

“Bioprospecting local ferns for phytoremediation of heavy and radioactive metals”

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Executive Summary

Collection and taxonomical identification of ferns:

1. The study was conducted by collection of various fern flora available in this part of the region. Particularly, the ferns were collected from Yedmaru village of Udupi district which is adjacent to the UPCL Thermal Power Plant. Western Ghats possess a large extent of biodiversity over the ecology and biogeography of Peninsular India and extensive sampling was done from the iron ore rich region of Western Ghats - Kudremukh National Park, Karkala. ferns were also collected from the Charmadi Ghats which connects the western part of Dakshina Kannada district to Chikmagalur district. The availability of pteridophytes varies significantly throughout the year and therefore the collection was made in three phases- the pre-monsoon, monsoon and post monsoon season of the year. Around 42 different fern species were taxonomically identified and confirmed and a Book on Pteridophytes of Western Ghats titled "A Guide on Ferns of Kudremukh National Park" was released.

Among the Pteridophytes, 22 ferns based on their availability were used for the preliminary study.

Soil profiling-Physical and Chemical parameters:

The soil samples were collected from the industrially contaminated site and also from the iron ore rich Kudremukh National Park. Various tests to determine its fresh weight, dry weight, pH, Organic Carbon Content, Atomic Absorption Spectroscopy (GBC 6000) for different toxic and non toxic elements like Cadmium (Cd), Chromium (Cr), Lead (Pb), Iron (Fe), Zinc (Zn) and Copper (Cu).

pH of the soil collected showed a range from 4.55 to 6.66. Based on the standards for identifying the soil types, the soil sample collected from Kudremukh showed the minimum stating its extremely acidic nature since it is less than 4.6. The soil samples from the farm were moderately acidic in nature. Overall the soil samples of both regions were acidic in nature.

It is generally assumed that on an average organic matter contains about 58% organic carbon. The % organic content present in 1 g of soil varied from 0.23 to 4.93. The soil samples of the industrially polluted area showed the least organic carbon content indicating poor soil fertility.

The soil samples from both the locations have shown significant amounts of Chromium, Iron and Zinc. The concentration of Cr in soil varied from 1.003 -1.065; Fe: 0.339-0.42 and Zn: 0.372-2.689 ppm. There is no trace of the other three elements tested i.e., Cd, Cu and Pb. The presence of chromium in the soil indicates the extensive pollution with this heavy metal.

Determination of percentage dehydration and heavy metal detection of the ferns under abiotic stress:

Since the soil samples showed the presence of heavy metal like Cr, 22 ferns were checked for their physiological changes in the system. For this, the foliar region (in triplicates) was sampled for the determination of fresh weight, dry weight, percentage hydration, and AAS for the similar elements as that of soil i.e., Cadmium (Cd), Chromium (Cr), Lead (Pb), Iron (Fe), Zinc (Zn) and Copper (Cu).

The ferns showed preferential absorption of some heavy metals. Among the heavy metals Chromium was the most preferred and among the non toxic ones, iron was the highly absorbed metal. The average foliar Cd content varied from 0 - 130 ppm; Cr from 41.63 -581.88 ppm; Pb from 0 -3.94 ppm; Fe from 20.42 to 2010 ppm; Zn from 45.29 – 817.38 ppm and Cu=0. 581.88 ppm/g dry wt. of Chromium had accumulated in the foliar region of *Tectaria coadunata* which was collected from UPCL site (Fig 4) and the highest accumulation of 1792.37 ppm /g dry wt. of iron was observed in an *Athyrium hohenackerianum* collected from iron rich soil of Kudremukh, Western Ghats (Fig 5). The fern samples collected from UPCL region showed preference for Zn and Cr and the ferns from Kudremukh region showed accumulation of Fe and Zinc in their fronds.

Based on the preliminary study, the findings were as follows:

Cr accumulation in the leaf ranged from 41.63 to 581.88 ppm.

- The maximum was 581.88 ppm in the foliar region of *Tectaria coadunata* which was collected from UPCL site.
- The other two good candidate ferns were *Nephrolepis hirsutula* - 324.44 ppm and *Drynaria quercifolia* - 235.42 ppm.

Cd accumulation ranged from 0 to 130 ppm.

- Maximum being 130 ppm in *T.coadunata*, 50.91ppm in *Acrostichum aureum* and 50.83 ppm in *Athyrium hohenackerianum*.
- Laboratory studies on *Pityrogramma calomelanos* had shown a good hyperaccumulation for cadmium upto 70 µg/g dry wt.

Candidate ferns that showed hyperaccumulation



A.hohenackerianum

T.coadunata

D.quercifolia

T.polymorpha





N.hirsutula



P.calomelanos

Publications:

Journal:

Sudha Sajeev, Pavithraraj, Adarsh DB and Smitha Hegde (2015) Phytoconstituents of *Nephrolepis hirsutula* and *Pityrogramma calmelanos*, two medicinal ferns of the Western Ghats. *Indian Fern J*, 2015; 32: 244-256 (SJIR impact factor 2.66)

Shaiesh Morajkar, Sudha Sajeev and Smitha Hegde (2015) “A DNA barcode for *Stenochlaena palustris*: An edible medicinal fern” *Indian Fern J*. 2015; 32: 124-131(SJIR impact factor 2.66)

Shaiesh Morajkar, Sudha Sajeev and Smitha Hegde (2015). Ferns a thriving group of urban dwellers. *Bionature*, 35 (1&2): 13-21.

Smitha Hegde (2014) An Intriguing flora, environmental and ethno botanical significance. A report on National Seminar on Pteridophyta 30:1-9

Book:

1. Smitha Hedge and Sudha Sajeev, 2013. A Guide on Ferns of Kudremukh National Park. Karnataka Forest Department, Government of Karnataka, India.

Book Chapters:

1. Sudha Sajeev, Ramya P.V, Sunitha B.C and Smitha Hegde. Phytoremediation of Cadmium using *Pteris sp.* Prospects in Bioscience: Addressing the Issues. Sabu, Abdulhameed; Augustine, Anu (Eds.) 16 (131) 2013. ISBN 978-81-322- 0809-9 Springer India Publications.
2. Sudha Sajeev, Aparna A, Rajani M. Nayak, Lobsang Tsultrim, Shimju K.T, Melo J.S, Smitha Hegde. Screening Native Ferns for Heavy Metal Accumulation-A Preliminary Study. Biodiversity Conservation: Challenges and Prospects, 2013. ISBN: 978-93-5104-266-2

Poster Presentations:

1. Ramya P.V, Sunitha B.C and Smitha Hegde, Phytoremediation of Cadmium Using *Brassica juncea* and *Nephrolepis spp.* National Seminar “Biocatalysts” 2012, Organized by Department of Biochemistry, St.Aloysius College, Mangalore, India.
2. Sudha Sajeev, Ramya P.V, Sunitha B.C and Smitha Hegde, Phytoremediation of Cadmium using *Pteris spp.* International Conference on Advances in Biological Sciences (ICABS), 15-17 March 2012, Kannur University, Kannur, India.
3. Renu Kashyap, Sudha Sajeev and Smitha Hegde, Identification of Juvenile Forms of Ferns Using Molecular Markers. A National Seminar on Pteridophyta: An Intriguing Flora.Environmental and Ethnobotanical Significance,8-9 February, 2013, St Aloysius College, Mangalore, India.
4. Sudha Sajeev, Aparna A, Rajani M. Nayak, Lobsang Tsultrim, Shimju K.T, Melo J.S, Smitha Hegde, Bioprospecting ferns of the local flora suitable for Phytoremediation of heavy metals. DAE-BRNS LIFE SCIENCES SYMPOSIUM 2012 (LSS-2012) on Trends in Plant, Agriculture and Food Sciences (TIPAFS), 17-19 December 2012, BARC, Mumbai, India.
5. Nikhil R.Halarnkar, Hermann Fernandes, Poojitha G, Sudha Sajeev and Smitha Hegde “ DNA Barcoding of Pteridophytes of Western Ghats” at National Seminar on Trends in Environmental Biotechnology and its Applications, 10-11 December 2013, pg 55. Organized by Departments of Biotechnology and Botany, St Aloysius College, Mangalore.

Research presentations as abstracts:

1. Smitha Hegde, Sudha Sajeev and L.D'Souza on Tissue culture of Ferns at 101st Indian Science Congress, Jammu, 3-7 February 2014. Organized by University of Jammu-The Indian Science Congress Association, Kolkata.
2. Raksha Thabitha Maben, Vranda Rao, Sudha Sajeev and Smitha Hegde "In-vitro germination and sporophyte induction studies of *P.calomelanos*-Candidate fern for phytoremediation" at National Seminar on Trends in Environmental Biotechnology and its Applications, 10-11 December 2013, pg 54, Organized by Departments of Biotechnology and Botany, St Aloysius College, Mangalore.

Trainings/Seminars attended:

1. International Conference on Advances in Biological Sciences (ICABS) organized by Kannur University, Kannur. 15-17th March 2012.
2. Training on Real Time PCR organized by Life Technologies-Applied Biosystem. 29-31st August, 2012.
3. Training programme on Genetic Diversity and Mapping Analysis Using Molecular Markers organized by CPCRI, Kasargod, Kerala. 15-20th October, 2012.
4. National Level Seminar on DNA Barcoding: Its Power and Practice organized by the Loyola Centre for Research & Development, St Xavier's College Campus, Ahmedabad. 7-8th December, 2012.
5. DAE-BRNS Life Sciences Symposium (LSS-2012) on Trends in Plant, Agriculture and Food Sciences (TIPAFS) organized by BARC, Mumbai. 17-19th December, 2012.
6. National Seminar on Pteridophyta : An Intriguing Flora. Environmental and Ethnobotanical Significance organized by Department of Botany and PG Department of Biotechnology, St Aloysius College in collaboration with Indian Fern Society. 8- 9 February, 2013.
7. Visit to Division of Nuclear Agriculture and Biotechnology (NA&BTD), BARC, Mumbai from 7-13 October, 2013 for experiments on Uranium absorption and Irradiation.
8. National Seminar on Trends in Environmental Biotechnology and its Applications, organized by Departments of Biotechnology and Botany, St Aloysius College, Mangalore, 10-11 December, 2013.