STRUCTURE AND FUNCTION OF THE CHLOROPLAST Ndh COMPLEX

The chloroplast DNA of the most of higher plants contains 11 genes *ndh* encoding components of the multi-polypeptidic Ndh complex of thylakoid, homologous to the complex I (EC 1.6.5.3) of the respiratory electron transport chain, which catalyses the reduction of plastoquinone (PQ) with NADH:

 $NADH + H^{+} + PQ \rightarrow NAD^{+} + H_2PQ$

By feeding electrons from NADH, the Ndh complex participates in a system that regulates the oxido-reduction level of intermediaries of the photosynthetic cyclic electron transport from which two additional reactions drain electrons: 1. Mehler reaction:

 $FeSP_{red} + O_2 \rightarrow FeSP_{oxid} + O_2^-$ (superoxide anion radical)

2. Thylakoid peroxidase reaction:

$$H_2PQ + H_2O_2 \rightarrow PQ + H_2O$$

Where hydrogen peroxide is generated by the superoxide dismutase (SOD) catalyzed reaction:

$$2 \cdot O_2^- + 2 H^+ \rightarrow 2 H_2O + O_2$$

Alternatively, an oxidase could oxidise H₂PQ to PQ with O₂.

By acting in a concerted way, the reactions cited above ensure that the intermediaries of the photosynthetic cyclic electron transport are not highly oxidised or reduced, therefore allowing high rates of electron transport, appropriate membrane potential and photophosphorylation.



The levels of the Ndh complex increase under photo-oxidative stress conditions and early during leaf senescence. The last increase of the Ndh complex is not accompanied by a similar increase of SOD. As a consequence, it determines a rise of reactive oxygen species (ROS) which trigger cell death. Through this way, the chloroplast plays a regulatory role of leaf senescence similar to that of mitochondria in the programmed cell death in other organs. Accordingly, transgenic tobacco plant defective in the Ndh complex ($\Delta ndhF$) shows a marked delay of leaf senescence

when compared with wild type (*wt*) tobacco.



wt

∆ndhF

Two main difficulties to investigate the thylakoid Ndh complex stem from its scarcity (one Ndh per 100 to 200 of each photosystem I and photosystem II complexes) and its low stability in purified fractions. Similarly to the complex I of the respiratory chain, the thylakoid Ndh complex seem to have an L shape with one hydrophobic arm imbibed into membrane and one hydrophyllic arm. This last, in chloroplasts, is sided to stroma where, plausibly, is the NQO1 subunit that binds NADH. Several experiments and the probable analogy with the respiratory complex I suggest the probable orientation in thylakoid of the Ndh complex shown bellow.



The nuclear genes that probably encode the subunits NQO1, NQO2 y NQO3 (in white) still remain to be identified. In contrast, the chloroplast genes encoding the other 11 subunits (in grey: NDH-A to NDH-K), have been identified in the most of higher plants investigated.

The NDH-F subunit probably contains 15 transmembrane helices, one conserved and phosphorylable 181-threonine and one also conserved 349-histidine which could be part of a proton channel as indicated in the following figure showing the topology of the NDH-F subunit in the thylakoid membrane.



The phosphorylation of the NDH-F subunit increases the catalytic activity of the Ndh complex. A large hydro-philic region (m) of the NDH-F subunit is sided to thylakoid lumen.

Active investigations in this field pursue the identification of the NQO subunits and their corresponding genes, transmembrane topology of plastid-encoded subunits other than NDH-F and interactions among the different subunits. Proteomic as well as point mutation analyses are promising tools in this and other fields. Therefore, point mutation analyses could give light on the functional relevance of the 181-Thr phosphorylation and of the proton channel.

The generation of transgenic plants showing delayed leaf senescence by modifying components of the Ndh complex is also a promising field in the biotechnology of post-harvest vegetables and plant productivity.