Pharmacognostic Investigations and Antimicrobial Activity on *Crepe Ginger*

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Abstract: Crepe Ginger is known as Cane-reed, Malay ginger, Spiral flag, spiral gingerin India the tree flowers at the end of the cold season and the commencement of the hot weather. In India some of the trees bloom the whole year round, biological named as Hellenia speciosa belonging to the family Costaceae. Crepe Ginger e grows to a height of 9-12 m. Stem is generally crooked wood intensely hard, the branches knotty and bark deeply furrowed. The dense crown of close-growing foliage gives the tree a rounded, compact, net appearance. Each leaf is composed of 2 or 3 pairs of smooth, stalk less leaflets arranged on a slender mid-rib. The leaflets are 6-13 cm in length. Present work was carried out on pharmacognostical studies on leaf of Crepe Ginger includes T. S, stomatal index, vein islet and termination no., palisade ratio, and powder analysis, moisture content, ash value, extractive value for leafy sample. Certain microscopical characteristics like stomata, trichomes, calcium oxalate crystals, starch grains, stone cells, fibers, vessels are important anatomical characteristics of organized drugs. The powder characters were Parenchyma Cells, Oil globules, Non glandular trichomes, Glandular trichomes, Epidermal Cells, stomata, Phloem Fibers, Spiral xylem vessel. Plant shows vital role in Antimicrobial activity in aqueous extract of the plant.

Keywords: Crepe Ginger, Pharmacognostical studies, Powder microscopy, Antimicrobial activity

1. Introduction

Propagation of Crepe Ginger is by dividing the Rhizome. Plant the Rhizome in well-drained soil and semi-shaded location. Keep the soil moist. Fertilize periodically. Crepe ginger is only a distant relative of the edible ginger family. It is a tall and dramatic landscape plant with large dark green leaves arranged on the stalk in a spiral. This Costus can grow to 10 ft tall in frost-free areas, but is typically small as a potted plant. The flowers appear in late summer or early fall, and are quite unusual looking. They form on red 4 in cone-shaped bracts, with several 2 in pure white crinkled flowers protruding from each cone. The flowers look like crepe paper-thus the common name of crepe ginger. After the flowers fade away, the attractive red cone-shaped bracts remain. The large creepy object is not the petal, but the stamens-the three true petals of each flower are inconspicuous, and arealmost hidden by the bell shaped stamen.



Leaf

Stem/ Bark

Crepe Ginger

Medicinal uses: Crepe Ginger has many historical uses in Ayurveda, where the rhizome has been used to treat fever, rash, asthma, bronchitis, and intestinal worms. It is mentioned in the Kama Sutra as an ingredient in a cosmetic to be used on the eyelashes to increase sexual attractiveness.

2. Materials and Methods

Plant material the leaves, stem & Bark (Aerial part of the plant) were collected from the species of Costus. The air dried and powered drug (500g) were extracted with petroleum ether for 3-4 hours at $60-80^{\circ}$ C in a Soxhlet apparatus. Different concentrations of plant extracts were

evaluated for its Pharmacognostic study & antimicrobial activity.

Pharmacognostic study: Authenticated samples of dried leave, stem bark of Crepe Ginger were subjected to morphological and Microscopical examination. Quantitative standards like Moisture content, Total Ash value, Acid insoluble ash value, water soluble ash value, alcohol soluble extractive values and water soluble extractive values were determined.

Morphology: Leaves are membranous or chartaceous, ovate or orbicular, 20-60 x 15-70 mm, apex acute, base cordate, rounded, truncate or shortly cuneate, not decurrent on petiole, margin regularly or irregularly serrate, sparsely to densely hispid both sides, with minute sessile glands beneath, reticulate veins prominent beneath, petioles 5-60 mm long, hispid, peduncle sessile to 15 mm long, pubescent with short hairs, 4-5 mm long, obliquely campanulate in fruit, 8-12 mm long. The leaves showed multicellular trichomes, non-glandular trichomes, vascular bundles, collenchyma cells, parenchyma cells and laminar region.

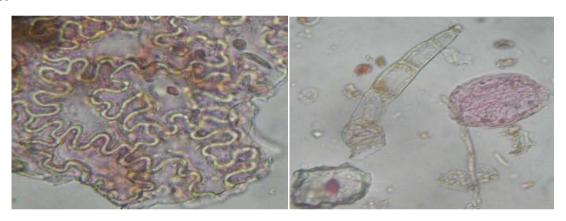
3. Methodology & Results

Table 1: Physico chemical parameters								
Sample	Moisture	Total Ash	Acid insoluble	Water soluble	Water soluble	Alcohol soluble		
Identity	Content %	%	ash %	ash %	Extractive value %	Extractive value%		
Leaves	3.25	8.98	1.00	6.5	9.13	8.86		

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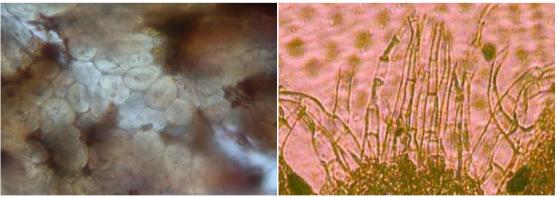
Table 2: Leaf constant values							
Sample Identity	Stomatal no.	Stomatal index	Vein islet no.	Vein termination no.	Palisade ratio		
Leaves	5	20-21	5	6	5		

Microscopy of Leaves (T. S.)



Epidermal Cells (Surface view)

Trichomes and stomata

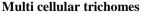


Phloem Fibers

Anti Microbial Study:

Test organisms

organisms Pseudomonas aeruginosa, Four test Staphylococcus aureus, Escherichia coli, Bacillus subtilis, Streptococcus pyogenes where obtained from RB Science, Bhopal, M. P. Cultures of test organisms were maintained on nutrient agar slants and sub-cultured in petri dishes prior to



testing. The readymade medium (Hi-media, 23g) was suspended in distilled water (1000ml) and heated to boiling until it dissolved completely. The medium and petridishes were autoclaved at 15Ps if or 20min. Stock solutions were prepared by dissolving plant extract in DMSO (dimethylsulphoxide) and different concentrations were made (50-2000µg).

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Minimum inhibitory concentration (MIC)

Test MIC test was performed by broth dilution method. Different concentrations ($200\mu g/ml$, $400\mu g/ml$, $600\mu g/ml$, $800\mu g/ml$, $1000 \ \mu g/ml$) of aerial part of the plant extract were prepared in 1ml working solution of extract (2mg/ml) was added to the test tube1, containing1ml of nutrient broth. After thorough mixing, 1ml of solution was transferred to second test tube and the process was continued for succeeding transfers. The last test tube received no test solution and served as control (DMSO). For both tests, culture was kept in an incubator at $26^{\circ}c$ and results were observed after 24 h and 3-4 replicates were maintained for each treatment.

Anti microbial Activity:

For antimicrobial activity Agar well diffusion assay (zone inhibition assay) was used. This is the most widely used method for determining the susceptibility of organisms to antimicrobials. In case of plant extract screening this method is widely applicable because turbidometry is very difficult to do when the extracts impart color to the broth. The extracts obtained from the plants were used for studying their antimicrobial activity. A loop full of bacterial strain was inoculated in 30 ml of Nutrient broth in a conical flask and incubated for 24 hrs for bacterial culture at 37°C to get

active strain. In the molten Mueller Hinton agar around 0.2 ml (OD 0.04) of test strains were inoculated, Care was taken to ensure proper homogenization. The media was poured in sterile petriplates. After the medium solidified, a well was made in the plates with sterile borer (7 mm). The extract compound (100 µl) was introduced into the well and plates were incubated at 37°C for 24 hrs for bacterial cultures at 37° C. Aseptic conditions were maintained by swabbing and cleaning the work area with alcohol and the entire procedure was carried out in between the burners. All samples were tested in triplicates. The plates were kept for pre-diffusion before keeping them for incubation. A control with DMSO was kept for all test strains as it was the diluents used for reconstituting the extracts so as to ascertain that it is not responsible for the activity demonstrated if any. The plates were observed for the presence of inhibition of bacterial growth that was indicated by the clear zone around the wells. The size of the zone of inhibition was measured and the antibacterial activity was expressed in terms of the average diameter of zone inhibition in millimeters. The absence of zone inhibition was interpreted as the absence of activity.

4. Observations

Name of Microorganism	200 µg/ml	400 µg/ml	600 µg/ml	800 µg/ml	1000 µg/ml	Ctrl DMSO
Pseudomonas aeruginosa	11	11	13	14	14	
Staphylococcus aureus	8	8	9	10	12	
Escherichia coli	8	10	11	16	18	
Bacillus subtilis	8	8	9	9	11	
Streptococcus pyogenes	6	7	7	8	9	

5. Conclusion

Present work was carried out on Pharmacognostical studies on leaf of Crepe Ginger, T. S, powder microscopy of leaf, physicochemical parameters and Anti-microbial study were carried out.

6. Future Plans

Though several of its constituents have been isolated and characterized, there is ample scope for further isolation from the plant.

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