Evidence of proteoid roots in Chinar (*Platanus orientalis* L.): taxonomic implications

Oriental plane or Chinar (*Platanus orientalis*) is grown as an avenue tree in the Kashmir valley, India because of its canopy and autumn tint. It is a monoecious tree with encircling stipules, infrapetiolar buds, and usually palmately lobed and veined leaves. It carries pendant spike of heads bearing unisexual flowers and fruits of bristly achenes[1]. Though the tree tolerates a degree of drought, it prefers moist soils; it cannot grow in shade[2]. *Platanus* is the only extant genus belonging to the family Platanaceae that encompass eight known species[3]. While the family has traditionally been placed in the order Hamamelidales[4,5], recent phylogenetic studies suggest that it belongs to Proteales[6].

In the Kashmir valley, Chinar flowers during early spring, in March. Male inflorescences appear slightly earlier than the female ones. The globular assemblage of one-seeded achenes hangs on the tree until seed dispersal. Seeds mature in January of the following year, but seed dispersal starts in March. Though there are about 75,000 Chinar trees growing in the valley, natural regeneration is almost non-existent. Therefore, we examined whether the seeds were viable or not. If the seeds were viable, we explored further, whether they were dormant for the period between seed maturity (January) and seed dispersal (March).

While studying seed germination, we observed an interesting phenomenon: proteoid roots in the germinating seedlings (Figure 1). In light of this finding, we explore the relationship of Platanaceae with its sister family Proteaece.

For the study, we selected three sexually mature (diameter at breast height; dbh range: 179.8–320.1 cm), middle-aged trees growing in the Faculty of Forestry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir (SKUAST-K), Ganderbal, Kashmir. Seeds from these trees were collected twice, in January and March. The germination test was conducted immediately after seed collection. A replicated germination trial was laid out in a walk-in plant growth chamber (Blue Star; model no: BS-153) in January, immediately after the first seed collection. Nine hundred seeds from each of the three trees were selected randomly. A total of 2700 seeds were placed in petri dishes (27 in number), each containing 100 seeds on moist germination paper. We placed these in the growth chamber at 25° ± 2°C. The same procedure was followed for seeds collected in March.

We recorded germination count daily for one month. Seeds germination started from the fifth day onwards. Seedlings continued to emerge till the 18th day (Table 1). On an average, about 60% seeds germinated (Table 1). It shows that the seeds were viable and had good germination potential. Germination was slightly higher in the January seed-lot than in the March seed-lot. However, the difference was statistically non-significant ($P = 0.581$). This implies that the seeds were ready for germination as soon as they matured, thus indicating the absence of seed dormancy.

During the emergence of radicals, we noticed white filamentous growth covering it (Figure 1). Initially, it appeared as some fungal growth, but soon we identified these as proteoid roots. These roots were ephemeral and persisted on the radicals only for 8–10 days (Table 1).

Proteoid roots, also known as cluster roots, form clusters of closely spaced short lateral rootlets. Engler first noticed such roots in 1894 in the plants of the family Proteaceae. Purnell coined the term 'proteoid root'. She examined 44 species from ten Proteaceae genera (order: Proteales) and found proteoid roots in every genus, except *Persoonia*[7]. So far, proteoid roots are reported in about 1800 species belonging to ten families[8] (figure 4 of Lambers *et al.*[9]).

Proteoid roots have not been reported in any other species of Plantaceae so far. Even while growing over a year in low phosphorus nutrient solution, *Platanus hybrida* never produced proteoid roots[9]. They have not been reported in any other family of Proteales, except Proteaceae[8,9]. Proteoid roots are ephemeral and physiologically active for a little more than a few weeks[10,11]. The density of rootlets produced per unit root axis is far greater in the Proteaceae than in the other families[9]. Proteoid roots provide Proteaceae a competitive advantage over non-proteoid rooted species[12]. These plants can grow in soils deficient in nutrients, especially phosphorus-deficient soils[7].

As these roots are adaptive features of the plants that grow in nutrient-deficient soils, this finding may help in the study of adaptive behaviour in *P. orientalis*. Proteoid roots increase the surface area by over 140 times, and soil volume explored 300 times compared to an equivalent non-proteoid root. This increases the exudation of carboxylates, phenolics, solubilization of minerals and organic nutrients, thereby enhancing the uptake

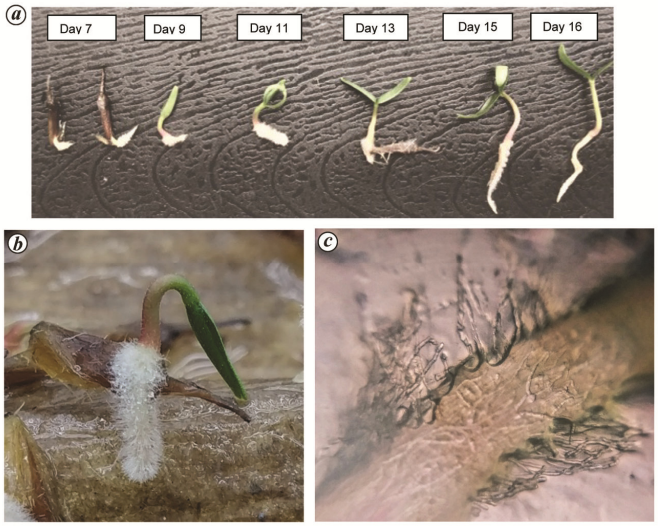

Figure 1. Proteoid roots in *Platanus orientalis*: (***a***) growth stages, (***b***) growth on 11th day after keeping the seed for germination and (***c***) longitudinal section (image magnification 400×).

Table 1. Seed germination and seedling emergence in *Platanus orientalis* in Kashmir, India

	Germination per cent		Germination progress		Duration of proteoid roots	
Seed source	January	March	Initiation	Termination	Appearance	Disappearance
Tree 1 (dbh: 179.8 cm)	59.67	68.33	7th day	18th day	7th day	15th day
Tree 2 (dbh: 202.4 cm)	63.33	44.33	6th day	17th day	6th day	15th day
Tree 3 (dbh: 320.1 cm)	61.67	61.33	5th day	18th day	6th day	16th day
Mean germination	61.56	58.00				
Overall mean	59.78					

of inorganic nutrients, amino acids and water per unit root mass[13,14]. These roots exude organic acids that solubilize phosphate associated with iron and aluminium in the rhizosphere[15]. The roots can help absorb inorganic phosphate (Pi) faster than non-proteoid roots[16,17]. In most species, clusters of proteoid roots decline as phosphorus availability to the roots increases, and when its supply is restricted, the clusters become prominent[18].

Presence of proteoid roots in *Platanus* has taxonomic implications as well. Before 1998 (pre-APG I phase), the Platanaceae was placed in the order Hamamelidales[4,5,19]. In 1998, it was transferred to Proteales[20]. Though all the subsequent APG classifications consistently placed Platanaceae in Proteales, the Thorne classification continued with the earlier scheme[21]. Finally in 2007, a revision of the Thorne classification accepted the change[22]. By 2009 (Angiosperm Phylogeny Group (APG) III), the position of Platanaceae got firmly established in Proteales[6]. Both these reports based their conclusions on molecular evidence. The molecular evidence based on gene sequences established the sister relationship of Platanaceae and Proteaceae[23,24]. The paucity of morphological evidence could be due to the ancient nature of the families. *Platanus* is considered a living fossil[25]. It is the only extant genus of the family Platanaceae[26], represented by as few as eight known extant species[3]. Though Doyle and Endress[27] mentioned orthotropous ovules, free nuclear endosperm development and large embryos as similarities between Platanaceae and Proteaceae, these were not sufficient to club them together in one order[20].

The molecular evidence for the closeness of the relationship between Platanaceae and Proteaceae has promoted the search for previously obscure morphological synapomorphies. Carpenter *et al.*[24] found similarity of trichome base structure in the two families. They interpreted it as another morphological similarity – the first non-reproductive trait – linking the two families. Our finding of proteoid roots in *P. orientalis* provides one more such evidence.

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