-causing RNA genome, hence assumed to have no danger, which should be strictly confirmed during laboratory testing.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

REFERENCES

1. Zhang H, Penninger JM, Li Y, Zhong N, Slutsky AS. Angiotensin-converting enzyme 2 (ACE2) as a SARS-CoV-2 receptor: molecular mechanisms and potential therapeutic target. *Intensive care medicine* [Internet]. 2020;2. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/32125455>
2. Sharma D, Saini A, Singh DD. SARS-CoV-2 whole-genome classification analysis, protein homology modelling and elucidation of associated protein functions [Internet]. OSF Preprints; 2020. Available from: osf.io/9afp3
3. Pinto D, Park Y, Beltramello M, Walls AC, Tortorici MA, Bianchi S, et al. Cross-neutralization of SARS-CoV-2 by a human monoclonal SARS-CoV antibody. *Nature* [Internet]. 2020; Available from: <http://dx.doi.org/10.1038/s41586-020-2349-y>
4. Abbott TR, Dhamdhare G, Liu Y, Lin X, Goudy L, Zeng L, et al. Development of CRISPR as an Antiviral Strategy to Combat SARS-CoV-2 and Influenza. *Cell* [Internet]. 2020;181(4):865-876.e12. Available from: <https://doi.org/10.1016/j.cell.2020.04.020>