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BRIEF COMMUNICATION

Microtubule-like structures in developing wheat-leaf chloroplasts

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Abstract.

Microtubule-like structures were observed in mesophyll cell chloroplasts during the ontogeny of the second leaf of wheat cultured in full nutrient solution as well as under nitrogen deficiency.

Microtubule-like structures (MTLS) in the chloroplasts described up to now in about two dozens of papers (*e.g.* Bartels and Weier 1967, Lawrence and Possingham 1984) remain still somewhat controversial plastid structures. Recently, Artus *et al.* (1990) established that the formation of MTLS in greening etioplasts within first leaves of wheat was not inhibited by microtubule inhibitors and so MTLS presumably did not consist from tubulin. Little is known about the MTLS occurrence in chloroplasts during the whole life span of the leaf; only Lawrence and Possingham (1984) studied the chloroplast MTLS during spinach leaf development.

Triticum aestivum L. cv. Grana plants were grown in a modified Knop's nutrient solution with or without nitrogen (+N or -N plants, Činčerová 1990) in an air-conditioned room (16 h irradiation of about 150 μmol m^{-2} s^{-1}, 8 h dark). The samples for the transmission electron microscopy were taken from the middle third of the second leaf blades 9, 11, 15, 22, 29, and 36 d after plant sowing always in the middle of the light period. The samples were prepared by the conventional procedure: glutaraldehyde/osmium acid fixation, embedding in Spurr's low viscosity resin, contrasting with uranyl acetate and lead citrate (see Kutík *et al.* 1988).

During inspection of about five hundred sections of mesophyll cell chloroplasts, groups of small electron dense circles or bundles of electron dense strips of various lengths (sometimes with electron translucent core) were observed (see Fig. 1).

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They obviously represent the transverse or longitudinal sections of the MTLS bundles. The outer diameter of these MTLS was about 13 nm which is in agreement with the results of Artus *et al.*(1990) and Bartels and Weier (1967) who also worked with wheat and reported the outer diameters of MTLS of 13 ± 2 and 13 nm, respectively. A different diameter of MTLS was found *e.g.* by Lawrence and Possingham (1984) in spinach chloroplasts (23 nm). For cytoplasmic microtubules, the diameter of about 25 nm has been reported (*e.g.* Lloyd 1982).

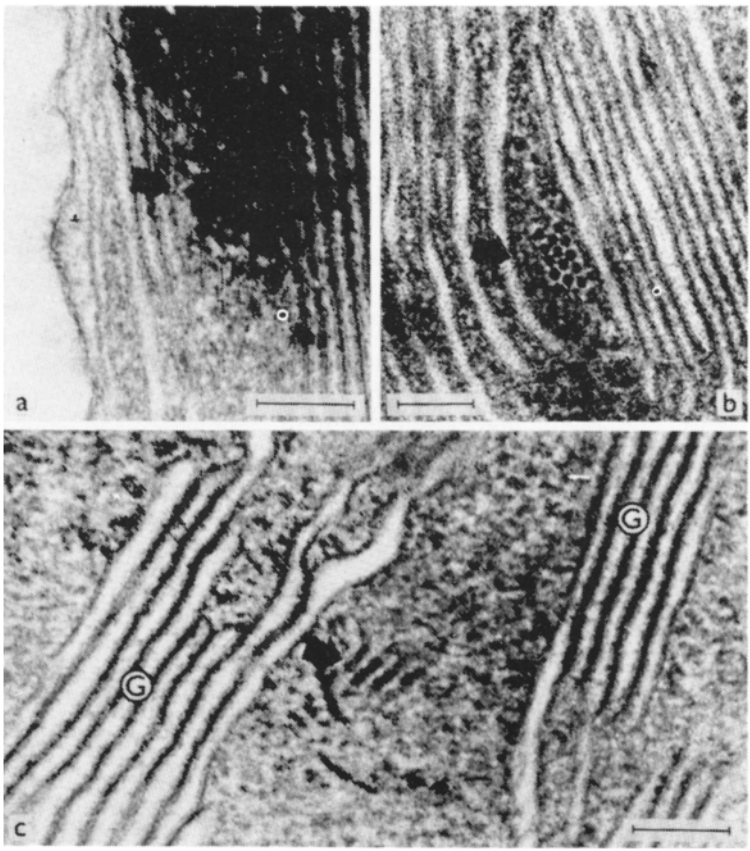

Fig. 1. Transverse (*a*) and longitudinal (*b*) sections of microtubule-like structures (MTLS; indicated by *arrows*) in the chloroplasts in mesophyll cells of the second wheat leaves: *a*) group of MTLS between unstacked (*left*) and stacked (*right*) thylakoids; *b*) a small bundle of MTLS situated between two granal stacks (*G*). Plants grown without nitrogen, 11 d after sowing; ultrathin sections; bar = 0.1 μm.

Lawrence and Possingham (1984) found a decreasing number of MTLS per a chloroplast section in the cells of palisade and spongy parenchyma with an increasing length of the growing spinach leaves; no MTLS were visible in the chloroplasts of mature leaves. This is principally in agreement with my experiment

studying the development of the second wheat leaves: MTLS were most frequent in young leaves, 9 and 11 d after sowing. They were found in about two thirds of the observed chloroplast sections, more frequently in +N plants. In +N plants, the frequency of MTLS did not decrease substantially until 22 d after sowing when the leaves and their chloroplasts reached the maturity (fully expanded leaves contained chloroplasts having the most developed system of thylakoids with large grana). In -N plants, the frequency of MTLS decreased with the age of leaves. After 22 d, their chloroplasts were smaller then those of +N plants and had conspicuous starch inclusions; less than one half of the chloroplasts contained MTLS. In senescent leaves, *i.e.* 29 d after sowing (and also 36 d after sowing in the +N plants), the chloroplasts showed a partial disorganization of the system of thylakoids and contained large plastoglobules. In these leaves, MTLS were very rare or absent in both the +N and -N plants.

Therefore, MTLS may play pivotal role in the controlling the growth and differentiation of the system of chloroplast thylakoids, the process advancing until the chloroplast senescence (*e.g.* Kutík *et al.* 1988).

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