

Phytomedicines: Extraction, Purification, In Vitro and In Vivo studies

24th – 30th July 2022

Sponsored by

Department of Science & Technology - Synergistic Training Program Utilizing the
Scientific and Technological Infrastructure (DST-STUTI)

Organized by

Department of Biochemistry, Kuvempu University.

Department of Biotechnology, Sahyadri Science College, Shivamogga

JSS Academy of Higher Education and Research, Mysore.

One week National workshop on Hands on training on **“Phytomedicines: Purification, characterization, in-vitro and in-vivo testing”** [“Phytomedicines: Purification, characterization, in-vitro and in-vivo testing”]



Brief Report

The Department of Biochemistry, Kuvempu University. The Department of Biotechnology, Sahyadri Science College, Shivamogga and JSS Academy of Higher Education and Research, Mysore. Organized a 7-day workshop on “**Phytomedicines: Purification, characterization, in-vitro and in-vivo testing**” from 24th – 30th July 2022. The Department of Science & Technology - Synergistic Training Program Utilizing the Scientific and Technological Infrastructure (DST-STUTI) sponsored the workshop. We had received more than **125** applications for the workshop, among which **34** participants were selected based their bio data, research interest, and recommendation letter and were sponsored to attend the workshop.



Inauguration of the 7 days national level workshop and release of the workshop handbook

The workshop was inaugurated on 24th July 2022 by **Prof. Rajeshwari N.** Principal Sahyadri Science College, Shivamogga. **Dr. Dinesh A. Nagegowda** Senior Principal Scientist & Scientist In-Charge CSIR Central Institute of Medicinal and Aromatic Plants Research Centre, Bengaluru. **Dr. Prashant Vishwanath** MBBS., MD., PhD Director (Research) & IQAC Coordinator & DST STUTI PMU Coordinator JSS Academy of Higher Education & Research, Mysore. **Dr. Subba Rao. M.V.S.S.T.** Professor, Cellular & Molecular Biology Center of Excellence in Molecular Biology & Regenerative Medicine (CEMR) Laboratory, Dept. of Biochemistry, JSS Medical College JSS AHER, Mysuru **Dr. Niranjana Pathappa** Organizing

secretary and Chairman, Dept. of PG Studies & Research in Biochemistry Kuvempu University, Shivamogga. **Dr. B.T. Prabhakar**, Organizing secretary and Associate Professor,

Dept. of Biotechnology, Sahyadri Science College, Shivamogga. **Dr. Vadlapudi Kumar**, Ph.D. Professor, Department of Biochemistry, Shiva gangothi, Davangere University. **Dr. G J Sathisha**, Prof. Dept. of PG Studies & Research in Biochemistry Kuvempu University, Shivamogga and **Dr. S E Neelgund** Prof. Dept. of PG Studies & Research in Biochemistry Kuvempu University, Shivamogga.

The 7-day workshop consisted of a series of lectures from various faculties as well as hands-on sessions on different techniques which are generally used in research and diagnostic laboratories. The first six days of the workshop were scheduled at the Department of PG Studies & Research in Biochemistry Kuvempu University and Department of Biotechnology, Sahyadri Science College, Shivamogga in Parallel. The participants were then divided into two groups of 17 each to engage them in both Department of PG Studies & Research in Biochemistry Kuvempu University and Department of Biotechnology, Sahyadri Science College, Shivamogga hands-on work in parallel. Batch 1 had stay at Kuvempu University for first three days and Batch 2 at Sahyadri Science College. For the next 3 days Batches were exchanged respectively.



Scientific Sessions by Dr. Dinesh A. Nagegowda and Dr. Vadlapudi Kumar

On the first day, **Dr. Dinesh A. Nagegowda** Senior Principal Scientist & Scientist In-Charge CSIR Central Institute of Medicinal and Aromatic Plants Research Centre, Bengaluru,

introduced the concepts of **High-value Phytochemicals: Biosynthesis and overproduction through molecular and metabolic engineering techniques**, in the 1stSession.

Dr. Vadlapudi Kumar Prof. & D.O.S. in Biochemistry, Davangere University, Shivagangothri, Davangere, addressed the gathering through his talk on **Approaches for the extraction, isolation and characterization of bioactive compounds in plants** to the participants in the second session. This was followed Qualitative analysis of phytochemicals: (carbohydrates, proteins, phenols and flavonoids). Saturation of paper / thin layer chromatography chamber in the research lab of Department of PG Studies & Research in Biochemistry Kuvempu University, Kuvempu University on the next day morning.

Phytochemicals extraction by millet seeds was performed by Mahadevaswamy (JSSAHER) for first batch students, followed by centrifugation and quantitative estimations of Protein and Reducing sugar by Dr. Kotresh K R and Dr. Avinash B at Department of Biochemistry, KVV Shankarghatta.



Hands on Session on Phytochemicals extraction from millet seeds

On the first day (24th July), **Dr. B T Prabhakar**, Associate Professor, Department of Biotechnology, Sahyadri Science College, Shivamogga. Introduced the concepts of hands on training planned in Molecular Biomedicine Laboratory to the participants. This was followed by brief introduction of the demonstrators to the participants and outline of the experimental plans. After the lunch session, Mrs. Banumathi and Mr. Siddesh B.M demonstrated the procedure of Cell lysate preparation. Ms. ShruthiShree and Ms. Hemavathi demonstrated the Tryphan Blue dye exclusion assay and the counting of the in-vitro cultured cells. Mr. Kiran B.K Mr. Ankith Sherapura and Mr. Ganapathi Yadav brief out the cell culture disciplines which included Good cell culture practice followed by the hands on training on basics of the

cell culture to the participants such as Cell thawing, Trypsinization, Seeding to T-Flask and 96 well plate for the MTT assay.



Participants performing Immuno blotting set up



Scientific session by Dr. S.E. Neelgund

On the second day afternoon session, students of batch 1, attended lectures by **Dr. S E Neelgund** Prof.Dept. of PG Studies & Research in Biochemistry Kuvempu University, Shivamogga. His lecture focused on **“Exploring the therapeutic bioactive molecules from medicinal plants”**.

On the Second day (25th July) of the training programme participants attended lecturing session by the resource person **Dr. Pradeepa K, Assistant Professor, Department of Biotechnology, Sahyadri Science College, Kuvempu University, Shivamogga**. On the suspension culture. The training programme was continued with a demonstration of SDS PAGE- gel preparation, protein estimation from cell lysates, followed by western blotting. Half-day was dedicated to hands on training of the western blotting technique, as instructed by Mrs. Banumathi, Mr. Siddesh B M, Mr. Ganapathi Yadav, Ms. Shruthi Shree and Ms. Hemavathi to the individual participants.

In the afternoon, participants were involved in the hands on training of the individual steps of the western blotting experiment such as transfer of the separated proteins to nitrocellulose membrane followed by blocking and incubation with primary antibody. Further participants also trained with cell culture technique of MTT assay like drug treatment by Mr. Kiran B.K. After the high tea break, **Dr. B.T. Prabhakar**. Associate Professor, Department of Biotechnology, Sahyadri Science College, Shivamogga-577 203. engaged the lecturing session on role of angiogenesis in the cancer progression and the role of good diet for the healthy



Participants performing CAM Assay

Day 3 started with the scientific lecture on **“Isolation, Purification and characterization of bioactive peptides/proteins from plants by conventional and recombinant DNA techniques “** by **Prof. G.J Sathisha**, Department of Biochemistry. Students of Batch 1 attended this session then followed by demonstration and hands on training about Protein purification by FPLC guided by Dr. Vasanth & Dr. Anitha at the research lab of Department of PG Studies & Research in Biochemistry Kuvempu University, Shankaraghatta.



Scientific session by Prof. G.J. Sathisha

3rd day (26th July) of the training programme in Sahyadri Science College initiated with the lecturing session by **Dr. Pramod S.N.** Invited speaker, Associate Professor, Department of Food Technology, Davangere University, Shivangotri, Davangere-577 007, Karnataka, India. After the session, participants were engaged in the continuation of the western blotting completion by secondary antibody incubation and blot development for the visualization of the protein bands by Mrs. Banumathi, Mr. Siddesh B M. After the tea break, participants were hands on trained with non-tumorigenic angiogenic model *In-Vivo* CAM assay demonstrated by Ms. ShruthiShree and Ms. Hemavathi. and the continuation of the MTT assay for the evaluation of the cytotoxic analysis and IC₅₀ value determination was assessed

by Mr. Kiran B.K. The afternoon session was completely engaged in primer design and qRT-PCR demonstration by BioRad kit method. After the demonstration part feedback was taken from the participants of batch-I.



Later the day ended by an interaction session with **Dr. Niranjana Pathappa** Chairman, Dept. of PG Studies & Research in Biochemistry Kuvempu University, Shivamogga on the topic of “Recombinant expression and purification of antimicrobial peptides”.



Participants preparing the cell lysate under the guidance of Dr. B.T. Prabhakar



Scientific session by Dr. Vadlapudi Kumar which is followed by an interactive session.

The next day (4th) started with a talk from **Dr. Vadlapudi Kumar** Prof. & D.O.S. in Biochemistry, Davangere University, Shivagangothri, Davangere, Karnataka, addressed the gathering through his talk on “ **Approaches for the extraction, isolation and characterization of bioactive compounds in plants** ” to the participants of **batch 2**.

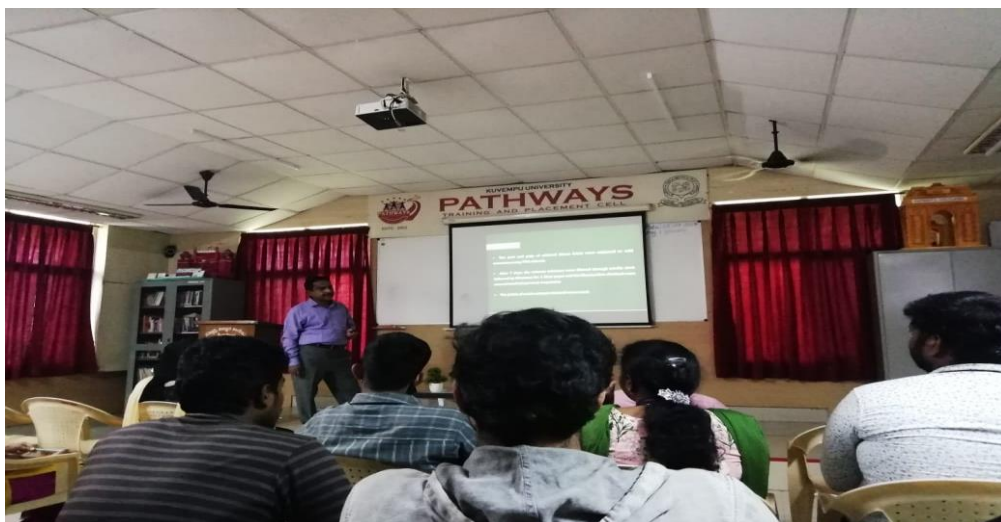
The Fourth day (27th July), training programme in Molecular Biomedicine Laboratory was initiated by new batch. the lecturing session was engaged by **Dr. Pramod S.N.** Invited speaker on “Plant based dietary lectins: promising future phytomedicines for immunotherapeutics ”. After the session, the participants were briefed out by **Dr. B T Prabhakar, Associate** Professor, Department of Biotechnology, Sahyadri Science College, Shivamogga regarding the concepts of hands-on training planned in Molecular Biomedicine Laboratory followed by brief introduction of the demonstrators to the participants and outline of the experimental plans. After the lunch session, participants were involved in hands on training on basics of the cell culture to the participants such as Cell thawing, Trypsinization, Seeding to T-Flask and 96 well plate for the MTT assay. Mrs. Banumathi and Mr. Siddesh demonstrated the procedure of Cell lysate preparation and hands on training in protein estimation. Ms. Shruthi Shree and Ms. Hemavathi demonstrated the Tryphan Blue dye exclusion assay and the counting of the in-vitro cultured cells to the participants individually.



An interactive session was held by Dr. Niranjana Pathappa, Chairman, Dept. of PG Studies & Research in Biochemistry Kuvempu University. In his research lab, he introduced the instruments and some of his novel works to all the students of batch 2 from 9 am to 10 am.



Experimental sessions on measurement antioxidant potential of the extract by DPPH and FRAP assays



Scientific session by Prof. C.K. Ramesh



Scientific session by Dr. Pradeepa K.

On the Fifth day (28th July) of the training programme participants attended lecturing session by the resource person **Dr. Pradeepa K.** The training programme was continued with a hands-on training experience of SDS PAGE- gel preparation, followed by western blotting. Half-day was dedicated to hands on training of the western blotting technique, as instructed by Mrs. Banumathi, Mr. Siddesh B M, Mr. Kiran B.K, Ms. ShruthiShree and Ms. Hemavathi to the individual groups. In the afternoon, participants were involved in the hands on training of the individual steps of the western blotting experiment such as transfer of the separated proteins to nitrocellulose membrane followed by blocking and incubation with primary antibody. Further participants also trained with cell culture technique of MTT assay like drug treatment by Mr. Kiran B.K. After the high tea break, Dr. B.T.Prabhakar engaged the lecturing session on role of angiogenesis in the cancer progression and the role of good diet for the healthy life style.



Participants performing immunoblot experiment assisted by Ms. Shruthishree and Ms. Hemavathi B.C.



Participants performing cell lysate preparation as assisted by Mrs. Banumathi.



Participants performing sonication process under the supervision of
Mr.Siddesh B.M



Dr.B.T. Prabhakar explaining the SDS-PAGE to the participants

The 6th day (29th July) of the training programme was initiated with the lecturing session by **Prof. C.K. Ramesh**, Professor, Department of Biotechnology, Sahyadri Science College, Kuvempu University, Shivamogga, on “**Plants as anticancer agents**”. the session included the information regarding use of the natural products isolated from the mother nature.

After the session, participants were engaged in the continuation of the western blotting completion by secondary antibody incubation and blot development for the visualization of the protein bands by Mrs. Banumathi, Mr. Siddesh B M and Mr. Ganapathi Yadav. After the tea break, participants were hands on trained with non-tumorigenic angiogenic model **In-Vivo CAM assay** demonstrated by Ms. ShruthiShree and Ms. Hemavathi. and the continuation of the MTT assay for the evaluation of the cytotoxic analysis and IC₅₀ value determination by Mr. Kiran B.K. The afternoon session was completely engaged in primer design and qRT-PCR demonstration by BioRad kit method. After the demonstration part feedback was taken from the participants.



Lecture by Dr.Mahantesha Naika B.N

The Day started with Lecture by Dr.Raghavendra C. K. about Automated Western Blot and other protein purification . Followed by the lecture by **Dr.Mahantesha Naika B.N**, Assistant Professor Dept. of Biotechnology and Crop Improvement Kittur Rani Channamma College of Horticulture, Arabhavi, University of Horticultural Sciences, Bagalkot, Karnataka, India. He talked about **“Bioinformatics resources and tools for phytochemicals”**



Dr. Sharangouda Assistant Controller of Patents & Designs.

Dr. Sharangouda Assistant Controller of Patents & Designs, the Patent office, Government of India, GST road, Guindy, Chennai-600032, Tamilnadu, India, Took over and delivered a talk on Patenting of inventions based on traditional knowledge- Indian perspective. These highly informative lectures lightened up the knowledge of students and probably gave a fitting end to the workshop sessions on a high note.



Post workshop Assessment Examination for the participants

Post Workshop Assessment Examination was conducted to every participant to assess their learning from workshop and also their helpful and valuable feedbacks were received.



1st & 2nd prize won by the participants Hrushikesh K and Anushka Agarwal respectively

The Day ended with distribution of certificates to the participants. Both participants and volunteers were presented with participation certificates.

SL.No	Name of the participants	Designation	Host Institution
1	Manjula kambi	M.Sc genetics	Karnatak University Dharwad.
2	ARIHANT JAYAWANT KADAPURE	Research scholar	Department of Biochemistry ,H14:H42 Karnatak University Dharwad
3	Mohammed Ali Manik	PG Student	Rani Channamma University Belagavi
4	Vinaykumar Tarikeri	Research scholar	Rani Channamma University Belagavi
5	Gayatri Pawar	PG Student	Padmashree group of institutions
6	R Mahesh	Research scholar	Padmashree institute of management and science(pims) kengeri
7	Yalpi Karthik	Research scholar	Mangalore University
8	Ningaraju S	Research scholar	Mangalore University
9	Srinivasa K	Research scholar	Jnana kaveri Post Graduation campus, Mangalore University, Kodagu
10	Brahmam Bheemisetty	Research scholar	Manipal college of pharmaceutical sciences
11	Pooja S. Rajaput	Research scholar	Garden City University, Bangalore
12	Deepthi S	Research scholar	Garden City University, Bangalore
13	Nagesh.Y.R	Research scholar	Kuvempu University
14	MAHANTHESHKUMAR G T	Research scholar	Sahyadri Science College, Kuvempu University, Shivamogga
15	Sharath K P	Research scholar	Dept. of Applied Botany, Kuvempu University, Jnana sahyadri, Shankaraghatta
16	Abdul shafiulla	Research scholar	Sahyadri Science College Shivamogga
17	Yogesh H Kannammanavar	UG Student	Sahyadri Science College Shivamogga
18	Santhosh CR	UG Student	DVS College of Arts and Science, Shivamogga
19	Hrushikesh K P	PG Student	Adichunchanagiri University
20	Dr. Srikanth Jupudi	Faculty	JSS College of Pharmacy, Ooty
21	Dr. Gomathi Swaminathan	Faculty	JSS College of Pharmacy, Ooty
22	Mr. Kannan R	Research scholar	Adichunchanagiri University
23	Anushka Agrawal	Research scholar	JSS Academy of Higher Education and Research, Mysuru
24	Veronica.A	PG Student	Maharani`s Science College for Women Mysore
25	Amala G	Research scholar	Maharani`s Science College for Women Mysore
26	Sheela.K.R	Faculty	DVS College of Arts and Science, Shivamogga
27	Pavan Kumar SV	Research scholar	Pondicherry University
28	YASHWANTH D	Research scholar	Post Graduation in Microbiology University of Mysore
29	Dr. Mukunda S	Faculty	S.R.N.M. National College of Applied Sciences
30	Amala Claret E	PG Student	CSIR-Central Food Technology Research Institute

