List of publications, conference presentations and awards

Publication

Devi, Y. D., Devi, A., Gogoi, H., Dehingia, B., Doley, R., Buragohain, A. K., Singh, C. S., Borah, P. P., Rao, C. D., Ray, P., Varghese, G. M., Kumar, S., & Namsa, N. D. (2020). Exploring rotavirus proteome to identify potential B- and T-cell epitope using computational immunoinformatics. Heliyon, 6(12), e05760. https://doi.org/10.1016/j.heliyon.2020.e05760.(published)

Devi, Y. D., Dey, U., Kumar, A., Singh, C. S., & Namsa, N. D. (2022). Genome Sequence of a Wa-Like G3P[8] Rotavirus from a 12-Month-Old Child with Diarrhea in Manipur, India. Microbiology resource announcements, 11(8), e0125421. https://doi.org/10.1128/mra.01254-21.

Conference presentations

Oral presentation on the title “Genetic Analysis of Rotavirus VP7 and VP4 Antigenic Epitopes Between Circulating Strains and Rotavac vaccine: Monitoring Vaccine efficacy” 44th Annual Conference of Indian Association of Medical Microbiologist (IAMM) ‘MICROCON 2021’ with the theme “Clinical Microbiology in the wake of emerging threats”

Speaker Certificate on the topic “Epidemiology of Group A Rotaviruses Among Under-five Children in Imphal, Manipur, India” at international conference “Tackling Global Viral Epidemics Conference 2021” from 16-18 June 2021 organized by World Society for Virology (WVS).

Poster presentation on the title “Hospital-based surveillance of rotavirus diarrhea among under-five children in RIMS, Imphal, Manipur” in ‘VIROCON 2017’ with the theme “Viruses to Viromes in Health and Disease" 2017 at NITTE University, Mangaluru, Karnataka, India.

Poster presentation on the title “Hospital-based surveillance of rotavirus diarrhea among under-five children in RIMS, Imphal, Manipur” in National seminar on “Diarrhoeal Disease Burden and Management: Special Reference to North-eastern India” and Continuing Medical Education on “Infantile Diarrhoea, Its Prevention and Management” organized by the Department of Molecular Biology and Biotechnology, Tezpur University.

Awards

2nd Best Oral Presentation Award in Medical Virology in oral presentation entitled "Exploring rotavirus proteins to design B- and T-cell multi-epitope chimeric antigen using in silico Approach" conferred at the 27Th International Conference of Virology "INTERVIROCON 2018" organized by Indian Virological Society (IVS).

3rd Best Poster Award in Medical Virology on the title "Hospital-based surveillance of rotavirus diarrhoea among under-five children in Imphal, Manipur, India" 28th International conference of Indian Virological Society (IVS) 'VIROCON 2020' on the theme "Evolution of Viruses and Viral Diseases".

AMERICAN SOCIETY FOR MICROBIOLOGY | Microbiology Resource Announcements

GENOME SEQUENCES

Genome Sequence of a Wa-Like G3P[8] Rotavirus from a 12-Month-Old Child with Diarrhea in Manipur, India

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ABSTRACT Rotavirus A (RVA) was detected in the stool of a 12-month-old child with diarrhea, mild fever, and vomiting. A viral metagenomic approach identified a Wa-like genotype G3P[8] strain named RVA/Human-wt/IND/RM25112/2016.

Rotavirus A (RVA) is the major etiologic agent of acute gastroenteritis in young children worldwide. Six structural proteins (VP1 to VP4, VP6, and VP7) and six nonstructural proteins (NSPs) (NSP1 to NSP6) are encoded by the 11-double-stranded RNA (dsRNA) genome (1). The VP7-VP4-VP6-VP1-VP2-VP3-NSP1-NSP2-NSP3-NSP4-NSP5/NSP6 genes of an RVA strain are denoted by the descriptor Gx-P[x]-Ix-Rx-Cx-Mx-Ax-Nx-Tx-Ex-Hx (x represents the genotype number) in the classification system (2). The Wa, DS-1, and AU-1 genotype constellations are described as G1-P[8]-I1-R1-C1-M1-A1-N1-T1-E1-H1, G2-P[4]-I2-R2-C2-M2-A2-N2-T2-E2-H2, and G3-P[9]-I3-R3-C3-M3-A3-N3-T3-E3-H3, respectively (3–6).

The presence of RVA antigen in stool was confirmed using the Premier Rotaclone enzyme-linked immunosorbent assay (ELISA) kit (Meridian Bioscience Inc., USA) and a rapid immunochromatographic test, the Rota+Adeno+Astro+Noro enzyme immunoassay (EIA) combo card kit (CerTest Biotec S.L., Spain). The sample was homogenized by vortex-mixing at room temperature for 5 min, centrifuged at 14,500 rpm for 12 min, and filtered through a 0.22-μm filter. Total RNA was extracted using the TRIzol method (catalog number 15596018; Invitrogen, Carlsbad, CA, USA). cDNA was prepared using SuperScript III reverse transcriptase and random hexamers (Invitrogen). The dsRNA was denatured at 95°C for 5 min. The reverse transcription reaction was carried out with thermal cycling steps at 25°C for 10 min and 55°C for 60 min. VP7 and VP4 nested multiplex PCR and sequencing confirmed the isolate as the G3P[8] genotype (7).

The TruSeq stranded total RNA library preparation kit (catalog number 20020594; Illumina) was utilized to prepare the library for whole-genome sequencing, and final libraries were quantified using a Qubit v.4.0 fluorometer and a Qubit DNA high-sensitivity (HS) assay kit (catalog number Q32851; Thermo Fisher Scientific). Paired-end (2 × 150-bp) sequencing was performed using an Illumina NovaSeq 6000 system. A total of 19.6 million raw reads were generated; after trimming of the adapters and barcodes, 17.4 million retained reads were utilized for the downstream analysis. The quality control of the raw data was carried out using FastQC v.0.11.9. The processed reads were submitted to Genome Detective (8) and CCMetagen (9) servers to determine the taxonomic assignment of the sequences. The cleaned reads were aligned with rotavirus reference genome sequences obtained from NCBI (GenBank accession numbers GCA_003156295.1, GCA_003259085.1, GCF_000864225.1, GCF_000864245.1, GCF_000880735.1, GCF_000890155.1, GCF_000907835.1, GCF_000910335.1, GCF_000973395.3, GCF_001343825.1, GCF_004117615.1, and GCF_013086085.1), and the alignment statistics were measured using Bowtie2 v.2.4.2 (10). The rotavirus-specific reads were extracted using CCMetagen and *de novo* assembled with MEGAHIT v.1.2.9 (11). All software was used with default settings.

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The authors declare a conflict of interest.

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Research article

Exploring rotavirus proteome to identify potential B- and T-cell epitope using computational immunoinformatics

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ABSTRACT

Rotavirus is the most common cause of acute gastroenteritis in infants and children worldwide. The functional correlation of B- and T-cells to long-lasting immunity against rotavirus infection in the literature is limited. In this work, a series of computational immuno-informatics approaches were applied and identified 28 linear B-cells, 26 conformational B-cell, 44 T_C cell and 40 T_H cell binding epitopes for structural and non-structural proteins of rotavirus. Further selection of putative B and T cell epitopes in the multi-epitope vaccine construct was carried out based on immunogenicity, conservancy, allergenicity and the helical content of predicted epitopes. An *in-silico* vaccine constructs was developed using an N-terminal adjuvant (RGD motif) followed by T_C and T_H cell epitopes and B-cell epitope with an appropriate linker. Multi-threading models of multi-epitope vaccine construct with B- and T-cell epitopes were generated and molecular dynamics simulation was performed to determine the stability of designed vaccine. Codon optimized multi-epitope vaccine antigens was expressed and affinity purified using the *E. coli* expression system. Further the T cell epitope presentation assay using the recombinant multi-epitope constructs and the T cell epitope predicted and identified in this study have not been investigated. Multi-epitope vaccine construct encompassing predicted B- and T-cell epitopes may help to generate long-term immune responses against rotavirus. The computational findings reported in this study may provide information in developing epitope-based vaccine and diagnostic assay for rotavirus-led diarrhea in children's.

1. Introduction

Rotavirus is the most common cause of acute gastroenteritis in infants and children worldwide. As per WHO reports of 2013 about 215 000 children under five-years of age die annually due to rotavirus infections mainly in low-income countries [1]. Rotavirus particles naturally excreted in the stools of infected children are transmitted mainly through the fecal-oral route, close-contact and fomites [2]. Rotaviruses are non-enveloped RNA viruses and belongs to the family *Reoviridae*. The mature infectious rotavirus particles is made up of three layers of capsid proteins: outer (proteins VP7 and VP4), middle (protein VP6), and inner (protein VP2). The dsRNA genome of rotavirus encodes for 6 structural proteins and 6 non-structural proteins [3]. Rotavirus infectivity is enhanced by cleavage of VP4 protein into two fragments, VP5* (facilitates cell membrane penetration) and VP8* (mediates cell attachment) [4]. Rotavirus VP4 and VP7 proteins that are commonly used for serotyping are equally important for vaccine development due to development of neutralizing antibodies to VP7, VP8*, and VP5* during natural rotavirus infection [5]. Rotavirus is further divided into nine serogroups (A-I) based on group specific viral antigen VP6 [6].

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