

UDC 581.174.2:582.734.3 + 29

PHYTOFERRITIN ACCUMULATIONS IN CHROMOPLASTS OF *SORBUS AUCUPARIA* L. FRUITS

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Received November 23, 1981

The massive accumulations of regularly arranged phytoferritin particles were found in the stroma of chromoplasts in ripe fruits of *Sorbus aucuparia*. The phytoferritin particles were not, or only very exceptionally, found in chloroplasts of green unripe fruits.

The origin and function of phytoferritin inclusions in plastids, especially in chromoplasts are discussed.

Introduction

Phytoferritin has been reported to occur in a variety of angiosperms (for literature see: Ljubešić 1976, Wildman and Hunt 1976), in ferns (Sheffield and Bell 1978), in fungi Peat and Banbury 1968, David and Easterbrook 1971) and in red algae (Pueschel and Cole 1980). Every phytoferritin particle contains a core, 5—6 nm in diameter, which possesses native electron density due to the presence of iron. A proteinaceous shell, conspicuous in negative stain, surrounds the core.

Conditions under which phytoferritin appears are manifold: e. g. senescence, viral infections, cellular differentiation, mutations etc. (see: Seckbach 1972). In general phytoferritin is present in plastids with reduced amount of photosynthetic membranes — in photosynthetically inactive or less active plastids. Phytoferritin is almost exclusively located in the plastid stroma, except in fungi where it is associated with lipid globules. Phytoferritin is present in many types of plastids: the proplastids of the male generative cell of *Pelargonium* (Knoth et al. 1980), the plastids from phloem of *Centrospermae* (Behnke 1977) and in the chloroplasts of different developmental or degradation stages (Ljubešić 1976, Wildman and Hunt 1976). However, the presence of phytoferritin in typical chromoplasts has not been described yet (Matienko 1973).

The present report is concerned with heavy phytoferritin deposits in the tubulous type of chromoplasts in *Sorbus aucuparia* fruits and discusses evidence that the disintegration of thylakoids is responsible for the formation of phytoferritin.

Material and Methods

Fruits of *Sorbus aucuparia* L. were collected from the trees growing in the Botanical Gardens of Zagreb University during the ripening period of the fruits (June — September). The subepidermal cell layers of the fruits were studied by light and electron microscopy. The material was fixed in 1% glutaraldehyde and postfixed in 1% OsO₄ (at 274 K). In some cases the postfixation was omitted. After dehydration in ethanol series the material was embedded in Araldite and cut with a Reichert Om U2 ultramicrotome and stained with uranyl acetate and lead citrate. The samples of ripe old fruits were prestained before dehydration with 2% aqueous uranyl acetate for 12 hours. The materials were observed in a Siemens Elmiskop I.

Results

The young unripe fruits of *Sorbus aucuparia* usually contain chloroplasts without phytoferritin particles (Fig. 1). However, smaller phytoferritin accumulation are rarely present (Fig. 2). The chloroplasts of young green fruits contain a few small grana consisting of up to 10 thylakoids. Globules (0.1 — 0.5 μm in diameter) are quite irregular in shape and heterogeneously osmiophilic.

During the process of ripening thylakoids gradually disappear. Plastoglobules become numerous and bigger. Among them only small remnants of thylakoids are present. There are massive accumulations of phytoferritin in each plastid (Fig. 3).

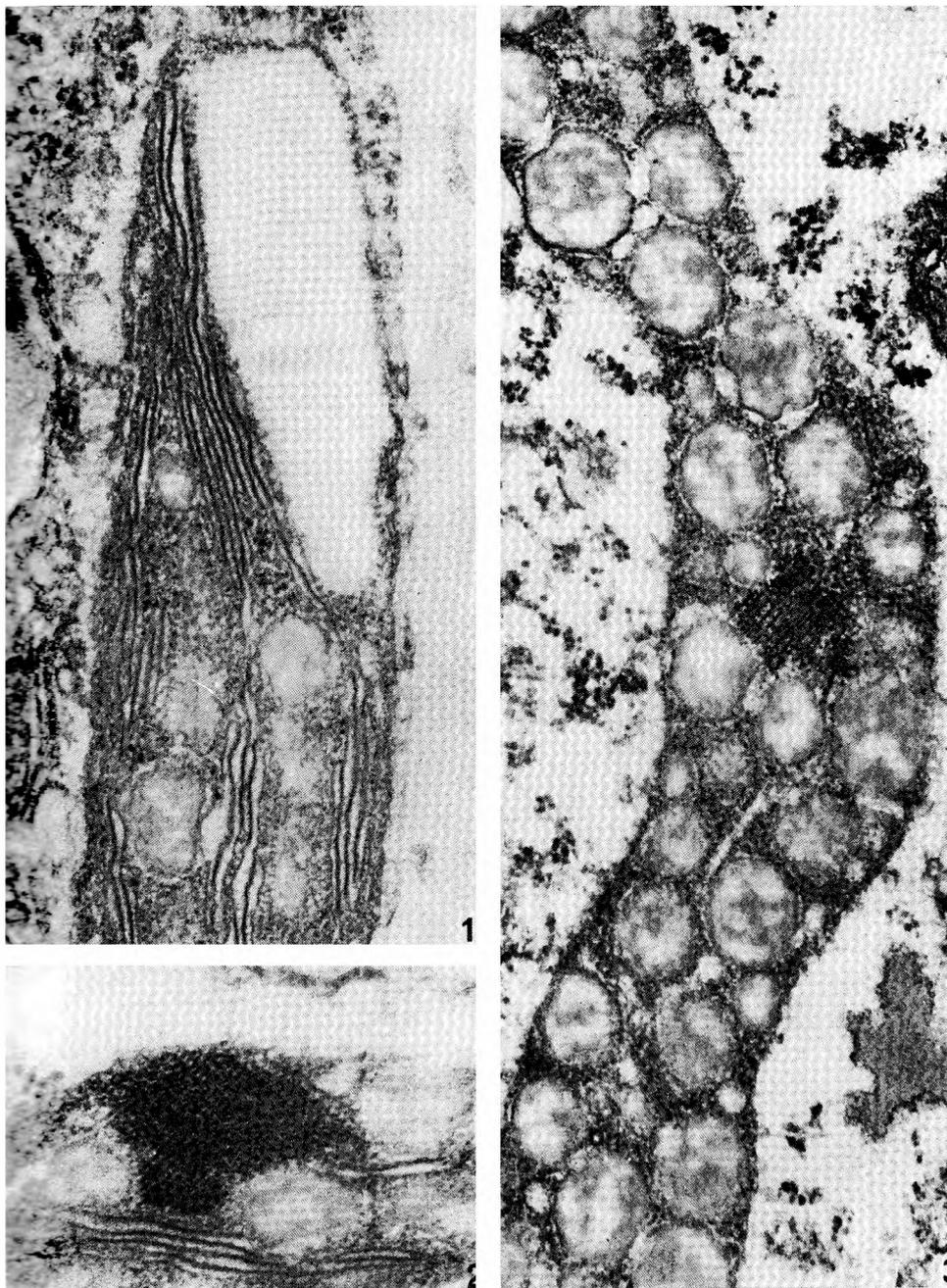
Fig. 1. Chloroplast from green unripe fruit. Starch, plastoglobules and small grana are present. 60,000 : 1.

Fig. 2. Part of chloroplast at the beginning of the ripening process with phytoferritin inclusion. 65,000 : 1.

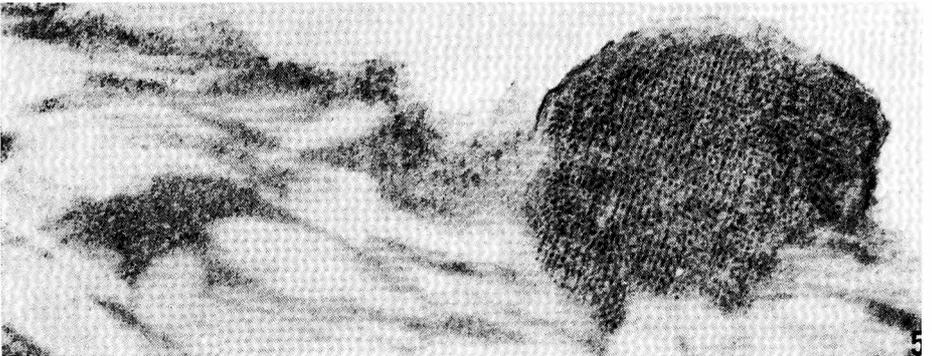
Fig. 3. Plastid from the ripening process of the fruit with phytoferritin accumulation. 50,000 : 1.

Fig. 4. Chromoplasts from the ripe fruit with large phytoferritin inclusions among the fibrils (tubules). Plastoglobules are big and heterogeneously osmiophilic. The upper phytoferritin inclusion shows its structure clearly only in the circular field where the exposure time of the photographic paper was shortened to reach the proper contrast. 52,000 : 1.

Fig. 5. Phytoferritin accumulation in chromoplast of ripe fruit. The material was prepared without OsO₄-postfixation. Unstained section. The electron dense points are the consequence of the presence of iron atoms in the core of each phytoferritin particle. 82,000 : 1.



Figs. 1—3.



Figs. 4—5.

Ripe fruits are intensely red. Subepidermal cell layers contain very long spindle-shaped chromoplasts. They are full of bundles of long, non-ramified fibrils (tubules). In each chromoplast there is a big accumulation of phytoferritin among the fibrils (tubules). The diameter of this accumulation is about 0.5 μm . The arrangement of phytoferritin particles is regular (crystalline) (Figs. 4, 5). Irregular arrays have not been noticed.

Phytoferritin inclusions remain unchanged in the chromoplasts of *Sorbus aucuparia* till the decay of the fruits.

Discussion

Phytoferritin inclusions are present in photosynthetically inactive or less active plastids. The formation of these plastids occurs generally in two basic ways. In the first case the plastids are originally photosynthetically inactive: as proplastids (Knoth et al. 1980), the plastids of photosynthetically deficient mutants (Röbbelen 1966), and the plastids from phloem (Behnke 1977). In these cases phytoferritin represents a storage substance of iron in a non-toxic form. If such plastids develop into normal chloroplasts the phytoferritin particles disappear.

The other and more frequent way is when the normal chloroplasts are secondarily transformed into photosynthetically inactive plastids. Senescence (Barton 1970, Ljubešić 1976) and lethal yellowing diseases (Maramorosch and Hirumi 1973, Wildman and Hunt 1976) are the most frequent examples of this process. The appearance of phytoferritin is a parallel process with the disintegration of the thylakoidal system. The appearance of phytoferritin inclusions is the result of the degradation process of iron-containing molecules such as ferredoxin and cytochrome (Pueschel and Cole 1980).

The majority of chromoplasts originates from chloroplasts, i. e. after the disintegration of thylakoidal system photosynthetically active chromoplasts. Although during this process the phytoferritin particles appear very often (Ljubešić 1976), such inclusions, as found in *Sorbus aucuparia* fruits have not been described yet in chromoplasts. It is still unknown why the chromoplasts of *Sorbus aucuparia* fruits contain phytoferritin, while the chromoplasts of other species do not.

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The author is indebted to Dr. M. Wrischer and Professor Z. Devidé for helpful discussions and critical reading of the manuscript.

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SAŽETAK

NAKUPINE FITOFERITINA U KROMOPLASTIMA PLODOVA VRSTE *SORBUS AUCUPARIA* L.

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U kromoplastima zrelih plodova vrste *Sorbus aucuparia* L. nađene su velike nakupine fitoferitinskih čestica. Kloroplasti nezrelih zelenih plodova sadrže ih samo u rijetkim i izuzetnim slučajevima. Autor raspravlja o porijeklu i ulozi fitoferitina u plastidima, napose u kromoplastima.

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