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REVIEW ARTICLE

MOLECULAR MECHANISMS AND THERAPEUTIC POTENTIAL OF RESVERATROL

Alejandra Torres-Flores[1,2], Ana-Lucia Novelo-Aguirre[1,2], Sofia-Joseline Sandoval-Villagrana[1,2], Tania González-Cortés[2], Iraís Castillo-Maldonado[2], Dealmy Delgadillo-Guzmán[1], Cecilia Hernández-Morales[3], Agustina Ramirez-Moreno[4], Erika Flores-Loyola[4], Joaquín Avalos-Soto[5], Miguel Ángel Téllez-López[5], Gladis Michel-Ramírez[2] and David Pedroza-Escobar*[2]

[1]Facultad de Medicina. Universidad Autonoma de Coahuila. Unidad Laguna. Torreon, Coahuila, 27000, Mexico., [2]Centro de Investigacion Biomedica. Universidad Autonoma de Coahuila. Unidad Laguna. Torreon, Coahuila, 27000, Mexico., [3]Facultad de Odontologia. Universidad Autonoma de Coahuila. Unidad Laguna. Torreon, Coahuila, 27000, Mexico., [4]Facultad de Ciencias Biológicas. Universidad Autonoma de Coahuila. Unidad Laguna. Torreón, Coahuila, 27275, Mexico. [5]Cuerpo Académico Farmacia y Productos Naturales, Facultad de Ciencias Químicas, Universidad Juárez del Estado de Durango, Gómez Palacio, Mexico
Email: dpedroza@uadec.edu.mx

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Abstract: Trans-resveratrol is a polyphenol that belongs to the stilbene family. In nature, resveratrol works primarily as a phytoalexin., it is synthesized by more than 70 plant species as a primary defense mechanism against environmental stressors, such as prolonged exposure to ultraviolet radiation or fungal and bacterial infections. Among the richest botanical sources is the *Vitis vinifera* plant whose fruits store this compound mainly in the skin of the grapes. Berries, blueberries and cranberries are a known source of resveratrol. *Polygonum cuspidatum* is a plant used as a source of resveratrol extraction for use in dietary supplements. Other sources of resveratrol include various trees like eucalyptus and floral species such as lilies. Several studies have documented different medicinal effects of resveratrol on human health, including its antioxidant, anti-inflammatory, cardioprotective, and neuroprotective properties. For human consumption, this antioxidant is available in a variety of everyday foods, especially red and purple fruits, peanuts, grapes and red wine. The maceration process of red wine, which includes the skin, seeds, and woody parts of the plant, facilitates a very high concentration of this compound, giving it great nutritional value. On a structural level, resveratrol is composed of two aromatic rings linked by a carbon-carbon double bond, allowing it to exist in cis and trans configurations. The trans form is the most stable and, therefore, the most used in health research. Its synthesis occurs through three condensation reactions mediated by the enzyme stilbene synthase. **Conclusions:** Trans-resveratrol is a naturally occurring polyphenolic stilbene synthesized by numerous plant species as part of their defense mechanisms against environmental stress conditions. It is found in human foods such as grapes, berries, peanuts, and red wine. Several studies have documented its potential benefits for human health.

Keywords: Medicinal properties, Resveratrol, Stilbene, Phenolics, Trans-resveratrol

INTRODUCTION

Resveratrol possesses multiple medicinal properties on human health. The five major biological activities are antioxidant, enhancer of enzyme activity, anti-inflammatory, cardioprotective and neuroprotective. First, it acts as a powerful antioxidant capable of neutralizing free radicals, inhibiting lipid peroxidation in cell membranes, and modulating oxidative stress at the mitochondrial level (Frémont *et al.*, 2000). Resveratrol enhances the activity of endogenous antioxidant enzymes and protects LDL and VLDL lipoproteins from oxidation (Draczynska-Lusiak *et al.*, 1998). It also has anti-inflammatory effects, as it inhibits the enzyme cyclooxygenase-2 (COX-2) and reduces the production of pro-inflammatory mediators such as nitric oxide and various cytokines such as TNF-α and interleukin-1β (Meng *et al.*, 2021., Martínez-Canabal *et al.*, 2005). The cardioprotective properties of resveratrol have been documented through several studies that report its ability of inducing vasodilation by increasing nitric oxide, preventing cardiac fibrosis, improving the lipid profile by reducing LDL cholesterol and triglycerides, and lowering blood pressure (Bonnefont-Rousselot *et al.*, 2016). Also, the neuroprotective properties are attributed to resveratrol since it directly combats brain oxidative stress and prevents the accumulation of pathological proteins associated with degenerative diseases, facilitating the cleaning of beta-amyloid peptides in Alzheimer disease and alpha-synuclein in Parkinson disease, in addition to activating Sirtuin 1 to improve synaptic plasticity (Rahman *et al.*, 2020).

*Corresponding Author

SOURCES OF RESVERATROL

Trans-resveratrol (3,4,5-trihydroxystilbene) is a polyphenol that belongs to the stilbene family (Soleas *et al.*, 1997). Resveratrol is an antioxidant produced by more than 70 plant species in response to situations of stress due to ultraviolet radiation or fungal infections (Maier-Salamon et. al., 2011). One of the richest sources is the *Polygonum cuspidatum* plant, whose root extracts are used in traditional Chinese and Japanese medicine., it has also been documented in trees such as eucalyptus, spruce, and the tropical deciduous tree *Bauhinia racemosa* (Soleas *et al.*, 1997).

Resveratrol is found in flowering plants such as lilies (*Veratrum grandiflorum* and *Veratrum formosanum*) and in the legume *Pterolobium hexapetalum* (Kumar *et al.*, 1988). A high content is present in the leaves when the plant is damaged by chemical treatments or subjected to stress situations (Soleas *et al.*, 1997., Maier-Salamon *et. al.*, 2011).

Resveratrol is present in several fruits that are part of the human diet, mainly in red and purple fruits such as blueberries (*Vaccinium spp.*), currant (*Vaccinum spp.*), and blackberries (*Morus spp.*). It can also be found in citrus fruits, apples, and peanuts (*Arachis hypogaea*). Another food rich in resveratrol is red wine (Maier-Salamon et. al., 2011), which provides the highest resveratrol content to human diet (Gambini *et al.*, 2013).

The resveratrol content in wine comes from the vines (*Vitaceae*), specifically from the skin, seeds, petioles, and woody parts (stems and peduncle), which are the richest in this compound (Gambini *et al.*, 2013). Because of this, red wine is richer in resveratrol than white wine, since the parts of the grape containing resveratrol are macerated during the red winemaking process (Gambini *et al.*, 2013).

CHEMICAL STRUCTURE OF RESVERATROL AND MECHANISMS OF ACTION

Resveratrol is a natural polyphenol. Figure 1 shows its structure which consists of two aromatic rings joined by a C-C double bond, generating 3,5,4′-trihydroxystilbene (Tian *et al.*, 2020). Because of the double bond, resveratrol has cis and trans configurations, the last one is the most used in health research, since this form is more stable (Zhang *et al.*, 2021).

In plants, resveratrol is synthesized as a defense mechanism mainly against fungal or bacterial infection or stress events (Koushki *et al.*, 2018). Furthermore, in plants it can be found in glycosylated form, which gives it bioavailability by having a better solubility and better partition coefficient, which facilitates the entry of polyphenols into enterocytes (Zhang *et al.*, 2021).

Resveratrol is a white powder that is synthesized through stilbene synthase via three consecutive condensation reactions between three molecules of malonyl-coenzyme A and one molecule of coumaroyl-coenzyme A (Perrone *et al.*, 2017).

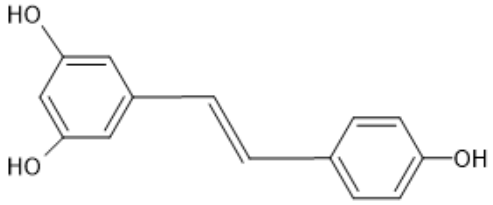

Figure 1. Chemical structure of resveratrol.

Some authors consider that the effects attributed to resveratrol on various pathological conditions, including skin aging, wound healing, fibrosis (Hecker *et al.*, 2022), endometriosis (Sienko *et al.*, 2023), and several types of cancer, are mainly associated with the antioxidant and anti-inflammatory properties of this compound. Delayed tissue repair, insufficient vascularization, infections, and prolonged inflammatory responses are among the major factors involved in these pathological conditions, all of which contribute to impaired wound healing. In this context, resveratrol has attracted considerable attention due to its potential to promote anti-inflammatory, antioxidant, and angiogenic effects (Hecker *et al.*, 2022).

Several studies suggest that resveratrol promotes the expression of vascular endothelial growth factor (VEGF) (Khanna *et al.*, 2002) and the activation of silent mating type information regulation 2 homolog 1 (SIRT1), both of which are regulatory molecules involved in angiogenesis (Huang *et al.*, 2019). In particular, in vitro and in vivo models (HUVECs and db/db mice) have demonstrated an overexpression of forkhead box O1 (FOXO1), a transcription factor regulated by SIRT1 (Huang *et al.*, 2019., Christovam *et al.*, 2019).

Regarding the mechanisms underlying the resveratrol antioxidant activity, the hydroxyl (OH) groups present in the aromatic rings of resveratrol play a fundamental role. These groups are capable of donating electrons or hydrogen atoms to stabilize free radicals before they induce oxidative damage to biomolecules such as lipids, proteins, or DNA. A simplified representation of this type of chemical reaction is shown below.

$$ROO\bullet + ArOH \rightarrow ROOH + ArO\bullet$$

where:

ROO• = peroxyl radical, one of the major reactive species involved in lipid peroxidation.

ArOH = aromatic phenolic group (Ar, i.e., aromatic ring., OH, i.e., hydroxyl group).

ROOH = stabilized hydroperoxide product.

ArO• = phenoxy radical stabilized by aromatic resonance.

In this antioxidant context, although the exact mechanism of action has not yet been fully elucidated, resveratrol has been associated with the regulation of superoxide dismutase (SOD), glutathione peroxidase (GPX), and catalase (CAT),

as well as with an increase in Nrf2 mRNA concentration and activation of the NF-E2-related factor 2 (Nrf2)-mediated antioxidant signaling pathway (Roshani *et al.*, 2022., Chiou *et al.*, 2011) as shown in figure 2.

Additionally, resveratrol has been associated with the modulation of inflammatory signaling pathways, particularly through inhibition of nuclear factor kappa B (NF-κB) (Xu *et al.*, 2018) activation and reduction of pro-inflammatory mediators such as tumor necrosis factor alpha (TNF-α) (Wang *et al.*, 2020), interleukin-1β (IL-1β) (Malaguarnera, 2019), nitric oxide (NO) (Xia *et al.*, 2014), and cyclooxygenase-2 (COX-2) (Yi *et al.*, 2011), thereby contributing to attenuation of chronic inflammatory responses.

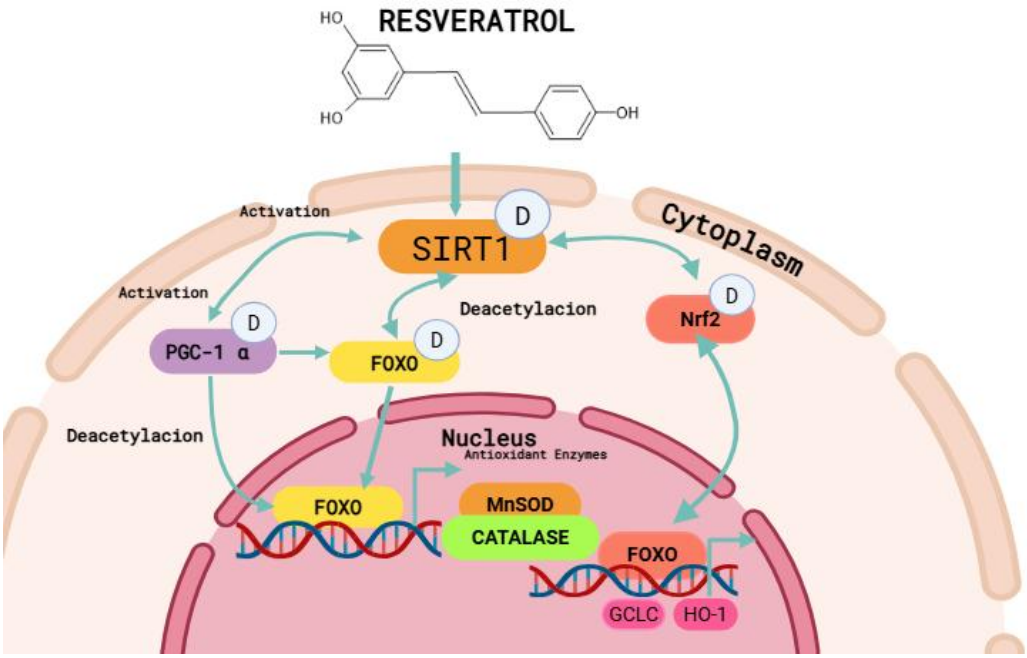

Figure 2. Resveratrol antioxidant pathway. Resveratrol exerts part of its antioxidant activity by activating SIRT1, an NAD+-dependent deacetylase that regulates multiple transcription factors related to cellular redox homeostasis. SIRT1 activation promotes the deacetylation of regulatory proteins such as FOXO, PGC-1α, and Nrf2, facilitating their transcriptional activity. Consequently, these factors migrate to the nucleus and induce the expression of various antioxidant genes, including manganese superoxide dismutase (MnSOD), catalase (CAT), heme oxygenase-1 (HO-1), and catalytic glutamate-cysteine ligase (GCLC). The result is an increase in cellular antioxidant capacity, a decrease in reactive oxygen species (ROS), and greater protection against oxidative stress.

MEDICINAL PROPERTIES OF RESVERATROL

Antioxidant

Resveratrol is recognized for its potent antioxidant activity, as its protective effect is based on its ability to neutralize free radicals and prevent oxidative tissue damage (Tan *et al.*, 2019).

One of its mechanisms is the inhibition of lipid peroxidation in cell membranes, which helps preserve their structural and functional integrity. By protecting membranes, resveratrol reduces the harmful effects of oxidative stress on living cells (Frémont *et al.*, 2000).

Additionally, resveratrol exerts antioxidant effects at the mitochondrial level. It has been shown in isolated brain mitochondria that compounds with estrogenic activity can inhibit mitochondrial respiration and the activity of complex III of the electron transport chain by competing with coenzyme Q. This effect is relevant, since this complex is an important source of reactive oxygen species (ROS). Therefore, its antioxidant action depends not only on the uptake of unpaired electrons but also on the direct modulation of free radical-generating sites (Zini *et al.*, 1999).

On the other hand, resveratrol can increase the activity of endogenous antioxidant enzymes and decrease the production of ROS, strengthening the cellular defense systems against oxidative stress. In this context, it has been observed that the oxidation of lipoproteins such as LDL and VLDL contributes to tissue damage and cell death due to their internalization into cytotoxic forms (Draczynska-Lusiak *et al.*, 1998).

Antiinflammatory

The inflammatory response is an adaptive mechanism that begins with the detection of molecular signals of exogenous and endogenous origin, which can be triggered by signs of danger, such as infections or tissue injury (Herold *et al.*, 2019). These molecular signals are recognized by

pattern recognition receptors (PRRs), such as Toll-like receptors (TLRs), triggering intracellular signaling cascades, such as kinases and transcription factors. These signaling pathways can promote the production of a variety of inflammatory mediators such as cytokines (interleukin (IL-1β) and tumor necrosis factor-α (TNFα)), thus promoting the development of inflammation (Meng *et al.*, 2021).

Within these mechanisms, a key enzyme is cyclooxygenase-2 (COX-2), an inducible isoform that is expressed in response to different inflammatory stimuli (Martínez-Canabal *et al.*, 2005). COX-2 catalyzes the conversion of arachidonic acid into prostaglandins, signaling molecules that regulate inflammation by acting as autocrine and paracrine messengers (Martínez-Canabal *et al.*, 2005., García Meijide *et al.*, 2000).

In this regard, bioactive compounds such as resveratrol exert anti-inflammatory effects by modulating different points of these pathways. Resveratrol has been reported to be able to inhibit the production of pro-inflammatory mediators, such as nitric oxide (NO) and tumor necrosis factor-alpha (TNF-α) in lipopolysaccharide-stimulated Kupffer cells, which contributes to attenuating the inflammatory response associated with oxidative damage (Kawada *et al.*, 1998).

Likewise, experimental studies have shown that resveratrol supplementation can regulate cytokine expression, promoting an anti-inflammatory profile. In *Fabius Maximus* fish, the administration of 500 mg/kg of feed increased the levels of transforming growth factor β (TGF-β) mRNA while inhibiting the expression of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-8 (Meng *et al.*, 2021).

Finally, recent in silico evidence suggests that resveratrol can be biotransformed by the gut microbiota into metabolites such as stilbendiol and stilbenol, which not only retain their biological activity but could also have a greater affinity for the COX-2 enzyme, a relevant therapeutic target in chronic inflammatory diseases (Niño Sánchez *et al.* 2025).

Cardioprotective

Among cardioprotective effects attributed to resveratrol is its ability to attenuate pathological cardiac remodeling. This effect has been associated with the prevention of cardiac fibrosis and maladaptive hypertrophy by preserving regulatory proteins such as phosphatase and tensin homologue (PTEN) and the modulation of mitogen-activated protein kinase (MAPK) signaling pathways, including extracellular signal-regulated kinase (ERK1/2), thereby limiting abnormal growth and dysfunction (Bonnefont-Rousselot *et al.*, 2016).

Resveratrol can improve vascular health by regulating nitric oxide. Evidence shows a low incidence of cardiovascular diseases in the French population even though the French diet is high in saturated fats., this phenomenon is attributed to the continued moderate consumption of red wine rich in polyphenols such as resveratrol (Bonnefont-Rousselot *et al.*, 2016).

It is important to highlight that resveratrol has the ability to increase the availability of nitric oxide, which consequently promotes vasodilation, as well as having an antisclerotic effect since it modifies the lipid profile by decreasing triglycerides and LDL cholesterol, even by increasing HDL, it also inhibits the migration of vascular smooth muscle cells, thus reducing the formation of atheromatous plaque (Gal *et al.*, 2021). Several human studies have shown that at high doses (>150 mg/day), resveratrol reduces systolic blood pressure (Gal *et al.*, 2021).

Neuroprotective

Resveratrol has antioxidant and anti-inflammatory properties and therefore acts as a neuroprotectant by reducing the accumulation of toxic proteins and oxidative stress in the brain (Rahman *et al.*, 2020).

The antioxidant action, in which enzymatic modulation occurs, reduces the concentrations of inducible nitric oxide synthase and increases the production of hemooxygenase-1 (HO-1), strengthening the antioxidant defenses of the neurons (Rahman *et al.*, 2020).

The modulation of pathological proteins of various diseases such as Alzheimer disease facilitates the cleaning of beta-amyloid peptides, reduces the formation of senile plaques, and promotes the non-amyloidogenic cutting of the amyloid precursor protein. In Parkinson disease resveratrol prevents the accumulation of alpha-synuclein, a protein that affects neuronal movement and function. In multiple sclerosis, resveratrol helps to maintain healthy nerve cells and contributes to myelin repair (Sánchez-Nieto *et al.*, 2023).

Resveratrol has been associated to the activation of the enzyme Sirtuin 1, which contributes to reducing apoptosis in hippocampal cells, facilitating neurogenesis, and improving synaptic plasticity, as well as improving mitochondrial function, providing greater energy efficiency to neurons (Vivar-Altamirano *et al.*, 2024).

Anti-aging

Resveratrol has been reported to have protective effects against UVB-induced photoaging by reducing the expression of matrix metalloproteinases (MMPs) and inflammatory factors. This is achieved through the inhibition of MAPK and COX-2 signaling pathways, as well as the inhibition of caspase activation and by acting on VEGF-β to induce anti-apoptotic effects (Cui *et al.*, 2022).

Similarly, resveratrol restores the cell cycle by activating autophagy and reducing reactive oxygen species (ROS) through adenosine monophosphate-activated protein kinase (AMPK) phosphorylation to mitigate the effects of UVA-induced photoaging (Xia *et al.*, 2024).

One of the effects of aging is the loss of bone volume, which is reflected in a greater likelihood of

bone damage and fractures. It has been found that the administration of resveratrol in old rats increases bone volume, as well as a greater number of bone trabeculae and less space between them. Consequently, resveratrol has been reported to promote osteoblast proliferation (Fernández-Tresguerres *et al.*, 2014).

On the other hand, although resveratrol is widely recognized as an anti-aging agent, its bioavailability is low, which is why modifications to resveratrol have been explored. Resveratrol modified with a 4-hydroxy-3′-trifluoromethoxy group has been reported to have increased accumulation in inflammatory cells. In a group of mice aged by D-galactose, mice treated with resveratrol modified with a 4-hydroxy-3′-trifluoromethoxy group were shown to improve age-related changes in the hippocampus and cholinergic system dysfunction by inhibiting NO production, reducing ROS, and inhibiting oxidative cytotoxicity (Liang *et al.*, 2024).

CONCLUSION

Resveratrol is a pleiotropic molecule with immense therapeutic potential against chronic diseases.

Resveratrol has the ability of intervening in multiple cellular targets, from the direct modulation of inflammatory pathways to the protection of mitochondrial function and the activation of enzymatic pathways linked to cellular longevity, such as Sirtuin 1.

However, the documented medical literature also reveals a fundamental biomedical challenge for its large-scale clinical implementation due to low natural bioavailability in the body. Although its glycosylated form partially improves cellular absorption, its rapid metabolism limits its *in vivo* effects. Ultimately, resveratrol represents an invaluable bridge between natural nutrition and the biotechnological development of the future.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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RESEARCH ARTICLE

DIMERIA RAJANIANA (POACEAE): A NEW SPECIES FROM THE COASTAL WETLANDS OF NORTHERN KERALA, INDIA

M.K. Ratheesh Narayanan[1]*, C.N. Sunil[2], Sidharth Sathyan Nair[3], K. Chaithra[3] and M.K. Lakshmi Nandana[4]

[1]Department of Botany, Payyanur College, Edat P.O., Kannur – 670 327, Kerala, India
[2]S.N.M. College, Maliankara P.O., Ernakulum – 683 516, Kerala, India
[3]PG and Research Department of Botany, Sir Syed College, Karimbam P.O, Taliparamba, Kannur – 670 142, Kerala, India (Affiliated to Kannur University)
[4]School of Agriculture, Lovely Professional University, Phagwara, Punjaab, IN-144411
Email: drratheeshpoduval73@gmail.com

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Abstract: A new species of the genus *Dimeria* from northern Kerala, India, *Dimeria rajaniana* Sunil, Ratheesh & Sidharth, is described. A comprehensive taxonomic account, including its distribution, ecology, and comparison with related taxa, is provided, along with photographic plates of the habit and dissected floral parts to facilitate identification.

Keywords: Coastal, *Dimeria*, Kannur, Kerala

INTRODUCTION

The genus *Dimeria* R.Br. (1810:204), belonging to the family *Poaceae* (Tribe Andropogoneae), is a relatively less explored grass genus distributed across tropical and subtropical regions of the Old World. Members of the genus occur widely in Tropical Asia, extending from India to China, Japan, Korea and the Philippines, and also in Sri Lanka, Northern Australia and Madagascar (Bor, 1953; Teerawatananon *et al*., 2014; Kiran Raj *et al.*, 2015; Naik *et al*., 2016). According to Plants of the World Online (POWO, 2025), the genus currently comprises about 57 accepted species worldwide.

India represents one of the major centres of diversity for the genus, particularly the Peninsular region, where a large proportion of the species occur. Recent taxonomic assessments indicate that about 39–42 species are distributed in India, nearly half of which are endemic to the country (Prasanna *et al*., 2020; Nagaraju *et al.*, 2023). The Western Ghats region, especially the state of Kerala, contributes significantly to this diversity, with about 23 species along with several infraspecific taxa reported from the state.

Species of *Dimeria* are morphologically characterized by their distinctive inflorescence consisting of usually paired, divergent racemes bearing solitary, laterally compressed spikelets. The spikelets are with a cylindrical callus, keeled glumes and margins lacking auricles at the apex (Sreekumar & Nair, 1991; Kellogg, 2015; Kiran Raj *et al*., 2015a). Based mainly on rachis and pedicel characters, Bor (1953) proposed an infrageneric classification of the genus into three sections: *Dimeria* sect. *Annulares* Bor., sect. *Capillares* Bor. and sect. *Loriformes* Bor., covering species from India, Sri Lanka and Myanmar. Subsequently, Kiran Raj *et al.* (2015b) established an additional section, *Dimeria* sect. *Dimeria*, based on the type species *Dimeria acinaciformis* R.Br.

Despite several taxonomic treatments and regional floristic accounts (Hooker, 1897; Fischer, 1934; Blatter & McCann, 1935; Cooke, 1958; Bor, 1952, 1960; Kulkarni, 1988; Bhat & Nagendran, 2001), the genus remains incompletely explored, particularly in the diverse ecosystems of the Western Ghats. Continued floristic explorations in this region frequently reveal populations that do not conform to any previously described taxa.

MATERIALS AND METHODS

During recent botanical explorations in the coastal mangrove-associated wet paddy fields of northern Kerala, on 6th January 2026, the authors collected an unusual population of *Dimeria*. Detailed examination of the specimens, together with a critical review of relevant taxonomic literature and comparison with herbarium collections including MH, CALI, CAL and K indicated that the material represents a distinct

*Corresponding Author

and previously undescribed species belonging to *Dimeria* sect. *Loriformes*. Fresh materials were collected from coastal wetlands of Edat of Kerala State, India and herbarium specimens were prepared following international standards (Vogel, 1987; Forman and Bridson, 1989). Additionally, the specimens were preserved in FAA solution for subsequent microscopic examinations. All measurements and morphological character assessments were carried out using fresh samples. Photomicrographs were taken with trinocular stereo zoom microscope (Nikon, SMZ-45T). The species is described and illustrated here as *Dimeria rajaniana*, and its morphological characters are compared with closely allied species.

RESULT

Taxonomic treatments: ***Dimeria rajaniana*** Sunil, Ratheesh & Sidharth ***sp. nov***. (Fig. 1).

Type: — INDIA, Kerala, Kannur Distr., Poruni Vayal: Kunhimangalam, 12°5'39" N 75°13'49" E, alt. ±1 m MSL), 06 January 2026, *Ratheesh, Sidharth & Sunil* 0051 (Holotype: MH!; Isotypes CALI!, SNMH!).

Diagnosis:

The new species is morphologically similar to *Dimeria copeana* Sreek. *et al.*, but differs in its annual habit (vs. perennial); rachis of raceme 0.8–1 mm wide (vs. 0.25–0.5 mm wide); pedicel glabrous (vs. margins scabrid and bristly hairy at apex); callus glabrous in the spikelets towards lower half of the raceme and sparsely hairy in those towards upper half, with hairs up to 0.2 mm long (vs. callus densely hairy throughout the raceme, with hairs ca. 0.5 mm long); spikelets towards lower half of the raceme either unawned or with an imperfect awn, while those towards upper half perfectly awned (vs. all spikelets perfectly awned with a column); upper glume glabrous or scabrid (vs. long ciliate along the keel, sides and margins); and upper lemma of spikelets towards lower half of the raceme acute at apex either unawned or bearing an imperfect awn, where as those towards upper half bifid at the apex, with a well-developed perfect awn from the sinus (vs. upper lemma of all spikelets throughout the raceme bifid at the apex and bearing a perfect geniculate awn).

Description

Annual tufted herbs. Culms slender, branched, 20–60 cm tall; upper nodes bearded. Leaves distributed along the culms; sheaths up to 5 cm long, rounded on the back, glabrous or sparsely hairy towards the apex; blades 3.5–8 × 0.2–0.4 cm, linear-lanceolate or lanceolate, 9–11-nerved, base more or less truncate, apex acuminate; margins long-ciliate with bulbous-based hairs, the hairs up to 4 mm long; ligule membranous, hyaline, ca. 1 mm long, more or less truncate and prominently fimbriate at the apex. Racemes 2–4, well exerted from the spatheole, 3.5–8 cm long; rachis 0.8–1 mm wide, trigonous, glabrous or scabrid along the angles. Spikelets 2–3.2 × 0.5–1 mm, oblong or oblong-elliptic, apex rounded to acute; those towards lower half of the raceme unawned or with a rudimentary awn, those towards upper half with a well-developed geniculate awn; pedicels short, 0.1–0.2 mm long, concave at the apex, glabrous; callus short, 0.1–0.2 mm long, glabrous in the spikelets towards lower half of the raceme and glabrous or sparsely hairy in those towards upper half, with hairs up to 0.2 mm long. Lower glume 1.8–2.8 × 0.25–0.3 mm, linear-oblong, apex acuminate, acute or obtuse, keeled on the back; keel wingless or narrowly winged towards the apex; surfaces glabrous or sparsely scabrid on the sides. Upper glume 2–3 × 0.5–0.8 mm, oblong, apex rounded, obtuse or acute, chartaceous, keeled on the back; keel scabrid and narrowly winged; sides chartaceous, glabrous or scabrid. Lower lemma hyaline, 1.5–2 × 0.4–0.6 mm, oblanceolate to obovate, apex rounded to acute; margins ciliate. Upper lemma hyaline, 1.7–2.2 × 0.4–0.5 mm; in spikelets towards lower half of the raceme oblong to oblong-elliptic with an acute apex, unawned or with a rudimentary awn; in spikelets of the upper half oblanceolate with a bifid apex and bearing a well-developed awn from the sinus; awn geniculate, up to 6 mm long, with a column ca. 2 mm long. Lodicules 2, small, wedge-shaped. Stamens 2; filaments 0.5–1 mm long; anthers 1.6–2 mm long, oblong, yellow. Ovary ca. 0.3 × 0.2 mm, ellipsoid; style 0.4–0.6 mm long; stigmas 2, ca. 1 mm long, plumose, creamy-yellow.

Flowering & fruiting: November–January.

Etymology: The specific epithet '*rajaniana'* is proposed in honour of Mr. P. P. Rajan, an environmentalist from northern Kerala, India, who dedicated his life to the conservation of mangrove ecosystems and the lateritic hill landscapes of the region. Through his persistent efforts in environmental awareness, habitat protection, and community engagement, he significantly contributed to safeguarding these ecologically fragile habitats and their unique biodiversity.

Table 1. Comparison of distinguishing characters of *Dimeria copeana* and *D.rajaniana*

Characters	***Dimeria copeana***	***Dimeria rajaniana***
Habit	Perennial herbs, stoloniferous or with thick root stock	Annual herbs without stolones or thick root stock
Rachis of raceme	0.25–0.5 mm wide	0.8–1 mm wide
Pedicel	Scabrid on the margins, with one or more bristly hairs towards the apex	Glabrous
Callus	Densely hairy throughout, hairs ca. 0.5 mm long	Glabrous towards the lower half of racemes; glabrous or sparsely hairy towards the upper half, hairs up to 0.2 mm long
Spikelets	All with a perfect awn, scaberulous column present	Unawned or with an imperfect awn towards the lower half of racemes; imperfectly or perfectly awned towards the upper half of racemes
Upper glume	Long ciliate along the keel, sides, and margins	Glabrous or scabrid along the keel and sides
Upper lemma	Apex bifid, with a perfect geniculate awn arising from the sinus; awn ca. 8 mm long	Entire at apex with or without an imperfect awn towards the lower half of racemes; bifid at apex with a perfect awn from the sinus towards the upper half; awn up to 6 mm long

Figure 1: Map showing the Type Locality

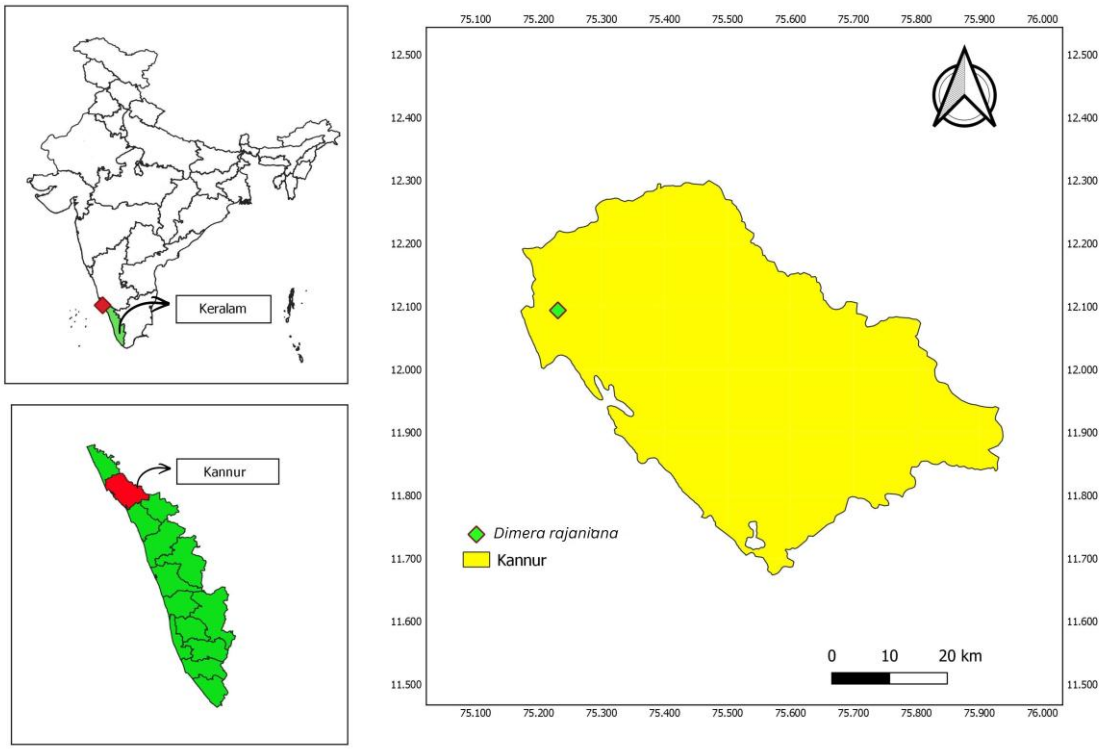

Figure 2: **A**. Habitat; **B**. Habit; **C**. Inflorescence; **D**. Node

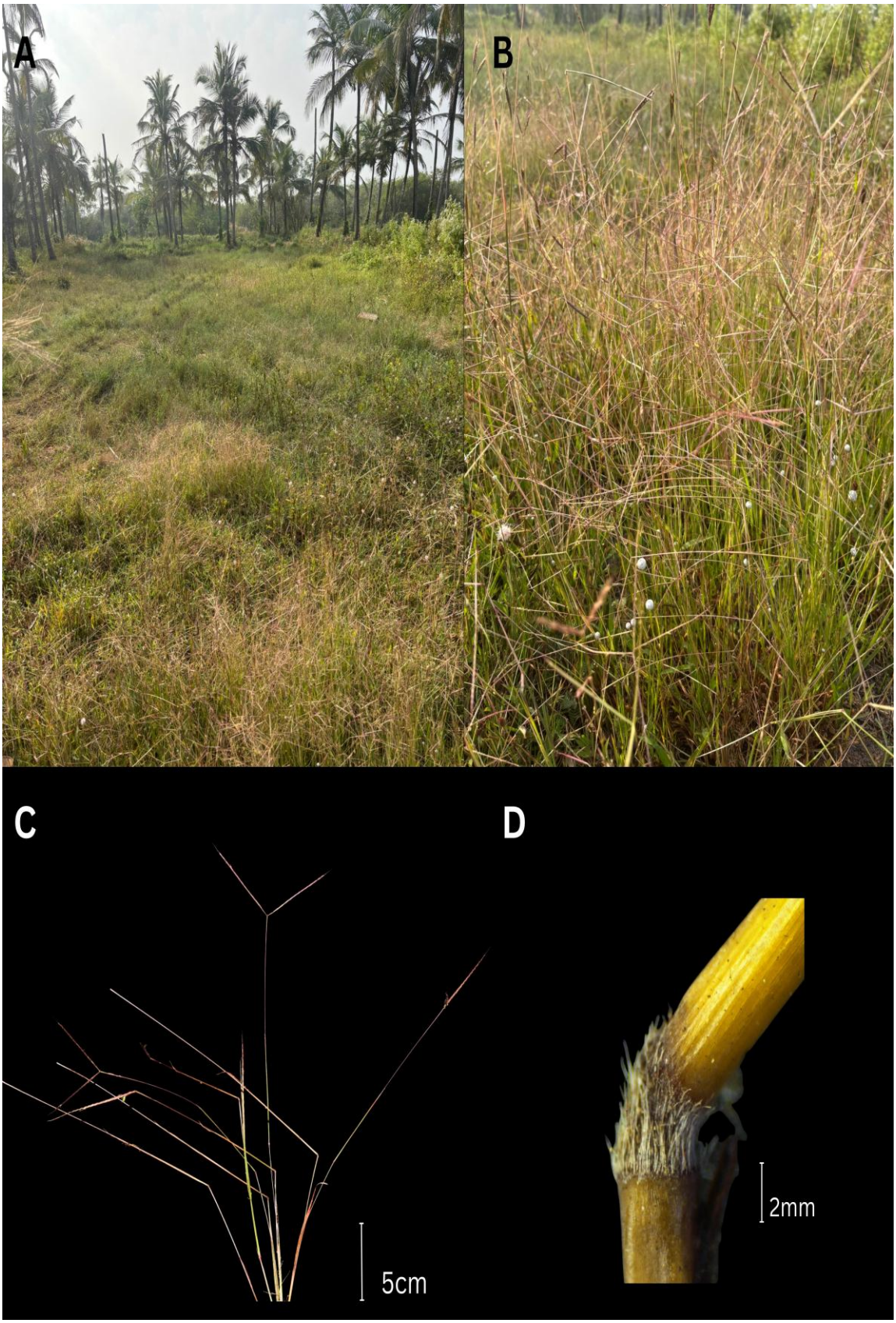

Figure 3: Morphology of the plant parts

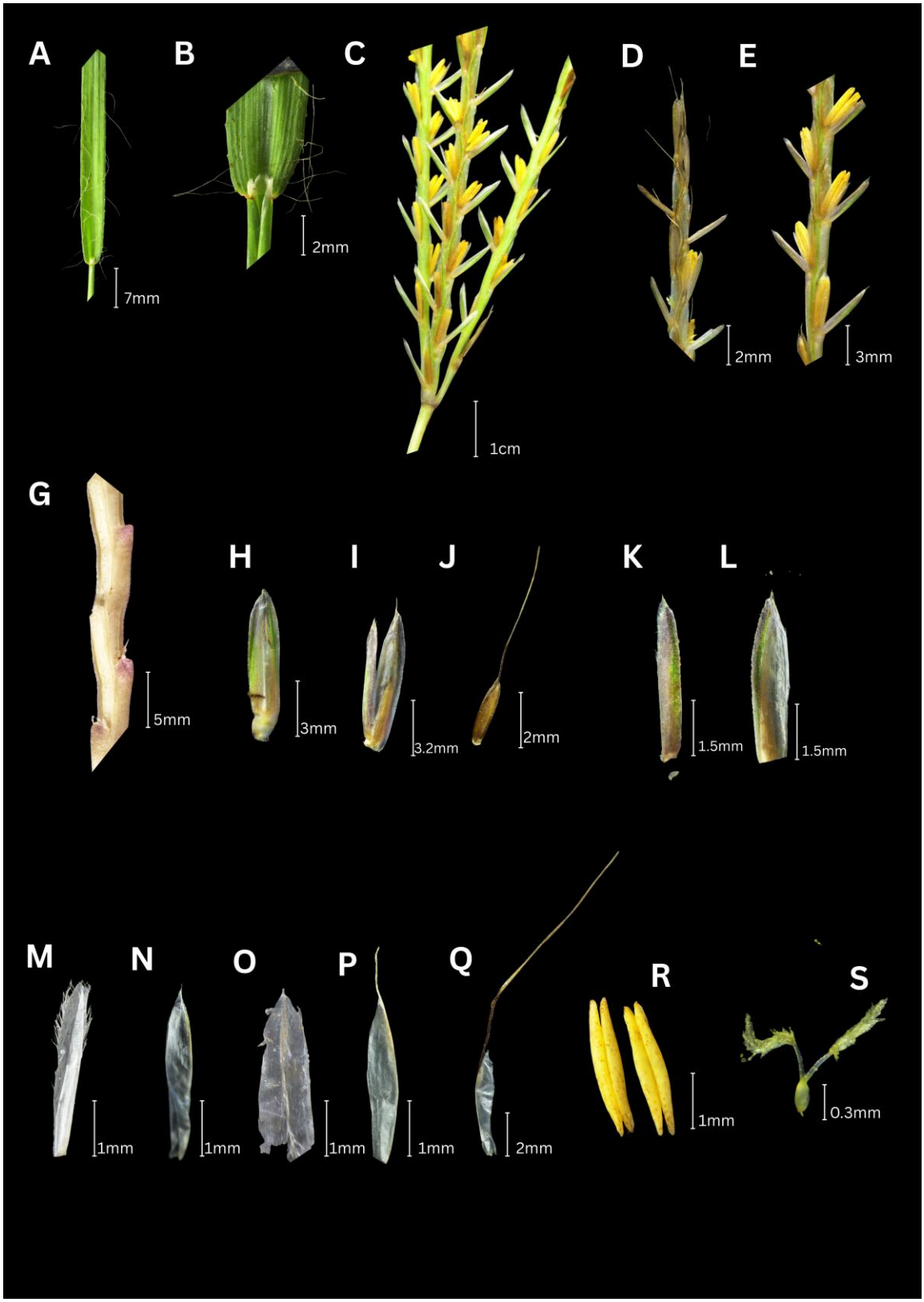

Fig 3: A. Leaf blade adaxial view; **B.** Leaf showing ligule; **C.** Base of inflorescence showing three raceme with awnless spikelets; **D.** Apex of a raceme with awned spikelets; **E.** Basal portion of a raceme showing awnless spikelets; **G.** Rachis adaxial view; **H.** Basal awnless spikelet; **I.** Spikelet with rudimentary awn; **J**. Awned spikelet; **K**. Lower glume; **L**. Upper glume; **M.** Lower lemma; **N & O.** Awnless upper lemma; **P**. Upper lemma with rudimentary awn; **Q.** Upper lemma with perfect awn; **R.** Anthers; **S.** Pistil

DISCUSSION

Dimeria rajaniana sp. nov. can be placed under section *Dimeria* Kiran *et al*. of the genus, as it possesses a trigonous, wingless rachis and closely packed spikelets. The spikelets of this new species exhibit marked morphological variation from the base to the apex of the raceme. Spikelets at the base are awnless; moving upwards, the awn gradually develops from an arista to a rudimentary awn without a column, eventually forming a perfect geniculate awn with a well-developed column towards the apex. The number of spikelets per raceme ranges from 32 to 53, of which 14–22 at the base are either awnless or bear a rudimentary awn. Spikelet size also varies along the raceme: those at the lower portion are larger (2.5–3.2 × ca. 1 mm) and gradually decrease in size towards the upper portion (2–2.5 × 0.5–0.6 mm). The callus is glabrous in the lower spikelets of raceme and becomes sparsely hairy towards the upper portion. Similarly, spikelets are glabrous at the base but become scabrous towards the apex. The upper lemma of basal spikelets are unawned with an acute apex; this gradually transitions to an aristate form, then to an imperfect awn towards middle portion, and finally becomes bifid at the apex with a perfect geniculate awn arising from the sinus in the upper portion of the raceme.

Among the three Indian species of *Dimeria* with an imperfect (i.e., without a column) or awnless upper lemma, the new species shows some similarity to *D. lehmannii* (Steud.) Hack. However, *D. lehmannii* differs markedly in several characters, including the ciliate margins of the rachis and pedicel, larger spikelets (4–4.5 mm long), a ciliate lower glume, and a larger upper glume (4–4.5 mm long) with a ciliate winged keel. In addition, all upper lemmas in *D. lehmannii* are either imperfectly awned or completely awnless. Therefore, the new species appears to represents a connecting link between taxa with a fully developed (perfectly awned) upper lemma and those with an awnless or imperfectly awned upper lemma.

CONCLUSION

At present, the species is known only from the type locality. It occurs in coastal wet paddy fields associated with mangrove ecosystems, where it grows in association with common herbs such as *Hygrophila auriculata* (Schumach.) Heine, *Limnophila repens* (Benth.) Benth., *Marsilea quadrifolia* L., *Fuirena capitata* (Brum. *f*.) Koyama, *Eriocaulon sexangulare* L., *Scleria caricina* (R.Br.) Benth., *Ischaemum ciliare* Retz., *Xyris pauciflora* Willd. etc.

At present, the species is known only from the type locality. According to IUCN (2024) criteria, it is categorized as “Data Deficient” (DD) due to the lack of sufficient information on its geographic distribution and population size.

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RESEARCH ARTICLE

WOOD-DECAYING CRUST FUNGI OF SAL FORESTS: AN ANNOTATED CHECKLIST FROM KODERMA WILDLIFE SANCTUARY, JHARKHAND

Arvind Parihar[1]* and Manoj Emanuel Hembrom[2]

[1]Botanical Survey of India, Arid Zone Regional Centre, Jodhpur, Rajasthan, 342014
[2]Botanical Survey of India, Industrial Section Indian Museum, Sudder Street, Kolkata, West Bengal, 700016
Email: arvind_peace@rediffmail.com

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Abstract: Corticoid fungi are not a natural and well-defined group but assemblage of many ecological life forms which are kept together for the convenient of survey, collection, characterization, identification, documentation and preservation. In the present communication an annotated checklist of 20 taxa falling under 11 genera belonging to 9 families showing similar ecological life forms i.e. crusty basidiomata, also known as corticoid fungi is being provided.

Keywords: Crust fungi, India, Jharkhand, Koderma, Taxonomy

INTRODUCTION

Kingdom Fungi represents one of the most taxonomically diverse kingdoms. Fungi are multicellular, organized as simple as mycelia, invisible to naked eye (micro-fungi) to complex and well developed, tiny to large sized and variously shaped fruiting bodies, which can be termed as "Macrofungi". Corticioid fungi are a diverse and heterogeneous group of fungi which are usually consist resupinate form of Basidiomata. However, Basidiomata of these groups of fungi are simple and consist only generative hyphae in most of the species but, more or less sclerified hyphae are also found in some species. The Hymenophore in these groups of fungi may be smooth or sometimes with different variations to increase the spore production area such as rugose, tuberculate, aculeate, merulioid, folded, or poroid hymenial surfaces. Micro-morphologically these fungi surprise us with a great diversity of forms of basidia, cystidia, spores and other microscopic elements (Hjortstam et al. 1987). For survival they are largely depends upon the wood and completing their life cycle by causing either white- or brown-rot on the host wood.

Koderma Wildlife Sanctuary, situated in the northern Part of Jharkhand, India (24°25′–24°38′ N and 85°25′–85°40′ E), with an area of 150 km² of predominantly dry deciduous forest within the Chotanagpur Plateau (Figure 1). Macrofungi constitute a vital component of the sanctuary's trophic architecture, functioning as saprotrophs, parasites, and mycorrhizal symbionts. These fungi facilitate nutrient cycling, organic matter decomposition, and symbiotic interactions with vascular plants, thereby enhancing forest resilience and productivity. The sanctuary's fungal diversity remains underexplored, but a comprehensive study of wood rotting Fungi by BSI indicates a high representation of Basidiomycetes, particularly corticioid and polyporoid taxa associated with decaying wood and leaf litter.

MATERIALS AND METHODS

During the year 2013-2019 Corticoid fungus were contineoously monitored and collected in the months of June to November in order to fulfill the objectives of survey and collection from various regions of Koderma Wildlife Sanctuary, Jharkhand. Habitat and habit images of each taxon were taken with the help of Digital Camera followed by assigning a unique field number to each specimen. Essential informations like locality name, GPS coordinates, habitat type, substratum or host, and macro-morphological features along with suitable reference measuring tool of the fruiting bodies were noted down and specimens were kept in brown paper bags with a unique collection number for every specimen. Fresh samples were macroscopically examined on the same day upon return to the base camp. Dried specimens were stored in airtight zip-lock bags.

In the laboratory, macro-morphological features were re-examined using an Olympus SZ51 stereo-zoom dissecting microscope. Micro-morphological observations including measurements of

*Corresponding Author

taxonomically important structures are based upon on free-hand sections of dried material stained with 5% KOH and phloxin, and mounted in 30% glycerol. Cyanophilic reactions and amyloidy tests were checked by using Cotton Blue and Melzer's reagent. Microscopic structures like hyphae, cystidia, setae, basidia, and other sterile elements were measured and examined under 100X magnification.

RESULTS

During the survey more than 50 localities of the study area were surveyed and 38 specimens of Corticoid fungi were collected. After two folds critical examination of morphological characterization in the field and laboratory we assigned them into 20 taxa. In this study we successfully recovered 9 families, 14 genera and 20 species each is having unique combination of macro- and micro-scopic features and adopted to either saprophytic or parasitic mode of nutrition. Phanerochaetaceae is having three genera namely *Phanerochaete* and *Phlebiopsis* are represented by two species each whereas, *Porostereum* is represented by one species. Hymenochaetaceae is a unique family which is characterized by the Xanthochoric reaction in which taxa are reactive to KOH solution and permanently changes into black color is being represented by one genus i.e. *Hymenochaete* and four species showing highly diversified genus. In the family *Peniophoraceae* three genera namely *Duportella*, *Peniophora* and *Scytinostroma* each with single species is recorded. Whereas, *Cerrenaceae*, *Corticiaceae*, *Irpicaceae*, *Phleogenaceae*, *Polyporaceae* and *Serpulaceae* circumscribes single genera each with single species.

Duportella tristicula (Berk. & Broome) Reinking, *Philipp. J. Sci*. **17**: 364 (1920).

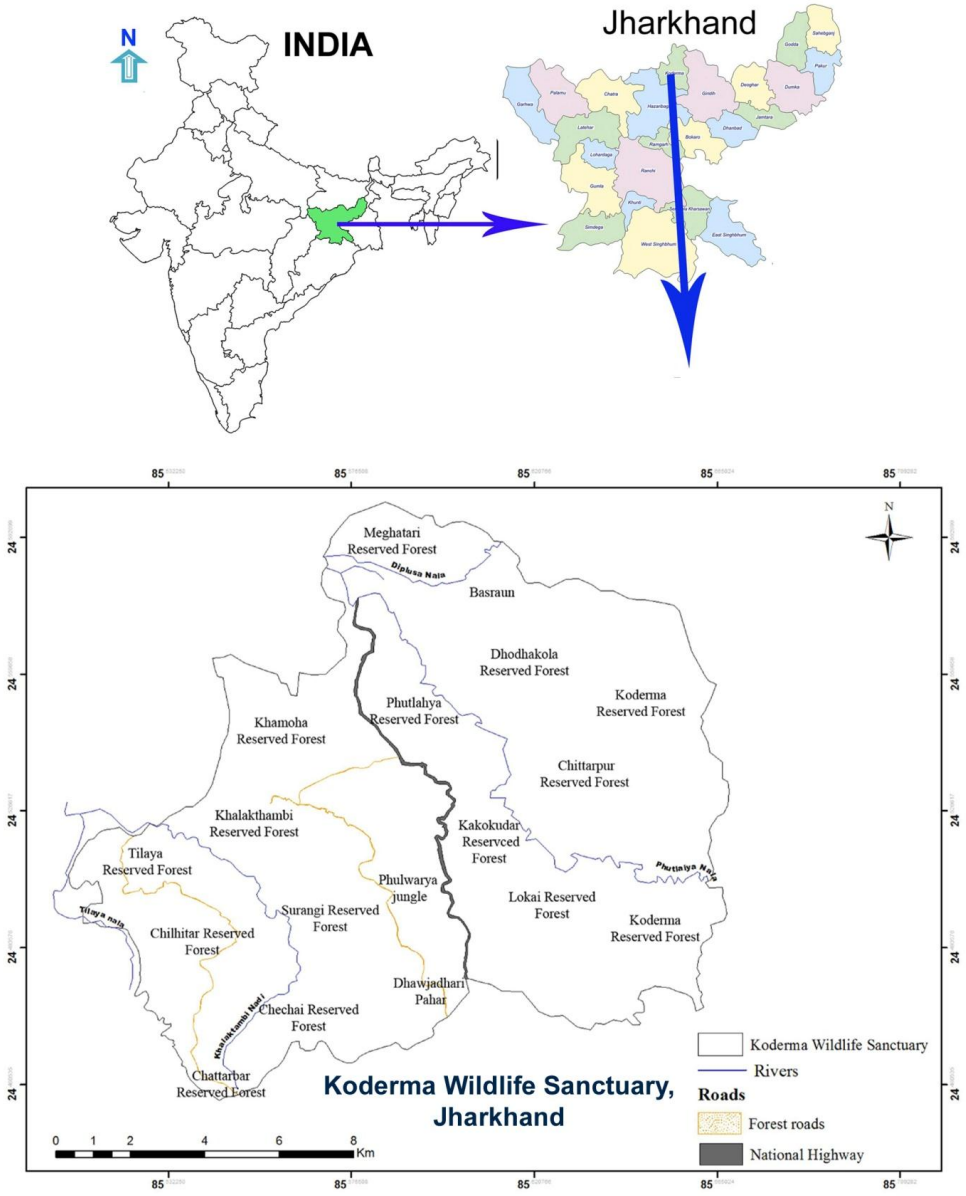

Figure 1: Map of Koderma Wildlife Sanctuary, Jharkhand

Figure 2: A. *Phlebiopsis friesii*; B. *Peniophora lycii*; C. *Gyrodontium sacchari*; D. *Hymenochaete conchata*; E. *Scytinostroma duriusculum*; F. *Hymenochaete rubiginosa*; G. *Hymenochaete villosa*; H. *Pseudolagarobasidium subvinosum*; I. *Hymenochaete rheicolor*; J. *Flavodon flavus*; K. *Lyomyces sambuci*.

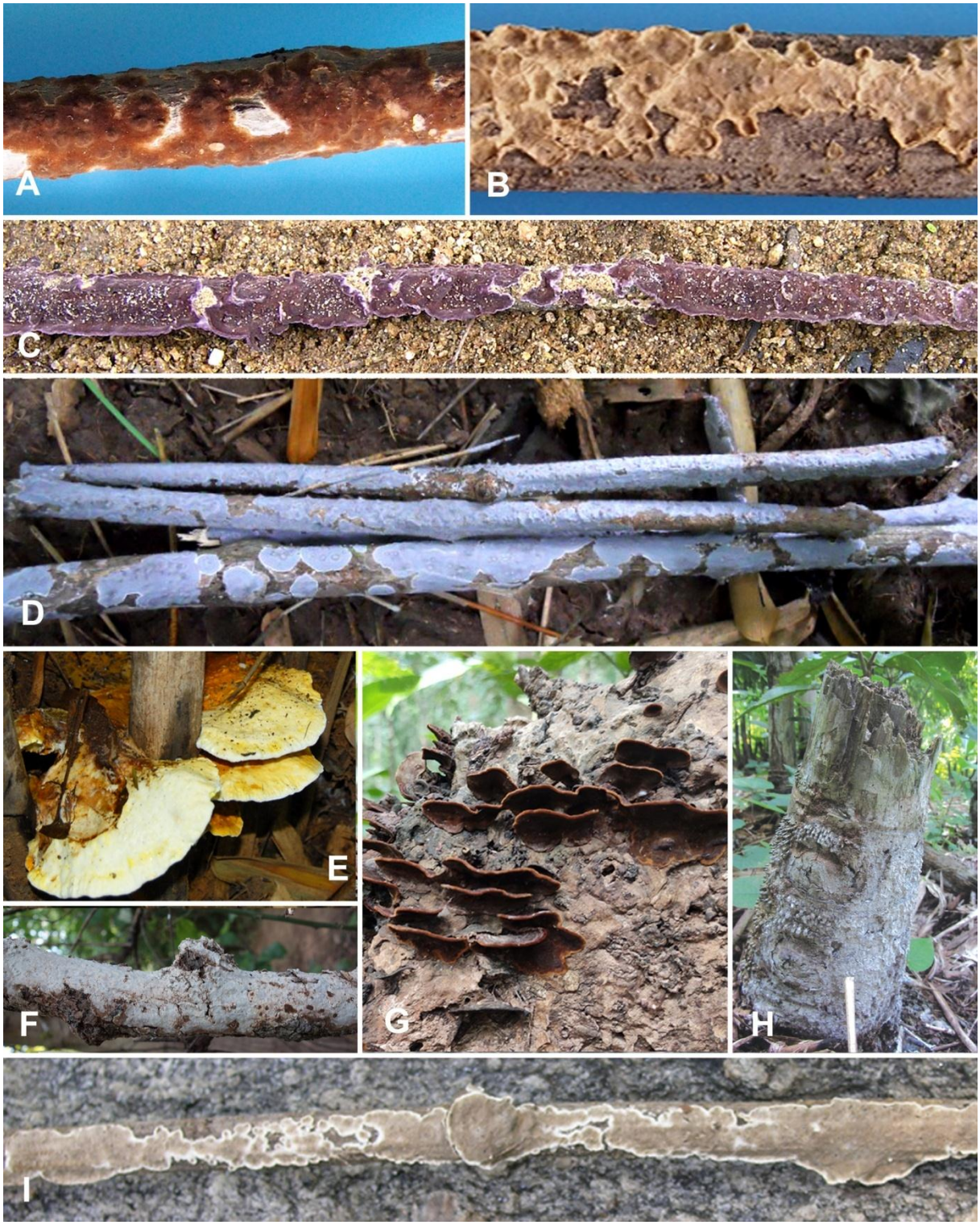

Figure 3: A. *Duportella tristicula*; B. *Phlebiopsis crassa*; C. *Porostereum spadiceum*; D. *Grammothele fuligo*; E. *Serpula similis*; F. *Phanerochaete affinis* G; *Hymenochaete variegata*; H. *Helicogloea globosa*; I. *Phanerochaete velutina*

LEGENDS FOR FIGURES

Figure 1: Map of Koderma Wildlife Sanctuary, Jharkhand.

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G; *Hymenochaete variegata*; H. *Helicogloea globosa*; I. *Phanerochaete velutina.*

Specimen examined: INDIA, Jharkhand, Koderma, KWLS, near phulwaria road, alt. 368 m, 06 Oct., 2012, N24°29′42.2″ E85°35′20.0″, on dead thin branches, *Arvind Parihar*, AP 6569.

Notes: This species is widely distributed especially on the dead thin branches along the road sides. Most specimens were observed to be sterile, particularly the fresh collections, which displayed a dark brown, velvety hymenial surface with a distinct golden-brown margin and a more or less leathery basidiomata. However, a few specimens were fertile, showing mature basidiomata that were papery-thin, brittle, and ochraceous to greyish-white in colour, with an indistinct margin on macroscopic examination. Microscopically, gloeocystida filled with encrusted contents and apically encrusted pseudocystidia are the combination of features to identify the young basidiomata as *D. tristicula*.

Flavodon flavus (Klotzsch) Ryvarden, *Norw. Jl Bot*. **20**(1): 3 (1973); *Irpex flavus* Klotzsch, *Linnaea* **8**(4): 488 (1833). Figure 1(J)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Meghatari, Diplaswa nala, alt. 300 m,N24°35′32.8″ E85°34′51.8″, on a dead wood log, 13 Sept.2010, *Arvind Parihar*, AP 44976; ibid., KWLS, Meghatari, Suggi point, alt. 300 m, N24°35′32.8″ E85°34′51.4″, on a dead standing log, 15 Aug., 2011, *Arvind Parihar*, AP 45053; ibid., KWLS, Meghatari, Durwasha rishi pahar, alt. 300 m, N24°35′19.6″ E85°34′531.0″, on a dead wood log, 16 Aug., 2011, *Arvind Parihar*, AP 45077; ibid., KWLS, Meghatari, Durwasha rishi pahar, alt. 300 m, N24°35′19.68″ E85°34′31.0″, on a dead wood log, 04 Oct., 2012, *Arvind Parihar*, AP 6529, AP 6534; ibid., KWLS, Khalakhtambhi, alt. 310 m, N24°31′59.1″ E85°33′55.8″, on a dead thin branches, 10 Oct., 2012, *Arvind Parihar*, AP 6602; ibid., KWLS, Chatarbar, Mahuatad, alt. 330 m, N24°28′22.8″ E85°34′07.6″, on a dead twigs, 12 Oct., 2012, *Arvind Parihar*, AP 6624; ibid., KWLS, Taraghati, alt. 290 m, N24°32′05.1″ E85°35′42.1″, on a living shrub, 18 Sept., 2013, *Arvind Parihar*, AP 6635.

Notes: Almost all kind of economically important dead woods either fallen in forest or kept in various prmises and houses are severely attacked by this species in the entire areas except monocots/bamboos. Present species a well known white rot causing agent can easily be identified with the help of bright yellow to egg yellow coloured polymorphic hymenophore which have tendency to form angular to circular pores to iripicoid to hydnoid to irregular teeth like structures. So far it's basidiomata shapes are concerned they are equally variable like their hymenophore ranging from resupinate to effused reflexed to pileate forms depending upon the place and position of the host however, dimitic hyphal system with non clamped generative hyphae and cylindrical clavate encrusted cystidia are the constant features to identify this species.

Grammothele fuligo (Berk. & Broome) Ryvarden, *Trans. Br. mycol. Soc.* **73**(1): 15, 1979; *Polyporus fuligo* Berk. & Broome, *J. Linn. Soc*., Bot. **14**(no.73): 53, 1875. Figure 2(D)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Lokai, alt. 337 m, N24°29′35.8″ E85°57′33.4″, on a dead thin branch, 08 Oct., 2012, *Arvind Parihar*, AP 6583

Note: Growing exclusively on woody monocots and causes white rot on them. Morphologically, its grayish brown to bluish white hymenophore with minute pores almost invisible to the naked eyes makes it easy to identify in the field. Microscopically, presence of dendrohyphidia and hymenophore being limited at base of tubes are significant to identify.

Gyrodontium sacchari (Spreng.) Hjortstam, *Mycotaxon* **54**: 186 (1995); *Hydnum sacchari* Spreng.:Fr. *Syst. Mycol*. **I**: 416 (1821). Figure 1 (C)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 369 m, N24°30′17.1″ E85°35′30.8″, on dead bamboos, 08 Sept., 2010, *Arvind Parihar*, AP 44958

Notes: Present species is widely distributed in the tree inhibiting continents. This is a monotypic species under the genus though, designated by various names by various workers (www.indexfungorum.org). But detailed and critical studies of these misidentified taxa (Hjortstam, 1995) reveal them as *Gyrodontium sachari*. Features like resupinate to effused-reflexed basidiomata which is easily separable from its host, thick and yellowish white subiculum, distinctly hydnoid olive-green to sulphurecolured hymenophore, thick-walled septate generative hyphae, absence of cystidia and strongly dextrinoid and cyanophilic basidiospores are identifying and unique for this species.

Helicogloea globosa Chee J. Chen & Oberw., *Mycotaxon* **76**: 280 (2000). Figure 2(H)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Basraun, alt. 378 m, N24°44'51.9″ E85°42'34.6″, on a dead twig, 06 Sept., 2010, *Arvind Parihar*, AP 44939

Notes: Genus *Helicogloea* is sharing similar habit and habitat like other corticoid macrofungi and characterized by widely effused, resupinate basidiomata. However, when microscopically investigated it shows peculiar features including variable thickness of basidiomata, gelatinous subhymenium, probasidia with lateral sacs and usually stalked metabasidia (Baker, 1936 and Chen and Oberwinkler, 2000).

Hymenochaete conchata L.W. Zhou, *Phytotaxa* 273(3): 202 (2016). Figure 1(D)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 369 m,

N24°36′58.8″ E85°59′30.3″, on the living *S. robusta* trunk, 17 Sept., 2010, *Arvind Parihar*, AP 44997

Notes: It usually found to grow exclusively on dead branches, stumps and living trees of *Shorea robusta* throughout the study areas. Morphologically, presence of duplex subiculum with context, abundant subulate hymenial setae, dense setigerous layer and oblong ellipsoid basidiospores are identifying features of this species.

Hymenochaete rheicolor (Mont.) Lév., *Annls Sci. Nat*., Bot., sér. 3 **5**: 151 (1846); *Stereum rheicolor* Mont., *Annls Sci. Nat*., Bot., sér. 2 **18**: 23 (1842). Figure 1(I)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 389 m, N24°29′18.7″ E85°35′50.1″, on a dead wood log, 08 Sept., 2010, *Arvind Parihar*, AP 44959; ibid., KWLS, Meghatari, Suggi point, alt. 369 m, N24°33′54.7″ E85°34′55.2″, on a dead wood log, 08 Sept., 2010, *Arvind Parihar*, AP 45043.

Notes: Its pileus (reflexed part which itself is quite distinct in the field upper surface remain silky fibrous weakly zonate and concentrically sulcate; can be easily folded without breaking seems good field characters to identify. Microscopically, longest (among known species of *Hymenochaete* from the study area) cylindrical to narrowly ellipsoidal and guttulate basidiospores (6.5–7.5 µm long) are unique identifying characters.

Hymenochaete rubiginosa (Dicks.) Lév., *Annls Sci. Nat*., Bot., sér. 3 **5**: 150 (1846); *Helvella rubiginosa* Dicks., *Fasc. pl. crypt. brit*. (London) **1**: 20 (1785). Figure 1(F)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Mughalamaran, alt. 369 m, N24°30′59.7″ E85°34′29.4″, on a living tree, 03 Sept., 2010,*Arvind Parihar*, AP 44907; ibid., KWLS,Basraun, alt. 359 m, N24°34′48.9″ E85°37′44.8″, on a dead standing log, 06 Sept., 2010,*Arvind Parihar*, AP 44938; ibid., KWLS, Koderma enclosure, alt. 339 m, N24°29′18.7″ E85°35′50.1″, on a dead log, 08 Sept., 2010,*Arvind Parihar*, AP 44953; ibid., KWLS,Kodermaenclosure, alt. 329 m, N24°29′42.9″ E85°35′63.8″, on a living tree, 11 Aug., 2011,*Arvind Parihar*, AP 45020; ibid., KWLS, Meghatari, Suggi point, alt. 319 m, N24°33′54.7″ E85°34′55.2″, on a dead standing log, 15 Aug., 2011,*Arvind Parihar*, AP 45055; ibid., KWLS, Dhodhakhola, alt. 328 m, N24°30′36.6″ E85°39′30.1″, on a dead tree, 17 Aug., 2011,*Arvind Parihar*, AP 45084; ibid., KW1LS, Meghatari, Sambhu bhura mines, alt. 310 m, N24°35′02.3″ E85°35′29.6″, on a dead standing log, 03 Oct., 2012,*Arvind Parihar*, AP 6517; ibid., KWLS, Meghatari, Suggi point, alt. 319 m, N24°33′54.7″ E85°34′55.2″, on a dead standing log, 15 Aug., 2011,*Arvind Parihar*, AP 6546.

Notes: Present species is widely distributed in tropical India (Cunningham, 1957; De, 2008; Sharma, 1995, 2012). Macroscopic features of present species like effused-reflexed, woody pileate basidiomata with villose zonate pilear surface are good identifying charecters.

Hymenochaete variegata Bres., *Hedwigia* **56**: 301 (1915). Figure 2(G)

Specimen examined: INDIA, Jharkhand, Koderma, KWLS, Chatarbar, Mahuataan, alt. 230 m. 12 Oct., 2012, N24°27′59.3″ E85°33′56.7″, on a dead wood log, *Arvind Parihar*, AP 6623.

Notes: The thin, shiny, confluent, flexible and yellowish basidiomata with smaller hymenial setae and basidiospores differetiate it from *H. rubiginosa*.

Hymenochaete villosa (Lév.) Bres., *Annls mycol*. **8**(6): 588 (1910); *Stereum villosum* Lév., *Annls Sci. Nat*., Bot., sér. 3 **2**: 212 (1844). Figure 1(G)

Specimens examined: INDIA; Jharkhand, KWLS, Khalakhtambhi, alt. 356 m, N24°31′59.1″ E85°33′55.8″, on the fallen branches of unknown wood, 10 Oct., 2012, *Arvind Parihar*, AP 6604.

Notes. Present species is characterized by resupinate basidiomata with date brown to umber brown smooth hymenial surface with light yellow to corn colored floccose to fibrous marginin. Microscopically, hymenial setae which arise from hymenium and subhymenium are very characteristic. Scrutiny of literature (Cunningham, 1957 and De, 2008) reveals the report of basidiomata of *H. villosa* as effused-reflexed but only resupinate forms of specimens were recovered from present survey and collection. Microscopic features like organization of subiculum (presence of context near base), hyphal and setal dimension and orientation were found agreeable with the earlier report.

Lyomyces sambuci (Pers.) P. Karst., *Bidr. Känn. Finl. Nat. Folk* **37**: 153 (1882); *Corticium sambuci* Pers., *Neues Mag. Bot*. **1**: 111 (1794). Figure 1(K)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Baghitan road, Lokai, alt. 370 m, N24°19′42.8″ E85°25′10.8″, on a decaying tree log, 10 Aug., 2011, *Arvind Parihar*, AP 45015

Notes: Present species was known earlier from temperate regions of North West Himalaya (Prashar, 2015) and southern peninsular regions of India (Natarajan and Kolandavelu, 1998). *Lyomyces sambuci* is characterized by chalky white resupinate, sparsely tuberculate hymenophore, clamped generative hyphae and projecting papillate cystidia which is projected beyond hymenium (Prashar, 2015). However, Natarajan and Kolandavelu (1998) have mentioned smooth nature of cystidia whereas, encrusted cystidia are found in present observation that completely agree with other extralimital materials (Eriksson and Ryvarden, 1976; Wu, 1990; Bernichia and Gorjón, 2010 and Yurchenko and Wu, 2016).

Peniophora lycii (Pers.) Höhn. & Litsch., *Sber. Akad. Wiss. Wien*, Math.-naturw. Kl., Abt. 1 **116**: 747 (1907) [1906]; *Thelephora lycii* Pers., *Mycol. eur*. (Erlanga) **1**: 148 (1822). Figure 1(B)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 360 m,

N24°29′28.3″ E85°38′36.1″, on dead thin branches, 08 Sept., 2010, *Arvind Parihar*, AP 44954.

Notes: *Peniophora lycii* is characterized by wide globose encrusted cystidia and presence of dendrohyphidia (Lambevska, 2015) an ecological adaptation against drought (Bernicchia and Gorjón, 2010). At a glance the present species is microscopically appears peculiar due to presence of richly branched dendrohyphidia, obovate to subglobose encrusted cystidia and acute to obtuse sulphocystidia. Its, whitish blue grey hymenophore with sparsely distributed tubercules/papillae in basidiomata macroscopically coupled with dimensions of basidiospores length and other sterile structures microscopically are identifying features of the present species.

Phanerochaete affinis (Burt) Parmasto, *Consp. System. Corticiac*. (Tartu): 84 (1968); *Peniophora affinis* Burt, *Ann. Mo. bot. Gdn* **12**: 266 (1926) [1925]. Figure 2(F)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 388 m, N24°29′18.7″ E85°35′50.1″, on a dead thin branch, 14 Sept., 2010, *Arvind Parihar*, AP 44986.

Notes: Previously this species is known to occure at North Western Himalaya (Prasher, 2015) and Southern Peninsular India (Natarajan and Kolandavelu, 1998). Macroscopically, white to cream, smooth, resupinate to effused-reflexed basidiomata with floccose to fibrous margin are striking feature for identification, whereas, microscopically subiculum is characteristic because it bears loose crystals imbedded on it and possess thin- to moderately thick-walled generative hyphae in which width doesn't remain uniform.

Phanerochaete velutina (DC.) P. Karst., *Kritisk Öfversigt af Finlands Basidsvampar*, (Basisiomycetes; Gastero- andHymenomycetes) (Helsingfors) **3**: 33 (1898); *Thelephora velutina* DC., in de Candolle and Lamarck, *Fl. franç*., Edn 3 (Paris) **5/6**: 33 (1815). Figure 2(I)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, near Phulwaria. 388 m, 06 Oct., 2012, N24°29′34.2″ E85°35′33.7″, on a dead thin branch, *Arvind Parihar*, AP 6562.

Notes: Earlier this species is known to be distributed in Western Ghats (Ranadive et al., 2011) and Western Himalaya (Prasher, 2015) only. The resupinate basidiomata which is yellowish white to pale yellow when fresh and becoming yellowish brown on drying are the notable features of this species. Microscopically abundant encrusted cystidia which remains subcylindrical, comparatively thick-walled (up to 3 µm thick) and much longer projection (upto 50 µm) beyond hymenium are characteristic features for this species (Bernichia and Gorjón, 2010 and Prasher, 2015).

Phlebiopsis crassa (Lév.) D. Floudas and Hibbett, *Fungal Biology* **119**: 710 (2015); *Thelephora crassa* Lév., *Annls Sci. Nat*., Bot., sér. 3 **2**: 209 (1844). Figure 2(B)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Meghatari, Durwasha rishi pahar, alt. 308 m, 04 Oct., 2012, N24°35′29.5″ E85°34′43.3″, on dead thin branches, *Arvind Parihar*, AP 6532; ibid., KWLS, Chatarbar,Dumariya tan, alt. 386 m, 21 Sept., 2013, N24°32′20.0″ E85°35′01.0″, on dead thin branches, *Arvind Parihar*, AP 6653.

Notes: This is a common and easily recognized species in the field due to it's brown to purple colored basidiomata with narrowly reflexed margin. But, confused with *Hypoxylon haematostroma* Mont. (a carbon and cushion fungi commonly occupying similar habitat) as both of them possess similar macromorphology (pale purple colored corticoid fruiting bodies). But, present species still can be distinguished because it doesn't bear any papillate ostiole like *H. haematostroma* (belongs to the phylum Ascomycota).

Phlebiopsis friesii (Lév.) Spirinand Miettinen, in Miettinen, Spirin, Vlasák, Rivoire, Stenroos and Hibbett, MycoKeys 17: 25 (2016); *Thelephora friesii* Lév., *Syst. Verz*.: 17 (1854). Figure 1(A)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma NRF, alt. 388 m, N24°39′28.30″ E85°56′59.60″, on dead wood log, 13 Aug., 2011, *Arvind Parihar*, AP 45034.

Notes: Macromorphologically, turning (with the application of KOH) of basidiomata to violet is a worth mentioning macroscopic feature for identification. Apart from leathery nature of basidiomata with pale cinnamon brown wrinkled to folded hymenophore, coarsely encrusted hyaline cystidia are its characteristic microscopic features (Hjortstam and Ryvarden, 1990 and Tiwari et *al*., 2010).

Porostereum spadiceum (Pers.) Hjorstam & Ryvarden, *Syn. Fung.* (Oslo) **4**: 51 (1990); *Lopharia fulva* (Lév) Boidin, *Bull.mens. Soc. linn.Soc. Bot.* Lyon **28** (7): 213 (1959). Figure 2(C)

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Meghatari, Durwasha rishi pahar, alt. 348 m, 04 Oct., 2012, N24°35′29.5″ E85°34′43.3″, on dead thin branches, *Arvind Parihar*, AP 6535.

Notes: Morphologically brown coloured resupinate to effused-reflexed basidiomata with pseudo-dimitic hyphal system (basal part of skeletocystidia appear as skeletal hyphae) and encrusted hyaline cystidia are unique characters to identify the present species. Possession of another important feature i.e. origin of skeletocystidia from basal part of subiculum is also on conformity with the earlier statement made by Wu and Chen (1992).

Pseudolagarobasidium subvinosum (Berk. & Broome) Sheng H. Wu, Acta bot. fenn. 142: 113 (1990); *Hydnum subvinosum* Berk. & Broome, *J. Linn. Soc., Bot.* **14**(no. 73): 60 (1873) [1875]. Figure 1(H).

Specimens examined: INDIA, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 358 m, N24°39′21.7″ E85°35′48.4″, on a dead tree trunk branch, 07 Oct., 2012, *Arvind Parihar*, AP 6577.
Notes: Closely adnate basidiomata with obtuse to acutely toothed greyish violet to bluish violet hymenophore which being developed from meruloid surface when young are characteristic in the present species. Microscopically, presence of gloeocystidia is an additional identifying feature within family. Earlier, this species was recorded from Kerala growing on similar host (Sankaran and Sharma, 1986) but unfortunately not included in any checklist and references of Indian mycobiota.
Scytinostroma duriusculum (Berk. & Broome) Donk, *Fungus* **26**: 20, 1956; *Stereum duriusculum* Berk. & Broome, *J. Linn. Soc*. Bot. **14**: 66, 1873. Figure 1(E)
Specimens examined: **Specimen examined**: India, Jharkhand, Koderma, KWLS, Koderma enclosure, alt. 390 m N24°29′18.7″ E85°35′50.1″, on a dead tree log, 16 Sept., 2010, *Arvind Parihar*, AP 44988.
Notes: Whitish cream to yellowish white, leathery resupinate basidiomata with smooth hymenial surface is distinguishing macro-morphological feature of this species. Microscopically, dimitic hyphal systems with septate generative hyphae and thick-walled dextrinoid skeletal hyphae (dichohyphidia) and globose amyloid basidiospores are the important characters of this species.
Serpula similis (Berk. and Broome) Ginns, *Mycologia* **63**(2): 231, 1971; *Merulius similis* Berk. & Broome, *J. Linn. Soc*., Bot. **14** (no. 73): 58, 1873. Figure 2(E)
Note: This species was found to grow exclusively on dead and living bamboo culms in the entire study area. Its glutinate leathery resupinate to effused-reflexed to pileate basidiomata with chalky consistency, folded and pale brown to orange yellow hymenophore are quite characteristic. In the field young basidiomata of *Gyrodontium sacchari* is often mistaken as present species because both attains almost similar habit and habitat i.e. similar shape and size of pileus and association with bamboos; although, *G. sacchari* prefers to grow on other dicot hosts as well. Moreover, presence of toothed hymenophore in *G. sacchari* easily distinguishes it from the present species.

DISCUSSION

Ecologically, *Hymenochaete conchata* and *Serpula similis* were found host specific and showing pathogenic nature causing brown rot on *Shorea robusta* and Bamboos successively in the entire study areas. But, rest eighteen species are successfully recovered from various other angiospermic woods showing the saprophytic nature of habit. *Grammothele fuligo* is also host specific particularly affecting abundantly the monocots species belonging to the Bamboos and Palms in the Koderma Wildlife Sanctuary.
So far hymenial configurations is concerned most of the species are having smooth hymenophore like *Duportella tristicula, Helicogloea globosa, Hymenochaete conchata*, *Phanerocahete affinis, P. velutina*, *Phlebiopsis crassa*, *P. freisii*is. *Grammothele fuligo* is having minutely shallow pores whereas *Serpula similis* is having meruloid hymenophore configuration. Some of the species like *Xylodon sambuci*, *Peniophora lycii* are having papillate hymenophore whereas *Flavodon flavus* and *Gyrodontium sacchari* are having distinct toothed hymenophore.
All these species caused various types of rots to the trees of Koderma Wildlife Sanctuary, Jharkhand and plays an important role in the Ecology of the forest ecosysytem. The majority of the species—including *Duportella tristicula*, *Flavodon flavus*, *Grammothele fuligo*, *Pseudolagarobasidium subvinosum*, and other members of the genera *Hymenochaete*, *Phanerochaete*, *Phlebiopsis*, and *Peniophora* were responsible for causing white rot fungi. These organisms are characterized by their ability to degrade lignin alongside cellulose and hemicellulose, often producing oxidative enzymes such as laccases and peroxidases. However, *Serpula similis* and *Gyrodontium sacchari*, were classified as a brown rot fungus and decay nature caused by *Helicogloea globbosaa* is still unclear.

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RESEARCH ARTICLE

IMPACT OF MONSOON ON WATERLOGGING AND GROUNDWATER QUALITY IN HISAR DISTRICT

Santosh Rani[1]*, Mukesh Kumar[1], Kapil Rohilla[2], Naveen Kumar[3], Rajkumar[1] and Baljeet Singh Gaat[1]

1- Department of Soil and Water Engineering, COAE&T, CCSHAU, Hisar – 125004
2- Haryana Space Application Centre, Hisar – 125004
3- Department of Agricultural Engineering, SKNCOA, SKNAU, Jobner - 303329

Email: snjangra94@gmail.com

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Abstract: The study was conducted to assess and map the extent of waterlogging and groundwater quality in Hisar district through Geo-spatial techniques. The maps of water table depth and groundwater quality for pre and post monsoon seasons were generated for the years 2013 to 2018. LANDSAT-8 satellite image was effectively used for determining surface waterlogged area which found to be 7.76 per cent in Hisar district. Analysis of water table depth maps for both pre and post monsoon of Hisar district generated in GIS environment for the years 2013 to 2018 reveals that in post monsoon area (average 0.80 %) under fully waterlogged (< 1.5 m) condition was increased as compared to pre monsoon (average 0.30 %).

Keywords: Geo-spatial techniques, Groundwater quality, Waterlogging, Water table

INTRODUCTION

Waterlogging and salinity are the dual problems which are mainly found in irrigated area of developing countries like India. In India, the area under waterlogging and salt affected land found to be 2.46 and 7.0 million hectares (mha) (Kanga and Singh, 2017). Generally waterlogging is followed by salinization in arid and semi-arid regions. Hence, it is considered as dual problem. Prolonged stagnation of water over the surface or saturation of root zone due to poor drainage and shallow water table causes restriction in air diffusion in the soil and reduced oxygen level leads to waterlogging condition. Waterlogging causes change in cropping pattern which ultimately leads to inefficient use of land. Land becomes partially unavailable for part of the year or fully unavailable throughout the year, leading to overall poor production and performance of agricultural sector. Due to excess of irrigation water in poorly drained areas, waterlogging and secondary salinization appeared and caused losses in productivity for rice (42%), wheat (38%) and sugarcane (61%) crops (Samra *et al.*, 2006).

Haryana state is situated in the middle of the flat, topographically featureless, Trans-Gangetic alluvial plain. In this plain, drainage systems to manage groundwater tables are almost absent. Thick calcareous layers irrigated by canal and tube well in these areas caused secondary soil salinization and waterlogging is caused by thick calcareous layers due to irrigation from canal and tube well (Mandal *et al.*, 2017). The waterlogging and salinity are becoming big problems due to over use of water in rice-wheat cropping system. Therefore, proper management strategies are urgently required to manage salinity and waterlogging. Waterlogging and salinity have adverse effect on the crop and land productivity which occurs mainly due to uprising of salts with water table in root zone (Datta *et al.*, 2002). It is the major factor in land degradation which occurs mainly due to blockage of natural drainage with the construction of transport network (e.g. roads, railway line and other structure) responsible for stagnating monsoon runoff (Goyal *et al.*, 2005). Hence, assessment of rigorousness and extent of the waterlogging and salinity is necessary. Extent of waterlogging is not only determined by field condition but also influenced by the situation of neighbouring area of the site. Thus, mapping of available features of waterlogged and saline area is required in planning and management of the problem. The common tactics of mapping waterlogged area such as ground survey has very limited scope due to high human power and time

*Corresponding Author

required, shortage of data available and costs involved. Further, the problem of waterlogging is dynamic which limits the usefulness of traditional methods for data collection. Remote sensing technology coupled with GIS tools is reliable tool in mapping and analysing different spatial descriptions of the study area. Field data used in conjunction with information derived from satellite images can overcome the limitations of traditional approach of survey due to less time, cost and high effectiveness.

MATERIALS AND METHODS

The study was conducted for Hisar district which is located in west central part of Haryana and lies in semi-arid region between latitude 28º 56'00"N to 29º 38'30" N and longitude 75º 21'12"E to 76º 18'12" E having mean maximum temperature 43˚C (May & June) and mean minimum temperature 5˚C (January). The area is mainly irrigated by shallow tubewells, network of Bhakhra and Western Yamuna Canal Systems. The net irrigated area in Hisar district is 272000 ha out of which 63000 ha is irrigated with tubewells/borewells (Anonymous 2013).

In the study area, to assess the extent of subsurface waterlogging and groundwater quality fluctuations, data on water table depth maps and groundwater quality for pre and post monsoon from 2013 to 2018 were attached into ArcMap to create point features and interpolated using Inverse Distance Weighted (IDW). This interpolation method follows the concepts of proximity to Thiessen polygons with gradual change of the trend surface. Maps of subsurface waterlogging were classified using reclassify tool in the ranges, fully waterlogged (depth < 1.5 m), waterlogged (1.5-3 m), potentially waterlogged (3-10 m) and safe (> 10 m) (Paliwal, 2017). Similarly, groundwater quality classified into four different regions i.e. good (0 - 2 dS/m), marginally saline (2 - 4 dS/m), saline (4 - 8 dS/m) and highly saline (>8 dS/m) (Paudyal *et al*., 2016).

LANDSAT-8 satellite data (Table 1) was used to delineate surface waterlogged area. Layer stacking and mosaicking of the collected images was performed and it was clipped to the district boundary in order to obtain the final False Colour Composite (FCC) (Red, Green and Near Infrared) image (spatial coordinate system World Geodetic System (WGS) 1984 UTM 43N projection was used). The image was visually analysed to identify surface waterlogged area according to shape, size, texture, tone and association in Hisar district. The dark blue to blackish red mixed tone with irregular shape and coarse with mottled texture were considered as surface waterlogged area and digitized manually on standard FCC image. Dark black tone within regular shape was considered as water bodies and not digitized as surface waterlogged area.

RESULTS AND DISCUSSION

Subsurface Waterlogging

The groundwater data reveals that the average water table depth in whole district during pre-monsoon was 7.73, 7.55, 7.81, 7.94, 7.67 and 7.74 m in June 2013, 2014, 2015, 2016, 2017 and 2018, respectively whereas during post-monsoon, it was 7.29, 7.56, 7.61, 7.65 and 7.64 m in October 2013, 2014, 2015, 2016 and 2017, respectively. The extent of subsurface waterlogged area for both pre and post monsoon seasons (2013-18) was assessed and mapped as critical areas in term of ground water level near to ground surface (< 1.5 m) as shown in figures 1 and 2, respectively. The maximum area covers under the water table depth 3.0 - 10.0 m. In the pre monsoon, area under fully waterlogged condition was estimated as 3.92, 18.03, 12.29, 5.56, 23.68 and 9.92 km^2 in June 2013, 2014, 2015, 2016, 2017 and 2018, respectively while in post monsoon, area under fully waterlogged condition was found to be 44.53, 25.31, 20.04, 24.04 and 47.83 km^2 in October 2013, 2014, 2015, 2016 and 2017, respectively. This reveals that in post monsoon season waterlogged area under shallow water table depth increases as compared to pre monsoon. The change in area under shallow water table depth is clearly visible in figure 1 and 2. Likewise the area under safe zone in pre monsoon estimated 809.23, 814.86, 881.18, 1010.36, 1011.85 and 1121.39 km^2 in June 2013, 2014, 2015, 2016, 2017 and 2018, respectively while in post monsoon area found to be 737.22, 840.79, 999.45, 958.76 and 966.57 km^2 in October 2013, 2014, 2015, 2016 and 2017, respectively. The area under different water table depth (0-1.5, 1.5-3.0, 3.0-10.0 and > 10.0 m) for pre and post monsoon is presented in table 2. The villages which mainly come under shallow water table depth (<1.5 m) are Sorkhi, Sisar, Mohla, Puthi, Kumba (South-East of Hisar), Mirka, Satrod Kalan, Satrod Kurd, Dhani Raipur, Kaimri (centre of Hisar district), Ghirai, Garhi, Dhani Mirdad, Matloda and Rajli (South of Hisar district).

Groundwater Quality

The ground water quality maps of Hisar district during pre and post monsoon season are shown in figure 3 and 4, respectively. It was found that the average groundwater quality in whole district was 2.96, 3.04, 3.17, 3.25, 3.35 and 3.34 dS/m in June 2013, 2014, 2015, 2016, 2017 and 2018, respectively whereas during post-monsoon, it was 2.86, 3.11, 3.17, 3.27, 3.18 and 3.34 dS/m in October 2013, 2014, 2015, 2016, 2017 and 2018, respectively. Analysing groundwater quality fluctuations in study area (electrical conductivity (EC)), it was observed that the maximum area of Hisar district covers groundwater quality 2.0-4.0 dS/m. In pre monsoon

season, area under highly saline (> 8.0 dS/m) category was estimated to be 29.56, 26.16, 27.39, 29.41, 30.67 and 31.60 km^2 in June 2013, 2014, 2015, 2016, 2017 and 2018, respectively. While in post monsoon, area under highly saline (> 8.0 dS/m) category was observed to be 27.35, 26.62, 27.88, 29.70, 25.12 and 32.26 km^2 in October 2013, 2014, 2015, 2016 and 2017, respectively.

There is only small variation in area of highly saline category for both pre and post monsoon in the Hisar district. The patch of highly saline area is found in the same regions for pre and post monsoon of the district as shown in figures 3 and 4. From table 3, it is observed that area under saline category (4.0 - 8.0 dS/m) increased from 2013 to 2015 and afterward it is almost stabilized. Similarly, area under good quality (0 - 2.0 dS/m) was in decreasing trend for both pre and post monsoon but for the year 2017, there was sudden increase in area under this category. The villages in Hisar district mainly Dhani Raipur, Ludas, Kamari (centre of Hisar district), Durjanpur P/T, Landhari P/T (North of Hisar district) comes under highly saline categories.

Surface Waterlogged Area

This was revealed that Hisar district faces both surface and subsurface waterlogging problem. The surface waterlogged area was found maximum in central and south-eastern part comprising nearly 7.76 percent (315.30 km^2) of the total area, indicates severe problem of waterlogging during *rabi* season as shown in figure 5. In FCC image, surface waterlogged area was easily identified as dark blue to blackish red mixed tone (higher absorption of electromagnetic radiation in visible and near infrared band), irregular in shape and coarse with mottled in texture. The standing surface waterlogged area reveals as dark blue to black in tone depending on the water depth that is in small proportion as compared to blackish red mixed tone which was identified as waterlogged area (also verified from water table depth maps of 2017) with supportive active growth of aquatic crops.

CONCLUSION

Analysis of water table depth maps for both pre and post monsoon of Hisar district generated in GIS environment for the years 2013 to 2018 reveals that in post monsoon area (average 0.80 %) under fully waterlogged (< 1.5 m) condition was increased as compared to pre monsoon (average 0.30 %). The area under highly saline ground water increased in the recent years. LANDSAT-8 satellite image was effectively used for determining surface waterlogged area which found to be 7.76 percent in Hisar district.

Table 1: Detail of specification of satellite data

Satellite	Sensor	Spectral Resolution (µm)	Spatial Resolution	Path No.	Row No.
LANDSAT-8	Operational Land Imager	Band 3: 0.533 - 0.590 Band 4: 0.636 – 0.673 Band 5: 0.851 – 0.879	30 m	147	40

Table 2: Area distribution under different water table depth categories in Hisar district during 2013-18

Year	Pre and Post monsoon	Area under different water table depth (km^2)			
		0-1.5 m	1.5-3.0 m	3.0-10.0 m	> 10.0 m
2013	June	3.92	123.57	3125.33	809.23
	October	44.53	338.73	2,941.21	737.22
2014	June	18.03	305.75	2922.85	814.86
	October	25.31	298.38	2,897.26	840.79
2015	June	12.29	227.81	2940.65	881.18
	October	20.04	266.64	2,775.26	999.45
2016	June	5.56	200.24	2845.61	1010.36
	October	24.04	294.85	2,784.16	958.76
2017	June	23.68	331.64	2694.82	1011.85
	October	47.83	376.98	2,670.55	966.57
2018	June	9.92	283.57	2646.75	1121.39

Table 3: Spatial distribution of groundwater quality in Hisar district during 2013-18

Year	Pre and Post monsoon	Area under different groundwater quality (km^2)			
		0-2.0 dS/m	2.0-4.0 dS/m	4.0-8.0 dS/m	> 8.0 dS/m
2013	June	753.28	2711.59	568.46	29.56
	October	806.52	2685.51	541.96	27.35
2014	June	439.57	3043.68	551.71	26.16
	October	334.37	3106.30	594.28	26.62
2015	June	258.32	3136.62	639.35	27.39
	October	207.89	3172.96	653.07	27.88
2016	June	188.92	3119.68	723.65	29.41
	October	186.93	3116.27	729.35	29.70
2017	June	193.11	3003.88	834.27	30.67
	October	328.45	3022.96	685.20	25.12
2018	June	201.54	2966.28	862.08	31.60
	October	201.76	2955.03	872.58	32.26

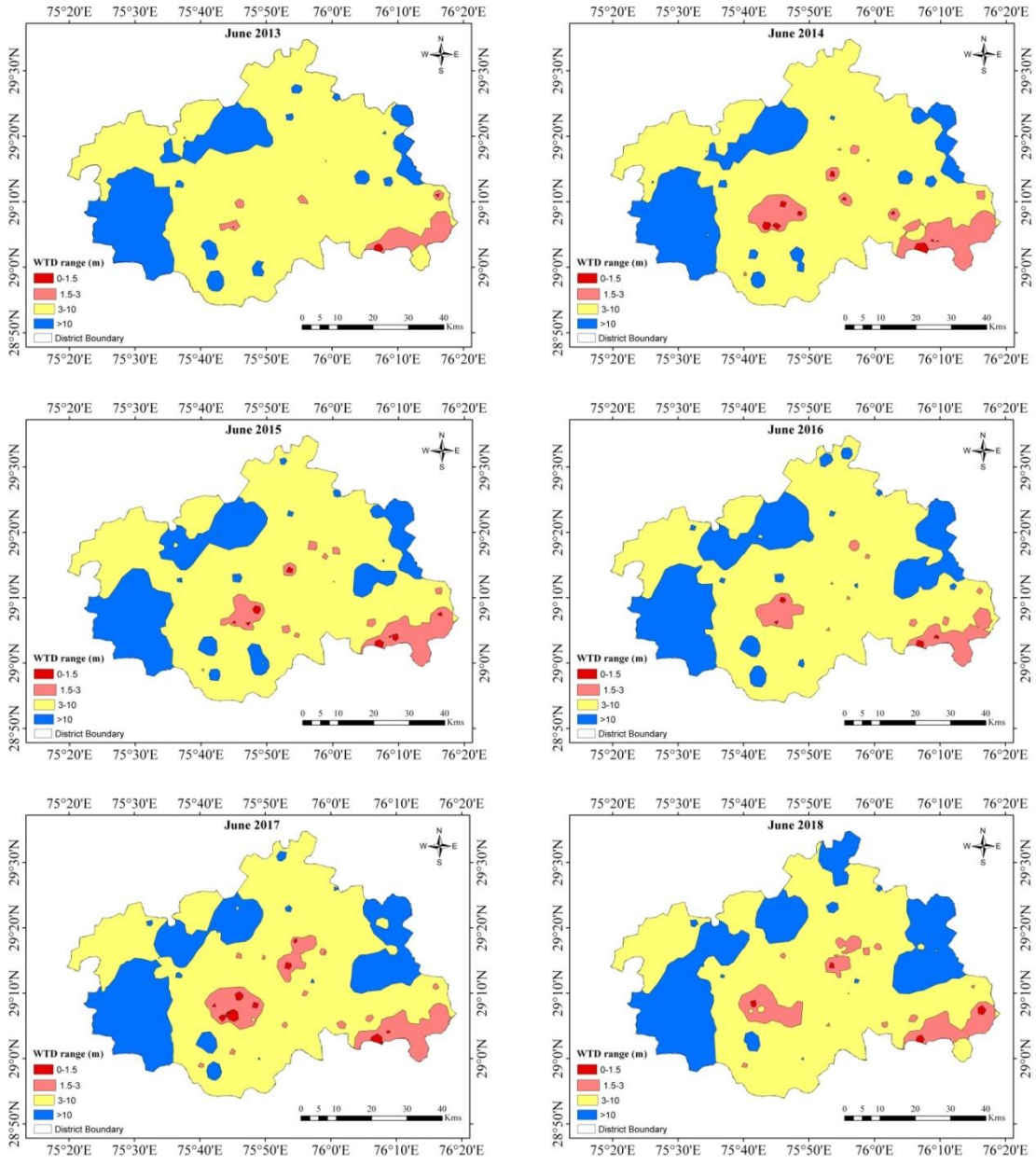


Figure 1: Water table depth maps of Hisar district for Pre-Monsoon (2013-2018)

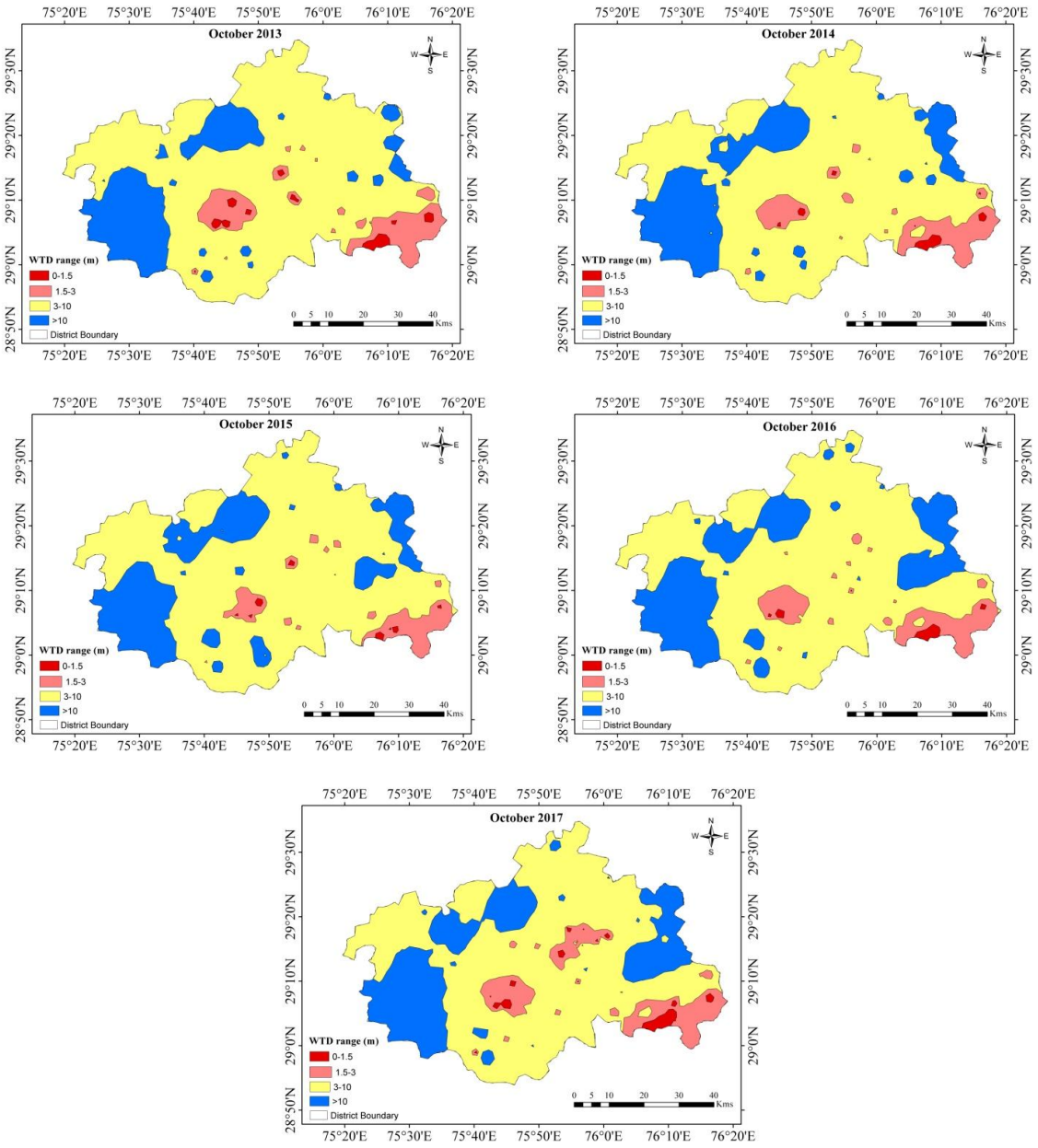
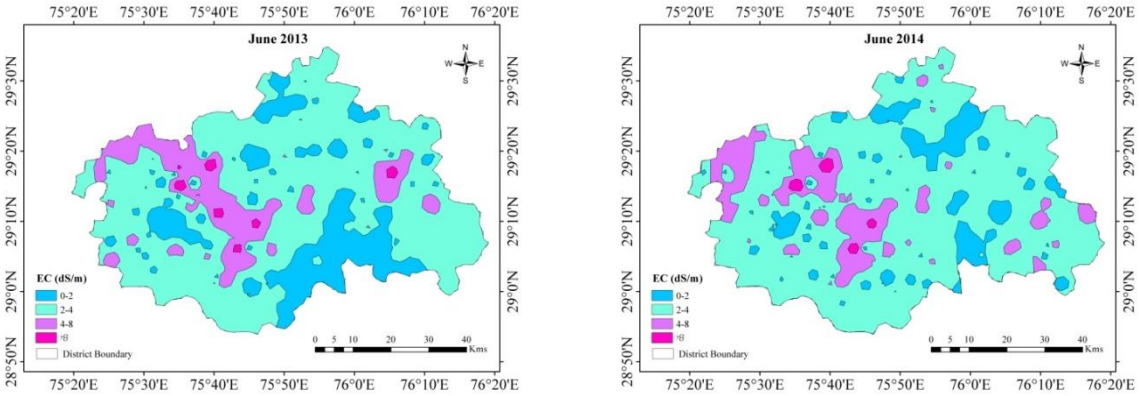


Figure 2: Water table depth maps of Hisar district for Post-Monsoon (2013-2017)



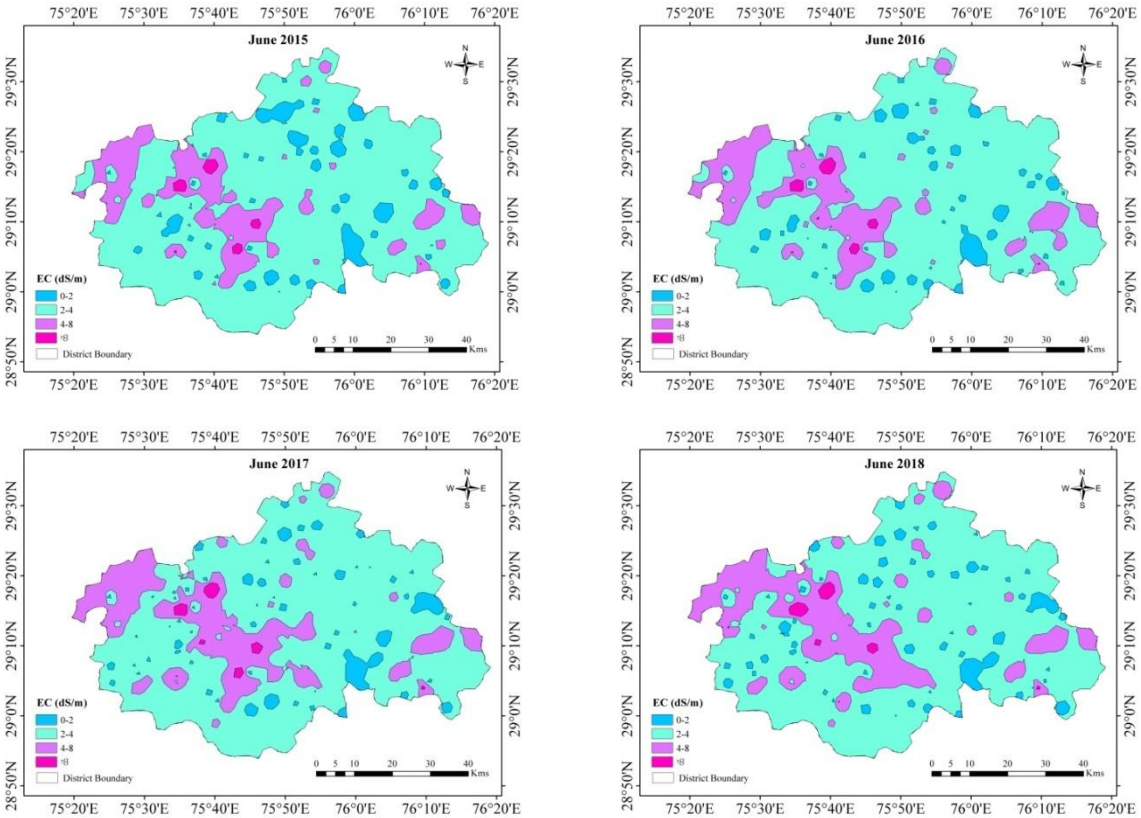
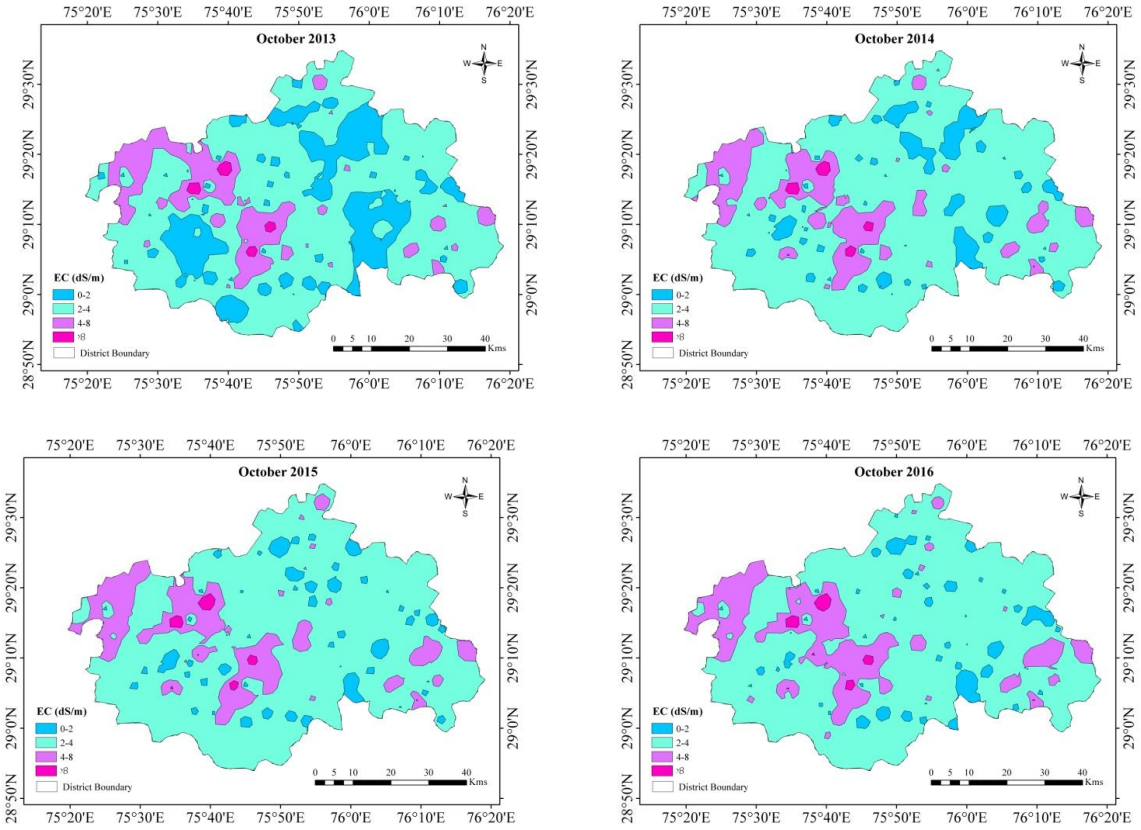


Figure 3: Groundwater quality maps of Hisar district for Pre-Monsoon (2013-18)



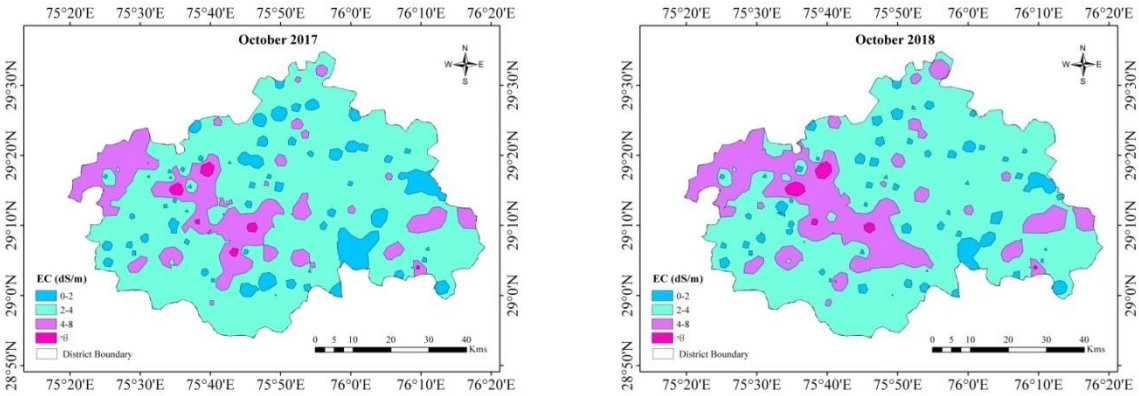


Figure 4: Groundwater quality maps of Hisar district for Post-Monsoon (2013-18)

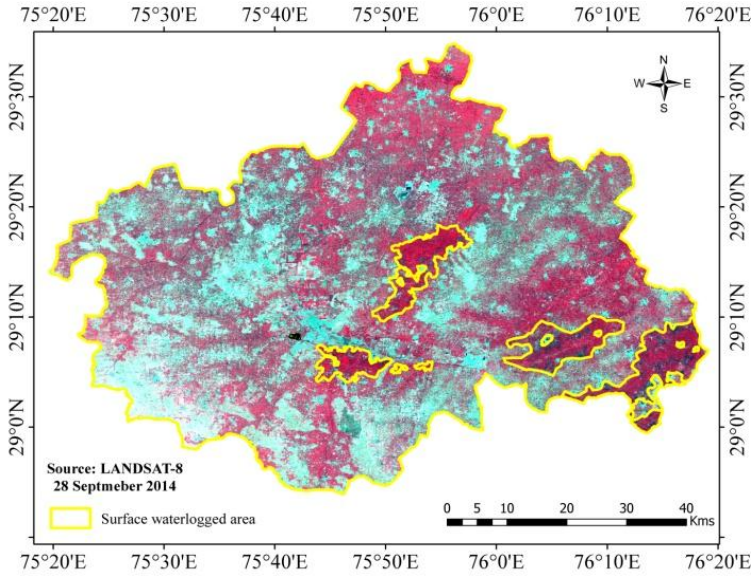


Figure 5: Surface waterlogged area in Hisar district delineated using LANDSAT-8 satellite data of 28th September 2014

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RESEARCH ARTICLE

FOREST-BASED LIVELIHOODS IN TRANSITION: A STUDY OF THE HALAKKIVAKKALIGA COMMUNITY IN UTTARA KANNADA DISTRICT, KARNATAKA

Rajeshwari N.*[1] and Nagaratna C. Kurbetta[2]

[1,2]Department of Extension and Communication Management, College of Community Science, UAS, Dharwad, Karnataka
Email: rajeshwarinavaneeth@gmail.com

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Abstract: Forest-based livelihoods have historically supported the subsistence, nutrition and cultural identity of indigenous communities, but these systems are increasingly shaped by agriculture, wage labor and market integration. This study examines the livelihood transition of the Halakki Vakkaliga community in Uttara Kannada district, Karnataka, using an ex post facto design and data collected from 500 respondents through a structured interview schedule. The findings show that the sample was largely middle-aged and older, with 29.20% in the 51–60 age group and 43.20% illiterate; most households had small landholdings of 0.10–0.60 acres (75.40%) and medium annual income (75.40%). Agriculture remained the dominant occupation for 81.60% of families, but 44.60% combined agriculture with wage earning, indicating livelihood diversification. Forest dependence remained strongest for fuelwood, used by 100% of respondents, while use of wild fruits such as jamun (38.40%), karonda (31.40%) and amla (29.80%) persisted at moderate levels. The study concludes that the Halakki Vakkaliga community is experiencing a transition from diversified forest dependence toward agriculture and wage-based livelihoods, with important implications for ecological knowledge, cultural continuity and livelihood security.

Keywords: Forest livelihoods, Halakki Vakkaliga, Indigenous community, NTFPs, Livelihood transition, Uttara Kannada

INTRODUCTION

Forests constitute one of the most important natural resource bases supporting the livelihoods, culture and identity of indigenous communities across the world. In India, forests have historically sustained the subsistence economy of tribal communities by providing a wide range of timber and non-timber forest products (NTFPs), including edible fruits, leafy vegetables, mushrooms, medicinal plants, fibres, honey, bamboo, resins and fuelwood. Beyond their economic significance, forests are deeply embedded in the cultural, spiritual and social life of indigenous societies, influencing traditional beliefs, rituals, folklore, healthcare systems and community institutions.Forests remain indispensable for the livelihoods of more than 275 million forest-dependent people in India, particularly tribal households that rely on forest resources to supplement agricultural production, improve household nutrition, generate seasonal income and reduce vulnerability during periods of economic hardship [Singh *et al*, (2024)].

However, increasing demographic pressure, agricultural expansion, infrastructure development, conservation regulations, market integration and climate variability have significantly altered patterns of forest dependence over the past few decades. Consequently, many tribal communities are experiencing a gradual transition from predominantly forest-based livelihoods towards diversified livelihood systems that combine agriculture, wage labour, livestock rearing and small-scale enterprises. Livelihood transition refers to the dynamic process through which households modify their economic activities, resource-use strategies and livelihood portfolios in response to ecological, economic, institutional and socio-cultural changes. Rather than representing a complete abandonment of traditional practices, livelihood transition often involves diversification of income sources while retaining selected components of traditional ecological knowledge and forest-resource use. For indigenous communities, this process is strongly influenced by changing access to common property resources, government development programmes, market opportunities, educational attainment and evolving aspirations of younger generations.

Within this broader context, the Western Ghats represent one of the world's most significant biodiversity hotspots, characterized by exceptional species richness, high levels of endemism and remarkable ecological heterogeneity. The ecological significance of the Western Ghats extends beyond

*Corresponding Author

biodiversity conservation, as these forests provide essential ecosystem services, including watershed protection, carbon sequestration, climate regulation and soil conservation. Simultaneously, they support the livelihoods of thousands of rural and tribal households whose survival remains closely linked to forest resources.

Among the districts located within the central Western Ghats, Uttara Kannada occupies a unique ecological and cultural position because of its extensive forest cover, diverse vegetation types and rich indigenous heritage. The district contains tropical evergreen, semi-evergreen, moist deciduous and coastal ecosystems that harbour considerable plant and animal diversity. Rural households in Uttara Kannada have traditionally depended upon forests for fuelwood, fodder, medicinal plants, wild edible species, construction materials and numerous non-timber forest products. Nevertheless, rapid socio-economic transformation, improved road connectivity, agricultural commercialization, educational expansion, migration and changing employment opportunities have influenced traditional livelihood systems, resulting in measurable changes in patterns of forest-resource utilization.

The HalakkiVakkaliga community represents one of the distinctive indigenous communities inhabiting the coastal and forested landscapes of Uttara Kannada district. Traditionally recognized for their intimate association with agriculture, forests and indigenous cultural practices, the HalakkiVakkaligas have developed extensive knowledge regarding local biodiversity, medicinal plants, wild foods, seasonal resource availability and sustainable harvesting practices. Their livelihood systems have historically integrated rainfed agriculture with the collection of forest products, livestock rearing, artisanal activities and reciprocal community labour. These studies highlight the community's role in maintaining biocultural diversity within the Western Ghats landscape.

Despite this rich ecological knowledge, evidence suggests that the nature of forest dependence among the Halakki Vakkaligas is undergoing significant transformation. Against this background, the present study investigates the transition of forest-based livelihoods among the Halakki Vakkaliga community in Uttara Kannada district of Karnataka. Specifically, the study examines the extent of dependence on forest resources, identifies major forest products contributing to household livelihoods, analyses emerging livelihood diversification strategies and interprets these changes within the broader framework of livelihood transition and sustainable rural development. By documenting contemporary patterns of forest-resource utilization and their relationship with changing socio-economic conditions, the study contributes empirical evidence to the growing discourse on indigenous livelihoods, traditional ecological knowledge and biocultural sustainability in the Western Ghats. The findings are expected to inform policies aimed at strengthening sustainable forest management, promoting diversified livelihood opportunities, conserving indigenous knowledge systems and enhancing the long-term well-being of forest-dependent tribal communities.

MATERIALS AND METHODS

The study adopted an ex post facto research design to examine livelihood transition and forest dependence among the Halakki Vakkaliga community in Uttara Kannada district, Karnataka. A structured interview schedule was used to collect information on demographic characteristics, livelihood patterns, forest resource use and cultural participation from 500 respondents.

The study was conducted in Belambar, Badiger, Hegde and Jaddigadde villages of Uttara Kannada district in Karnataka state. Uttara Kannada is a geographically diverse district along the western part of Karnataka, extending from the Arabian Sea coast to the forested foothills and higher reaches of the Western Ghats; the district is widely known for its high forest cover, ecological diversity and strong dependence of rural communities on forest and farm resources [Census of Karnataka, 2011; Ground Water Department, 2012; Centre for Ecological Sciences, IISc, 2013; Census of India, 2001]. The district’s landscape includes coastal, transitional and hilly forested zones, which create varied livelihood settings for indigenous and agrarian communities.

The selected villages represent rural settlements where Halakki Vakkaliga households maintain close links with agriculture, wage labor and forest-based resource use. These villages provide an appropriate setting for studying livelihood change because they are embedded in a landscape where farming, forest access and cultural practices continue to interact in daily life. Census and district sources confirm the presence of villages such as Jaddigadde and Hegde in Uttara Kannada, supporting their relevance as field sites for community-level research [Census of India, 2001].

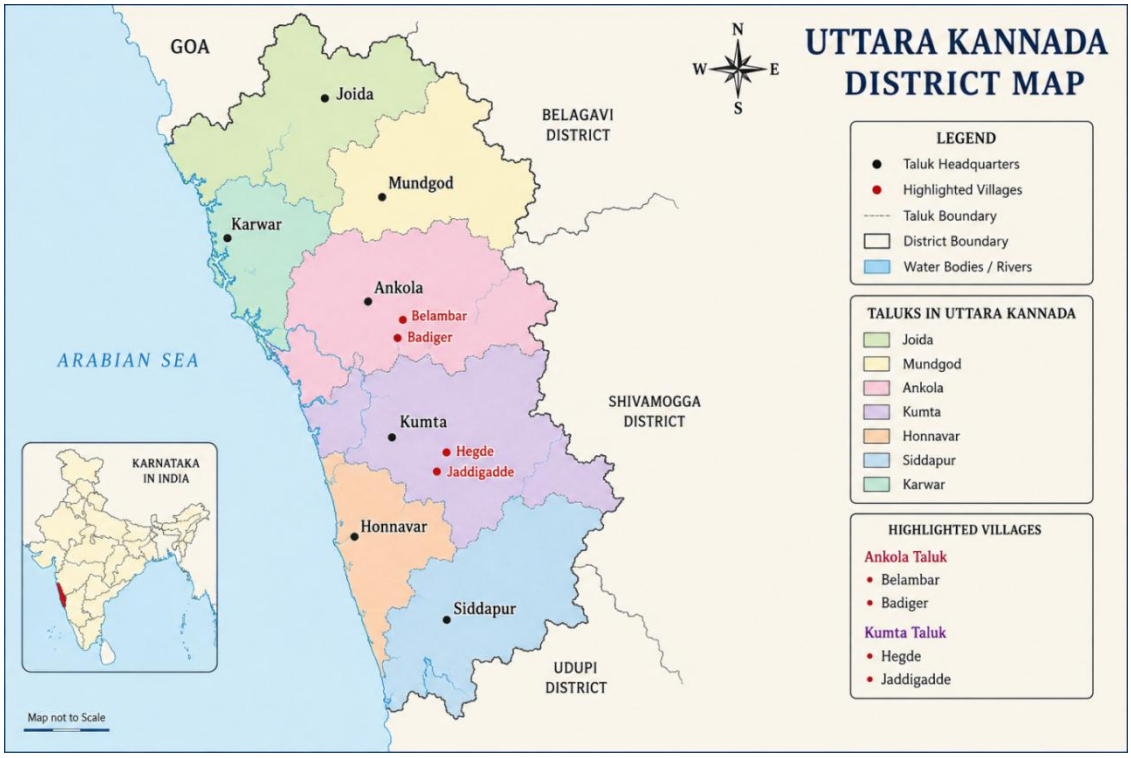

Figure 1: Map of Uttara Kannada district showing the study area

A total of 500 respondents belonging to the Halakki Vakkaliga community were selected from the four villages. The respondents were interviewed using a pretested schedule to obtain data on age, education, family type, marital status, house type, landholding, family occupation, annual income and forest use. The sample was distributed across the villages to capture the livelihood conditions of households in the study area. Primary data were collected through household interviews. The information obtained was coded, tabulated and analyzed using simple statistical tools such as frequency and percentage. The results are presented in tabular form to describe demographic characteristics, livelihood patterns and cultural participation. Interpretation focused on identifying livelihood transition, changes in forest dependence and continuity of cultural practices.

RESULTS AND DISCUSSION

Demographic characteristics of the Halakki Vakkaliga community

The demographic profile (Table 1) indicates that the majority of respondents (29.20%) belonged to the 51–60 years age group, followed by 41–50 years (23.40%), 61–70 years (20.80%), 30–40 years (20.20%), while only 6.40 per cent were aged between 71 and 80 years. The predominance of middle-aged respondents suggests that economically active adults continue to play the principal role in agricultural production, household management and preservation of indigenous ecological knowledge. Older members of indigenous communities often possess extensive traditional ecological knowledge accumulated through lifelong interaction with forests and agroecosystems. However, younger generations increasingly seek wage employment and urban opportunities, reducing their participation in traditional livelihood systems. Similar demographic trends have been documented among tribal communities of the Western Ghats, where middle-aged adults constitute the major custodians of indigenous knowledge while younger populations exhibit gradual occupational diversification

Educational attainment revealed that 43.20 per cent of respondents were illiterate, 34.40 per cent had completed primary education, 9.20 per cent had higher primary education, 8. 60 per cent had studied up to high school, while only 4.60 per cent had attained pre-university or higher education. The low educational attainment reflects historical socio-economic marginalization, limited access to educational infrastructure in forest-fringe villages, poverty and dependence on family labour. Poor educational attainment among tribal populations has consistently been linked with geographical isolation, inadequate educational facilities and opportunity costs associated with schooling

The predominance of nuclear families (57.00%) over joint families (43.00%) indicates gradual transformation in household structure. Increasing nuclearization may be attributed to land fragmentation, changing occupational aspirations, migration of younger members and government housing programmes encouraging independent households. Similar shifts from joint to nuclear families have been reported among rural and tribal

communities across India due to modernization and socio-economic transition

Regarding marital status, 76.20 per cent of respondents were married, while 20.40 per cent were widowed and only 3.40 per cent were unmarried. The relatively high proportion of widowed respondents may be associated with the inclusion of elderly respondents in the sample and greater female life expectancy. Marriage remains the predominant social institution among tribal societies, facilitating family labour sharing, inheritance and transmission of traditional knowledge.

Housing conditions demonstrate moderate economic development, with 56.40 per cent residing in semi-pucca houses, 23.60 per cent in kutcha houses and 20.00 per cent in pucca houses. The predominance of semi-pucca houses suggests gradual improvement in living standards while indicating continued economic constraints. Expansion of rural housing schemes such as the Pradhan Mantri Awas Yojana and state-sponsored housing initiatives has contributed to the transition from traditional kutcha dwellings toward semi-pucca structures in rural India

Landholding distribution reveals that 75.40 per cent possessed only 0.10–0.60 acres, while 10.20 per cent were landless, 11.60 per cent owned between 0.61–1.32 acres and only 2.80 per cent possessed more than 1.35 acres. Such predominance of marginal holdings reflects historical land fragmentation, inheritance practices and ecological limitations characteristic of the Western Ghats. Small landholdings restrict agricultural productivity and compel households to diversify into wage labour and forest-based activities. Similar findings have been reported by Spoorthi K.N. (2023).

Agriculture constituted the principal family occupation for 81.60 per cent of respondents, followed by daily wage labour (17.40%) and service employment (1.00%). The dominance of agriculture reflects continued dependence on rainfed farming despite limited land resources. However, increasing participation in wage labour indicates livelihood diversification as an adaptive response to declining agricultural profitability and seasonal unemployment. Income distribution further supports this pattern, with 75.40 per cent belonging to the medium-income category, 12.80 per cent to the low-income category and only 11.80 per cent to the high-income group. The predominance of medium-income households suggests moderate economic stability despite limited productive assets. Agricultural income supplemented by wage employment likely contributes to maintaining household consumption while preventing extreme poverty.

Livelihood pattern of the Halakki Vakkaliga community

Table 2 demonstrates that livelihood diversification is a defining characteristic of the Halakki Vakkaliga economy. Nearly 44.60 per cent combined agriculture with wage earning, while 34.80 per cent depended exclusively on agriculture and 16.20 per cent relied primarily on wage labour. Very small proportions combined agriculture with government service (2.40%), animal husbandry (0.60%), agriculture-animal husbandry-wage earning (0.80%), or agriculture-animal husbandry-horticultural marketing (0.60%).

The predominance of agriculture combined with wage labour reflects adaptive livelihood diversification among marginal farmers. Limited landholdings, seasonal fluctuations in agricultural employment, uncertain rainfall and declining profitability encourage households to supplement farm income through wage employment under public works, plantation labour, construction activities and other rural occupations. The Food and Agriculture Organization has emphasized that diversification improves resilience among smallholder farmers facing climate variability and market uncertainties (FAO, 2018).

The limited engagement in animal husbandry and horticultural enterprises suggests inadequate access to capital, veterinary services, markets and extension support. Studies conducted in the Western Ghats indicate that tribal households frequently face institutional constraints limiting diversification into higher-value enterprises despite possessing suitable ecological conditions.

Overall, the livelihood profile illustrates that agriculture remains the economic foundation of the community, while wage employment functions as an essential supplementary income source for maintaining household resilience.

Cultural participation among the Halakki Vakkaliga community

Table 3 demonstrates exceptionally high participation in ceremonial and religious activities. Marriage ceremonies were attended regularly by 56.80 per cent of respondents, while the remaining 43.20 per cent participated occasionally, indicating universal community involvement in kinship-based ceremonies.

Participation in traditional festivals such as Deepavali, Tulasi Habba, Ashada Habba, Suggi Habba, Sankranthi and Amavasya exceeded 95 per cent regular participation, while Regular Poojas recorded 88.60 per cent regular observance. Only Bandi Habba exhibited relatively lower participation, with 59.40 per cent participating regularly and 35.80 per cent reporting non-participation.

The consistently high participation in religious festivals reflects the central role of cultural traditions in maintaining collective identity, social cohesion and indigenous ecological knowledge. Many tribal festivals coincide with agricultural seasons, monsoon cycles, harvest periods and forest resource management practices. Berkes (2012) argued that traditional rituals and cultural ceremonies often function as mechanisms for conserving ecological knowledge across generations. In contrast,

recreational activities demonstrated considerably lower participation. Only 3.80 per cent regularly attended village fairs (Jatras), while 68.80 per cent participated occasionally and 27.40 per cent never attended. Participation in drama performances was even lower, with only 1.60 per cent regular attendance. The limited participation may reflect increasing occupational demands, changing entertainment preferences, migration, or declining availability of traditional folk performances.

The contrast between high participation in religious ceremonies and lower engagement in recreational events indicates that cultural practices closely linked to social identity and religious beliefs remain resilient despite ongoing socio-economic transformation. This pattern has been widely documented among indigenous communities where ritual traditions exhibit greater continuity than recreational folk arts.

Forest resource used by the Halakki Vakkaliga community

Table 4 indicates that forest dependence is highly selective rather than comprehensive. Fuelwood emerged as the only universally utilized forest product, with 100 per cent of respondents reporting its use. Fuelwood continues to constitute the primary domestic energy source because of easy accessibility, affordability and limited access to cleaner cooking fuels in remote rural settlements. Similar findings have been consistently reported across forest-dependent communities in India (FAO, 2020).

Among edible forest products, Jamun (38.40%), Karonda (31.40%) and Amla (29.80%) were the most commonly collected fruits. These wild fruits provide important seasonal nutritional supplements rich in vitamins, antioxidants and micronutrients. Only 10.00 per cent collected fodder from forests, while bamboo (1.00%), grass for broom making (1.00%), Ayurvedic medicinal materials (0.80%), gum (0.40%), leaf litter (1.20%) and leaves for meal plates (0.40%) were used by very few households. Honey collection was reported by only 3.60 per cent of respondents, whereas Pongamia seed collection was completely absent.

The universal use of fuelwood is a central result because it shows that forests still remain essential for everyday domestic energy. In the Halakki case, fuelwood appears to be the most stable remaining forest resource, while other resources have become secondary or marginal. This is a clear sign of transition rather than complete disengagement from forests.

The moderate use of jamun, karonda and amla indicates that the community still retains some edible forest knowledge. This finding is consistent with the plant-diversity work of Bhat and Rajanna (2017), which documented the Halakki community's role in maintaining useful species in home gardens and agro forestry systems. It also resonates with the ethnomedicinal study by Shanbhag and Venkatesh (2012), which showed that Uttara Kannada communities retain knowledge of selected plants for health and household use. However, the very low use of bamboo, honey, medicinal plants, gum and leaf-based products suggests that the broader NTFP system is weakening, likely because of reduced knowledge transmission, market constraints and changing livelihood priorities, as emphasized in by National Institute of Rural Development and Panchayati Raj, (2026).

The cultural data add another important dimension. High participation in major festivals and poojas shows that cultural identity remains strong. The continuity of events such as Deepavali, Tulasi Habba and Suggi Habba suggests that livelihood change has not erased ritual life. However, the low participation in Jatras and drama indicates that some forms of collective recreation and public performance are declining.

The limited extraction of most non-timber forest products (NTFPs) suggests substantial transformation in forest-based livelihoods. Increased market availability of substitute products, declining access to forests due to conservation regulations, changing occupational preferences and improved availability of commercial goods have reduced dependence on traditional forest products. Similar declines in NTFP dependence have been observed across many tribal regions following livelihood diversification and improved rural infrastructure. Nevertheless, continued reliance on fuelwood and wild edible fruits demonstrates that forests remain important for household subsistence and nutritional security. Forest ecosystems therefore continue to provide critical ecosystem services despite declining commercial extraction.

Table 1: Demographic characteristics of the Halakki Vakkaliga tribes in Uttara Kannada district of Karnataka (n=500)

Sl. No.	Variables	Category	Frequency	Percentage
1	**Age**	30-40	101	20.20
		41-50	117	23.40
		51-60	146	29.20
		61-70	104	20.80
		71-80	32	6.40

2	**Education**	Illiterate	216	43.20
		Primary (1st–4thStd)	172	34.40
		Higher Primary (5th–7thStd)	46	9.20
		High School (8th–10thStd)	43	8.60
		Pre-University College	14	2.80
		Graduate and above	9	1.80
3	**Family Type**	Nuclear	285	57.00
		Joint	215	43.00
4	**Marital Status**	Unmarried	17	03.40
		Married	381	76.20
		Widowed	102	20.40
5	**House Type**	Kutcha	118	23.60
		Pucca	100	20.00
		Semi-pucca	282	56.40
6	**Land Holding**	Landless	51	10.20
		0.10–0.60 acres	377	75.40
		0.61–1.32 acres	58	11.60
		1.35–2.00 acres	14	2.80
7	**Family Occupation**	Agriculture	408	81.60
		Daily Wage Labour	87	17.40
		Service	05	1.00
8	**Annual Income**	Low (Less than 10755)	64	12.80
		Medium (10755 to 34922)	377	75.40
		High (more than 34922)	59	11.80

Table 2: Livelihood pattern of Halakki Vakkaliga tribes in Uttara Kannada district of Karnataka (n=500)

Sl. No.	Occupational Category	Frequency	Percentage
1	Agriculture	174	34.80
2	Wage Earning	81	16.20
3	Agriculture + Wage Earning	223	44.60
4	Agriculture + Govt./Private Service	12	2.40
5	Agriculture + Animal Husbandry	03	0.60
6	Agriculture + Animal Husbandry + Wage Earning	04	0.80
7	Agriculture + Animal Husbandry + Vegetable/Flower/Fruit Selling	03	0.60

Table 3: Cultural participation of Halakki Vakkaliga tribes in Uttara Kannada district of Karnataka (n=500)

Sl. No.	Activities	Regular F (%)	Occasional F (%)	Never F (%)
I.	**Ceremonial Activities**			
1.	Marriage	284 (56.80)	216 (43.20)	0 (0.00)
II.	**Festivals**			
2.	Deepavali	476 (95.20)	24 (4.80)	0 (0.00)
3.	Tulasi Habba	476 (95.20)	24 (4.80)	0 (0.00)
4.	Ashada Habba	476 (95.20)	24 (4.80)	0 (0.00)

5.	Ugadi	475 (95.00)	24 (4.80)	1 (0.20)
6.	Bandi Habba	297 (59.40)	24 (4.80)	179 (35.80)
7.	Suggi Habba	476 (95.20)	24 (4.80)	0 (0.00)
8.	Sankranthi	476 (95.20)	24 (4.80)	0 (0.00)
9.	Amavasya	476 (95.20)	24 (4.80)	0 (0.00)
10.	Regular Pooja	443 (88.60)	57 (11.40)	0 (0.00)
2. Recreational Activities				
1	Fairs (Jatras)	19 (3.80)	344 (68.80)	137 (27.40)
2	Drama	8 (1.60)	352 (70.40)	140 (28.00)

Table 4: Non-Timber Forest Products (NTFPs) used by the Halakki Vakkaliga tribes in Uttara Kannada district (n=500)

Sl. No.	**Products**	**Yes**	**No**
		F (%)	**F (%)**
1.	Leaves for meal plate	2 (0.40)	498 (99.60)
2.	Pongamia seeds	0 (0.00)	500 (100.00)
3.	**Fruits:**		
	i. Karonda	157 (31.40)	343 (68.60)
	i. Jamun	192 (38.40)	308 (61.60)
	i. Amla	149 (29.80)	351 (70.20)
	v. Any other	18 (3.60)	482 (96.40)
4.	Fuelwood	500 (100.00)	0 (0.00)
5.	Bamboo	5 (1.00)	495 (99.00)
6.	Honey collection	18 (3.60)	482 (96.40)
7.	Grass for brooms	5 (1.00)	495 (99.00)
8.	Gum collection	2 (0.40)	498 (99.60)
9.	Ayurvedic (bark, leaf, roots, flowers, seeds, etc.,)	4 (0.80)	496 (99.20)
10.	Fodder	50 (10.00)	450 (90.00)
11.	Leaf litter	6 (1.20)	494 (98.80)

CONCLUSION

Overall, the findings indicate that the Halakki Vakkaliga community is transitioning from a predominantly forest-dependent livelihood system towards a diversified rural economy without completely abandoning its traditional knowledge systems and cultural values. This coexistence of socio-economic transformation and cultural continuity underscores the adaptive capacity of the community in responding to environmental, economic and institutional changes.

The study highlights the need for policy interventions that simultaneously promote sustainable livelihood enhancement and cultural conservation. Strengthening agricultural productivity through climate-resilient farming practices, improving access to education and skill development, promoting value addition and marketing of locally available forest and agricultural products, supporting women's livelihood enterprises and documenting indigenous knowledge systems can significantly enhance community resilience. Furthermore, participatory forest management and biodiversity conservation programmes should actively incorporate the traditional ecological knowledge of the Halakki Vakkaliga community, recognizing indigenous people as key stakeholders in achieving sustainable development and conserving the unique biodiversity of the Western Ghats.

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SHORT COMMUNICATION

IN VITRO CALLUS INDUCTION AND SHOOT REGENERATION FROM LEAF EXPLANTS OF *BARLERIA LUPULINA* LINDL.

Manju M. George*

Department of Botany, Union Christian College (Autonomous), Aluva, Ernakulam District, Kerala – 683 102, India
Email: manju@uccollege.edu.in

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Abstract: *Barleria lupulina* Lindl. is a medicinally important shrub native to Southeast Asia, valued in Thai folk medicine for anti-inflammatory, anti-amoebic, and diuretic properties. Overexploitation and limited natural regeneration have created an urgent need for reliable *in vitro* propagation protocols. In the present study, callus was induced from leaf, petiole, and stem explants on Murashige and Skoog (MS) medium supplemented with combinations of 6-benzylaminopurine (BAP), indole-3-butyric acid (IBA), and α-naphthaleneacetic acid (NAA). Leaf explants showed the highest callus induction frequency. Established callus was transferred to MS medium containing BAP and/or kinetin (Kn), alone or with low concentrations of auxins, to initiate shoot organogenesis. The present study reports the first systematic account of callus-mediated shoot regeneration in *B. lupulina* and establishes a framework for clonal multiplication, germplasm conservation, and future genetic improvement of this species.

Keywords: *Barleria lupulina*, Callus culture, Acanthaceae, Plant growth regulators

INTRODUCTION

The genus *Barleria* L. comprises approximately 300 species distributed across tropical and subtropical regions, with greatest diversity in eastern Africa and Southeast Asia [Sudheer and Praveen, 2021]. The genus is widely recognised for pharmacological potential, with documented antibacterial, anti-inflammatory, and immunomodulatory activities [Sawarkar *et al.*, 2023], [Lekhak, *et al.,* 2022]. *Barleria lupulina* Lindl., commonly known as "Sornomukhi," is a perennial shrub distributed across South and Southeast Asia and occupies a prominent place in Thai traditional medicine, where it is applied externally for inflammation, insect and snake bites, herpes simplex and varicella zoster lesions, and is taken for amoebic infection and diuresis [Sawangjaroen *et al*., 2006], [Mandal *et al*., 2014].

Phytochemical investigations have identified a rich profile of bioactive compounds in *B. lupulina* including iridoid glycosides, phenylethanoid glycosides, and terpenoidal compounds [Lee *et al.*, 2016], [Kim *et al.,* 2015]. Pharmacological studies confirm antibacterial activity against multiple human pathogens including *Bacillus pumilus*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* [Sarmad *et al*., 2012], [Kumari and Dubey, 2016]; significant antioxidant and immunomodulatory effects in animal assays [Kumari *et al.*, 2016]; anti-inflammatory activity via Nrf2 pathway activation in microvascular endothelial cells [Senger *et al.,* 2016]; and cytotoxic effects on cancer cell lines [Kumari *et al.*, 2016]. This documented bioactivity has fuelled escalating commercial harvest, placing pressure on wild populations and reinforcing the need for reliable propagation technology.

Conventional propagation of *B. lupulina* is constrained by poor seed viability and slow vegetative growth. *In vitro* micropropagation bypasses these limitations by enabling rapid, year-round clonal multiplication of elite material, free from pathogens, and genetically uniform [Loberant and Altman, 2010], [Kaviani, 2015]. Within Acanthaceae, micropropagation protocols have been established for several related species — *Barleria prionitis* var. *dicantha* (75.5% bud break on 8.88 µM BA; rooting on 2.46 µM IBA) [Singh *et al.*, 2015], *Justicia gendarussa* (87% shoot response on 17.7 µM BA; 92% callus-mediated regeneration on BA + NAA) [Thomas and Hoshino, 2010], *Adhatoda beddomei* (5–10 shoots per explant on 3.0 mg L^{-1} BAP + 1.0 mg L^{-1} IAA) [Sudha and Seeni, 1994], and *Centratherum punctatum* (100% shoot induction, ~30 shoots per explant on 4.5 mg L^{-1} BAP + 4.0 mg L^{-1} Kn) [Talan *et al*., 2023]. Published information specific to tissue culture of *B. lupulina* is restricted to a preliminary callus establishment study [Singh *et al*., 2017].

*Corresponding Author

The present study aims to: (i) establish callus from leaf, petiole, and stem explants; and (ii) regenerate shoots from established callus using BAP and Kn combinations, with the objective of developing a reproducible propagation protocol for this medicinally important species.

MATERIALS AND METHODS

Plant Material and Explant Preparation

Plants of *Barleria lupulina* Lindl. were maintained in the greenhouse of the Department of Botany, Union Christian College, Aluva, Kerala. Young leaves, petioles, and stem segments up to the 5th node from actively growing shoots were used as explant sources. Explants were washed under running tap water for 15 min, treated with 2% (v/v) liquid detergent for 5 min, and rinsed thoroughly.

Surface Sterilization

All sterilization was performed in a laminar air flow cabinet. Explants were immersed in 70% (v/v) ethanol for 30 s, rinsed with sterile distilled water, and then surface-sterilized with 0.1% (w/v) mercuric chloride ($HgCl_2$) for 3 min (leaves and petioles) or 5 min (stem segments). Explants were rinsed 4–5 times with sterile distilled water, trimmed to approximately 1.0–1.5 cm, and inoculated immediately.

Culture Media and Growth Conditions

MS basal medium [20] supplemented with 3% (w/v) sucrose and solidified with 0.8% (w/v) agar was used throughout. The pH was adjusted to 5.8 ± 0.1 before autoclaving (121°C, 15 psi, 20 min). Plant growth regulators (PGRs) — BAP, Kn, IBA, and NAA — were filter-sterilized (0.22 µm) and added to cooled (~50°C) medium. Cultures were maintained at 25 ± 2°C under a 16-h photoperiod (40–50 µmol m^{-2} s^{-1}, cool-white fluorescent lamps) with 60–70% relative humidity.

Callus Induction

Explants were cultured on MS medium supplemented with BAP (0.5, 1.0, 2.0, 3.0 mg L^{-1}) in combination with IBA (0.5, 1.0, 2.0 mg L^{-1}) and NAA (0.5, 1.0, 1.5 mg L^{-1}). Each treatment included 15–20 explants and experiments were replicated three times. Callus induction percentage, colour, and texture were recorded at 4 weeks.

Shoot Regeneration

Well-established calli (4–6 weeks old) were transferred to MS medium containing BAP (1.0, 2.0, 3.0, 4.0 mg L^{-1}) or Kn (1.0, 2.0, 3.0, 4.0 mg L^{-1}) alone or in combination with NAA or IBA (0.5 mg L^{-1}). Shoot regeneration frequency, number of shoots per callus, and shoot length were recorded after 6–8 weeks.

Statistical Analysis

Experiments were arranged in a completely randomized design (CRD) with three replications. Percentage data were arcsine-transformed before analysis of variance (ANOVA). Means were compared using Duncan's Multiple Range Test (DMRT) at $p < 0.05$. Data are presented as mean ± SE.

RESULTS AND DISCUSSION

Callus Induction from Different Explant Types

Surface contamination, a primary challenge in medicinal plant tissue culture, was effectively controlled by the $HgCl_2$ protocol; contamination rates are presented in Table 1. Callus initiation was observed within 10–14 days of culture, appearing first at cut edges before spreading across explant surfaces.

Table 1. Effect of BAP, IBA, and NAA combinations on callus induction from leaf, petiole, and stem explants of *B. lupulina* on MS medium (4 weeks)

Treatment No.	BAP (mg L^{-1})	IBA (mg L^{-1})	NAA (mg L^{-1})	Explant Type	Callus Induction (%)	Callus Colour	Callus Texture
T1	0.5	0.5	0.5	Leaf	48.3 ± 2.1 c	Cream-white	Compact
T2	1.0	0.5	0.5	Leaf	63.4 ± 2.8 b	Pale yellowish	Friable
T3	2.0	1.0	0.5	Leaf	82.5 ± 3.2 a	Yellowish-green	Friable
T4	3.0	1.0	1.0	Leaf	60.7 ± 2.5 b	Dark brown	Compact
T5	2.0	1.0	0.5	Petiole	68.3 ± 2.9 b	Cream-yellow	Friable
T6	2.0	1.0	0.5	Stem	51.7 ± 2.4 c	Cream	Compact

Values are mean ± SE of three replications (n = 15–20 explants per treatment). Means followed by different letters within a column differ significantly (DMRT, $p < 0.05$). [Author to insert experimental observations]

Leaf explants produced the highest callus induction frequency across all PGR combinations, followed by petioles and stem segments. This hierarchy is consistent with reports in other Acanthaceae species: leaf-derived callus outperformed nodal explants in *Justicia gendarussa* Thomas and Hoshino, 2010. The superior response of leaf explants is attributed to higher meristematic cell density and more favourable endogenous hormone ratios [Skoog and Miller, 1957]. Friable, yellowish-green to cream-coloured callus — optimal for subsequent organogenesis — was obtained at mid-range BAP concentrations combined with IBA and NAA, while high-auxin treatments yielded compact, dark-brown callus with poor growth. This pattern mirrors findings in *Barleria prionitis*, where callus colour and texture varied markedly with auxin concentration [Singh *et al.*,2015].

The synergistic requirement for cytokinin and auxin in callus induction supports the Skoog–Miller model [Jain and Häggman, 2007], wherein the relative ratio of these hormones determines developmental fate. Callus formation and organogenesis in *Acanthaceae* species consistently depend on fine-tuning this ratio rather than application of either hormone class alone [Thomas and Hoshino, 2010], [Sudha and Seeni, 1994].

Shoot Organogenesis from Callus

Shoot bud differentiation was observed within 3–4 weeks of transfer to regeneration medium. Multiple shoot primordia emerged from the callus surface and elongated progressively over 2–3 weeks to reach 2–4 cm, suitable for excision and subculture.

Table 2. Effect of BAP and kinetin concentrations on shoot regeneration from *B. lupulina* callus on MS medium (8 weeks)

Treatment No.	BAP (mg L^{-1})	Kn (mg L^{-1})	Auxin (mg L^{-1})	Shoot Regeneration (%)	Shoots per Callus (mean ± SE)	Mean Shoot Length (cm)
R1	1.0	0	—	25.0 ± 3.2 d	4.2 ± 0.5 d	1.8 ± 0.2 c
R2	2.0	0	—	43.3 ± 2.8 bc	8.6 ± 0.8 bc	2.6 ± 0.3 b
R3	3.0	0	—	56.7 ± 3.5 a	12.4 ± 1.2 a	3.2 ± 0.4 a
R4	4.0	0	—	36.7 ± 3.0 c	7.3 ± 0.7 c	2.2 ± 0.3 b
R5	0	2.0	—	30.0 ± 2.5 cd	5.8 ± 0.6 cd	2.0 ± 0.2 bc
R6	2.0	1.0	NAA 0.5	50.0 ± 3.3 ab	14.7 ± 1.5 a	3.5 ± 0.4 a
R7	2.0	2.0	IBA 0.5	46.7 ± 3.1 b	11.3 ± 1.1 ab	3.0 ± 0.3 ab

Values are mean ± SE of three replications. Means followed by different letters within a column differ significantly (DMRT, $p < 0.05$). [Author to insert experimental observations]

Cytokinin-dominant media effectively induced shoot organogenesis, supporting the well-established role of BAP and Kn in promoting shoot differentiation in callus cultures. Moderate BAP concentrations yielded the highest shoot regeneration frequency and number of shoots per callus. Addition of low-concentration auxin (NAA or IBA at 0.5 mg L^{-1}) in combination with BAP further enhanced shoot number in some treatments, consistent with synergistic cytokinin–auxin interactions noted in *Adhatoda beddomei* [Sudha and Seeni, 1994] and *Centratherum punctatum* [Talan *et al*., 2023]. However, elevated auxin concentrations suppressed shoot formation and promoted root formation, as expected from the Skoog–Miller balance.

For comparative context, shoot regeneration frequencies and shoot numbers reported in related Acanthaceae species range widely: *J. gendarussa* achieved 92% regeneration from callus at 17.7 µM BA + 5.4 µM NAA Thomas and Hoshino, 2010; *C. punctatum* achieved 100% shoot induction with ~30 shoots per explant at 4.5 mg L^{-1} BAP + 4.0 mg L^{-1} Kn [Talan *et al*., 2023]; *Phlogacanthus thyrsiflorus* produced a mean of 13 shoots per explant at 2.0 mg L^{-1} BA [Singh *et al*., 2017]. The response of *B. lupulina* in the present study falls within this reported range and is consistent with the general Acanthaceae pattern of responsiveness to cytokinin-based regeneration systems.

The present study provides the first peer-reviewed account of callus-mediated shoot organogenesis in *B. lupulina*, addressing a gap noted in the genus-level review by Sudheer and Praveen [2021]. The establishment of this regeneration system has direct applications in: (i) clonal multiplication for conservation and commercial planting-material supply; (ii) establishment of a transformation competent tissue system for introducing disease-resistance or metabolite-enhancing transgenes; and (iii) callus-based secondary metabolite production studies targeting the iridoid and phenylethanoid glycosides responsible for the plant's

pharmacological activities [Leet *et al*., 2016], [Kim *et al.*, 2015], [Senger *et al*., 2015].

***In Vitro* Rooting of Regenerated Shoots**

Excised shoots (2–3 cm) were transferred to half-strength MS medium supplemented with different concentrations of IBA and NAA for rooting. Root initiation was first observed within 8–12 days of culture, and fully rooted plantlets with well-developed root systems were obtained within four weeks. Results are summarised in Table 3.

Table 3. Effect of IBA and NAA on in vitro rooting of regenerated B. lupulina shoots on half-strength MS medium (4 weeks)

Treatment No.	IBA (mg L^{-1})	NAA (mg L^{-1})	Rooting Frequency (%)	Roots per Shoot (mean ± SE)	Mean Root Length (cm)	Days to Root Initiation
Ro1	0	0	13.3 ± 1.8 e	1.2 ± 0.3 d	0.8 ± 0.1 d	18.3 ± 1.2 a
Ro2	0.5	0	46.7 ± 3.2 d	3.8 ± 0.5 c	1.5 ± 0.2 c	14.5 ± 1.0 b
Ro3	1.0	0	66.7 ± 3.5 c	5.3 ± 0.6 bc	2.1 ± 0.3 b	12.3 ± 0.8 bc
Ro4	2.0	0	86.7 ± 4.1 a	8.7 ± 0.9 a	3.2 ± 0.4 a	10.8 ± 0.7 c
Ro5	3.0	0	73.3 ± 3.8 b	6.4 ± 0.7 b	2.6 ± 0.3 ab	11.5 ± 0.8 c
Ro6	0	0.5	40.0 ± 2.9 d	3.2 ± 0.4 c	1.3 ± 0.2 cd	15.2 ± 1.1 b
Ro7	0	1.0	56.7 ± 3.3 cd	4.6 ± 0.5 bc	1.8 ± 0.2 bc	13.7 ± 0.9 bc
Ro8	2.0	0.5	80.0 ± 3.9 ab	7.2 ± 0.8 ab	2.9 ± 0.4 ab	11.2 ± 0.7 c

Values are mean ± SE of three replications (n = 15 shoots per treatment). Means followed by different letters within a column differ significantly (DMRT, $p < 0.05$). Ro1 = auxin-free control; IBA = indole-3-butyric acid; NAA = α-naphthaleneacetic acid.

CONCLUSION

A complete in vitro regeneration protocol for *Barleria lupulina* Lindl. was successfully established through three sequential stages. Callus induction was most efficient from leaf explants on MS medium supplemented with BAP 2.0 mg L^{-1} + IBA 1.0 mg L^{-1} + NAA 0.5 mg L^{-1} (T3), yielding 82.5 ± 3.2% callus induction with friable, yellowish-green callus optimal for subsequent organogenesis. Shoot organogenesis was maximised on MS medium with BAP 3.0 mg L^{-1} alone (R3; 56.7 ± 3.5% regeneration; 12.4 ± 1.2 shoots per callus), while supplementation with Kn 1.0 mg L^{-1} + NAA 0.5 mg L^{-1} to BAP 2.0 mg L^{-1} (R6) further elevated shoot number to 14.7 ± 1.5 per callus with mean shoot length of 3.5 ± 0.4 cm. In vitro rooting of regenerated shoots was most effective on half-strength MS supplemented with IBA 2.0 mg L^{-1} (Ro4), achieving 86.7 ± 4.1% rooting frequency with 8.7 ± 0.9 roots per shoot and a mean root length of 3.2 ± 0.4 cm within four weeks. Leaf explants were consistently the most responsive source material across all culture stages. Together, the three-stage protocol provides a reproducible foundation for clonal multiplication and germplasm conservation of this overexploited medicinal species, and constitutes a direct prerequisite for future genetic transformation programs targeting the iridoid and phenylethanoid glycosides responsible for the pharmacological activities of *B. lupulina*.

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