Dr. Amit Sarat.

## University of Mumbai



Academic Planning and Development Section No. APD/ICD/2019-20/762 17<sup>th</sup> March, 2020

Sub: Minor Research Grant Project 2019-20

Sir/Madam,

I am directed to inform you that the said proposal has been considered by the University and the research grant as quoted above is sanctioned to the researcher.

The sanctioned amount will be disbursed in two installments. The first installment of 40% of the sanctioned amount will be disbursed within the month of March. The remaining 60% amount will be disbursed up to 31<sup>st</sup> December, 2020.

The researcher is expected to spend 60% amount initially from his/her own resources to carry out the work.

Further, I am to inform you that the researcher will have to utilize the 40% sanctioned amount on or before 31<sup>st</sup> March, 2020 and submit original bills/vouchers of the expenditure along with Utilization Certificate duly certified by the Principal/Director/Head/Institute/University Department/College to the Accounts Section of the University.

Please note that 60% balance amount, out of sanctioned grant will be released after Poster Presentation & final approval of the committee. Therefore you need to submit of utilization certificate after presentation of your research including bills/vouchers/receipts in original through University Account Section.

The report of the research work carried out by the concerned researcher will have to be submitted to the University on or before 31<sup>st</sup> December, 2020.

The Principal/Head of the Institute are requested to inform the researcher accordingly and arrange to forward his/her undertaking immediately to enable this office to release first installment of the research grant.

Yours faithfully,

Deepak V. More Assistant Registrar (APD Section)

712	DR. PAWAR UMESH RAMCHANDRA	South Ratnagiri District Shikshan Prasarak Mandal�s Shri Pancham Khemraj Mahavidyalaya	45000
713	DR AMIT YADAORAO SARAF	Government of Maharashtra Ismail Yusuf College of Arts Science & Commerce	40000
714	Dr. Ajit Abasaheb Kengar	The Kelkar Education Trust's Vinayak Ganesh Vaze College of Arts Science and Commerce	35000
715	DR. DETHE UTTAM LAXMAN	South Ratnagiri District Shikshan Prasarak Mandal�s Shri Pancham Khemraj Mahavidyalaya	4000
716	Mr. Nitin Devram Shelake	Government of Maharashtra Ismail Yusuf College of Arts Science & Commerce	3500
717	Dr. Lal Sahab Yadav	Smt. Chandibai Himathmal Mansukhani College	4500
718	Gopal N. Deshpande	Padmashri Annasaheb Jadhav Bharatiya Samaj Unnati Mandal's Bhiwandi Nizampur Nagarpalika Arts Science & Commerce College	4000
719	DR.Asmita. S .Mestry	Bharatiya Vidya Bhavan's M.M. College of Arts N.M. Institute of Science and Haji Rashid Jaffer College of Commerce (Bhavan's College)	3500
720	Dr. Arun Nivrutti Chandore	Rayat Shikshan Sansthas Abasaheb Marathe Arts and New Commerce Science College	3000
721	Dr. Vikram Pralhad Masal	Dapoli Education Society's Dapoli Urban Bank Senior Science College	3000
722	Dr. Raghunath Laxman Ghalme	Dapoli Education Society's Dapoli Urban Bank Senior Science College	3500
723	Dr. Bindu Gopalkrishnan	Shri Vile Parle Kelvani Mandal`s Mithibai College of Arts Chauhan Institute of Science and Amrutben Jivanlal College of Commerce and Economics	3500
724	Dr. Jai Knox	Wilson College	3500
725	Dr. Alpa K.Patel,	South Indian Children's Education Society's Arts Science & Commerce College	3500
726	Dr. Vinit Vaidya	Thakur Educational Trust's Thakur College of Science & Commerce	3500
727	Mr. Alok Gude	St. Xavier's College	3000
728	Shri. Suresh. S. Kale	Parle Tilak Vidyalaya Association's Sathaye College	4500
729	Leela Chauhan	Parle Tilak Vidyalaya Association's Sathaye College	3500
730	Dr. Vijaya Lobo	St. Xavier's College	3500
731	Dr. V. G. Chari	Seva Sadan's R.K.Talreja College of Arts Science and Commerce	4500
732	Vijay Kisan Hile	Bharatiya Vidya Bhavan's Hazarimal Somani College of Arts and Science Jayaramdas Patel College of Commerce & Management Studies	4000
733	Dr. Sarita S. Hajirnis	Satish Pradhan Dnyansadhana College Thane (Arts Science and Commerce )	4000
734	Dr. Dhanashree Sagar Patil	Br. Balasaheb Khardekar College	3500
735	Dr. Ritu L. Jain	G. M. Momin College	3500
736	Dr. Deepa Ramabhilash Varma	Visnu Vaman Thakur College, Virar	3000
737	Dr. Rajbhoj Balaji Govindrao	Sundarrao More College of Arts, Comm. & Sci., Poladpur	350





### Government of Maharashtra Ismail Yusuf College of Arts, Science and Commerce Jogeshwari (East), Mumbai 400 060

NAAC "A" Grade (CGPA 3.14)

Tel.no. (office)022-28352881

principaliyc@yahoo.co.in

Fax(principal) 022-28202188

Ref. no.: MRP/2022/02

Date: 28th April 2023

#### **UTILIZATION CERTIFICATE**

Certified that the sanctioned grant of Rs. 40,000/- (Rupees Forty Thousand only) and received amount of first installment of Rs. 28,000/ (Rupees Twenty-eight Thousand only) from the University of Mumbai under the scheme of support for Minor Research Project entitled "Structural elucidation and profiling of phytosterols from Costus igneus (Nak.) by HPTLC and LC-MS/MS" by 'Dr. Amit Yadaorao Saraf' has been fully utilized for the purpose for which it has been sanctioned and in accordance with the terms and conditions laid down by the University.

Project No: 713

University Letter. Ref. No.- APD/ICD/2019-20/762

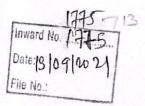
Dated on- 17th March, 2020

Principal Investigator

Principal
PRINCIPAL
Government of Mahareshtra's
Ismail Yusuf College of
Arts, Science & Commerce.
Jogeshwari (East), Mumbai -400 060

## University of Mumbai





de 13.9.21

NAME OF THE RESEARCHER LECTURE IN AMOUNT SANCTIONED CO-INVESTIGATOR Research Project No: 7/3

Botany 40,000/

> Ref No. AAMS/ICD/106 of 2021 23rd July, 2021

To,

Government of Maharashtra Ismail Yusuf College, of Arts, Science & Commerce, Jogeshwari (East). Mumbai - 400 060.

Sub: - Minor Research Grant Project.

Sir/Madam,

I am directed to inform you that the said proposal has been considered by the University and the research grant as quoted above is sanctioned to the researcher.

The sanctioned amount will be disbursed in two installments. The first installment of 70% of the sanctioned amount will be disbursed within the month of August. The remaining 30% amount will be disbursed up to 31st December, 2021.

The researcher is expected to spend 30% amount initially from his/her own resources to carry out the work.

Further, I am to inform you that the researcher will have to utilize the 100% sanctioned amount on or before 31st December, 2021 and submit original bills/vouchers of the expenditure alongwith Utilization Certificate duly certified by the Principal/ Director/ Head/Institute/University Department of the College to The Deputy Registrar, Accounts Section, Mahatma Phule Bhavan, Vidyanagari, Kalina Campus, University of Mumbai, Mumbai - 400 098.

The report of the research work carried out by the concerned researcher will have to be submitted to the University on or before 31st December, 2021.

The Principal/Head of the Institute are requested to inform the researcher accordingly and arrange to forward his/her undertaking immediately to enable this office to release first installment of the research grant.

Yours faithfully,

Assistant Registrar (Academic Planning & Development Section)

Name:	Dr. Amit Yadaorao Saraf
Title of the project:	Structural elucidation and profiling of phytosterols from
	Costus igneus (Nak.) by HPTLC and LC-MS/MS
Subject:	Botany
Project Number:	713
Year:	2019-2020
Amount sanctioned:	40,000/-
Amount released:	28,000/-
Amount utilised:	28,000/-

## **Project Report:**

#### 1. Introduction

#### 1.1 Background

Costus igneus Nak., commonly known as the insulin plant, fiery costus, or spiral flag, is a medicinal plant from the Costaceae family. Traditionally used in India to manage diabetes, this plant has garnered interest due to its purported hypoglycemic properties. Phytosterols, plant-derived sterols with structural similarities to cholesterol, are believed to play a role in the medicinal efficacy of Costus igneus.

#### 1.2 Objectives

The primary objectives of this study are:

- 1. To profile and elucidate the structure of phytosterols in Costus igneus.
- 2. To conduct comparative fingerprinting and profiling using High-Performance Thin-Layer Chromatography (HPTLC) following ICH guidelines.
- 3. To separate phytosterols using a Thin-Layer Chromatography-Mass Spectrometry (TLC-MS) interface.
- 4. To perform Mass Spectrometry (MS) studies for structural elucidation of the separated phytosterols.
- 5. To identify and characterize potential biomarkers associated with the plant.

#### 2. Materials and Methods

#### 2.1 Plant Material

Fresh leaves of *Costus igneus* were collected and authenticated at The Department of Botrany, Ismail Yusuf College. The plant material was dried, ground into a fine powder, and stored under controlled conditions.

#### 2.2 Preparation of Extracts

Ethanolic and Methanolic extracts were prepared by dissolving 100 mg of dried sample in 10 ml of solvents. The extracts were sonicated for 15 min in bath sonicator.

#### 2.3 HPTLC Analysis

The crude extract was analyzed by HPTLC to establish a comparative fingerprint and profile of phytosterols. The HPTLC method was optimized according to ICH guidelines for resolution and reproducibility. The analysis was performed by applying  $2\mu L$ ,  $5 \mu L$  and  $10 \mu L$  of extract on  $20 \times 10$  cm Merck, HPTLC Silica gel  $60 \, F_{254}$  plate with a mobile phase Toluene: Chloroform: Ethanol (4:4:1). Detection was carried out under long wave UV light  $366 \, \text{nm}$ .

#### 2.4 TLC-MS Interface and Phytosterol Separation

The phytosterol extract was applied to a TLC plate and developed under optimized conditions. The plate was interfaced with a Mass Spectrometer to enable the direct elution and analysis of separated compounds. The TLC-MS interface allowed for the in situ detection and characterization of phytosterols.

#### 2.5 Mass Spectrometry Analysis

The separated phytosterols were analyzed using MS to determine their structural characteristics. MS parameters were optimized for sensitivity and specificity. Data were interpreted to elucidate the molecular structures and identify the phytosterols present in *Costus igneus*.

 $\beta$ -sitosterol, this peak is observed at m/z 414. Molecular Ion Peak (M<sup>+</sup>) at 414 confirms the molecular weight of  $\beta$ -sitosterol. The most intense peak at 399 is often due to the loss of water or other small groups from  $\beta$ -sitosterol. Fragment Ions reveals additional peaks at m/z 381, 369, and 345 which provides information on the structure of the side chain and steroid ring.

#### 2.6 Identification and Characterization of Biomarkers

The identified phytosterols were compared with known standards to confirm their identity. Additionally, potential biomarkers associated with the plant's medicinal properties were characterized based on their spectral signatures and biological activity.

#### 3. Results

#### 3.1 HPTLC Fingerprinting

HPTLC analysis provided a detailed fingerprint of phytosterols present in *Costus igneus*. The profiles showed distinct spots corresponding to various phytosterols, with clear separation and reproducibility.

#### 3.2 TLC-MS Separation

The TLC-MS interface effectively separated and detected phytosterols from the plant extract. The interface enabled the precise elution and subsequent analysis of phytosterols by mass spectrometry, revealing distinct peaks for each compound.

#### 3.3 Mass Spectrometry Findings

MS analysis provided structural elucidation of the phytosterols. β-sitosterol was identified based on their mass-to-charge ratios and fragmentation patterns.

#### 3.4 Biomarkers Identification

The study identified several phytosterols as potential biomarkers for *Costus igneus*. Their profiles were compared with reference standards, and their medicinal relevance was discussed.

#### 4. Discussion

#### 4.1 Phytosterol Profile

The profiling of phytosterols from *Costus igneus* revealed a diverse range of sterols with potential medicinal properties. The HPTLC and TLC-MS methods provided complementary information, enhancing the accuracy of phytosterol identification.

#### 4.2 Structural Elucidation

MS studies confirmed the structures of key phytosterols, providing insights into their chemical nature and potential biological activities. The results align with previous studies on phytosterols, supporting their role in the plant's medicinal efficacy.

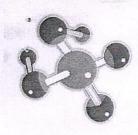
#### 4.3 Biomarker Potential

The identified phytosterols hold promise as biomarkers for the medicinal properties of *Costus igneus*. Further studies are needed to validate their effectiveness in clinical applications and their role in diabetes management.

#### 5. Conclusion

This study successfully profiled and elucidated the structure of phytosterols in *Costus igneus* Nak. Through HPTLC and TLC-MS analysis, we obtained a comprehensive understanding of the phytosterol content and identified key compounds with potential medicinal significance. The results contribute valuable information for the development of therapeutic agents and the characterization of biomarkers related to the plant's medicinal properties.

NO	NEEV RESEARCH PRODUCTS Shop No.7, Kalpaka CHS., Bldg. No. 1, Siddharth Nagar, Road No. 16, Goregaon (W), Mumbai - 400 104. Mobile: 99200 18225
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# **NEEV RESEARCH PRODUCTS**

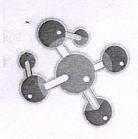
Dealers In: Laboratory & Research Chemicals, Glassware & Instruments Indian & Imported Shop No.7, Bldg, No. 1 Kalpaka CHS. Ltd., Siddharth Nagar, Road No. 16, Goregaon (W), Mumbai - 400 104. Mob. 99200 18225 • Email. neevresproducts07@gmail.com | GSTIN: 27BGSPS0165D1Z6

#### Tax Invoice

To, The Principal, Ismail Yusuf College Jogeshwari East Mumbai - 400 060

Tax Invoice No: 168 / 21-22 Date: 31/03/2022 PO No:-Dtd.

Sr No.	Particulars	HSN Code	Qty	Rate (Per)	Total
1	Sulphuric Acid 98%, Molychem make, Cat no,26400-8 x 500ml	28070010	01	2,080.00	2,080.00
2	N-N Dimethyl Formamide, Molychem make, Cat no. 23035-500ml	29211110	01	468.00	468.00
3	2-Methylpropanol, iso-BUTANOL AR Molychem make, Cat no. 22393 -500ml	29051490	01	351.00	351.00
4	n-buthyl Acetate, Molychem make, Cat no. 22392-500ml	29153300	01	520.00	520.00
5	Ethyl Methyl Keton, Loba make, Cat no.00145	29141200	01	603,00	603.00
6	Ammonia 25%, Molychem make, Cat no.21400-500ml	28142000	01	172.00	172.00
7	Di isopropyl ether, Molychem make, Cat no.60101-500ml	29091900	01	367.00	367.00
8	Nitric Acid, Molychem make, Cat no.24440 - 8x 500ml	28080010	01	2,236.00	2,236.00
9	Sodium Sulphate Anhydrous, Molychem make, Cat no. 26100-500ml	28331990	01	251.00	251.00
10	Petroleum Ether 60-80°, Molychem make, Cat no.32120-2.5 ltr	27101990	01	975.00	975.00
11	Ethyl Acetate , Molychem make, Cat no.23175 - 2.5 ltr	29153100	02	1,105.00	2,210.00
12	n-Butanol, Molychem make, Cat no. 22391-500ml	29051300	05	351.00	1,755.00
13	Acetonitrile, Molychem make, Cat no.21060-500ml	29269000	05	504.00	2,520.00
14	Glacial Acetic Acid, Molychem make, Cat no.21020 -500ml	29152100	10	331.00	3,310.00
15	Parafin Oil, Molychem make, Cat no.16710-500ml	27101990	10	314.00	3,140.00
16	Phenyl Hydrazine, Molychem make, Cat no. 24683-500ml	29280090	.01	3,705.00	3,705.00
				C/F	24,663.00



# **NEEV RESEARCH PRODUCTS**

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#### Tax Invoice

To, The Principal, Ismail Yusuf College Jogeshwari East Mumbai - 400 060

Tax Invoice No: 168 / 21-22 Date: 31/03/2022 PO No:-

Dtd.

Sr No.	Particulars	HSN Code	Qty	Rate (Per)	Tota
				B/F	24,663.00
17	Ortho phenyl Diamine, Molychem Make, Cat no. 16790-250gm	29215110	01	563.00	563.00
18	Mintroleum, Molychem make, Cat no.24290-2.5 ltr	27101990	02	813.00	1626.00
19	Heating Mantle 1000ml, Supertek make	8198990	01	3010.00	3010.00
20	50mm Funnel, Supertek make, Cat no. 15.304.0050	70179090	10	83.30	833.00
21	75mm Funnel, Supertek make, Cat no.15.304.0075	70179090	10	100.00	1001.00
22	10ml Flaks, Supertek make, Cat no.15.108.0010	70179090	10	88.00	875.00
23	25ml Flask, Supertek make, Cat no.15.108.0025	70179090	10	70.00	700.00
24	Pipette Stand, to hold 24 pipettes, Supertek make, Cat no.CH11855/1	39269099	01	427.00	427.00
GSTII	N/UIN: 27BGSPS0165D1Z6		Total		33,698.00
Our Bank A/c No: 002220110000268			CGST @ 9%		3,032.82
Bank	Name: Bank Of India Goregaon (W) Branch			GST @ 9% 3,032	3,032.82
			Less Round Off		0.36
	es Thirty Nine Thousand Seven Hundred a		Final To	tal	39,764.00

Terms: 1) Chemical sold as per bill are for laboratory use only and not for medical.

2) Interest @ 18% will be charged if payment in not received within 30 days.

3) Goods once sold will not be taken back.

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college

Date: 2-0-3-202

Sr. No.	Description	Qty.	Rate	Amount
1	Airtight contenar,	1	236	236
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Annual Communication of the Co				- A - P
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Authorised Signatory