



***Coniambigua diorygmatis* and *Distopyrenis graphidis*: Two new lichenicolous fungi on Graphidaceae from India**

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Abstract

Two new lichenicolous fungi, *Coniambigua diorygmatis* and *Distopyrenis graphidis*, are described from India. *Coniambigua diorygmatis* is characterized by the black sporodochia producing dark brown, ellipsoidal, non-septate conidia and was found on the thallus of *Diorygma junghuhnii*, whereas *D. graphidis* is distinguished by perithecioid ascomata with an apical ostiole, 1-septate brown ascospores on *Graphis stenotera*.

Keywords – *Coniambigua* – *Distopyrenis* – India – Lichenicolous fungi – new species

Introduction

Lichenicolous fungi, those that inhabit, parasitize, or form intricate associations with lichens, represent a taxonomically and ecologically diverse group of mycobiota. These fungi may be biotrophic, necrotrophic, or commensalistic, and are known for their high degree of host specificity and remarkable morphological adaptations (Lawrey & Diederich 2003, Hawksworth 2003). They often exhibit reduced or specialized structures, including pycnidia, perithecia, or immersed ascomata, enabling them to persist within or upon their lichen hosts with minimal external manifestation. The interactions between lichenicolous fungi and their hosts can range from benign cohabitation to pronounced pathogenicity, occasionally resulting in significant morphological alterations or suppression of the host's reproductive structures (Lawrey & Diederich 2003). Although more than 2,300 species of lichenicolous fungi are known worldwide (Diederich et al. 2025), their true diversity is likely far greater, constrained by their inconspicuous habit and the taxonomic challenges posed by reduced morphological features (Hawksworth & Rose 1976). India, with its vast array of climatic zones and topographic complexity, harbors a vibrant lichen flora, particularly within Himalayan biodiversity hotspots (Singh et al. 2015, 2018). Despite this floristic richness, lichenicolous fungi remain poorly explored in the Indian subcontinent. Earlier records are fragmentary, and only a modest number of taxa have been formally reported (Joshi et al. 2015, Sharma et al. 2016, Singh et al. 2017, Singh & Singh 2019, Sharma et al. 2023). Systematic surveys focusing on lichenicolous assemblages have been sporadic, and many regions remain underexplored, underscoring the likelihood that numerous undescribed species await discovery.

During recent lichen biodiversity surveys in Himachal Pradesh, part of the western Himalayan region, two previously undocumented lichenicolous fungi were discovered. The first, *Coniambigua diorygmatis*, was found colonizing on the thallus of *Diorygma junghuhnii*, while the second, *Distopyrenis graphidis*, was collected from *Graphis stenotera*. Both specimens possess distinct morphological and anatomical features that justify their recognition as new species. In this study, we provide detailed descriptions of these taxa based on meticulous morphological and anatomical examination, host association data, and comparative analyses with similar species. Their taxonomic placement and ecological significance are discussed in the broader context of lichenicolous fungal diversity in the Indian Himalaya.

Materials & Methods

Specimens were collected from Himachal Pradesh, Western Himalayas, India. Host lichens were identified using standard taxonomic keys and relevant literature. Fungal structures were examined under a dissecting stereomicroscope (NIKON SMZ 7451) and a compound light microscope (Nikon 50i, Leica DM 3000). Freehand sections were prepared and mounted in water, 10% KOH, and Lugol's iodine for diagnostic observations. Morphometric data were obtained from at least 30 mature ascospores or conidia per species. Voucher specimens are deposited in the herbarium of the Botanical Survey of India, Dehradun (BSD), and are available for future reference.

Results

Taxonomy

Coniambigua diorygmatis Pushpi Singh & Aptroot sp. nov. (Fig. 1. A–F)

MycoBank number: MB862231

Diagnosis – Colonies form discrete patches on the thallus and ascomata of the host. It is differentiated from other lichenicolous *Coniambigua* species in having erumpent, convex black sporodochia, dark brown, ellipsoidal, non-septate, $3.5\text{--}5.6 \times 3\text{--}4\ \mu\text{m}$, K– conidia.

Etymology – The epithet *diorygmatis* is derived from *Diorygma*, the genus of the host lichen.

Typus — India, Himachal Pradesh, Mandi district, on the way to Prashar, forest section, alt. c. 1000 m, deciduous forest, on the thallus of *Diorygma junghuhnii*, 25 April 2025, Pushpi Singh 137796 (BSD – holotypus).

Description – Sexual morph: unknown. Asexual morph: Colonies lichenicolous on *Diorygma junghuhnii*, forming erumpent, minuscule, convex sporodochia, black, rounded, $60\text{--}150\ \mu\text{m}$ diam., some confluent. Vegetative mycelium indistinct, hyaline to brown, immersed in the host thallus. Conidiophores and Conidiogenous cells not very distinct. Conidia aseptate, round to ellipsoid, pale brown to dark brown at maturity, K–, smooth-walled, $3.5\text{--}5.6 \times 3\text{--}4\ \mu\text{m}$, ($n = 40$), with $\sim 0.5\ \mu\text{m}$ thick walls.

Distribution and hosts – This species was known only from the type locality in India on the thallus of *Diorygma junghuhnii*.

Remarks – The genus *Coniambigua* was established by Etayo & Diederich (1995), with *C. phaeographidis* Etayo & Diederich designated as the type species, which was described on *Phaeographis* species. The genus is characterized by stromatic, sporodochial to pseudopycnidial conidiomata, immersed to slightly erumpent, dark brown to black, irregularly opening; walls reduced or absent. Conidiogenous cells are hyaline to pale brown. Conidia dark brown, 0(–1)-septate, smooth to verruculose, subspherical, ellipsoidal, subcylindrical, or polyhedral; conidiomata aggregated or indistinct (Diederich et al. 2024). Globally, four species of *Coniambigua* have been reported to date, three of which are associated with members of the Graphidaceae (Diederich et al. 2024). These include *C. aptrootii* (Diederich) Diederich on *Fissurina dumastii*, *C. graphidis* Kukwa & Diederich found on *Graphis scripta*, *C. phaeographidis* Etayo & Diederich, on *Phaeographis* species, and *C. ochrolechia* Diederich on *Ochrolechia*. During the study of graphidoid lichens in India, a new species of *C. diorygmatis* was discovered colonizing on the thallus of *Diorygma junghuhnii*. It is

characterized by its erumpent black convex sporodochia; dark brown, non-septate, smooth-walled, ellipsoidal conidia, measuring $3.5\text{--}5.6 \times 3.0\text{--}4.0\ \mu\text{m}$. This species closely resembles *C. aptrootii* and *C. graphidis* in non-septate, smooth-walled conidia (Diederich et al. 2024). However, *C. aptrootii* (on *Fissurina dumastii*) is characterized by the presence of 1-septate conidia, dividing in aseptate, often triangular-shaped part-conidia, and *C. graphidis* (on *Graphis scripta*) is characterized by smaller, smooth, K⁺ olivaceous conidia and occurs on different hosts. Comparison with the remaining known species of *Coniambigua* is presented in Table 1.

Table 1 *Coniambigua* species growing on Graphidaceae (Diederich et al. 2024).

Species	Host	Ascomata	Conidia	Country reported
<i>C. diorygmatis</i> sp. nov.	<i>Diorygma junghuhnii</i>	Stromatic sporodochial, erumpent, 50–100 μm	Conidia smooth, mainly ellipsoidal, aseptate, brown to dark brown $3.5\text{--}5.6 \times 3\text{--}4\ \mu\text{m}$, K [–] .	India
<i>C. aptrootii</i>	<i>Fissurina dumastii</i>	Stromatic sporodochial, immersed, later erumpent 50–100 μm	Conidia smooth, ellipsoidal or angular, often triangular-shaped, aseptate, medium to dark brown $5\text{--}6.5 \times 4\text{--}5\ \mu\text{m}$, K [–] ;	America
<i>C. graphidis</i>	<i>Graphis scripta</i>	stromatic, sporodochial to pseudopycnidial, immersed or erumpent; 50–150 μm diam.	Conidia smooth, subspherical to ellipsoidal or polyedric, aseptate, medium to dark brown, $3\text{--}5 \times 2.5\text{--}4\ \mu\text{m}$, K ⁺ olivaceous	Poland
<i>C. phaeographidis</i>	<i>Phaeographis</i> s. lat	stromatic, sporodochial to pseudopycnidial, immersed or erumpent, dark brown to black, 60–100 μm diam.	Conidia verruculose, aseptate or rarely 1-septate; rounded to cylindrical or polyedric, $7.5\text{--}12(16) \times 5\text{--}8\ \mu\text{m}$, K [–] .	America
<i>C. ochrolechia</i>	On <i>Ochrolechia</i>	sporodochium-like agglomerations of conidia, black, irregular in form, 20–100 μm diam.	Conidia ellipsoidal or angular, aseptate, dark brown, $4.8\text{--}6.0(7.3) \times 4.1\text{--}5.1\ \mu\text{m}$, K [–] .	N. America (Mexico)

Distopyrenis graphidis Pushpi Singh & Aptroot sp. nov.

(Fig. 2. A–D)

MycoBank number: MB862232

Diagnosis – Colonies form discrete patches mainly on the thallus of the host and rarely on the ascomata. Distinguished from other *Distopyrenis* species by perithecial ascomata with apical ostiole, brown fusiform, transversely 1-septate ascospores ($11\text{--}14 \times 4.5\text{--}5.5\ \mu\text{m}$), and its strict association with the thallus of *Graphis* species.

Typus – India, Himachal Pradesh, Mandi district, on the way to Prashar, alt. 1000 m, on the thallus of *Graphis stenotera* (Graphidaceae), 25 April 2025, Pushpi Singh 137795 (BSD, holotype).

Etymology – The species epithet *graphidis* denotes its association with the lichen genus *Graphis*.

Description – The fungus occurs on the thallus of the host lichen (*Graphis stenotera*). *Ascomata* perithecial, black, subspherical, erumpent, 120–150 μm in diameter, mostly solitary or in loose groups; perithecial wall, ca. 20 μm thick in lateral zones and to 40 μm in upper zones; *Ostiole* apical, sunken, simple; periphyses simple or sparsely branched, 1–1.5 μm wide; *Hamathecium* hyaline, non-inspersed; *Paraphyses* simple to sparsely branched; *Asci* clavate, 8-spored, bitunicate, narrowly cylindrical, young ones with thickened lateral wall, mature with a cylindrical ocular chamber, I–, KI–

, $45\text{--}55 \times 9\text{--}10\text{ }\mu\text{m}$; *Ascospores* arranged uni to biserially, brown, fusiform, transversely 1-septate, with 2 angular lumina, I–, $11\text{--}14 \times 4.5\text{--}5.5\text{ }\mu\text{m}$.

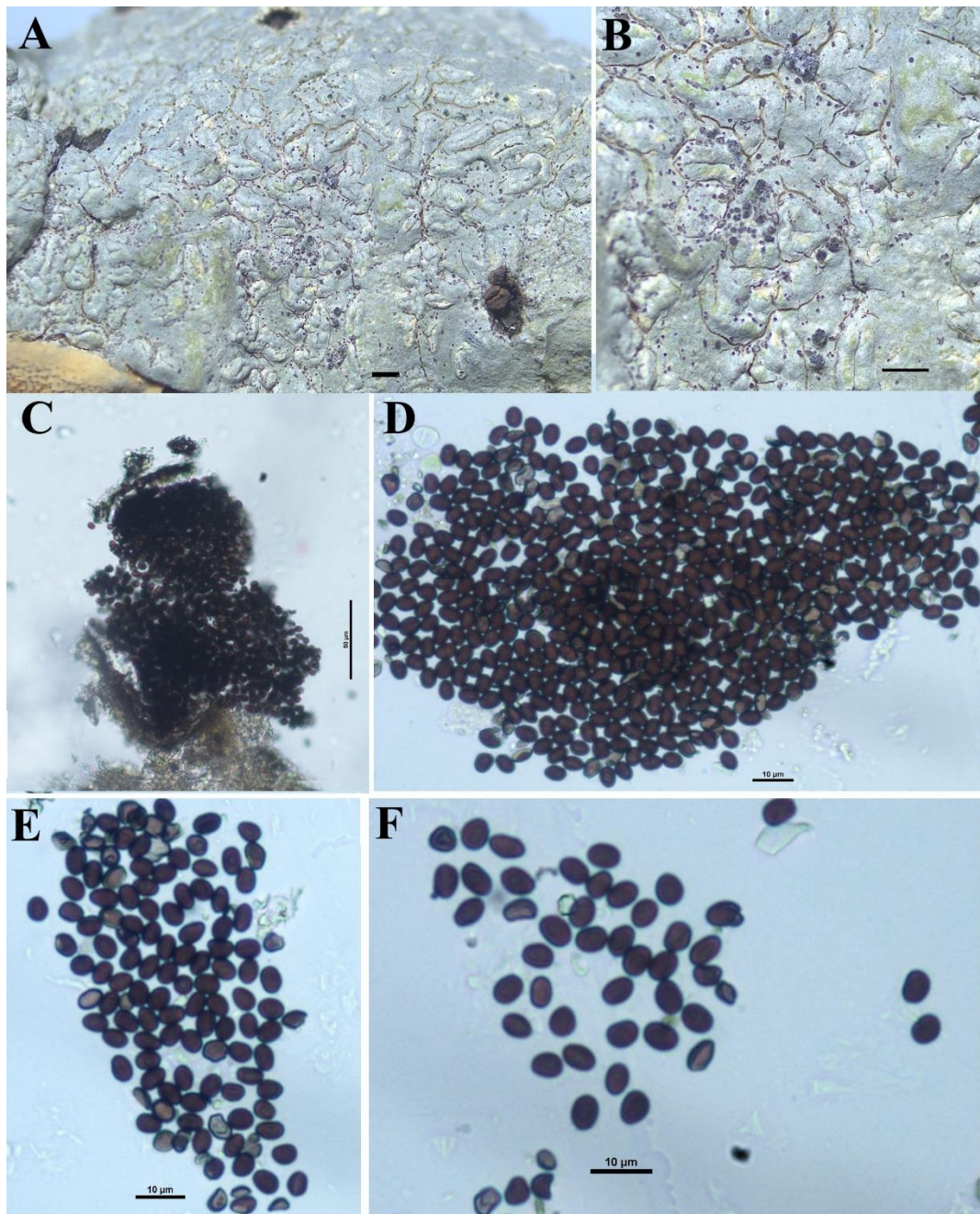


Fig. 1 – *Coniambigua diorygmatis*. A Sporodochia over the thallus of *Diorygma junghuhnii*. B Black punctiform conidiomata. C Mature conidioma. D–F Dark brown conidia. Scale bars: A, B = 1 mm, C–F = 10 μm .

Host and distribution – Known only from the type locality, colonised on the thallus of *Graphis stenotera*.

Remarks – *Distopyrenis*, established by Aptroot (1991), comprises nine species worldwide (Aptroot 2021, Index Fungorum 2025), two of which are lichenicolous, but none have been reported from India. *Distopyrenis* is distinguished by its erumpent perithecia or compound ascomata, carbonized exciple, inconspicuous apical ostioles, 8-spored bitunicate asci, and brown, 1-distoseptate ascospores. Species of *Distopyrenis* typically occur in tropical and subtropical regions, often on crustose lichens. Most are asymptomatic or weakly parasitic, occasionally inducing mild

discoloration or swelling of the host thallus. During the study of graphidoid lichens of India, a new species, *D. graphidis*, was discovered. This species closely resembles *D. japonica* Harada in several anatomical features, such as ascospore size, non-inspersed hymenium, but differs in having solitary to confluent ascomata with an apical ostiole and slightly smaller ascospores ($11\text{--}14 \times 4.5\text{--}5.5 \mu\text{m}$), compared to the ascospores of *D. japonica* ($12\text{--}15 \times 5\text{--}6 \mu\text{m}$) (Harada 2000). In general appearance, this species closely resembles *D. epidoriyoma* Etayo & van den Boom (van den Boom et al. 2022) but differs in having an inspersed hymenium and citriform ascospore. *Distopyrenis graphidis* appears to grow commensally on *Graphis stenotera*, indicating a lichenicolous habit. Within the genus *Distopyrenis*, this represents the third species of lichenicolous fungi to be documented. *Distopyrenis* is an important genus of lichenicolous fungi, yet it remains under-documented, especially in tropical regions, emphasizing the need for integrative taxonomic study.

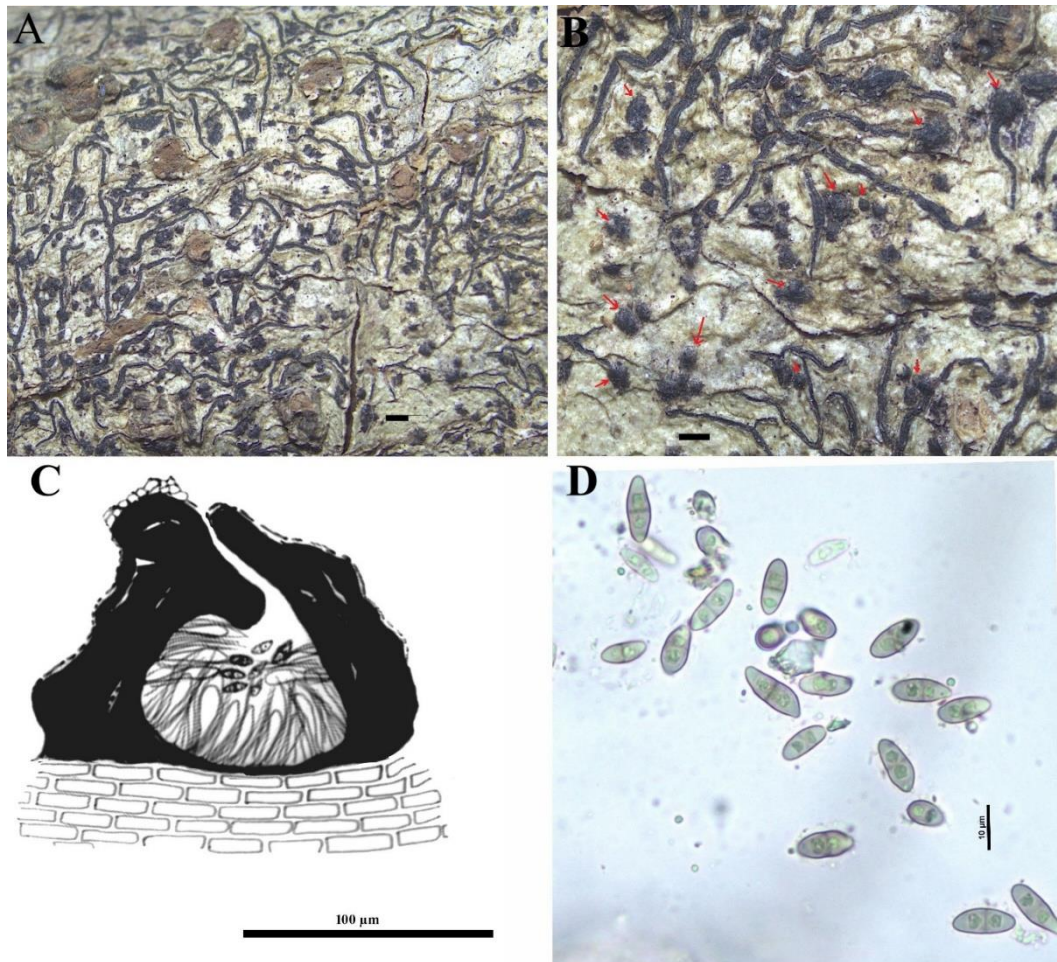


Fig. 2 – *Distopyrenis graphidis*. A Habit. B perithecia of *D. graphidis* associated with lirelliform ascomata of *Graphis stenotera* indicated by the red arrow mark. C Vertical section of perithecium. D Ascospores. Scale bars: A, B = 1 mm, C = 100 μm , D = 10 μm .

Table 2 All *Distopyrenis* species growing on Graphidaceae.

Species	Host	Ascomata	Hymenium	Ascospore size	Country reported
<i>D. graphidis</i>	<i>Graphis stenotera</i>	Ostiole apical	clear	$11\text{--}14 \times 4.5\text{--}5.5 \mu\text{m}$	India
<i>D. japonica</i>	<i>Graphis</i> sp.	Ostiole lateral	clear	$12\text{--}15 \times 5\text{--}6 \mu\text{m}$	Japan
<i>D. epidoriyoma</i>	<i>Diorygma</i> sp.	Ostiole apical	inspersed	citriform, $10\text{--}12 \times 6\text{--}7.5 \mu\text{m}$	S. America

Discussion

The two newly described taxa, *Coniambigua diorygmatis* and *Distopyrenis graphidis*, can be readily distinguished from previously known species in their respective genera by a distinct combination of morphological characters and host associations. *Coniambigua diorygmatis* is characterized by black sporodochia with ellipsoidal, dark brown, aseptate conidia, on *Diorygma*, while *D. graphidis* differs in its perithecioid ascomata with an apical ostiole, 1-distoseptate ascospore, and occurrence on *Graphis* species, representing the third lichenicolous member of the *Distopyrenis* reported from India. Both taxa are associated with graphidoid hosts of the Graphidaceae.

Lichenicolous fungi exhibit a broad spectrum of relationships with their hosts, ranging from parasitism, which induces visible damage or deformation, to near-commensalism, where little impact is observed (Hawksworth 2003, Diederich & Lawrey 2007). Many of these fungi display high degrees of host specificity, sometimes restricted to a single lichen genus or species. Nevertheless, their global diversity remains vastly underestimated, particularly in tropical and subtropical regions where lichenicolous fungi are presumed to be diverse but remain poorly documented.

The discovery of *Coniambigua diorygmatis* and *Distopyrenis graphidis* adds to the known diversity of lichenicolous fungi associated with graphidoid lichens. The description of *C. diorygmatis* represents an additional species within the small genus *Coniambigua*, which currently comprises only a few described taxa, and extends the documented morphological and host-associated variation within the genus. Likewise, the recognition of *D. graphidis* as the third lichenicolous species of *Distopyrenis* reported from India contributes to the growing knowledge of the genus and its occurrence on graphidoid lichen hosts.

Observations on host association in both species are based on the examined material and are presented here without implying strict host specificity or ecological preference. As these taxa are presently known from limited collections, broader conclusions regarding host range or distribution patterns cannot yet be drawn. Further studies based on additional collections from multiple localities, together with detailed morphological comparisons and molecular phylogenetic analyses, will be necessary to better understand the diversity, host associations, and distribution patterns of these lichenicolous fungi. The present study formally documents two previously undescribed taxa, thereby strengthening the taxonomic framework and providing a basis for future research on lichenicolous fungal diversity.

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