Holzforschung
47 (1993) 465–472

The Occurrence of Calcium Oxalate Crystals in the Cell Walls of the Secondary Phloem of Taxodiaceae

By Takao Itoh[1] and Kyung Duck Kang[2]

[1] Wood Research Institute, Kyoto University, Uji, Kyoto 611, Japan
[2] Department of Biology, College of Natural Sciences, Chonbuk National University, Chonbuk, Chonju 560–756, Korea

Keywords
Calcium oxalate
Cell wall
Phellogen
Secondary phloem
Rhytidome
Taxodiaceae
Cryptomeria japonica

Summary

Crystals were found in the intercellular layer or middle lamella of the radial walls of the secondary phloem in all species examined in Taxodiaceae. Deposition of the crystals is initiated in the cell walls of immature phloem cells adjacent to the cambium and they are distributed throughout the living secondary phloem. The crystals are absent from rhytidome except in *Glyptostrobus*, *Metasequoia* and *Taxodium* in which crystals were often found in the innermost tissue layer of rhytidome. The seasonal changes in the distribution of the crystals in the cell walls of *Cryptomeria japonica* indicate that the crystals disappear from the oldest annual ring of the living secondary phloem which is destined to change into rhytidome in June. These phenomena suggest the involvement of phellogen activity in the disappearance of the crystals from the cell walls of Taxodiaceae. The amount of such crystals varies from species to species; the largest amounts of crystals were observed in *Taxodium distichum*. Energy-dispersive X-ray analysis and acid treatment demonstrated that the crystals are composed of calcium oxalate.

Introduction

Crystals of many different shapes are observed in wood tissue. The crystals are present as styloid crystals, crystal sand, acicular crystals, variously shaped prisms, druses and raphides (Esau 1969; Franceschi and Horner 1980). Most of the crystals in wood tissue are composed of calcium oxalate (Watterdorf 1969; Frink 1991). In some cases, the crystals consist of calcium carbonate (Lee *et al.* 1985) or silicic acid anhydride, that is silica (Amos, 1952). Such crystals usually occur in the lumen of axial and/or ray parenchyma cells of both xylem and phloem tissue. Different forms of crystals are often found in a single species. Also, crystals with different constituents can be found in a single species (Lee *et al.* 1985; Taniguchi *et al.* 1987). The crystals are probably formed after the completion of metabolism of a cell before cell death. It has been pointed out that crystals of calcium oxalate can occur in the cell wall (Esau 1969; Franceschi and Horner 1980). More recently, Fink (1991) described the distribution of calcium oxalate crystals in various conifer needles. Such crystals occurred in the vascular bundles extracellularly in the xylem and phloem walls, extracellularly on the outside of the walls of mesophyll cells which face the intercellular spaces, and as numerous small crystals within the cell walls of the epidermal cells. However, the exact location, seasonal change of the crystals and the extent to which crystals are distributed in the secondary phloem tissue of coniferous species has not yet been demonstrated. It has been reported that many small crystals can be found in the cell walls of secondary phloem tissue of *Cryptomeria japonica* (Itoh 1971; Miyakawa *et al.* 1973). Thereafter, the present authors observed crystals in the radial walls of the secondary phloem tissue in six genera of Taxodiaceae. Seasonal changes in the distribution of the crystals in the cell wall were also examined in an effort to understand the appearance and disappearance.

Materials and Methods

The materials used in the present investigation included six genera of Taxodiaceae: *Cryptomeria japonica* D. Don, *Cunninghamia lanceolata* Hooker, *Glyptostrobus pensilis* K. Koch, *Metasequoia glyptostroboides* Hu *et* Cheng, *Sequoia sempervirens* Endl. and *Taxodium distichum* Rich. These species that belongs to 30 to 40 years old were used for the study of the distribution of crystals in cell walls. *Cryptomeria japonica* was used for examination of the appearance and disappearance of crystals in the cell walls of the secondary phloem tissue. The bark tissue of these species of Taxodiaceae, with the exception of *Cryptomeria japonica*, were collected in the Botanical Gardens of Osaka City University in the winter season of December. The bark tissues of *Cryptomeria japonica* were collected once a month from April to December in the experimental field of Wood Research Institute and fixed in 3.7% glutaraldehyde in phosphate buffer, pH 7.2. Some of the tissue blocks were further fixed with 1% osmium tetraoxide. Then these tissue blocks were embedded in Epoxy resin after dehydration in an alcohol series. Semi-thin sections of about 2–5 μm in thickness were cut on an ultramicrotome from the tissue blocks that has been fixed only in glutaraldehyde solution. These sections