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P504.29 (526)
Identification of Major SARS CoV-2 Variants in India using in silico PCR-RFLP analysis
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Purpose: As of June 30, 2021, WHO reported 181,344,224 confirmed cases worldwide and 3,934,252 deaths due to SARS CoV-2. India accounts almost 10% of the total mortality. The mutated Variants of Concern (VoC) and Variants of Interest (VoI) has acquired non-synonymous amino acid substitutions in Spike (S), ORF1a, ORF1b, Nucleocapsid (N), Membrane (M), Envelope (E), ORF6, ORF7a, ORF3a and ORF8 regions exhibiting more virulence and higher transmission rate. The S gene displaying nucleotide polymorphisms are studied for identification of major SARS CoV-2 variants circulating in India using in silico PCR-RFLP analysis.

Methods & Materials: DNA sequences of major SARS CoV-2 variants [alpha (B.1.1.7), beta (B.1.351), delta (B.1.617.2), epsilon (B.1.429+B.1.427), eta (B.1.525) and zeta (P.2)] prevailing in India retrieved from GISAID database were annotated using VIPR-VIGOR4 genome annotator tool. The S gene of variants were aligned using CLUSTALW algorithm in MEGA-X with default parameters and analyzed for sequence identity using Geneious Prime v2019.2.1. Primers were designed using Primer-BLAST tool, proof-read using FastPCR v6.7.46 and PCR-amplified using Snapgene v3.2.1. Unique restriction sites in S amplicons of each variant were subjected to online Restriction Analyzer tool. The S amplicons were digested with restriction endonucleases and the band profile of each variant was visualized in the gel simulation tool using Snapgene v3.2.1.

Results: The percentage of identical sites present in S region among SARS CoV-2 variants was found to be 98.6%. The amplified products were in the length of 3,689 and 3,698 bp. Out of 400 restriction endonucleases identified, 14 buffer-compatible enzymes were selected for single-step restriction digestion to generate unique RFLP profiles for each variant. The BsaXI-XcmI-Acul triple digest showed unique banding pattern identities on 2% agarose gel for individual variants including alpha, eta, and zeta. The ApalI-BsmI double digest produced distinct band profiles for beta and epsilon variants.

Conclusion: Our study strongly suggests PCR-RFLP analysis of Spike region can differentiate major SARS CoV-2 variants that are circulating in India. Further, quadruple digestion based wet lab experiments are underway to explore the possibility of surveillance of the major variants using a single-tube reaction followed by agarose gel-based profiling.

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P504.30 (734)
Public Search Interests Related to COVID-19: Insights from Google Search Trends in Bangladesh
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Purpose: Public response monitoring is critical to reducing COVID-19 infections and developing effective public health strategies. This study explored Google search trends to understand public responses to COVID-19 concerns in Bangladesh.

Methods & Materials: We used country-level Google search trends data to examine the association between Google search terms related to COVID-19 deaths, face masks, and COVID-19 vaccines and the actual and one-week lagged actual COVID-19 death counts from February 2, 2020, to December 19, 2020, in Bangladesh.

Results: Search terms related to COVID-19 deaths, face masks, and COVID-19 vaccines increased and peaked during March and April, but then began declining gradually after June 2020. The mean relative search volume for face masks (35 points) was higher than for death information (8 points) and vaccines (16 points) throughout the study period, and searching for masks peaked (100 points) during the third week of March. Search interests for death information and face masks were negatively correlated with the actual and one-week lagged actual COVID-19 death counts.

Conclusion: In response to declining trends in COVID-19-related google search terms, policymakers should strengthen ongoing risk communication and preventive information dissemination programs to control and prevent COVID-19 cases and deaths.

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P504.31 (1013)
Detection and Contact Tracing of COVID-19 in Indonesia through a Red Cross Community Based Surveillance System
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Purpose: Epidemics and pandemics begin and end in communities. Communities play a critical role in prevention, detection, and response to infectious disease threats when meaningfully engaged. Surveillance and contact tracing are core components of effective public health preparedness and response efforts. But, surveillance systems that rely on case detection via healthcare facilities may fail or be slow to detect and trace many cases. Communities are often the first to identify unusual disease patterns in humans and animals. Community-Based Surveillance (CBS) gives a voice to communities, enables them to communicate unusual health events to authorities, and complements facility-based surveillance systems.

Methods & Materials: Since 2019, Indonesia Red Cross (PMI) has piloted CBS in 8 villages within four provinces. Initiated for passive surveillance, the volunteers trained to report ‘alerts’ of epidemic-prone diseases to health authorities. The concept was then adapted to an active approach focussing on COVID-19 and scaled to 74 villages across eight provinces. Partnering with local authorities, volunteers supported COVID-19 screening, detection, tracing, and local response, all while implementing volunteer safety standards. The reporting system was created and coordinated by a structure within PMI.

Results: From August 2020-June 2021, community volunteers made 1,079 alerts: 99% of the alerts were reported to health authorities, with 71.1% followed up within 24 hours. After alert detection, volunteers sensitized family members on preventive measures while individuals awaited follow-up visits for diagnosis and management by health authorities. 64% of alerts were PCR positive for COVID-19, 12% were negative, 14% did not undergo testing, and test results for 10% of the alerts were unavailable. Of 979 contacts traced by volunteers, authorities followed up 99%, confirming
29% positive and 34% negative for COVID-19; the remaining 37% included those recommended to isolate without testing and those for whom test results were unavailable.

**Conclusion:** CBS has supported early detection and contact tracing of COVID-19 in Indonesia. The community case definition used to identify potential alerts was sensitive and easily understood by non-health workers. The system proved readily scalable and adaptable, enabling a shift from passive to active surveillance and making it feasible to be implemented anywhere in the community.

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**Topic 05: COVID-19 Diagnostics and Therapeutics**

**OP05.01 (564)**

**Colorimetric reverse transcriptional loop-mediated isothermal amplification for rapid detection of SARS-CoV-2**

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**Purpose:** With the increasing incidence of a novel coronavirus SARS-CoV-2 causing COVID-19 cases, accurate and early detection infection is need of the hour for effective prevention and management. Therefore, the aim of this study was to develop a colorimetric reverse transcriptional loop–mediated isothermal amplification for rapid detection of SARS-CoV-2.

**Methods & Materials:** Inactivated SARS-CoV-2 virus samples were procured from the National Institute of Virology, Pune, India. Various genes were targeted for primer design, such as nucleoapсид, spike, RNA dependent RNA polymerase, and envelope genes region of SARS-CoV-2. In-vitro synthesised viral RNA was used for the standardisation of the RT-LAMP. RT-LAMP products were visualised by the naked eye using hydroxy naphthol blue dye. The sensitivity of RT-LAMP assay was performed by diluting in-vitro synthesised viral RNA at a different concentration such as 5 ng/μL, 25 ng/μL, 50 ng/μL, 200 ng/μL. Additionally, the RNA copy number was estimated and tested with RT-LAMP. *In-silico* analysis was carried to calculating the percentage of mismatch using various viral sequences, including SARS-CoV-2, other coronaviruses, and other related RNA virus sequences available at GenBank.

**Results:** RT-LAMP assay was standardised using in-vitro synthesised viral RNA. Temperature and time standardisation revealed, all the targets i.e., E, S, N, and RdRp gene regions had an optimum temperature of 63°C and time, 60 min. The sensitivity of all the target genes were ten copies of viral RNA. RT-LAMP amplified products were visualised by the naked eye using hydroxy naphthol blue dye and verified by agarose gel electrophoresis. All the primers used for the RT-LAMP assay showed a zero percent mismatch with SARS-CoV-2 sequences available at GenBank.

**Conclusion:** Colorimetric reverse transcriptional loop–mediated isothermal amplification assay developed in this study could provide a visual and faster alternative to the RT-qPCR assays.

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**OP05.02 (711)**

**COVID-19 in People Living with HIV**

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**Purpose:** The current COVID-19 pandemic affects all strata of the population. Particular interest is the study of the course of this pathology in people with comorbidity.

The purpose of our research is to study the features of the course of a COVID-19 people living with HIV (PLHIV).

**Methods & Materials:** We conducted a retrospective analysis of the epidemiological, clinical and laboratory data of 121 patients with HIV infection treated for COVID-19 in 2020. Among PLHIVS, 87.6% were treated with antiretroviral therapy for HIV infection an average of 5.8 years.

**Results:** It was found that 45.5% were women and 54.5% were men. The average age was 41 years old (IQR: 20–78 years). The greatest number of cases was observed among both men and women in the age category of 30–49 years (74.2% and 72.7%, respectively). In the studied group, in 63.4%, in addition to HIV infection, patients suffered from other comorbid diseases, the most frequent of which were chronic lung diseases (22.3%), hypertension (18.2%), metabolic disorders (13.2%). In all cases, the COVID-19 disease occurred with a clinical manifestation. The most common symptoms were fever (76.0%), cough (63.6%) and sore throat (56.2%), loss of taste and smell was detected in 49.6% of cases. In 48.8%, COVID-19 in PLHIV was mild. In 88.1%, the duration of the disease did not exceed 14 days. The most common symptoms were fever (76.0%), cough (63.6%) and sore throat (56.2%), loss of taste and smell was detected in 49.6% of cases. In 48.8%, COVID-19 in PLHIV was mild. In 88.1%, the duration of the disease did not exceed 14 days. Moderate forms were noted in 40.5% of cases. 10.7% of cases have severe form of COVID-19. The fatal outcomes were recorded in 8 patients, while in the group of patients receiving antiretroviral therapy, the mortality rate was 3.8%, and in the group without antiretroviral therapy - 26.7%.

**Conclusion:** We estimated that the proportion of deaths in patients with HIV infection is higher than in the general population (2.2%). PLHIV are a vulnerable group in relation to the risk of death from COVID-19.

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**OP05.03 (55)**

**Assessment of High-Titer Convalescent Plasma as an Adjunctive Therapy in the Reduction of Mortality Rate and Viral Load in Patients with Severe COVID-19: A Meta-Analysis**

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