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PAXILLUS SWATICUS (BOLETALES; PAXILLACEAE): A NEW ALDER ASSOCIATED SPECIES WITHIN *P. RUBICUNDULUS* COMPLEX AND THE FIRST CONFIRMED REPORT OF THE GENUS FROM PAKISTAN

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Abstract

Based on morphological characteristics and molecular evidences from the ITS and LSU regions of the nrDNA, *Paxillus swaticus* is described here as a new species. This discovery also represents the first confirmed report of the genus *Paxillus* in Pakistan. The new species belongs to the well-known *P. rubicundulus* complex within *P.* subgenus. *Alnopaxillus,* which includes alder associated taxa. Colored photographs of fresh basidiomata along-with line drawings from the microscopic observations are provided and comparison with the associated species are discussed. This report will hopefully bridge the existing knowledge gaps and understanding of the diversity and phylogenetic relationship within *Paxillus.*

Key words: District Swat; First record; Molecular data; Morphology; New species

Introduction

Paxillus Fr. is a mushroom forming genus of basidiomycetes within the family Paxillaceae Lotsy. of the order Boletales E.-J. Gilbert. Species within this genus possesses decurrent and split lamellae, depressed or funnel-shaped pilei, brown spore prints, and a hymenophore which is separable from the pileus (Dessì & Contu, 1999). They grow in a variety of ecosystems primarily in temperate regions in the Northern hemisphere, dwelling wetlands and even polluted or degraded sites (Cagigal & Sánchez, 2017; Liu *et al.,* 2022). The genus is ectomycorrhizal, forming associations with variety of tress including species of *Alnus* Mill., *Betula* L., conifers and other higher plants (Brun *et al.,* 1995; Jargeat *et al.,* 2016). Although, approximately 69 legitimate names are registered in indexfungorum.org, the exact number of species within the genus remain unclear, partly because of the overlapping morphology of the species and the cryptic taxa, and partly because of the limited sampling (Jargeat *et al.,* 2014; Gelardi *et al.,* 2014).

Based on the molecular data from the ITS and LSU regions of the nuclear DNA, Jarosch & Bresinsky (1999) revealed the existence of two different clades within the genus. Further multi-locus analyses (Hedh *et al.,* 2008), confirmed the existence of the two clades which were later on raised to the rank of sub-genera, viz. *Paxillus* subg. *Alnopaxillus* Vizzini and Gelardi and *Paxillus* subg. *Paxillus* (Gelardi *et al.,* 2014).

The subg. *Alnopaxillus* contains alnicolous taxa which encompasses the well-known *P. rubicundulus* complex and *P. orientalis* Gelardi, Vizzini, E. Horak & G. Wu. They are mainly distributed in Asia and Europe and are characterized by squamulose pileal surface, yellowish to deep yellow context, color change (red or purple) of the hymenophore on bruising, and smaller basidiospores with an average length of < 8 µm (Hahn, 2000). Despite the fact that, Asia is the center of origin of alder, it remains poorly sampled and several phylogenetically recognized taxa still lacking formal descriptions (Jargeat *et al.,* 2016; Põlme *et al.,* 2016). Further investigation and species descriptions from this region is required to fully understand the co-evolution of *Paxillus* and their hosts.

Paxillus rubicundulus complex of the *P.* subg. *Alnopaxillus* is a well-known species complex that includes several phylogenetically distinct but morphologically similar taxa (Jargeat *et al.,* 2016). Morphological traits are overlapping and it is very difficult to draw a line based on morphology. Among the different morphological characteristics, basidiospore size and shape are considered as the most reliable traits for species delimitation within the complex. Spore print color can also be used as marker especially to separate *P. adelphus* J.-P. Chaumeton, Gryta, Jargeat & P.-A. Moreau and *P. olivellus* P.-A. Moreau, J.-P. Chaumeton, Gryta & Jargeat. Species within *P.* subg. *Alnopaxillus* are generally considered to be associated with *Alnus*. However, this association may be ambiguous for some species, such as *P. adelphus* and *P. olivellus*, which predominantly fruits in habitats where *Salix* and *Populus* sp. also occur (Jargeat *et al.,* 2016).

By contrast, species within subg. *Paxillus* circumscribe the *P. involutus* complex, species of which are characterized by a smooth pileus surface, whitish to light yellow context and basidiospores which on average are longer than 8 µm (Hahn, 2000). They are mainly distributed in Europe and North America and usually associated with a wide range of host trees showing no specificity (Hedh *et al.,* 2008; Vellinga *et al.,* 2012; Gelardi *et al.,* 2014).

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Alnus nitida (Spach) Endl, also known as the west Himalayan alder is the only alder species found in Pakistan. In Pakistan it is distributed in temperate regions of Swat and Dir, along the banks of streams and rivers at an altitude range of 1000–2900 m (Khan *et al.,* 2022). *Alnus* species are obligatory root symbionts with certain fungi and actinobacteria, which not only help them in obtaining mineral nutrients from soil but also fix atmospheric nitrogen (Yamanaka *et al.,* 2003). The ectomycorrhizal association of *A. nitida* in Pakistan were studied by Hanif & Khalid (2023), reporting 11 species of ectomycorrhizal fungi.

In a continuous study, dealing with the exploration of macrofungi in the northern areas of Khyber Pakhtunkhwa, Pakistan, several collections of an unknown *Paxillus* species were encountered. Based on their association with *Alnus* and morphology, the collections were primarily assigned to the *P.* subg. *Alnopaxillus*. Further detailed morphological investigations and molecular data from the ITS and LSU regions of the nuclear DNA revealed that these collections were yet to be described. In this paper we describe our collections of *Paxillus* representing a new species within subg. *Alnopaxillus,* which is also the first confirmed report of this genus in Pakistan.

Material and Method

Specimens collection and morphological description: From 2021-24, specimens of the present study were collected during spring (March-April) and autumn (September to November), in Ingaro Dherai and Takhtaband villages of Tehsil Babozai, District Swat, Khyber Pakhtunkhwa Province, Pakistan. Altitude of the collection sites ranges between 950–1000 m a.s.l. with deciduous trees including *Populus nigra* L.*, Alnus nitida* (Spach) Endl.*, Robinia pseudoacacia* L.*,* and *Salix tetrasperma* Roxb. The fruiting bodies were carefully excavated from the soil with the help of a sharp knife and photographed in their natural habitats. Each specimen was labeled with a unique tag number for proper documentation and future reference. Key morphological characteristics were noted in a field note book referring to McKnight *et al*., (2021). Colors were coded following to the Munsell soil color chart (Munsell, 1975). The properly collected specimens were sun dried in open air, packed in separate zipper bags, and kept in freezer for two weeks to kill any remains of insects and their larvae. The dried specimen were deposited as vouchers in the herbarium of University of Swat (SWAT). The new name has been registered at the Mycobank.org and a unique number has been generated.

Anatomical characterization: The anatomical features of the collected specimens were studied with a BM120 binocular microscope (BOECO, Germany) at the department of Botany, University of Swat**.** Thin sections of the basidiomata were prepared with the help of a sharp razor blade and mounted on a glass slides with a drop of 5% aqueous potassium hydroxide (KOH) solution. For contrast purpose, a drop of Congo red stain (1% aqueous) was applied following (Kiran *et al.,* 2025). From each collection, 30 basidiospores were measured to calculate the average dimensions. For basidiospores, values presented in parentheses denote extreme measurements. The parameter Q refers to the length/width ratio of individual spores, Qe is the average Q values, and Me represents the mean length and width of the spores (Kiran *et al.,* 2018). In addition, 20 measurements were taken for each of the other anatomical structures including the basidia, cystidia, and terminal elements of the pileipellis and stipitipellis.

DNA extraction and Polymerase chain reaction: For molecular phylogenetic analyses, DNA from the dried herbarium specimens was extracted by following the modified CTAB method of Gardes & Bruns (1993). Polymerase chain reactions (PCR) were performed in a 25 µl volumes with the concentrations and protocols mentioned in (Khan *et al.,* 2017). The PCR product were visualized in a UV illuminator using 1% Agarose gel added with Ethidium Bromide. The successful PCR products were sequenced through outsourcing at Beijing Genomic Institute, China and the sequence data was submitted to GenBank for future reference.

Sequence alignment and Phylogenetic analyses: Consensus sequences were generated by combining the forward and reverse reads in BioEdit software v. 7.2.5 (Hall, 1999). The generated consensus sequences were searched against GenBank sequences for homology using the Basic Local Alignment Search Tool (BLAST) at the National Center For Biotechnology Information (NCBI) website. Matching sequences, especially those published in Jargeat *et al.,* (2014; 2016) were downloaded for phylogenetic reconstruction. Separate datasets were used for the ITS and LSU sequences and the multiple sequences were aligned using the online MAFFT tool (https:// mafft.cbrc.jp/alignment/server/index.html). Maximum likelihood (ML) analyses were performed with RAxML-VI-HPC on XSEDE tool on the Cipres Science Gateway (Miller *et al.,* 2012) by employing RAxML-HPC v.8. rapid bootstrap analysis/search for best-scoring ML tree (Stamatakis, 2014). Branch supports values were calculated by using 1000 bootstrap replicates under the GTRGAMMA substitution model.

To estimate the evolutionary relationships among the studied taxa, Bayesian phylogenetic inference (BI) was performed with Bayesian Evolutionary Analysis Sampling Trees (BEAST v.1.2.8). The aligned ITS and LSU sequences were saved in NEXUS and XML formats for downstream analysis using Clustal X (Larkin *et al.,* 2007). Based on the AIC values and Bayesian Information Criterion (BIC), the GTR + Gamma was determined as the best-fit nucleotide substitution model. BEAST XML configuration files were prepared using the Bayesian Evolutionary Analysis Utility (BEAUti). A Yule speciation process was selected with uncorrelated relaxed lognormal clock, to accommodate rate heterogeneity among lineages. Markov Chain Monte Carlo (MCMC) analyses were run for 100 million generations. All priors were set at default settings unless otherwise justified. Log files were examined using Tracer to assess convergence, effective sample size (ESS), and parameter stability. Runs with ESS values >200 were considered reliable. A burn-in of 10–25% of the sampled trees was discarded to eliminate early, unconverged states. Posterior distributions of trees were summarized using TreeAnnotator to generate a maximum clade credibility (MCC) tree with mean node heights. The annotated trees were visualized and edited in FigTree v1.4. Posterior probability values were used to assess node support.

Results

Molecular Phylogenetic analyses figures 1 & 2: All the generated ITS consensus sequences of our *Paxillus* collections showed 98% similarity with *Paxillus rubicundulus* P.D. Orton (KU163493) in BLAST search. Sequences of *P. adelphus* (KU163464) and *P. filamentosus* (Scop.) Fr., (EU084664) were 95.44% and 95.30% identical respectively. Similarly, the consensus LSU sequences of our collections showed 90–92% resemblance with the sequences of *P. filamentosus* (AF167680), *Paxillus olivellus* (OR602341), and *P. rubicundulus* (OM238150).

For phylogenetic analyses, a total of 52 ITS sequences were used with *Tapinella atrotomentosa* (Batsch) Šutara (MN622802) and *T. panuoides* (Fr.) E.-J. Gilbert (MN954754) used for rooting purpose. The aligned ITS dataset comprised of 676 sites having 369 conserved, 303 variable and 233 parsimony informative sites. In the LSU based phylogenetic analysis, 27 sequence were included with *Tapinella Panuoides* (DQ071746) as outgroup taxon. In the final aligned dataset, there were a total of 781 sites in which 670 were conserved while 111 and 54 were variable and parsimony informative respectively. In the Bayesian and Maximum Likelihood analyses of both the ITS and LSU datasets, the sequences of *Paxillus* resolved in two well supported clades representing the two subgenera including *P.* subg. *Alnopaxillus* and *P.* subg. *Paxillus* (Gelardi *et al.,* 2014). The newly generated sequences of this study clustered sister to *P. rubicudulus* with a strong bayesian posterior probability and bootstrap support in both the ITS (1/99) and LSU (1/100) based analyses (Figs. 1 and 2).

Description

Paxillus swaticus: Sinan and J. Khan sp. nov. (Figs. 3 & 4)
Diagnosis: Differ from the related taxa by the presence of smaller basidiospores, lageniform, fusiform, or mucronate type of pleuro- and cheilocystidia, and growth in both early spring and autumn.
Etymology: The specific epithet "*swaticus*" is taken from the district of the occurrence of the new species.
MycoBank No. MB861011.
Holotype: Pakistan Khyber Pakhtunkhwa province, District Swat, Takhtaband**,** SK1 (SWAT001379, ITS=PX460695; LSU=PX460703), Sinan Khan and Junaid Khan, 10th Aug, 2024, on soil under *Alnus nitida* (Spach) Endl., scattered to gregarious, 900 m. a.s.l.

Taxonomic description

Basidiomata medium-sized. ***Pileus*** 40–85 mm in diam., initially convex to plane, becoming depressed to infundibuliform with age, often lobed, margin decurved at first, later involute to uplifted, occasionally irregular or lobed, surface dry, tomentose to finely scaly in young stages, smooth or depressed scaly in older basidiomata, when present the scales radiates from the center towards margins, color light olivaceous (2.5Y 5/6) in young stages, brown (7.5YR 4/4) to dark brown (5YR 3/2) later on, pileal context cream colored, darkening / reddening (2.5YR 3/2) on cutting, thickest at the center (3–5 mm), gradually thinning outward and ≤1 mm at the extreme margins. ***Hymenophore*** lamellate, more or less decurrent, lamellae close, forked, with frequent anastomoses at the stipe, yellowish-brown (7.5Y 9/4) when young, brownish (10YR 5/6) to dark yellowish brown (10YR 4/4) with age, turning dark brown on bruising or handling and aging, lamellulae present, often in 2-tiers. ***Stipe*** 20–40 × 5–12 mm, centric to off-centric, cylindrical to slightly tapering downward, often curved; surface concolorous the pileus or slightly pale, dry, fibrillose, base sometimes with adhering mycelial mat, stipe context firm, pale to light yellow colored especially in the upper parts, yellowish brown in the basal with reddish brown extreme base, unchanging on exposure or cutting. ***Spore print*** rusty brown.

Basidiospores **(**5.7) 6.4–6.6 (7.2) × (4) 4.4–4.5 (4.9) µm, Q = (1.3) 1.4–1.5 (1.6), Me = 6.5 × 4.4 µm, Qe = 1.5, short cylindrical with a slight allantoid tendency, to ovoid thick walled (0.2–0.3 µm) with a single large gutuule, apiculus prominent. ***Basidia*** 25–40 × 6–8.5 µm, bisporic or tetrasporic, clavate to more or less cylindrical, thin walled, with abundant oil droplets. ***Cheilocystidia*** 35–52 × 9–12 µm, lageniform, fusiform, or mucronate, thick walled, some with visible yellow brown oily contents. ***Pluerocystidia*** 40–90 × 7–12 µm, lageniform, fusiform, or mucronate with capitate heads. ***Pileipellis*** a weak ixocutis, made of cylindrical hyphae measuring 4–7 µm in diam., amber yellow in KOH, some hyphae faintly encrusted. **Stipitipellis** compose of long cylindrical hyphae measuring 5–10 µm in diam, Caulocystidia absent ***Clamp connections*** present (Table 1).

Ecology and distribution: on soil in scattered to dense clusters along the banks of rivers and stream under *Alnus nitida* from spring to autumn. At present, only reported from different locations in district Swat, Khyber Pakhtunkhwa, Pakistan.

Additional materials examined: Pakistan Khyber Pakhtunkhwa province, District Swat, Takhtaband, SK2 (SWAT001380, ITS=PX460694; LSU=PX460704), Sinan Khan, 15th October 2024, on soil under *Alnus nitida*, 950 m. a.s.l. Same locality, (TB013, ITS=PX460696; LSU=PX460705), Junaid Khan, 10th April 2021, on soil under *Alnus nitida* 975 m a.s.l.

Table 1. Comparative table showing morphological characteristics of *Paxillus swaticus* and associated species.

Character / Species	*P. adelphus*	*P. olivellus*	*P. rubicundulus*	*P. swaticus*
Basidiospores size (µm)	(5.5)6.1–7.3(8.3) × (3.7)4.0–4.7(5.2)	(5.7)6.4–7.9(10.2) (3.8)4.3–5.1(5.8)	(6.4)6.6–8.5(9.1) × (4.0)4.4–5.1(5.6)	**(**5.7)6.4–6.6(7.2) × (4)4.4–4.5(4.9)
Q values	(1.28)1.42–1.54–1.64 (1.85)	(1.30) 1.42–1.53–1.64 (1.96)	(1.32) 1.42–1.57–1.73 (1.95)	(1.3) 1.4–1.5–1.54 (1.6)
Spore shape	Short cylindrical to slightly allantoid	Short cylindrical to slightly allantoid	Cylindrical without allantoid tendency	Short cylindrical with a slightly allantoid
Basidia	25–37 × 6.5–8	22–32 × 5.5–8	18–38 × 5.5–8	25–40 × 6–8.5
Cheilocystidia	35–85 × 9–15, cylindrical to fusiform	40–55 × 5.5–11, narrowly ellipsoidal to slightly fusiform	35 × 7 (base) ×3.5 (apex), cylindrical to fusiform	35–52 × 9–12, lageniform, fusiform, or mucronate
Pleurocystidia	35–60 (90) × 8–12, cylindrical to fusiform or lageni-fusiform	62–68 ×11–16, lageni-fusiform	70 × 7 (base) × 4 (apex), cylindrical to fusiform	40–90 × 7–12, lageniform, fusiform, or mucronate

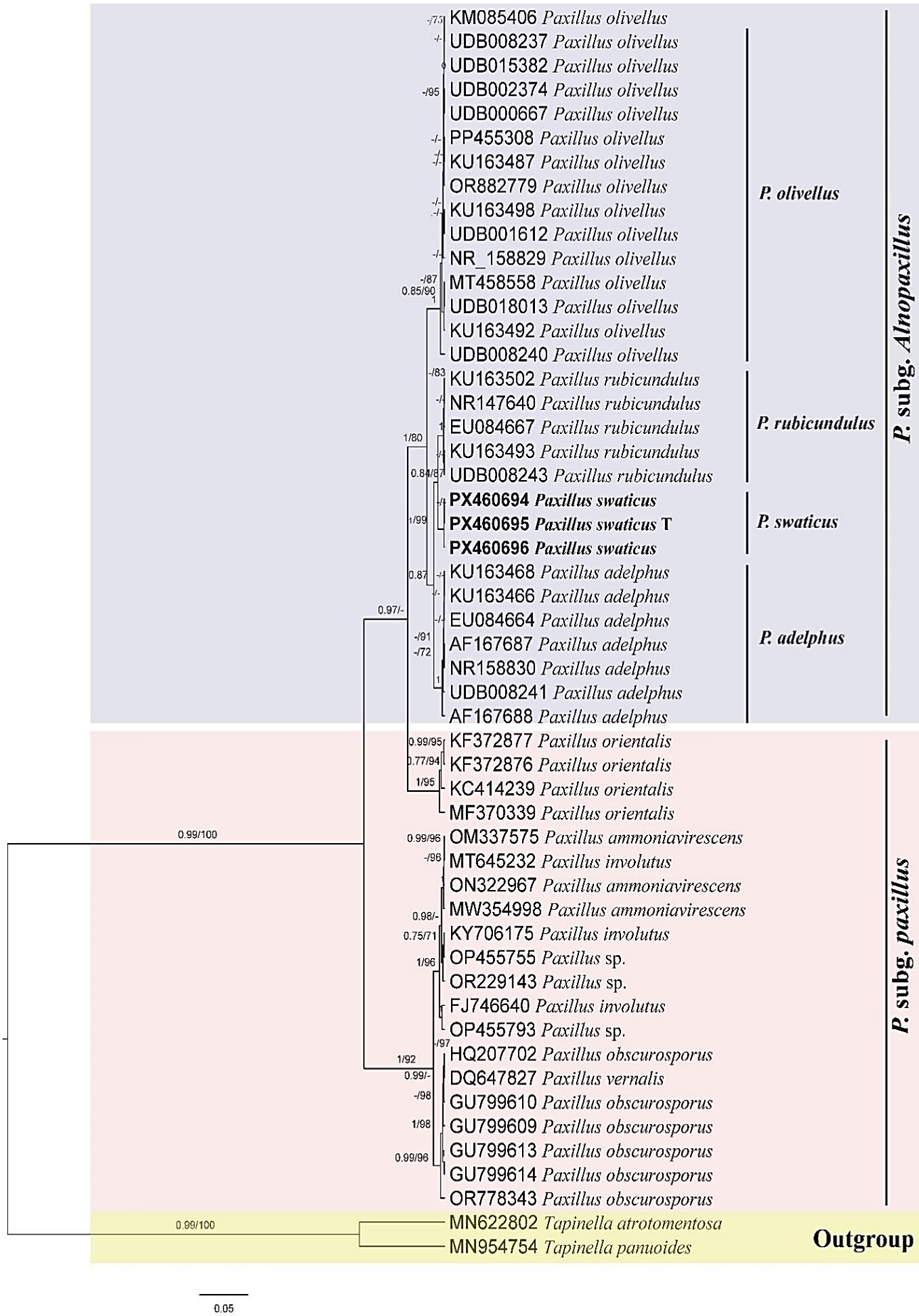


Fig. 1. Bayesian tree of *Paxillus swaticus* and associated taxa inferred from ITS dataset. The newly generated sequences of the present study are highlighted in bold. Bootstrap support values (≥70%) and Bayesian posterior probabilities (≥70%) are given above the nodes.

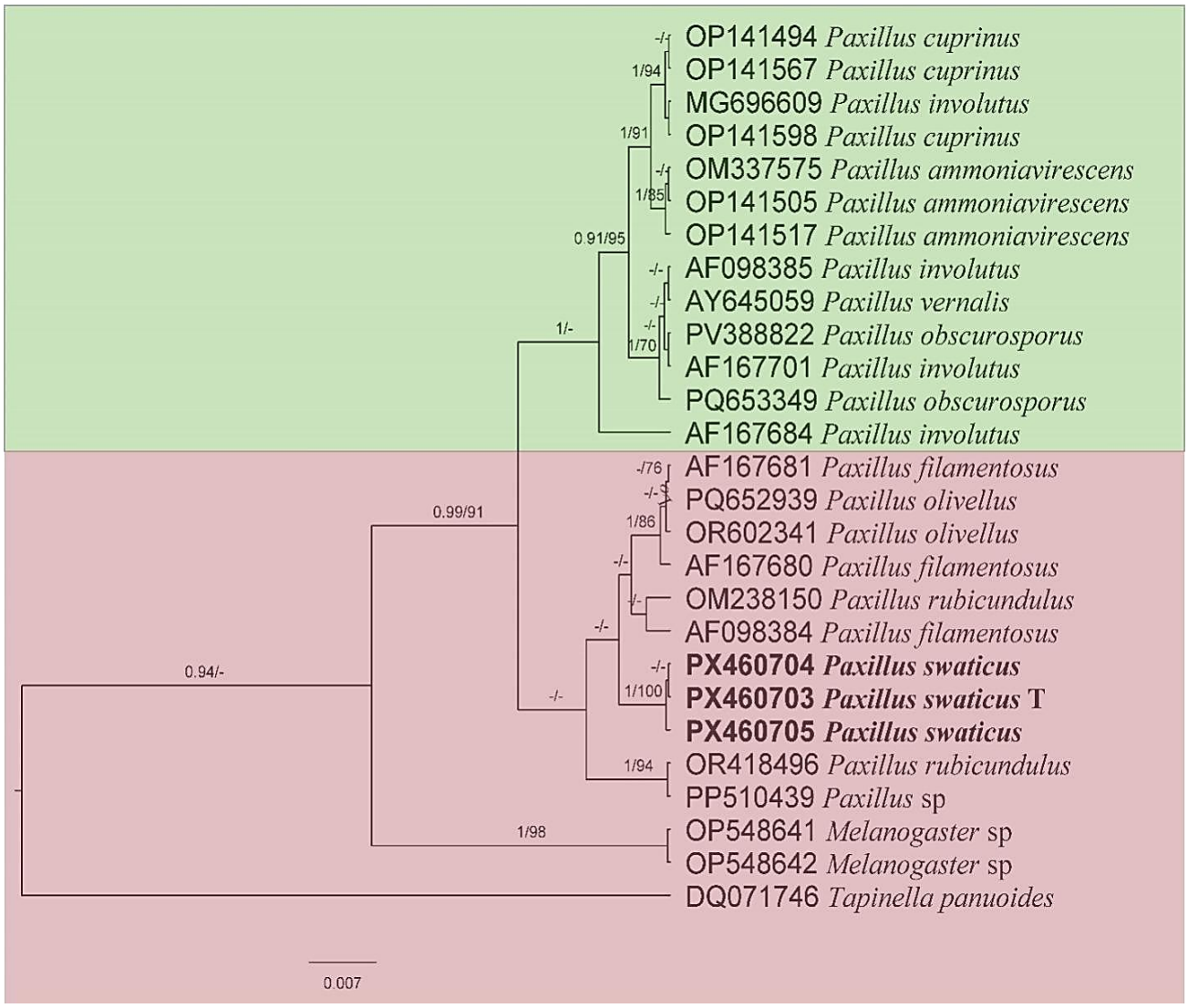


Fig. 2. Bayesian tree of *P. swaticus* and associated taxa inferred from LSU dataset. The newly generated sequences of the present study are highlighted in bold. Bootstrap support values (≥70%) and Bayesian posterior probabilities (≥70%) are given above the nodes. The *P.* subg. *Paxillus* is highlighted in light blue while *P.* subg. *Alnopaxillus* in light pink color.



Fig. 3. Colored photographs of different collections of *Paxillus swaticus*. **A & C**) SWAT001379 (holotype). **B**): SWAT00130. **D**) TB013. **E**) A close up of anastomosing lamellae. Scale bars= 10 mm for **A–E**.

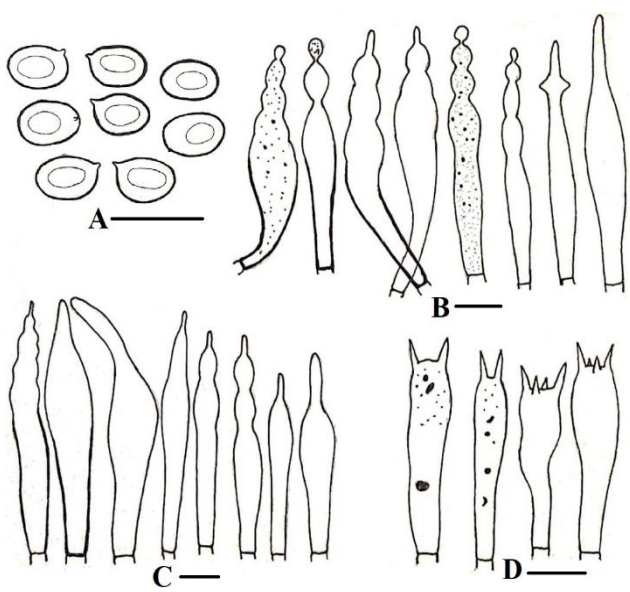


Fig. 4. Anatomical features of *Paxillus swaticus*. **A**) basidiospores. **B**) Pleurocystidia. **C**) Cheilocystidia. **D**) Basidia. Scale Bars=10 µm for A–D.

Discussion

Due to the overlapping morphological characters and presence of cryptic species, delimitation of different *Paxillus* species is a challenging task (Hahn & Agerer, 1999). The use of molecular data along-with the macro-morphological characteristics is therefore needed for correct species identification. Using the conjoint molecular and morphological approaches, well-known species (*P. rubicundulus* and *P. involutus*) complexes within *Paxillus* have been resolved to a larger extent with the description of new species (Jargeat *et al.,* 2016).

During the present investigation, *P. swaticus* is described as a new species which falls within the alder associated *P.* subg. *Alnopaxillus.* Along-with the collections mentioned in this study, many other collections were also encountered, majority of which were occurring in mixed stands of deciduous trees including *Alnus, Populus,* and *Salix* sp., however, some were collected from pure stands of *Alnus nitida* suggesting their association with alders.

Paxillus swaticus is well distinguished from other species of the complex by smaller basidiospores dimensions with slight allantoid tendency. It also has distinctive lageniform to fusiform or mucronate pleurocystidia and cheilocystidia and grows in early spring and late autumn. *Paxillus adelphus* is the closest species which also contains short cylindrical basidiospores with allantoid tendency (Jargeat *et al.,* 2016). However, it has cylindrical to fusiform cheilocystidia and fruit during later summer and autumn. *Paxillus olivellus* has a distinct olivaceous color, larger basidiospores and narrowly ellipsoidal to slightly fusiform cheilocystidia (Jargeat *et al.,* 2016). Similarly, *P. rubicundulus* has much larger basidiospores which are cylindrical in shape and without allantoid tendency (Orton, 1969; Jargeat *et al.,* 2016).

The current report on this new species is significant not only because it marks the first documentation of the genus *Paxillus* in Pakistan, but also contribute to bridging a gap in comprehending the evolution of species within the *P. rubicundulus* complex. Further investigations are recommended to fully explore the diversity of this genus in Pakistan.

References

Brun, A., M. Chalot, R. D. Finlay and B. Söderström. 1995. Structure and function of the ectomycorrhizal association between *Paxillus involutus* (Batsch) Fr. and *Betula pendula* Roth. I. Dynamics of mycorrhiza formation. *New Phytol.,* 129(3): 487-493.

Cagigal, E.F.M. and A.C. Sánchez. 2017. Influence of the culture media and the organic matter in the growth of *Paxillus ammoniavirescens* (Contu & Dessi). *Mycobiology*, 45(3): 172-177.

Dessi, P. and M. Contu. 1999. *Paxillus ammoniavirescens* spec. nov. con note sul genere Paxillus in Sardegna. *Micol. Veg. Mediterr.*, 13: 121-130.

Gardes, M. and T.D. Bruns. 1993. ITS primers with enhanced specificity for basidiomycetes-application to the identification of mycorrhizae and rusts. *Mol. Ecol.*, 2(2): 113-118.

Gelardi, M., A. Vizzini, E. Horak, E. Ercole, S. Voyron and G. Wu. 2014. *Paxillus orientalis* sp. nov. (Paxillaceae, Boletales) from south-western China based on morphological and molecular data and proposal of the new subgenus Alnopaxillus. *Mycol. Prog.*, 13(2): 333-342.

Hahn, C. 2000. Sind *Paxillus involutus* und *P. rubicundulus* anhand ihrer Sporenmabe eindeutig unterscheidbar? Zeitschrift fur. *Mykologie,* 66: 161-171.

Hahn, C. and R. Agerer. 1999. Studium zum *Paxillus involutus* Formenkreis. *Nova Hedwigia.,* 69: 241-310.

Hall, T.A. 1999. BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *In Nucleic acids symposium series*., 41: 95-98.

Hanif, M. and A.N. Khalid. 2023. Ectomycorrhizae of Himalayan alder (*Alnus nitida*) from the moist temperate forests of Pakistan. *Pak. J. Bot.,* 55(4): 1569-1577.

Hedh, J., P. Samson, S. Erland and A. Tunlid. 2008. Multiple gene genealogies and species recognition in the ectomycorrhizal fungus *Paxillus involutus*. *Mycol. Res.*, 112(8): 965-975.

Jargeat, P., J.P. Chaumeton, O. Navaud, A. Vizzini and H. Gryta. 2014. The *Paxillus involutus* (Boletales, Paxillaceae) complex in Europe: genetic diversity and morphological description of the new species *Paxillus cuprinus*, typification of *P. involutus* s.s., and synthesis of species boundaries. *Fung. Biol.*, 118(1): 12-31. https://doi.org/10.1016/j.funbio.2013.10.008

Jargeat, P., P.A. Moreau, H. Gryta, J.P. Chaumeton and M. Gardes. 2016. *Paxillus rubicundulus* (Boletales, Paxillaceae) and two new alder-specific ectomycorrhizal species, *Paxillus olivellus* and *Paxillus adelphus*, from Europe and North Africa. *Fung. Biol.*, 120(5): 711-728.

Jarosch, M. and A. Bresinsky. 1999. Speciation and phylogenetic distances within *Paxillus* s. str. (Basidiomycetes, Boletales). *Plant Biol*., 1(06): 701-705.

Khan, J., A. Majid, M. Nisar, A. Hazrat, N. Nazir, M. Zahoor and M. Yahya. 2022. Genetic diversity of *Alnus nitida* reported from Dir Lower, Khyber Pakhtunkhwa, Pakistan. *Sarhad J. Agri.,* 38(2): 489-496.

Khan, J., M. Kiran, S. Jabeen, H. Sher and A.N. Khalid. 2017. *Gymnopilus penetrans* and *G. swaticus* sp. nov. (Agaricomycota: Hymenogastraceae); a new record and a new species from northwest Pakistan. *Phytotaxa*, 312(1): 60-70.

Kiran, M., J. Khan, H. Sher, D.H. Pfister and A.N. Khalid. 2018. *Amanita griseofusca*: A new species of Amanita in section Vaginatae from Malam Jabba, Swat, Pakistan. *Phytotaxa*, 364(2): 181.

Kiran, M., Sana, A. Sattar and A.N. Khalid. 2025. A new species and a new record of *Mallocybe* (Inocybaceae, Agaricales) from Pakistan uncovered by integrated morphology and phylogeny. *Phytotaxa*, 720(1): 1-18.

Larkin, M.A., G. Blackshields, N.P. Brown, R. Chenna, P.A. McGettigan, H. McWilliam, F. Valentin, I.M. Wallace, A. Wilm, R. Lopez, J.D. Thompson, T.J. Gibson and D.G. Higgins. 2007. Clustal W and Clustal X version 2.0. *Bioinformatics.,* 23: 2947-2948.

Liu, L.R., G.S. Wang, L.K. Jia, J.Q. Kang, Z.L. Yang and Y.C. Li. 2022. Type studies on two *Paxillus* species (Paxillaceae, Boletales) described from China. *Phytotaxa*, 530(2): 177-188.

McKnight, K.B., J.R. Rohrer, K.M. Ward and K.H. McKnight. 2021. Peterson field guide to mushrooms of North America. Mariner Books.

Miller, M.A., W. Pfeiffer and T. Schwartz. 2012. The CIPRES science gateway: enabling high-impact science for phylogenetics researchers with limited resources. In: Proceedings of the 1st Conference of the Extreme Science and Engineering Discovery Environment: Bridging from the extreme to the campus and beyond. *Proceedings,* pp. 1-8.

Munsell, A.H. 1975. Munsell. Soil color charts. Baltimore.

Orton, P.D. 1969. Notes on British agarics. III. *Notes from the Royal Botanic Garden Edinburgh*, 29(1): 75-127.

Põlme, S., M. Bahram, T. Yamanaka, K. Nara, Y.C. Dai, T. Grebenc and L. Tedersoo. 2013. Biogeography of ectomycorrhizal fungi associated with alders (*Alnus* spp.) in relation to biotic and abiotic variables at the global scale. *New Phytol.,* 198(4): 1239-1249.

Stamatakis, A. 2014. RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, 30(9): 1312-1313.

Vellinga, E.C., E.P. Blanchard, S. Kelly and M. Contu. 2012. *Paxillus albidulus*, *P. ammoniavirescens*, and *P. validus* revisited. *Mycotaxon,* 119: 351-359.

Yamanaka, T., C.Y. Li, B.T. Bormann and H. Okabe. 2003. Tripartite associations in an alder: effects of Frankia and *Alpova diplophloeus* on the growth, nitrogen fixation and mineral acquisition of Alnus tenuifolia. *Plant and Soil,* 254(1): 179-186.