Fine Structure and Development of Laticifers in *Gnetum gnemon* L.¹

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Summary

Articulated laticifers are a regular component of stem cortex and pith of *Gnetum gnemon* L. Differentiation of laticifers starts very early and is completed within the very first centimeters adjacent to the apical meristem.

Ultrastructurally, young laticifers of *Gnetum* can be distinguished from surrounding cells by the presence of characteristic cytoplasmic inclusions, called spherule complexes and composed of 40–70 nm light spherules and up to 0.5 by 1.0 μm large particles. While there is no record on their first beginnings, a connection between rER and the large particles can be demonstrated during many steps of laticifer differentiation which includes an increase in spherule complex areas. On account of positive Sudan staining and responses to EM fixations and in comparison to other laticifers it is concluded that the spherule complexes are terpenoid-containing. A transition from spherules to larger particles (or *vice versa*) is discussed, but could not be documented.

Relative to the formation of the spherule complexes the other changes during laticifer maturation are inferior. The vacuolar system, in close connection to an extensive ER system, does largely expand and finally takes over the almost entire cell lumina. Nuclei do persist for an extended period, even after breakdown of the end walls and during disorganization of the cytoplasm. Plastids in all stages of laticifer ontogeny are very rarely encountered.

1. Introduction

*Gnetum gnemon* L. (*Gnetales, Cycadophytina*) is one of the rare examples of the occurrence of laticifers outside flowering plants: next to a few other *Gnetum* species the pteridophyte *Regnellidium* (*Marsileaceae*) is the only other positive genus. In *Gnetum* laticifers have been first described by Bower

¹ Supplemented part of an investigation presented in 1976 by S. H. to the Fakultät für Biologie der Universität Heidelberg in partial fulfilment of the requirements for the degree of a Diplom-Biologe.
Previous light and electron microscopic studies on the phloem of the same species (Palwal and Behnke 1973, Behnke and Palwal 1973) occasionally included observations on laticifers regularly occurring in cortex, pith and (more seldom) phloem tissues. Laticifers define several different single cells or series of interconnected cells (cf., Esau 1965) which generally function as depositories for the intracellular excretion of terpenoids, resins, tannins, alkaloids and others, altogether named latex. They are restricted to some 900 genera in about 30 families, among which the most prominent are Euphorbiaceae, Asteraceae, Apocynaceae, Asclepiadaceae, Papaveraceae (for detailed listing see Metcalfe 1967).

Ultrastructural investigations aimed at elucidating the origin and development of the different latex components demonstrated the synthesis of polyterpene particles in the cytoplasm (Schulze et al. 1967, Heinrich 1967), the location of alkaloids in vacuoles which most likely are derived from endoplasmic reticulum (Nessler and Mahlberg 1977) and the formation of lytic enzymes in autophagic vesicles (Marty 1971) which sometimes were named “lutoids” (Purjanisclie 1966) and in some species were identical with the alkaloid-containing vesicles (Matile et al. 1970). Despite obvious lytic activities of these vesicles, even in mature laticifers of many species the protoplasts are reported to remain intact and separated from the vacuoles by tonoplast membranes (Schnepp 1964, Schulze et al. 1967, Thureson-Klein 1970).

In view of the rare occurrence of laticifers outside angiosperms and many open questions concerned with the ontogeny of the latex it was worthwhile to follow up the structural development of laticifers in Gnetum gnemon.

2. Materials and Methods

Young stem parts, including apical meristems, were taken from a Gnetum gnemon L. tree growing in a greenhouse of the Botanical Garden Heidelberg and subjected to one of the methods described below. Although appropriate material was available throughout the whole year, for electron microscopy best results were obtained with spring and early summer material, only. The description of laticifer ontogeny, therefore, exclusively is using this material.

2.1. For light microscopy short stem pieces or thick hand sections were fixated during 24 hours at 4 °C in a mixture of formaldehyde-glutaraldehyde (Karnovsky 1965), washed,