

# Morphological characteristics of phytoliths from representative conifers in China

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## Abstract

Coniferous phytoliths in sediments are an effective tool for detecting the historical appearance of conifers. However, at the timberline in mountainous areas, such coniferous phytoliths are easily confused with grass phytoliths. This study analyses modern phytoliths from 17 conifer plants. Six common types and six rare types were identified. The conifers studied produce abundant blocky polyhedral and cubic (in the average 30–40  $\mu\text{m}$  size range), blocky scrobiculate (average 30–40  $\mu\text{m}$ ), tabular elongate unsculpted (length 50–100  $\mu\text{m}$ , width 10–20  $\mu\text{m}$ ), tabular elongate cavate (length 50–150  $\mu\text{m}$ , width  $\sim$ 10  $\mu\text{m}$ ), tabular elongate dendritic (50–100  $\mu\text{m} \times$  10–20  $\mu\text{m}$ ), and irregular oblong (20–40  $\mu\text{m}$ ) phytoliths. This paper aims to show morphological characteristics of coniferous phytoliths in China, and to show how the common coniferous phytoliths differ from similar grass phytolith types, such as blocky polyhedral coniferous phytoliths from silicified parallelepipedal bulliform cells produced by grass. Blocky polyhedral and cubic phytoliths are the commonest coniferous phytoliths found in the sediments, but need to be carefully distinguished from grass parallelepipedal bulliform cells. This study indicates that clearly protruding ridges and irregular inward edges are essential features of cubic and polyhedral morphotypes produced by conifers. Results of this paper might provide important material for the study of paleovegetation and paleoecology of mountainous areas, especially at the timberline.

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## 1. Introduction

Phytoliths are increasingly accepted as new vegetation, environment, and climate proxies (Lu et al., 2006, 2007; An and Lu, 2010; Barboni et al., 2010; Sánchez et al., 2010), which have now been used effectively in both archeology and paleoecology for strata dating back to the Cretaceous (Piperno and Sues, 2005; Prasad et al., 2005, 2011; Sánchez et al., 2010). To date, few studies have dealt with phytoliths from conifers (Rovner, 1971; Klein and Geis, 1978; Kondo and Sumida, 1978; Kondo et al., 2003; Kondo, 2010).

Phytoliths are solid silica deposits in higher plants, where phytoliths have retained a silicified cellular or intercellular structure. Phytoliths originate from  $\text{H}_4\text{SiO}_4$ , which is contained in

soil and absorbed into plant tissues by roots (Piperno, 1988; Sangster et al., 2001). Although pollen is the conventional vegetation indicator, it may provide a biased vegetation signal in mountains and alpine areas where gymnosperms are abundant. Indeed, the saccate pollen morphology of gymnosperms favors their transport over long distances (Lu et al., 2008). This deficiency in pollen as an indicator can be countered by using phytoliths, which have the advantage of gravitational and autochthonous deposition (Wang and Lu, 1992; Barboni et al., 2010). Phytoliths can also be a better proxy in oxidizing environments such as soils and lake sediments frequently exposed in the air, while pollen is apt to be degraded or even destroyed in these environments (WoldeGabriel et al., 2009; Barboni et al., 2010).

The accuracy of phytoliths as paleovegetation and paleoclimate indicators relies heavily on the accurate and detailed research into phytoliths extracted from modern plants. In order to reconstruct historical vegetation composition, it is crucial

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to understand phytolith types in modern forest trees (Kawano et al., 2007). For example, coniferous plants are widespread in the southern Qinghai–Tibet Plateau (hereafter abbreviated as QTP), an area most sensitive to global climate change (Zhong, 2002). Coniferous phytolith research is therefore of significance for reconstructing paleovegetation in this mountainous area, although coniferous phytolith content is much lower than grass content (Hodson et al., 2005). However, a large number of studies have focused on herbaceous, rather than woody, phytoliths from modern plants (Alexandre et al., 1997; Lu and Liu, 2003; Bremond et al., 2008; Blinnikov et al., 2013). Only a few studies of coniferous phytoliths have been conducted.

Early in 1971, Rovner analyzed phytoliths from five species of coniferous plants, and gave a general description of the content and morphology of phytoliths from each species. He found that polyhedrons are particular to conifers. However, he pointed out that phytolith analysis was poor at distinguishing conifer categories (Rovner, 1971). Klein and Geis (1978) quantified phytoliths for 15 coniferous species (six genera) from the US in detail, and classed phytoliths by their locations within cells. Klein and Geis attempted to differentiate coniferous phytoliths from grass phytoliths, but were largely unsuccessful, as knowledge of phytoliths produced by Poaceae was limited at that time. In order to explore the silicification of epidermic cells, mesophyll cells, stoma, epidermic hair, epidermic hair bases, and vascular bundles, Kondo and Sumida (1978) extracted phytoliths from 16 coniferous plants in Japan. They classified coniferous phytoliths into three main types, including polyhedral, tabular and oval. Kondo et al. (2003) made a detailed description of the production and morphology of phytoliths, using scanning electron microscopes. In Europe, Carnelli et al. (2004) described the phytoliths from six alpine conifer species (found at altitude 2000 m asl). These efforts laid the groundwork for the further study of coniferous phytoliths.

There are only a few studies of surface soils and sediments that used phytoliths to show coniferous phytoliths as a separate category (Sase et al., 1993, 1995; Hosono et al., 1995; Kawano et al., 2006; Lu et al., 2006, 2007). In order to improve the characterization of vegetation types dominated by conifers, such as subalpine forest in the QTP area, this paper undertook a study of the Himalayan Cedar, which is abundant in this region. Coniferous phytoliths may not have been separated from grass types due to poor identification and a paucity of available references, especially any detailing phytoliths in China. Therefore 17 representative common coniferous plants of China were collected as a whole for this phytolith research. This paper summarizes the morphology and taxonomic identification characteristics of phytoliths from typical mountain conifers, based on the results of this study. This research also attempts to distinguish coniferous phytoliths from similar grass types.

## 2. Material and methods

Conifer samples were collected from the Beijing Botanical Garden, Institute of Botany, Chinese Academy of Sciences. Sampling of all the 17 coniferous plants was conducted in October 2009, at the end of growing season. After each plant

was photographed, samples of mature stems and leaves were cut off, put into paper bags, labeled and recorded by name. Plant information, including botanical name, CVBG (Chinese Virtual Botanical Garden) number and field distribution, was noted on each label. Original records are detailed in Table 1.

Phytoliths were extracted using a conventional oxidation method with the exception of some specific adjustments (Pearsall, 2000; Piperno, 2006). The stems and leaves of each sample were first washed in distilled water in an ultrasonic cleaner. After this, samples were dried at 55 °C for 24 h. The dried samples were cut into small pieces ~1–5 mm and put into plastic test tubes to be oxidized by 68% nitric acid. Then samples were washed in distilled water until neutral. Finally, phytoliths were mounted on labeled slides in Canada balsam, and morphotypes were observed under a Leica microscope. Classification was conducted by following the International Code for Phytolith Nomenclature 1.0 (Madella et al., 2005). Photographs were taken using a Nikon camera.

## 3. Results

### 3.1. Phytolith types

Of the 17 plants sampled, only eight contained phytoliths (Table 1), which were classified into six rare types and six common types as follows (Table 2).

Blocky polyhedral and cubic phytoliths (Fig. 1a–d and f) often appear in *Picea* and *Abies* species. This type is characterized by smooth surfaces, sharp sections or sides, and is concave on one or both sides, with sharp edges, clear ridges and distinct corners, sometimes in clusters. The length of a side is 30–40 μm. Rovner (1971) called this shape an “irregular polyhedron”; Klein and Geis (1978) gave it the cytologic label “endodermal cell”; Strömberg (2004) referred to it as a blocky polyhedron; and Carnelli et al. (2004) called it “BLOCK-RIDGED”.

Blocky scrobiculate phytolith type (Fig. 1e) appears blocky with smooth edges, and has a psilate to irregular surface, sometimes with wrinkles or pits. The size of this type is usually ~30–40 μm. Carnelli et al. (2004) called it POLY CONIF, and stated that blocky polyhedrons transfusion cells are often pitted.

Unsculpted surfaces, straight edges and oblate cross-sections are features of tabular elongate unsculpted phytoliths (Fig. 1h, l). Such phytoliths are mainly found in *Cedrus* and *Larix*, and 50–100 μm long and 10–20 μm wide. In *Cedrus*, they exhibit as thin and fragile. This type of phytoliths corresponds to the “epidermal cell” of Klein and Geis (1978), and the “TRACH” of Carnelli et al. (2004).

Tabular elongate cavate (Fig. 1g, k) phytolith type is featured by a hole in the surface, as well as oblate cross-section. The length of tabular elongate phytoliths is usually 50–150 μm, and the width is ~10 μm.

Tabular elongate dendritic (Fig. 1i) is elongate with dendritic edges and oblate cross-sections. The length of tabular elongate dendritic phytoliths is ~50–100 μm, and the width is ~10–20 μm. Carnelli et al. (2004) called it “undulated hypodermal cell”.

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