

“An unprecedented saga of CUK
in societal service: From Dengue
virus research to COVID19
RTPCR testing for the public”

**MOLECULAR
VIROLOGY LAB**

COVID19 RTPCR TESTING
(Approved by ICMR, Govt of India,
March 2020)

Department of Biochemistry
&
Molecular Biology



**CENTRAL UNIVERSITY
OF KERALA (CUK)**



Objectives

- 1. To provide the high end molecular diagnosis for SARSCoV2 infection using RTPCR in a resource poor neighborhood of CUK.*
- 2. To obtain the timely diagnosis report so as to support the community and health authorities of the state in curbing the spread of pandemic*
- 3. To train the manpower to conduct the COVID19 RTPCR testing in the molecular virology lab of CUK approved by the ICMR.*
- 4. To provide diagnostic services with special emphasis on early detection of viral pathogen thereby supporting the healthcare system both at state as well as national level for the prevention and control.*
- 5. To conduct advanced research on viral genomic surveillance in collaboration with Dept of Health and Family Welfare, Govt of Kerala.*
- 6. To promote research in the area of viral diagnostics and antiviral drug development.*

“CUK- Committed for the educational upliftment and societal service”

Molecular Virology lab of CUK came into functioning on 28th Mar 2020 as the 8th COVID19 RTPCR testing lab in the Kerala State and the first of its kind in the university settings at national level, with the approval of ICMR.



**SUCCESSFULLY COMPLETED MORE THAN
2.1 LAKHS TESTS**

A concerted effort of the Dept of Biochemistry and Molecular Biology (BCMB), CUK for establishing Molecular Virology lab

➤ Dept of BCMB was instituted in 2010 as one of the first academic departments of CUK, under the Headship of eminent Biochemist, Prof P R Sudhakaran

➤ The department was functioning in the transit campus till 2018, however, the department began its research program during 2011 upon the recruitment of three faculty members, 1) Dr Sameer Kumar VB,

2) Dr Rajendra Pilankatta and 3) Dr Santosh R Kanade

➤ Three important research labs were initiated, focusing on Molecular Virology (Dr Rajendra Pilankatta), Angiogenesis (Dr Sameer Kumar VB) and Epigenetics (Dr Santosh R Kanade), involving advanced molecular biology as a practical tool. This led to the training of research scholars and master's students in the modern area of biology.

➤ The research and teaching strength was further augmented by the joining Prof D Govinda Rao (Immunology), Dr Aswati Nair (Plant Biochemistry) and Dr Thejaswini Venkatesh (Cancer Biology) in the year 2016.

➤ The Molecular Virology lab under the leadership of Dr Rajendra Pilankatta initiated basic research on Dengue Viruses and Adenoviruses in collaboration with the various research branches of the department and outside institutes. Following are the major focus of Molecular Virology research, 1) Virus host cell interactions, 2) Replication machinery of Dengue virus, 3) Antiviral drug screening, 4) Seroprevalence of Dengue viruses and Adenoviruses & its implications and 5) Antibody – Dependent Enhancement (ADE) of viral infections.

➤ Importantly, Dr MN Reddy (Former graduate student) and team explored the Occurrence

of concurrent infections with multiple serotypes of dengue viruses during 2013–2015 in the locale. This study established the standard molecular biology platform in the molecular virology laboratory for detecting the RNA virus infection in the human clinical samples using Nested RTPCR method. The study showed the existence of all four serotypes in the locale which is a great public health concern. The finding has been published in the leading international journal, Peer J with 39 citations yet¹.



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- As a continuation of this study, it was discovered by the lab that individuals with concurrent infection of multi-serotype of Dengue virus induces the enhancement of ApoA1 (Lipoprotein) in the blood, which is found to be involved in the enhanced infection of viruses into the cells in the human host. The findings have been published in the international peer reviewed journal, Tropical Medicine and International Health².
 - Similarly, the study on virus host cell interaction lead to the finding that the dengue virus infection upregulates several genes in the cell which is involved in the cell survival pathway known as Autophagy³. The mechanism can be targeted for the drug development against dengue virus. In continuation of this study, a novel nano-platform using ZnS QDs was established for the antiviral drug delivery impacting in the reduction of cytotoxicity⁴ in collaboration with Dr Swapna Nair, Dept of Physics.
 - Dr Rajendra Pilankatta received prestigious CV Raman Fellowship for conducting postdoctoral studies in the area of Dengue virus under the guidance of eminent virologist, Prof R Padmanabhan at Georgetown University, USA during 2014 – 2015. This visit had helped the department to build more strength in the area of molecular virology along with the publication of two important articles in the area of antiviral drug development against Dengue viruses as well as other related flaviviruses, targeting the viral protease⁵. The work also resulted in the identification of many curcuminoids as anti-dengue virus compounds⁶. The drug screening platform developed during this visit is being used by many PhD Scholars and Masters students in the laboratory for training and research purposes.
 - Meanwhile, the seroprevalence study on adenoviruses has resulted in the publication of a very novel method for the estimation of antibody dependent enhancement of virus entry into the cells⁷. The platform is currently being used for drug screening. Further, novel antiviral drugs that are targeting the replication complex bound proteins are being explored in the lab. Dr Rajendra Pilankatta worked as a visiting Associate Professor in collaboration with eminent Virologist, Prof Subhash Vasudevan, at DUKE NUS, Singapore in the area of isolation of Dengue viruses from clinical samples.
 - In the year 2018, the lab was shifted to permanent campus, Thejaswini Hills at Periyar along with the department. Thus, Molecular Virology Lab of CUK had been continuously working on both clinical as well as basic virological research questions since last one decade. As a recognition for the department as well as university, Dr Rajendra Pilankatta, was invited as expert committee member for the establishment of Institute of Advanced Virology (IAV) at Trivandrum by Govt of Kerala. Currently, he is also serving as Adjunct faculty at IAV, Trivandrum



Molecular Virology lab of CUK came into functioning as the 8th COVID19 RTPCR testing lab in the Kerala State and the first of its kind in the university settings at national level on 28th Mar 2020, with the approval of ICMR

COVID-19 pandemic is becoming a looming public health threat. Slowly, the human population is getting adjusted to live with the situation by careful approach following the COVID-19 preventive protocol, which has been shaped us during the year 2020. For any viral outbreak, the early diagnosis and

implementing the preventive measures becomes the ‘mantra’ to contain the spread of disease into the community. COVID-19 disease caused by SARSCoV2 infection became a toughest viral disease in the human history, to tackle due to its rapid pandemic spread. The Real Time Polymerase Chain Reaction (RT-PCR) based detection of genetic material of SARSCoV2 became the global technique for the detection of the early infection with more than 95% sensitivity, still it’s a gold standard for the diagnosis of SARSCoV2, even though several kind of alternative tool came into picture.

“RT-PCR testing for the SARSCoV2 infection in Kasaragod District!”, became a question in front of the health authorities of Govt of Kerala as well as public of the district, as it belongs to an historically backward region of northernmost Kerala. The testing demands a high end molecular biology lab with virus handling biosafety facility and above all highly skilled biochemist and molecular biologist. During Jan and Feb 2020, the district health authorities used to send samples to Pune or Alleppey virology lab and used to wait for weeks long to get the results, meanwhile, the virus was spreading every minute through mutual contact of the individuals. In this context, the Molecular Virology Lab of the Department of BCMB, CUK under the leadership of Dr Rajendra Pilankatta, teamed up with the Dr VB Sameer Kumar, an established cancer biologist along with the senior research scholars Mr Ranjeet, Ms Lathika, Mr Prajit and Mr Vishnu and Mr Ratheesh as technical staff to initiate the RT-PCR based SARSCoV2 testing on 28 th Mar 2020. ICMR gave the approval for the same as per the recommendation of Hon Vice Chancellor, CUK and Dept. of Health, Govt of Kerala for conducting the test. CUK was the first university in India to set up the SARSCoV2 RT-PCR testing eventhough it is only a decade old university, thereby CUK became a

model in the country by serving the community during the pandemic outbreak of deadly virus, SARSCoV2.

The team got expanded later on by the joining of four alumni researchers, Dr Sharath Shankar, Dr. Lincy Edatt, Dr. Prabitha Mohan and Dr. Sruthi M V. Further strength to the team was contributed by the joining of senior PhD Scholars, Ms Rabina, Ms Aswathi, Mr Rajesh, Mr Manoj, Mr Ashuthosh, Ms Anjali and Central University technical staff Mr Rajesh. Also, the team was fortunate obtain great support from the District Collector, Dr Sajith Babu, District Programme Manager, Dr Raman, District Medical Officer, Dr Ramdas, District Surveillance Officer, Dr Manoj in terms of providing NHM staffs as Lab Technician and Data entry operator. Engineering wing, CUK gave constant support and help for developing and maintaining lab facilities for conducting the COVID-19 RTPCR testing. Ambulance drivers Mr Praveen, Mr Vivekanandhan and Mr Sharath, Security staffs, electric staffs and cleaning staffs contributed a selfless service towards the same.

CUK as well as the testing team was highly fortunate to have Prof H Venkateswarlu as Vice Chancellor during the peak COVID-19 pandemic in Kerala, which accelerated the activity of the lab significantly. The lab provided an unprecedented support to the entire community of Kasaragod District as well as entire Kerala state during the COVID-19 pandemic and the process is being continued. Presently, the lab has completed more than 2.1 lakhs tests. The lab had initiated the testing with an average of 100 testing per day and currently the lab tests 2000 tests per day by working 3 shifts a day. The augmentation in terms of number of tests was due to the addition of new RTPCR machine (2 Nos) as well as automated RNA extractor (Thermo KingFisher) which was funded by the NHM and Dept of Health, Govt of Kerala. The



**Automated viral
RNA extractor**

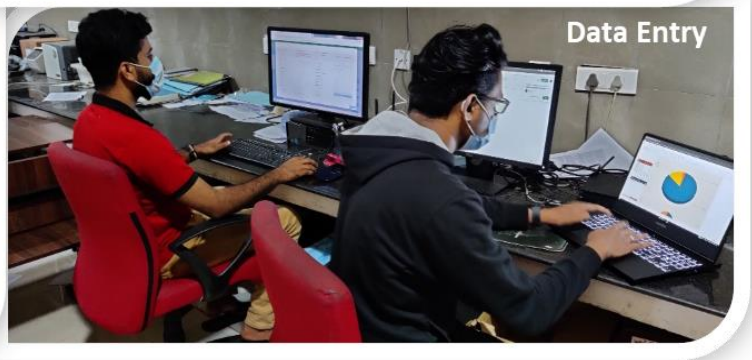
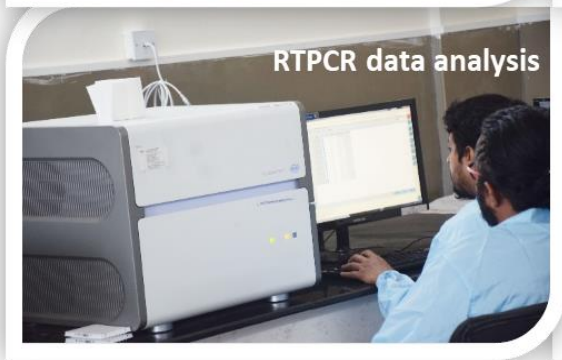
facility was further augmented by the addition of one more RTPCR machine and RNA extractor funded by UNICEF through ICMR, Govt of India.

Recently, the lab has entered into collaboration with the IGIB, New Delhi along with the Dept of Health, Govt of Kerala for the genetic surveillance of the SARSCOV2 wherein the whole viral genome is being sequenced for identifying the mutation in the SARSCOV2 in the locale. In this connection the lab has completed the sequencing of 1500 RNA samples in collaboration with IGIB, New Delhi. The northern most region of Kerala, Kasaragod suffers from serious lacunae of medical facilities. The services of COVID- 19 testing facility of CUK significantly decreased the time lag for obtaining the test results and hence augmented the quarantine procedure, which was instrumental in the controlling the spread of the disease in the locale.

In line with the expertise available in the field of virology and molecular biology, BIRAC, DBT, Govt of India grant was also awarded in Collaboration with MagGenome Technologies Pvt. Ltd. to develop a less invasive diagnostic tool to detect the presence of SARS-COV2 in saliva of the patient instead more invasive throat and nasal swab, and the same has been approved by ICMR. Antibody based SARSCOV2 diagnosis kit has been developed in collaboration with SCTIMST, Trivandrum and the same has been published in the international peer reviewed journal⁸. Further, research work was conducted in collaboration with the District Health authorities regarding the immunological aspects of SARSCOV2 re-infection, which has been published in the reputed Public Health Journal⁹. A manuscript is being prepared regarding the genomic surveillance of SARSCOV2, specifically on vaccine breakthrough infections in collaboration with IGIB, New Delhi. Currently, CUK is in the process of strengthening the existing COVID-19 RTPCR diagnostic laboratory by establishing a separate Virology lab within the campus by the support of National Health Mission and the Dept of Health, Govt of Kerala. The laboratory will cater the needs of the community by providing advanced molecular diagnosis for the emerging and re-emerging viral diseases apart from the ongoing COVID-19 diagnosis. In this regard, Hon'ble Vice chancellor of CUK Prof (Dr.) H. Venkateshwarlu has met Hon'ble Minister for Health and Social Justice, Smt. K K Shylaja and the Revenue Minister, Sri E Chandrasekharan on 29th Sept 2020 and decided to accelerate the process especially based on the current situation of increased rate of SARSCOV2 infection. An MoU was executed earlier in this regard between CUK and the Dept of Health, Govt of Kerala. Molecular virology lab in the CUK campus will be a milestone in the development of

Ksaragod District and aims to continue the service for the mankind beyond the geographical boundary.

COVID19 RTPCR TESTING INFRASTRUCTURE AT MOLECULAR VIROLOGY LABORATORY, BCMB, CUK



COVID19 RTPCR TESTING TEAM, MOLECULAR VIROLOGY LAB, BCMB, CUK



EXISTING MANPOWER

SL NO	NAME	DESIGNATION
1.	Dr RAJENDRA P	COVID 19 Lab in charge
2.	Dr SAMEER KUMAR V B	Quality Assessor
3.	ARATHI M	Lab Technician (NHM)
4.	SUNESH KUMAR KS	Lab Technician (NHM)
5.	VEENA	Lab Technician (NHM)
6.	JITHIN RAJ V	Lab Assistant (NHM)
7.	SHAHUL HAMEED SIMSAR	Lab Assistant (NHM)
8.	NIKHIL RAJ P	Data Entry Operator(NHM)
9.	SACHIN MP	Data Entry Operator(NHM)
10.	MUHAMMED RIZWAN AM	Data Entry Operator(NHM)
11.	MITHUN	Data Entry Operator(NHM)
12.	AISHWARYA P	Lab Technician(DMO)
13.	AL AKEEF	Lab Technician(DMO)
14.	ABHIJITH	Lab Technician(DMO)
15.	ADARSH	Lab Technician(DMO)
16.	PRAJIT J	Research Scholar
17.	VISHNU R	Research Scholar
18.	ASWATHI KS	Research Scholar
19.	RANJEET	Research Scholar
20.	RAJESH TP	Research Scholar
21.	MANOJ K	Research Scholar
22.	ANJALI J B	Research Scholar
23.	ASHUTOSH	Research Scholar
24.	RANJISHA	Research Scholar
25.	ANJALI KUNHIKRISHNAN	Research Scholar

MANPOWER TRAINED

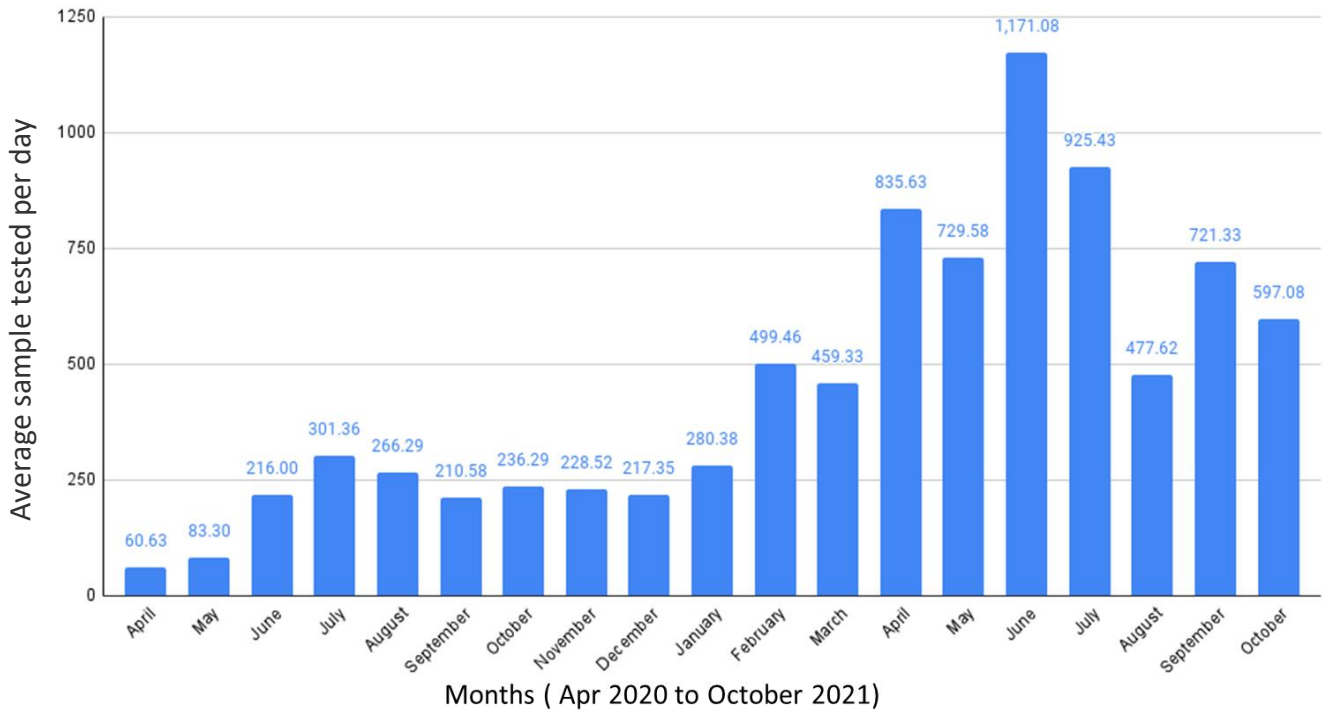
SL NO	NAME	CURRENT DESIGNATION	Working period	
			FROM	TO
1.	Dr. PRABHITHA MOHAN	Postdoctoral Fellow	01-05-20	05-09-20
2.	RAJESH R	Technical Assistant, BCMB, CUK	09-04-20	31-05-20
3.	RABINA P	Research Scholar, BCMB, CUK	10-05-20	31-08-20
4.	Dr. LINCY EDATT	Postdoctoral Fellow, USA	10-05-20	18-08-20
5.	LATHIKA	Research Scholar, BCMB, CUK	02-04-20	03-05-21
6.	Dr SHARATH SHANKAR	Postdoctoral Fellow, USA	01-07-20	10-09-20
7.	Dr SHAFEEQ HASSAN	Lab in Charge, Wayanad Dist	03-04-20	14-04-20
8.	KAVITHA KARUNAKARAN	Lab Technician (PSC)	01-04-20	09-04-20
9.	RATHEESH U	Lab Assistant (CUK)	02-04-20	30-07-20
10.	MUHAMMED IFRAN A I	Lab Technician (PSC)	01-04-20	11-04-20
11.	ABDUL MUEEZ PA	Lab Technician (PSC)	01-04-20	09-04-20
12.	AMALRAJ	Lab Technician (PSC)	10-04-20	27-10-20
13.	ASWIN PK	Lab Technician (PSC)	10-04-20	27-10-20
14.	SANIL KUMAR V	Lab Technician (PSC)	15-04-20	27-10-20
15.	AMALJITH	Lab Technician (PSC)	30-06-20	23-06-21
16.	BABY PRATHAP B	Lab Technician (PSC)	13-07-20	20-12-21
17.	KSHITHY M	Lab Technician (PSC)	07-05-20	28-05-20
18.	MANEESH MOHAN MD	Lab Technician (NHM)	07-05-20	15-07-20
19.	ASWATHI CM	Lab Technician at PHC	04-05-20	30-05-20
20.	ROOPESH K	Lab Technician at UAE	10-07-20	28-09-21
21.	SHAFEEQ	Lab Technician at UAE	10-05-21	07-10-21
22.	KRIJITH MV	Lab Technician , Pondicherry	04-05-20	18-09-21
23.	SUFAID MC	Lab Technician at UAE	04-05-20	18-04-21



MANPOWER TRAINED

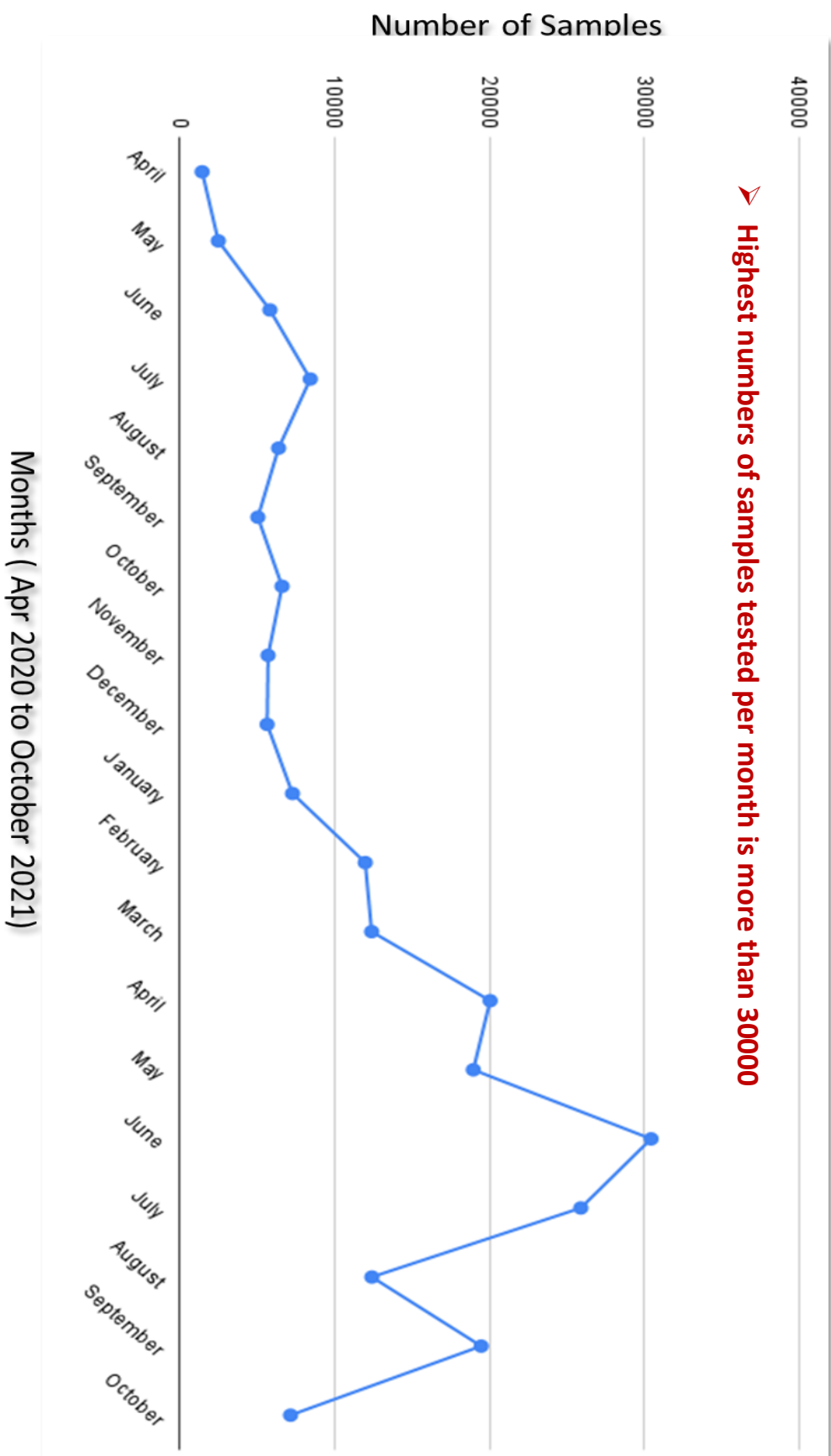
SL NO	NAME	CURRENT DESIGNATION	Working period	
			FROM	TO
24.	ROSHNA REMESAN	Lab Technician at Nileshwar	01-08-20	14-10-21
25.	ABDUL RAZAK	Staff Nurse at Mangalore	27-06-20	26-11-20
26.	ABHILASH	Lab Technician (NHM)	10-07-20	30-07-20
27.	NEETHU M	Lab Technician (PSC)	07-05-20	28-05-20
28.	ANAGHA	Lab Technician DH Kanhangad	09-08-21	25-08-21
29.	DARSHANA	Lab Technician DH Kanhangad	09-08-21	30-09-21
30.	ARPITHA YADHAV K	Data Entry Operator(E-Health)	10-08-20	31-12-20
31.	VARSHA AK	Data Entry Operator(E-Health)	25-08-20	23-01-21
32.	SUKANYA N	Data Entry Operator(E-Health)	12-09-20	28-09-20

**GRAPH SHOWING AVERAGE SAMPLES
TESTED PER DAY IN EACH MONTH**

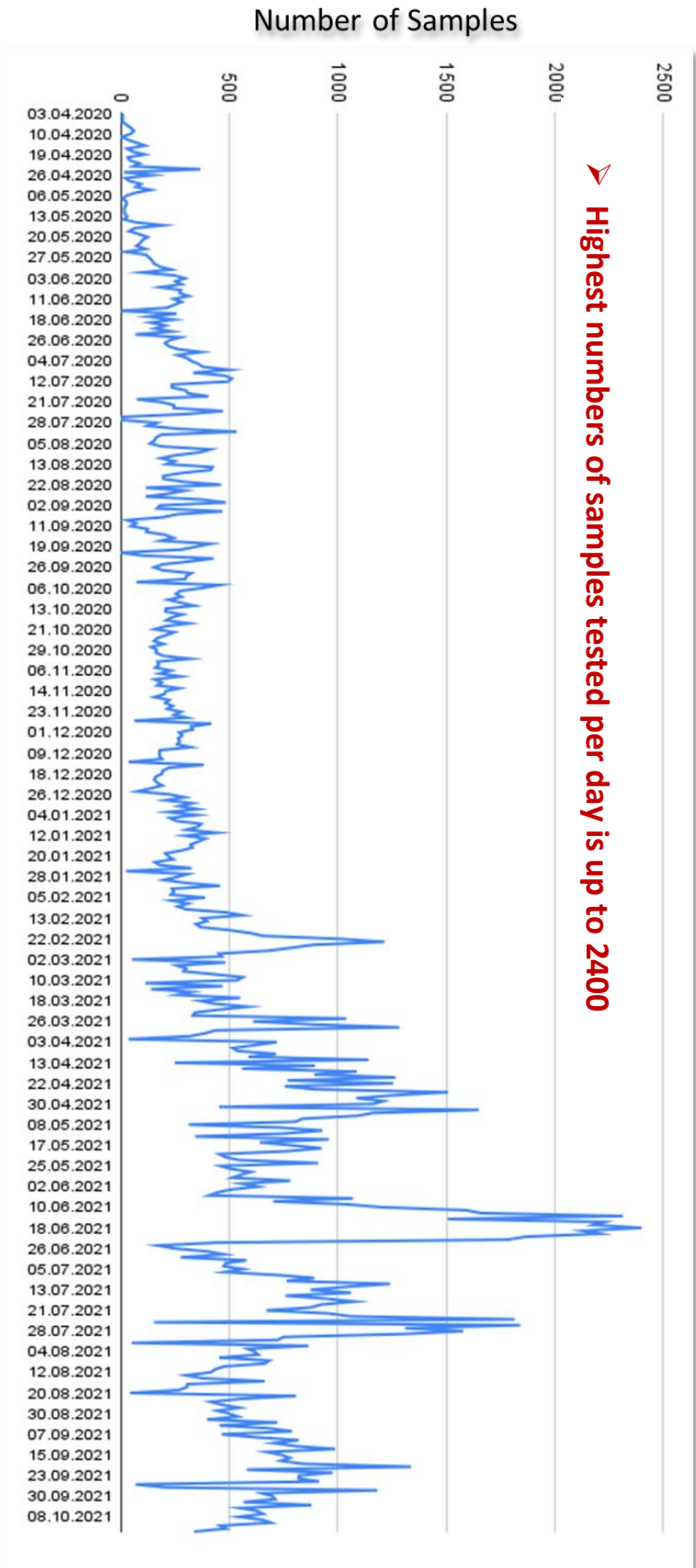


➤ **Average samples tested per day varies from 60 to 1171**

GRAPH SHOWING TOTAL NUMBER OF SAMPLES TESTED PER MONTH

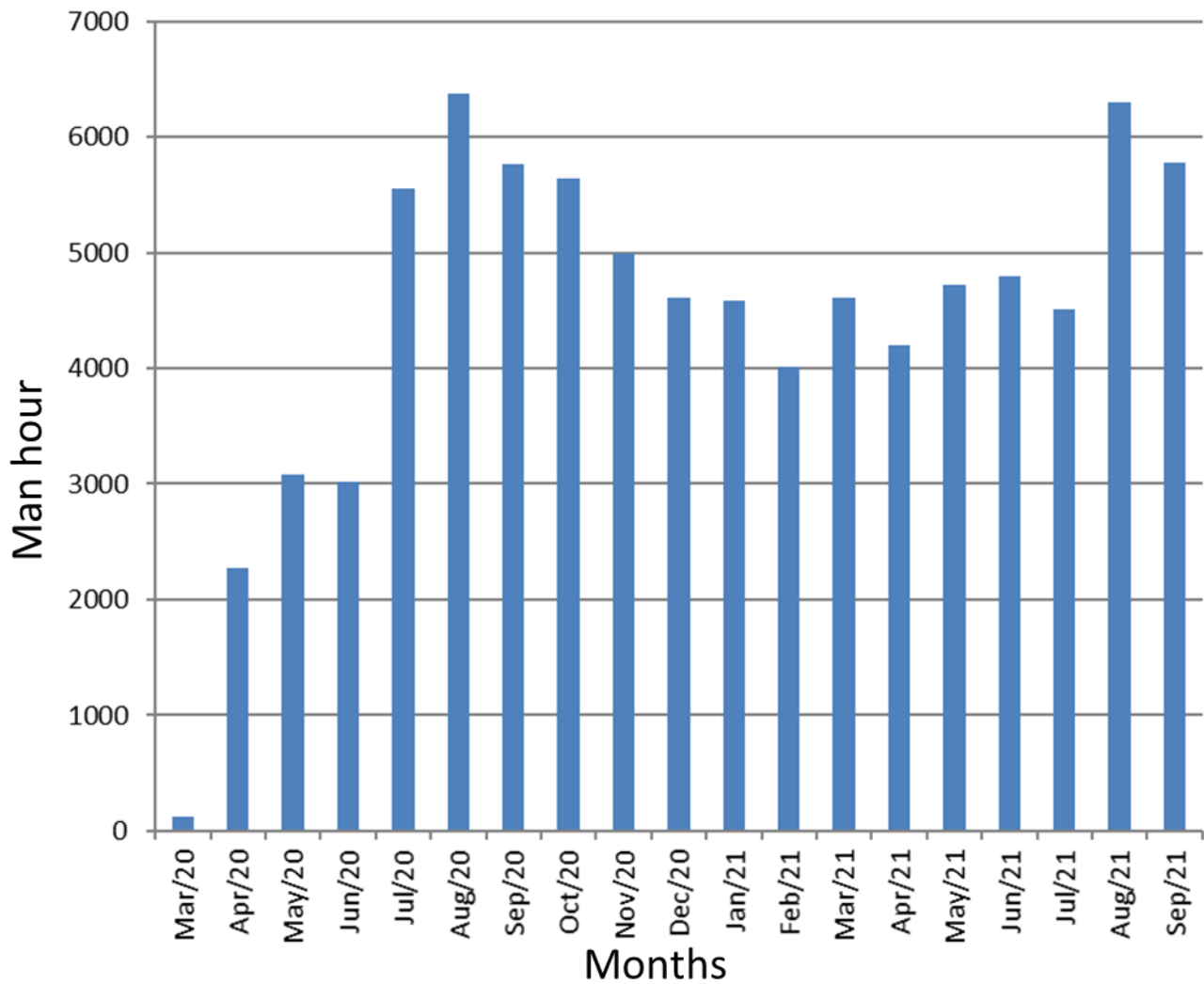


GRAPH SHOWING DAILY SAMPLE S TESTED



Days (Apr 2020 to October 2021)

GRAPH SHOWING MONTHLY MAN HOUR SPENT FOR TESTING



➤ **Monthly man hour spent for testing goes upto more than 6000**

RESEARCH AND DEVELOPMENT PROJECT UNDERTAKEN BY MOLECULAR VIROLOGY LAB, CUK IN THE AREA OF COVID19 AND OTHER VIRAL DISEASES OF PUBLIC HEALTH IMPORTANCE

PARTICIPATED IN THE GENOMIC SURVEILLANCE OF SARSCOV2 IN COLLABORATION WITH IGIB, NEW DELHI AND DEPT OF HEALTH AND FAMILY WELFARE, GOVT OF KERALA

#GENESCOV2

Genetic Epidemiology of SARS-CoV-2 in Kerala

A unique partnership between the Department of Health & Family Welfare, Government of Kerala, National Health Mission Kerala, CSIR Institute of Genomics & Integrative Biology (CSIR-IGIB) and various clinical and public health centers in Kerala aimed at providing insights into the genetic epidemiology of COVID-19 in Kerala

An initiative of Public Health relevance involving 12 Govt Medical Colleges, State Public Health & Regional Laboratories, Inter-University Centre for Biomedical Research, Central University of Kerala & 14 District Surveillance Units



Department of Health & Family Welfare
Govt of Kerala



National Health Mission
Kerala



CSIR Institute of
Genomics & Integrative
Biology (CSIR-IGIB)

Know More

[Updates on the programme and Interim Reports on the analysis](#)

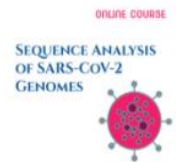
7500+ SARS-CoV-2 Genomes Sequenced
(Updated 30-08-2021)



Phylogenetic Map for
SARS-COV-2 Data



Science Blog



Online Course
2021

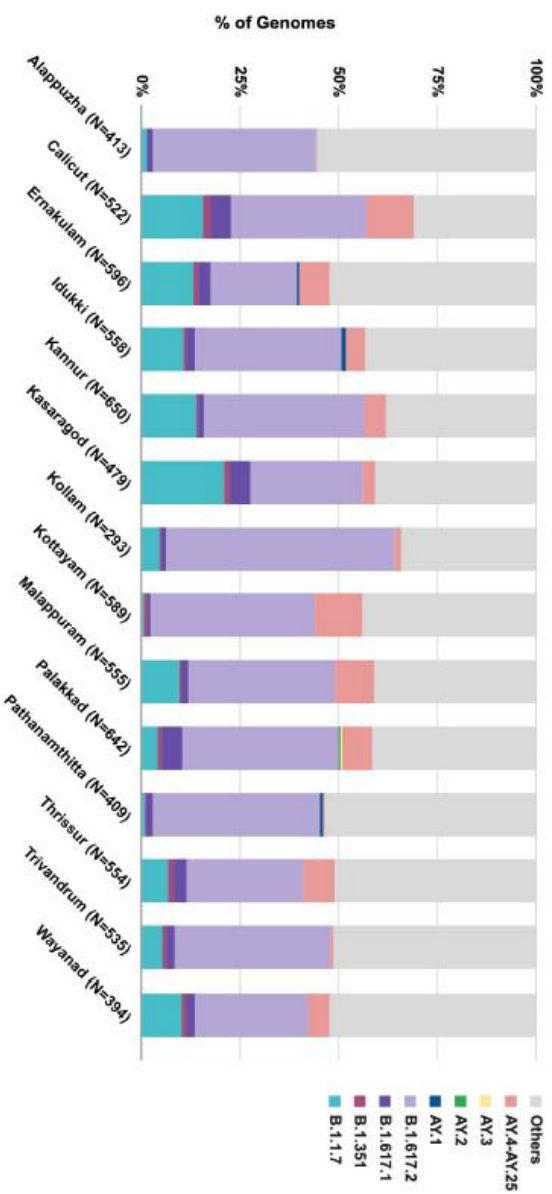
➤ **7500+ virus samples were sequenced statewide under this project**



GENOMIC SURVEILLANCE OF SARS-CoV-2: The data corresponding to Kasaragod District was supported by CUK

Last Updated : 29/08/2021

State-wide proportion of VOCs and Vols
(all data until date)



Genetic Epidemiology of SARS-CoV-2 in Kerala is a partnership between the Department of Health & Family Welfare, Government of Kerala, National Health Mission Kerala, CSIR Institute of Genomics & Integrative Biology (CSIR-IGIB) and funded by NHM-Kerala and CSIR India. More at: <https://genescov2.genomes.in>



**MOLECULAR VIROLOGY
LAB, CUKERALA
COLLABORATED WITH
MAGGENOME Pvt Ltd
FOR DEVELOPING
SALIVA BASED RTPCR
KIT FOR COVID 19
DIAGNOSIS FUNDED BY
BIRAC, DBT, GOVT OF
INDIA**



Biotechnology Industry Research Assistance Council
(A Government of India Enterprise)

To,

Date: 18th August, 2020

Dr. Tessa Iype
Principal Scientist
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And

Dr Padma Srikanth
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And

Rajendra Pilankatta
Associate Professor & HOD
Dept. of Biochemistry and Molecular Biology
Central University of Kerala
Email: praj74@gmail.com
Mob.: 9061877113

Ref: Project Title " Development of a diagnostic kit comprising sample collection buffer and RNA extraction kit for real time RT PCR based detection of SARS CoV-2." under strategic funding for Covid-19 Consortium.

Dear Dr. Iype, Dr. Srikanth and Dr. Pilankatta,

With reference to your above, mentioned proposal, we are pleased to inform that your proposal has been approved by BIRAC at a total cost of **Rs. 91.00 Lakhs (Rupees ninety one Lakhs only)** under strategic funding for Covid-19 Consortium with support of **Rs. 47.00 Lakhs (Rupees forty seven Lakhs only)** as Grant-in-aid to MagGenome Technologies Pvt. Ltd., support of **Rs. 15.00 lakhs (Rupees fifteen lakhs only)** as Grant-in-aid to Sri Ramachandra Institute of Higher Education and Research and support of **Rs. 19.00 lakhs (Rupees nineteen lakhs only)** as Grant-in-aid to Central University of Kerala. The amount put in by MagGenome Technologies Pvt. Ltd. is **Rs. 10.00 Lakhs (Rupees ten Lakhs only)** towards the Project for **8 months** as per the terms of the Grant in Aid Letter Agreement (GLA) enclosed.

If you are in acceptance with the terms of funding support as mentioned in the GLA, the following documents should be executed within a time period of **TWO WEEKS** from the date of issue of this communication.

- **Grant-in-aid Letter Agreement (GLA)** including all the Schedules: All pages to be clearly initialled by the Fund Recipient and the Company should affix the organisation official seal with Director Signatures on the signature page to the GLA. *Considering the current lockdown situation, Signatures of Competent Authority may be done and the GLA executed. The official seal could be affixed after your office resumes.*

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CIN No. : U73100DL2012NPL233152

SI No	Title	Funding agency	Duration	PI /Co-PI	Amount (Rs)
1	Seroprevalence Of Adenovirus serotype 5 and its implication in adenovirus vector based gene delivery	Kerala Biotechnology Commission, KSCSTE, Govt of Kerala	2014 to 2016	PI: Dr Rajendra Pilankatta	16.0
2	Role of dengue viral proteins in autophagy induction	DST – SERB , Govt of India	2014 to 2018	PI: Dr Rajendra Pilankatta	25.0
3	Role of copper-handling proteins in cisplatin drug resistance	DBT- Govt of India	2015 to 2019	PI: Dr Rajendra Pilankatta	25.0
4	Molecular docking and 3-D QSAR studies to evaluate the mechanism of antimicrobial action of fatty acids from virgin coconut oil and monolaurin	Kerala Biotechnology Commission, KSCSTE, Govt of Kerala	2016 to 2018	Co-PI: Dr Rajendra Pilankatta	14.0
5	Cutaneous microbiome profiling of Psoriasis and Elephantiasis using metagenomics	Kerala Biotechnology Commission, KSCSTE, Govt of Kerala	2017 to Ongoing	Co-PI: Dr Rajendra Pilankatta	25.3
6	Development of geometry and interface engineered lead free biocompatible magneto electric nanorod arrays for next generation self powered sensors, energy harvesters and futuristic implantable devices	DST –SERB (CRG), Govt of India	2020-Ongoing	Co-PI: Dr Rajendra Pilankatta PI: Dr Swapna S Nair, Dept of Physics	74.0

7	Targeting the host factor for inhibiting the secretion of Dengue virus NS1 lipoprotein complex: A novel strategy for anti-dengue viral drug development	ICMR – Govt of India	2021 (Sanctioned)	PI: Dr Rajendra Pilankatta	28.1
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**TABLE: LIST OF PROJECTS AWARDED TO MOLECULAR VIROLOGY LAB,
BCMB, CUK**

MOLECULAR VIROLOGY LAB, BCMB, CUK FACILITATED VACCINATION CAMPAIGN IN THE CAMPUS SINCE MARCH 2021, IN COLLABORATION DISTRICT ADMINISTRATION AND HEALTH CENTER OF CUK



➤ **COMPLETED 800 DOSES OF VACCINATION AS ON 22ND OCT, 2021**

PUBLICATIONS FROM THE MOLECULAR VIROLOGY LAB, CUK IN THE AREA OF DENGUE VIRUSES, ADENOVIRUSES AND COVID19 (Cited References)

1. Reddy MN, Dungdung R, Valliyott L, Pilankatta R. Occurrence of concurrent infections with multiple serotypes of dengue viruses during 2013-2015 in northern Kerala, India. PeerJ. 2017 Mar 14;5:e2970. doi: 10.7717/peerj.2970. eCollection 2017.



Occurrence of concurrent infections with multiple serotypes of dengue viruses during 2013–2015 in northern Kerala, India

Manchala Nageswar Reddy, Ranjeet Dungdung, Lathika Valliyott and Rajendra Pilankatta

Department of Biochemistry and Molecular Biology, Central University of Kerala, Kasargod, Kerala, India

ABSTRACT

Background. Dengue is a global human public health threat, causing severe morbidity and mortality. The occurrence of sequential infection by more than one serotype of dengue virus (DENV) is a major contributing factor for the induction of Dengue Hemorrhagic Fever (DHF) and Dengue Shock Syndrome (DSS), two major medical conditions caused by DENV infection. However, there is no specific drug or vaccine available against dengue infection. There are reports indicating the increased incidence of concurrent infection of dengue in several tropical and subtropical regions. Recently, increasing number of DHF and DSS cases were reported in India indicating potential enhancement of concurrent DENV infections. Therefore, accurate determination of the occurrence of DENV serotype co-infections needs to be conducted in various DENV prone parts of India. In this context, the present study was conducted to analyse the magnitude of concurrent infection in northern Kerala, a southwest state of India, during three consecutive years from 2013 to 2015.

Methods. A total of 120 serum samples were collected from the suspected dengue patients. The serum samples were diagnosed for the presence of dengue NS1 antigen followed by the isolation of dengue genome from NS1 positive samples. The isolated dengue genome was further subjected to RTPCR based molecular serotyping. The phylogenetic tree was constructed based on the sequence of PCR amplified products.

Results. Out of the total number of samples collected, 100 samples were positive for dengue specific antigen (NS1) and 26 of them contained the dengue genome. The RTPCR based molecular serotyping of the dengue genome revealed the presence of all four serotypes with different combinations. However, serotypes 1 and 3 were predominant combinations of concurrent infection. Interestingly, there were two samples with all four serotypes concurrently infected in 2013.

Discussion. All samples containing dengue genome showed the presence of more than one serotype, indicating 100% concurrent infection. However, the combination of serotypes 1 and 3 was predominant. To the best of our knowledge, this is the first report indicating the concurrent infection of dengue in the northern Kerala, India. The phylogenetic analysis of dengue serotype 1 identified in this study shows a close relationship with the strain isolated in Delhi and South Korea during the 2006 and 2015 epidemics respectively. Similarly this study indicates that the phylogeny of dengue serotype 3 of northern Kerala is more closely related to dengue isolate of Rajasthan state, India. The geographical and climatic conditions of Kerala favours the breeding of

Submitted 1 September 2016
Accepted 30 December 2016
Published 14 March 2017

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Rajendra Pilankatta,
praj74@cukerala.ac.in

Academic editor
Jerson Silva

Additional Information and
Declarations can be found on
page 13

DOI 10.7717/peerj.2970

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2. Manchala NR, Dungdung R, Pilankatta R. Proteomic analysis reveals the enhancement of human serum apolipoprotein A-1(APO A-1) in individuals infected with multiple dengue virus serotypes. *Trop Med Int Health*. 2017 Oct;22(10):1334-1342. <https://doi.org/10.1111/tmi.12931>

Proteomic analysis reveals the enhancement of human serum apolipoprotein A-1(APO A-1) in individuals infected with multiple dengue virus serotypes

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Abstract

OBJECTIVES Human serum protein profiling of the individual infected with multiple dengue virus serotypes for identifying the potential biomarkers and to investigate the cause for the severity of dengue virus infection.

METHODS Dengue virus NS1-positive serum samples were pooled into two groups (S2 and S3) based on the molecular serotyping and number of heterotypic infections. The pooled serum samples were subjected to two-dimensional gel electrophoresis (2DGE) to identify the differentially expressed proteins. The peptide masses of upregulated protein were detected by matrix-assisted laser desorption-ionisation time-of-flight MALDI-TOF mass spectrometry and analysed by MASCOT search engine. The results were compared with the control group (S1). The commonly upregulated protein was validated by quantitative ELISA and compared with control as well as single serotypic infected samples.

RESULTS Based on 2DGE, total thirteen proteins were differentially upregulated in S2 and S3 groups as compared to control. Some of the upregulated proteins were involved in mediating the complement activation of immune response. The apolipoprotein A-1 (APO A-1) was upregulated in S2 and S3 groups. Upon validation, APO A-1 levels were increased in line with the number of heterotypic infection of dengue viruses.

CONCLUSION Heterotypic infection of dengue viruses upregulate the serum proteins involved in the complement pathway in the early phase of infection. There was a significant increase in the level of APO A-1 in three different serotypic infections of dengue virus as compared to control. Further, the role of APO-A1 can be explored in elucidating the mechanism of dengue pathogenesis.

keywords dengue, serum, multiple, MALDI, 2DGE, APO A-1

Introduction

Dengue virus belongs to the family *Flaviviridae* within the genus *Flavivirus* that causes high mortality and morbidity. Dengue fever is a mosquito-borne viral disease, causing 100 million infections annually and 25 000 deaths mostly among children [1] and [2]. The global incidence of dengue cases is significantly increasing every year and becoming a major threat to the mankind. As per World Health Organization (WHO), there were 390 million cases of dengue fever reported worldwide [3], of which 100 million require proper medication. Dengue viral infection is caused by four distinct serotypes DENV-1, DENV-2, DENV-3 and DENV-4, which are varying antigenically in the envelope protein of the virus [4]. The capital city of India becomes the hot spot for dengue virus due to the climatic conditions favourable for *Aedes* mosquitoes breeding [5].

A spectrum of clinical manifestations is associated with dengue infection such as high fever, joint pains, headache, febrile illness, nausea, vomiting and characteristic skin rashes that can last up to a 4–7 days. During the secondary infection, dengue fever progresses to severe dengue haemorrhagic fever (DHF) and dengue shock syndrome (DSS) [6]. Dengue virus enters into the host cell by direct fusion with the cell membrane or by endocytosis [7]. Upon internalisation of the viral particle, the positive-strand RNA genome gets released into the cytoplasm, where the RNA gets translated. In humans, the immunity developed by any one of dengue serotype can potentially sensitise the individual to severe manifestation by exposure to a heterologous serotype [8]. The individual having DHF is characterised by severe bleeding, blood plasma leakage and an exceptionally low platelet count. However, DSS occurs due to low blood pressure, which may lead to a circulatory collapse (shock). The

3. Nageswar Reddy, Ranjeet Dung Dung, Pankaj Trivedi, Unnikrishnan Unniyampurath, Rajendra Pilankatta. Mycophenolic acid (MPA) modulates host cellular autophagy progression in sub genomic dengue virus-2 replicon cells. *Microb. Pathog.* 2019; 137, 103762. <https://doi.org/10.1016/j.micpath.2019.103762>

> *Microb Pathog.* 2019 Dec;137:103762. doi: 10.1016/j.micpath.2019.103762. Epub 2019 Sep 24.

Mycophenolic acid (MPA) modulates host cellular autophagy progression in sub genomic dengue virus-2 replicon cells

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PMID: [31560972](#) DOI: 10.1016/j.micpath.2019.103762

Abstract

Cellular autophagy (Macrophagy) is a self-degradative process, executed through the network of autophagy associated genes (ATGs) encoded proteins. Both in vitro and in vivo studies suggest that dengue virus (DENV) induces autophagy and supports the viral genome replication and translation. Therefore, the cellular autophagy induced by dengue virus can be a good target for antiviral drug development. The action of mycophenolic acid (MPA), a specific inhibitor of DENV replication, was investigated in the stable BHK-21/DENV2 replicon cells. The inhibition was mediated by enhanced degradation of autophagic substrates in stable BHK-21/DENV2 replicon cells as evidenced by a decrease in lipidated LC3 (LC3II) and p62 expression in the presence of MPA. In contrast, the results indicated that four gene sets, namely Transmembrane protein 74 (TMEM74), Unc-51-like kinase 2 (ULK2), Cathepsin D (CTSD) and Estrogen receptor 1 (ESR1) were upregulated in stable BHK-21/DENV2 replicon cells, due to the sustained dynamic replication of DENV2 genome. These ATGs involved in the pre-autophagosomal structure (PAS) formation, were suppressed in the presence MPA. Instead, MPA induced the expression of different set of autophagy genes such as ATG4, AKT1, APP, ATG16L1, ATG16L2, B2M and HPRT1. An enzyme involved in the nucleotide salvage pathway, HPRT1, was highly expressed in the presence of MPA. The study shows that DENV2 replication is dependent on PAS formation and is inhibited in the presence of MPA by enhancing the degradation of autophagic substrates and suppression of PAS formation. This study provides impetus in designing MPA analogues to effectively inhibit dengue viral replication.

Keywords: Autophagy; Dengue; LC3 II; MPA; Replication; Replicon; Viruses; p62.

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A slow, efficient and safe nanoplatform of tailored ZnS QD-mycophenolic acid conjugates for *in vitro* drug delivery against dengue virus 2 genome replication†

Ranjeet Dungdung,[‡] Manikanta Bayal,[‡] Lathika Valliyott,[‡] Unnikrishnan Unniyampurath,[‡] Swapna S. Nair^{†*} and Rajendra Pilankatta^{*‡}

Dengue is a major health concern causing significant mortality, morbidity and economic loss. The development of anti-dengue viral drugs is challenging due to high toxicity, as well as off-target/side effects. We engineered size tuned ZnS QDs as a platform for the efficient delivery of mycophenolic acid (MPA) against dengue virus serotype 2 (DENV2) to evaluate the drug efficacy and toxicity using the DENV2 sub-genomic replicon system in BHK21 cells. The results indicate that the Selectivity Index 50 (SI₅₀) of the ZnS QD-MPA conjugate was two orders higher than that of free MPA with lower cytotoxicity. The effect is attributed to the sustained release of MPA from ZnS QD-MPA. The conjugated MPA caused significant inhibition of the virus at the level of replication and viral protein translation. The study underpins the efficiency of the ZnS QD for the delivery of antiviral drugs against DENV2 with negligible toxicity and side effects.

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Introduction

Viral infections are one of the leading causes of mortality worldwide, having a global negative impact on healthcare and socio-economic development.¹ However, the lack of selective inhibitors against a multitude of medically important viruses, with a prominence of emerging and re-emerging RNA viruses (e.g.: Dengue, Chikungunya, SARS-CoV, MERS-CoV, and SARS-CoV2) has aggravated the scenario. Although it is impractical to develop drugs against each of these viruses within a short duration, the development of novel treatment strategies is the key to resolve the issue.² The adverse side effects/toxicity due to prolonged use of drugs and the rapid development of drug resistance in patients to the existing therapies make it a more serious public health concern.³

In this context, nanomedicine based strategies using biocompatible nanomaterials can be considered as a powerful tool to enhance the efficacy of antiviral drugs with the

possibility of a remarkable reduction in toxicity. Nanoparticles offer distinctive physical properties that can have associated advantages for drug delivery, mainly due to the small particle size, large surface area to volume ratio, and the tunable surface charge of the particle.⁴ The possibility of drug encapsulation, as well as the capability of nanoparticles to handle large drug payloads, can be attributed to the above properties. Thus nanoparticulate drug delivery systems are attractive candidates to improve the therapeutic effects of drugs.^{5,6} Several anti-cancer drugs including paclitaxel, doxorubicin, 5-fluorouracil, and dexamethasone have been successfully re-formulated using nano-material based delivery systems.⁷⁻¹⁰ Nano-medicines with different types of nano-formulations against HIV have been approved and are currently undergoing investigation for the treatment of viral infections.¹¹ It was reported that glucose coated gold nanoparticles (GNPs) attached to the drugs abacavir and lamivudine can act as a multivalent drug against HIV.¹²

Importantly, quantum dots (QDs) are promising zero-dimensional materials that can be designed/engineered for tailored applications. The use of QDs in medicine, and cell and molecular biology is one of the fastest emerging and most interesting interfaces of nanotechnology.¹³⁻¹⁵ The bio-distribution and the toxicity of QDs are determined by their surface coating and particle size.^{16,17} Zinc sulfide (ZnS), which belongs to the semi-conductor class II-VI, is one of the most ideal QDs which can be explored for biological applications due to its low toxicity levels.⁴ It has excellent potential for application in fields such as drug delivery, and bio-imaging and also

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5. Balasubramanian A, Manzano M, Teramoto T, Pilankatta R, Padmanabhan R. High-throughput screening for the identification of small-molecule inhibitors of the flaviviral protease. *Antiviral Res.* 2016 Oct;134:6-16. doi: 10.1016/j.antiviral.2016.08.014. PubMed PMID: 27539384;



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High-throughput screening for the identification of small-molecule inhibitors of the flaviviral protease

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Abstract

The mosquito-borne dengue virus serotypes 1-4 (DENV1-4) and West Nile virus (WNV) cause serious illnesses worldwide associated with considerable morbidity and mortality. According to the World Health Organization (WHO) estimates, there are about 390 million infections every year leading to ~500,000 dengue haemorrhagic fever (DHF) cases and ~25,000 deaths, mostly among children. Antiviral therapies could reduce the morbidity and mortality associated with flaviviral infections, but currently there are no drugs available for treatment. In this study, a high-throughput screening assay for the Dengue protease was employed to screen ~120,000 small molecule compounds for identification of inhibitors. Eight of these inhibitors have been extensively analyzed for inhibition of the viral protease in vitro and cell-based viral replication using *Renilla* luciferase reporter replicon, infectivity (plaque) and cytotoxicity assays. Three of these compounds were identified as potent inhibitors of DENV and WNV proteases, and viral replication of DENV2 replicon and infectious RNA. Fluorescence quenching, kinetic analysis and molecular modeling of these inhibitors into the structure of NS2B-NS3 protease suggest a mode of inhibition for three compounds that they bind to the substrate binding pocket.

Keywords

Protease inhibitors; High-throughput screening; *Renilla luciferase* reporter replicon; virus infectivity assays; therapeutic index; fluorescence quenching

Introduction

Dengue virus serotypes 1-4 (DENV1-4) cause the most frequent mosquito-borne infections to humans. According to a recent estimate by the World Health Organization, DENVs cause

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6. Balasubramanian A, Pilankatta R, Teramoto T, Sajith AM, Nwulia E, Kulkarni A, Padmanabhan R. Inhibition of dengue virus by curcuminoids. *Antiviral Res.* 2019 Feb;162:71-78. <https://dx.doi.org/10.1016%2Fj.antiviral.2018.12.002>.



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Inhibition of dengue virus by curcuminoids

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Abstract

The dengue virus is considered to be a globally important human pathogen prevalent in tropical and subtropical regions of the world. According to a recent estimate, the disease burden due to DENV infections is ~390 million infections per year globally in ~100 countries including the southern US, Puerto Rico and Hawaii, resulting in nearly ~25,000 deaths mostly among children. Despite the significant morbidity and mortality that results from DENV infections, there is currently no effective chemotherapeutic treatment for DENV infections. We identified curcumin as an inhibitor of DENV2 NS2B/NS3 protease in a previous high-throughput screening (HTS) campaign. We synthesized four analogues of curcumin (curcuminoids) and tested the *in vitro* protease activity and inhibition of replication by cell-based assays. The results revealed that curcumin is a weak inhibitor of the viral protease. However, the analogues exhibited more potent inhibition of DENV infectivity in plaque assays suggesting that the cellular pathway(s) required for viral replication and/or assembly are targeted by these compounds. Further analysis shows that inhibition of genes involved in lipid biosynthesis, and of actin polymerization by curcuminoids, are likely to be involved as their mode of action in DENV2-infected cells. Three of the curcumin derivatives possess good selectivity indices (SI) (>10) when compared to the parent curcumin.

1. Introduction:

Dengue virus (DENV) is a member of mosquito-borne flavivirus genus of *Flaviviridae* family which consists of > 70 human pathogens causing considerable morbidity and mortality in tropical and subtropical regions of the world (for reviews, see (Beasley, 2005; Gould and Solomon, 2008; Lindenbach et al., 2007)). There are four serotypes of dengue virus (DENV1–4) that together cause 390 million infections annually (Bhatt et al., 2013).

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7. Lathika Valliyott, Ranjeet Ddung, Rajendra Pilankatta. Semi-quantification of Antibody-Dependent Enhancement (ADE) in the Uptake of Adenovirus Serotype 5 Into THP-1 Cells. *Anal Biochem.* 2020 Feb 15;591:113568. doi: 10.1016/j.ab.2019.113568. Epub 2019 Dec 24.

> [Anal Biochem.](#) 2020 Feb 15;591:113568. doi: 10.1016/j.ab.2019.113568. Epub 2019 Dec 24.

Semi-quantification of antibody-dependent enhancement (ADE) in the uptake of Adenovirus serotype 5 into THP-1 cells

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Abstract

Replication defective recombinant Ad5 vectors (rAdV5) are extensively explored for its applications in gene therapy and vaccine delivery. Ad5 enter into monocytes and macrophages through CAR independent route as an immune complex termed as antibody-dependent enhancement (ADE). We developed an effective method for estimating the ADE of rAdV5 encoding GFP (rAdV5-GFP) into THP-1 cells, using fluorimetric semi-quantification of GFP. Initially, twenty numbers of human sera samples were screened in HeLa cells for anti-Ad5 antibody titer using neutralization assay. Uptake of rAdV5-GFP in THP-1 cells was observed only after pre-incubation with the serially diluted human sera which are attributed to ADE. The optimal dilution which showed the maximum GFP expression as per the fluorescence microscopic analysis in THP-1 cells was used for further analysis. Fluorimetric analysis of the THP-1 cell lysate showed a maximum GFP intensity of 17058 RFU, which was equivalent to the 0.397 pmoles of Alexa Fluor 488 under the same experimental condition. Similarly, immunoblot analysis of GFP in THP-1 cell lysate and HeLa cell lysate confirmed the entry of rAdV5-GFP into the cells. The assay can serve as a platform for understanding the molecular events involved in ADE for the uptake of viruses into immune cells.

Keywords: Antibody-dependent enhancement; Coxsackie and adenovirus receptor; Green fluorescent protein; Multiplicity of infection; Replication defective recombinant adenovirus 5 vector encoding GFP.

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ORIGINAL ARTICLE

Evaluation of diagnostic accuracy of developed rapid SARS-COV-2 IgG antibody test kit using novel diluent system

Vani Maya¹ · Jyothi Embekkat Kaviyil³ · Dinoop Koral Ponnambath³ · Renjith P. Nair² · Anugya Bhatt² · Rajendra Pilankatta⁴ · Lathika Valliyot⁴ · Ranjeet Ddungdung⁴ · Ramdas Athikkal Veettil⁵ · Manoj Gopi¹

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Abstract Immunochromatographic assay kits are used in primary diagnostics which is based on the principle of antigen and antibody interaction. These kits play pivotal role in rapid surveillance of infectious diseases at early stages as well as for the surveillance of the contagious diseases. The immunochromatographic test kits lacks sensitivity and specificity with certain diseases. In this study, our intention was to develop a rapid test kit for SARS-COV-2 with a novel diluent system to enhance the efficacy of antigen–antibody binding and thereby the improvement in the sensitivity outlined. Finally, IgG antibodies against SARS-COV-2 virus peptides were analyzed using 25 positive and 25 negative confirmed clinical samples. The sensitivity of the clinical studies showed 91% sensitivity and 100% specificity. Therefore, the authors propose that this assay will be a potential tool for efficient community or sentinel surveillance of SARS-COV-2 infection and additionally, for effective monitoring of convalescent sera therapy.

Keywords SARS-CoV-2 · Rapid test kit · IgG · Diluents · Surveillance

Introduction

In the immunological diagnostic/surveillance systems, antigen–antibody ligand formation plays a vital role in determining the sensitivity and specificity of a particular assay [1–3]. When a foreign antigen enters the human system, the host normally produces antibodies as part of the immune response, with subsequent formations of specific antigen–antibody complexes. Most infectious diseases are often diagnosed based on the detection of antigens and/or antibodies from the relevant biological samples [4, 5]. Due to certain conditions, such as low antibody and/or antigen titers or masking of the antibodies with other proteins, the detection of specific antibodies from the biological samples becomes complicated [6].

Timely detection of the etiologic agent is essential for appropriate diagnosis of the infectious diseases in order to aid targeted treatment. Also surveillance strategies in case of highly contagious diseases are important to contain spread of disease in population. We are currently in an era of rapid turnaround times [TAT] with immunochromatographic test kits playing a major role in aiding rapid and accurate diagnosis of some of the major infectious diseases [7]. Hence, developing newer immunochromatographic test devices incorporating novel diluent systems and other techniques that can enhance the sensitivity and specificity is essential in bringing about a better diagnostic ecosystem.

The SARS-CoV-2, causing COVID 19 infection and first isolated from China, has caused a pandemic across the globe [8, 9]. Infections with this virus can manifest with a wide range of symptoms—from asymptomatic to severe

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9. Raman Swathy Vaman, Rajendra Pilankatta , Adarsh MB. “Immune Response in SARS-CoV-2 Reinfection- A Friend or a Foe”- A Case Study from Kerala, India. *Int J Med. Public Health*. 2021; 11(2):124-125. DOI : 10.5530/ijmedph.2021.2.22

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Case Report

“Immune Response in SARS-CoV-2 Reinfection- A Friend or a Foe”- A Case Study from Kerala, India

Raman Swathy Vaman^{1,*}, Rajendra Pilankatta², Adarsh MB³

ABSTRACT

Covid-19 caused by the SARS-CoV-2 is a major public health threat affecting the entire world causing significant mortality. Few studies have demonstrated possible chances of reinfection with the SARS-CoV-2. This case study describes the features of reinfection with the virus thirty two days after RT-PCR negativity in a 20 year old female in Kerala. This case highlights the possible chance of reinfection and the shortened viremia in response to the mounted good immune response.

Key words: SARS-CoV-2, COVID-19, Reinfection, Kerala, India.

INTRODUCTION

COVID-19 pandemic is a looming public health threat which results in severe mortality across the globe owing to its high spreading rate, even though *per se* mortality is near 3 %. Studies by Tomassini *et al.*¹ and Batisse *et al.*² highlight the possibility of reinfection among patients with COVID 19. Both of them have emphasised the seroconversion these patients had and the probable reinfection among those with seroconversion. Among the cases reported by Tomassini *et al.* few had complete resolution of clinical symptoms with a negative PCR on follow up but turned positive again after a significant time duration. The possible reason they have suggested is that the antibodies wear off over a period and the host gets a reinfection. But this needs to be read with the fact that patients had worse outcomes with re infection in the series by Batisse *et al.* likely due to a hyperinflammatory response. There is currently no evidence that people who have recovered from COVID-19 and have antibodies are protected from a second infection.³ We had an interesting observation in this regard. Here we describe the features of a patient who developed clinical features suggestive of COVID-19, thirty-two days after being symptom free and negative for Real time Polymerase chain reaction (RT-PCR). She had a positive result with RT-PCR at this time but became negative within 48 hr with a good IgG response. This case is probably the first reported case of reinfection with the SARS-COV-2 in India. It highlights the possible chance of SARS-COV-2 reinfection and the shortened viremia in reinfection possibly due to the mounted good immune response. So, a reinfection may have an exuberant inflammatory response resulting in

worse outcome as in Batisse *et al.* study or may have a shortened viremia as in our case.

CASE STUDY

A 20-year-old female who was the primary contact of a COVID-19 super spreader developed mild fever and nasal discharge since 23/03/2020 and was tested positive for SARS- COV-2 on 24/03/2020. Her throat swab was positive for PCR for SARS-CoV-2 E (29.3), ORF1 (30.3) and RdRP (36.9) antigens. Her symptoms subsided within four days of onset and repeat throat swabs taken on ninth and fourteenth day of admission were all negative. There were no secondary cases from her and was released from home quarantine after 14 days.

She didn't travel anywhere or had any contact with confirmed or suspect cases since discharge. On 12/05/2020, 32 days after her discharge from the hospital she developed fever, rhinitis, mild cough and chest discomfort. Her RT-PCR was positive this time for both E (35.06) and S (34.52) antigens. But a repeat RT-PCR done 48 hr later became negative. Her Rapid antibody test was strongly positive for IgG. Her symptoms subsided by the fourth day of admission and the patient was discharged on 19/05/2020.

Pre-existing immunity due to the prior infection of SARS-CoV-2 might be the reason for the shortened secondary viremic phase associated with the enhanced IgG response as evidenced by the antibody test. The hyperinflammatory response seen in Betesse *et al.* study might have stemmed from the immune enhancement or antibody dependent enhancement (ADE) of virus uptake into immune cells during the secondary infection. These immune complexes are formed between the non-neutralizing cross

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AWARDS AND RECOGNITION FOR EXCELLENT PERFORMANCE IN PREVENTING AND CONTROLLING OF COVID19 PANDEMIC IN KASARAGOD DISTRICT



COVID19 RTPCR testing team received excellence award from Hon'ble Minister, Smt. K K Shylaja, Dept of Health and Family Welfare, Govt of Kerala and Hon'ble VC, Prof H Venkateshwarlu for excellent services in COVID19 prevention and control activities in the Kasaragod District.



COVID19 RTPCR Testing lab in charge, Dr Rajendra Pilankatta is briefing the activities of CUK in COVID19 prevention and control in the Kasaragod District in a District review meeting in the presence of Hon'ble Minister, Smt. K K Shylaja and Dr Raman, District Program Manager.

റോട്ടറി എക്സലൻസ് അവാർഡ് സമ്മാനിച്ചു

കാഞ്ഞങ്ങാട്: ഒടയാലിൽ റോട്ടറിയുടെ ഈ വർഷത്തെ വൊക്കേഷനൽ എക്സലൻസ് പുരസ്കാരം റോട്ടറി ഡിസ്ട്രിക്ട് ഗവർണർ ഡോ.പി.സി. ഹരികൃഷ്ണൻ നമ്പ്യാർ സമ്മാനിച്ചു.



ഒടയാലിൽ റോട്ടറി പുരസ്കാരം റോട്ടറി ഡിസ്ട്രിക്ട് ഗവർണർ ഡോ.പി.സി.ഹരികൃഷ്ണൻ നമ്പ്യാർ സമ്മാനിക്കുന്നു.

പെരിയ കേന്ദ്ര സർവകലാശാലയിലെ ബയോകെമിസ്ട്രി വിഭാഗം മേധാവി ഡോ. രാജേന്ദ്ര പിലങ്കട്ടയ്ക്കും പുടം കല്ലു താലൂക്ക് ആശുപത്രി സൂപ്രണ്ട് ഡോ.സി. സുകുമാറിനും പുരസ്കാരം സമ്മാനിച്ചു. ജില്ലാ സബ് ജൂനിയർ വോളിബോൾ ടീം ക്യാപ്റ്റൻ സാനാം ലഭിച്ച മീഥുൻ കൃഷ്ണനെ അനുമോദിച്ചു. ഒടയാലിൽ റോട്ടറി പ്രസിഡന്റ് എ.കെ രജീഷ് അധ്യക്ഷത വഹിച്ചു. റോട്ടറി മേഖലാ കോ ഓർഡിനേറ്റർ ഡോ. അ

നീൽ മേലത്ത്, അസി. ഗവർണർ ബി. മുക്യൻ പ്രജ, ടി.ജെ. സന്തോഷ്, എം.രാജൻ, എം. തമ്പാൻ, അനീൽകുമാർ ഫിലിപ്പ്, കെ. മോഹനൻ നായർ, ടി.ടി. സജി, വിനീഷ് രോഹിണി, കെ.എം. ബാബു എന്നിവർ സംസാരിച്ചു.

Dr Rajendra Pilankatta, COVID19 RTPCR testing lab in charge, was honored by vocational excellence award by the Rotary, Odayanchal, Kasaragod for the excellent service for the society in preventing COVID19 pandemic.

COVID19 RTPCR LAB OF CUK WAS WELL CITED BY PRINT AS WELL AS VISUAL MEDIA FOR SOCIETAL SERVICE

08 2020 മാർച്ച് 29 വാർത്തകൾ

പ്രതിരോധ കമ്പിയാങ്ങൾ സമാധാന സർക്കാർ ഉന്നത സ്തുതികൾ നേടിയതിനോടൊപ്പം സമാധാന സർക്കാർ ഉന്നത സ്തുതികൾ നേടിയതിനോടൊപ്പം സമാധാന സർക്കാർ ഉന്നത സ്തുതികൾ നേടിയതിനോടൊപ്പം...

ഉത്തരം ഇവിടെ തയ്യാറാകുന്നു

5 വർഷത്തിലേക്ക്

അഭിമാനം ഈ അറിവിൻ പ്രകാശം

സമാധാന സർക്കാർ ഉന്നത സ്തുതികൾ നേടിയതിനോടൊപ്പം...

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സമാധാന സർക്കാർ ഉന്നത സ്തുതികൾ നേടിയതിനോടൊപ്പം...

BRIEFING ABOUT THE COVID19 RTPCR TESTING ACTIVITY BEFORE THE LEADING VISUAL MEDIA BY Dr RAJENDRA PILANKATTA, Lab in charge



PLEASE LOGIN TO THE FOLLOWING LINK FOR FURTHER VISUAL MEDIA COVERAGE OF THE COVID19 RTPCR TESTING ACTIVITY OF CUK

1. <https://youtu.be/p7BfTQigHPM>
2. <https://youtu.be/A1claNuP51s>
3. <https://youtu.be/6TH9zP2Gj3M>
4. https://youtu.be/vfuhlPRPP_M

Thank You!

