

Healthy chloroplasts for healthy sports turf

Plant chloroplasts are large organelles that, like mitochondria, are bounded by a double membrane called the chloroplast envelope. In addition to the inner and outer membranes of the envelope, chloroplasts have a third internal membrane system, called the thylakoid membrane. The thylakoid membrane forms a network of flattened discs called thylakoids, which are frequently arranged in stacks called grana. Because of this three-membrane structure, the internal organization of chloroplasts is more complex than that of mitochondria. In particular, their three membranes divide chloroplasts into three distinct internal compartments: (1) the intermembrane space between the two membranes of the chloroplast envelope; (2) the stroma, which lies inside the envelope but outside the thylakoid membrane; and (3) the thylakoid lumen.

In addition to the inner and outer membranes of the envelope, chloroplasts contain a third internal membrane system: the thylakoid membrane. These membranes divide chloroplasts into three internal compartments.

The major difference between chloroplasts and mitochondria, in terms of both structure and function, is the thylakoid membrane. This membrane is of central importance in chloroplasts, where it fills the role of the inner mitochondrial membrane in electron transport and the chemiosmotic generation of ATP. The inner membrane of the chloroplast envelope (which is not folded into cristae) does not function in photosynthesis. Instead, the chloroplast electron transport system is located in the thylakoid membrane, and protons are pumped across this membrane from the stroma to the thylakoid lumen. The resulting electrochemical gradient then drives ATP synthesis as protons cross back into the stroma. In terms of its role in generation of metabolic energy, the thylakoid membrane of chloroplasts is thus equivalent to the inner membrane of mitochondria.

The Structure and Function of Chloroplasts

Plant Cell Chloroplast Structure

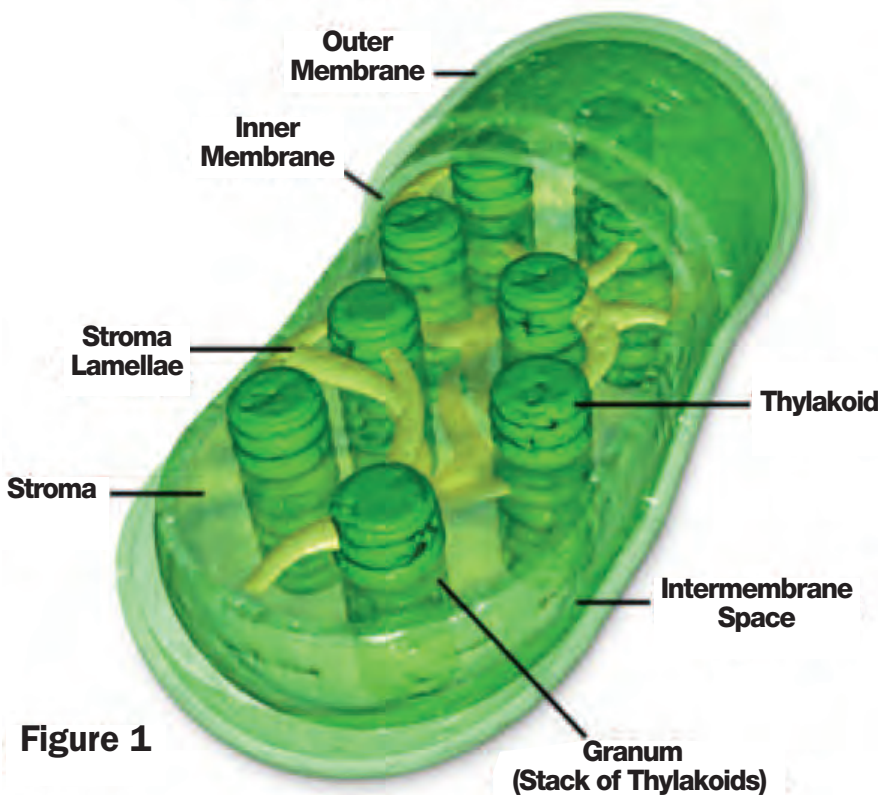


Figure 1

THE CHLOROPLAST GENOME

Like mitochondria, chloroplasts contain their own genetic system, reflecting their evolutionary origins from photosynthetic bacteria. The genomes of chloroplasts are similar to those of mitochondria in that they consist of circular DNA molecules present in multiple copies per organelle. However, chloroplast genomes are larger and more complex than those of mitochondria, containing approximately 120 genes.

The chloroplast genomes of several plants have been completely sequenced, leading to the identification of many of the genes contained in the organelle DNAs. These chloroplast genes encode both RNAs and proteins involved in gene expression, as well as a variety of proteins that function in photosynthesis. Both the ribosomal and transfer RNAs used for translation of chloroplast mRNAs are encoded by the organelle genome. These include four rRNAs (23S, 16S, 5S, and 4.5S) and 30 tRNA species. In contrast to the smaller number of tRNAs encoded by the mitochondrial genome, the chloroplast tRNAs are sufficient to translate all the mRNA codons according to the universal genetic code. In addition to these RNA components of the translation system, the chloroplast genome encodes about 20 ribosomal proteins, which represent approximately a third of the proteins of chloroplast ribosomes. Some subunits of RNA polymerase are also encoded by chloroplasts, although additional RNA polymerase subunits and other factors needed for chloroplast gene expression are encoded in the nucleus.

IMPORT AND SORTING OF CHLOROPLAST PROTEINS

Protein import into chloroplasts generally resembles mitochondrial protein import. Proteins are targeted for import into chloroplasts by N-terminal sequences of 30 to 100 amino acids, called transit peptides, which direct protein translocation across the two membranes of the chloroplast envelope and are then removed by proteolytic cleavage. As in mitochondria, molecular chaperones on both the cytosolic and stromal sides of the envelope are required for protein import, which requires energy in the form of ATP. In con-

trast to the pre-sequences of mitochondrial import, however, transit peptides are not positively charged and the translocation of polypeptide chains into chloroplasts does not require an electric potential across the membrane.

Protein import into the chloroplast stroma: Proteins are targeted for import into chloroplasts by a transit peptide at their amino terminus. The transit peptide directs polypeptide translocation through the Toc complex in the chloroplast outer membrane. Proteins incorporated into the thylakoid lumen are transported to their destination in two steps. They are first imported into the stroma, as already described, and are then targeted for translocation across the thylakoid membrane by a second hydrophobic signal sequence, which is exposed following cleavage of the transit peptide. The hydrophobic signal sequence directs translocation of the polypeptide across the thylakoid membrane and is finally removed by a second proteolytic cleavage within the lumen.

The goal of every turfgrass manager is to provide a playable surface and aesthetically pleasing green turfgrass. Achieving the latter involves a reciprocal balance between soil, fertility, moisture, temperature, humidity, grass species, mowing techniques, cultural practices and cooperation from Mother Nature. All these aspects have to be working in sync for turfgrass to perform properly and be appealing color wise.

Protecting and strengthening chloroplasts would seem like the logical action to take because this is where chlorophyll, a pigment that gives turfgrass its green appearance, is developed.

The most important characteristic of turf plants is their ability to photosynthesize: to make their own food by connecting light energy into chemical energy. This process is carried out in specialized organelles called chloroplasts. A photosynthetic cell contains anywhere from one to several thousand chloroplasts. The electrons from chlorophyll molecules in photosystem II replace the electrons that leave chlorophyll molecules in photosystem I.

Located inside the chloroplast are thylakoid membranes where light reactions take place. This is where chlorophyll is found, therefore, there's a synergistic relationship between keeping the chloroplasts and the thylakoid membranes as healthy as possible.

There are events that can be harmful to chloroplasts and thylakoid membranes, as well as necessary components that can prevent damage to them.

FREE RADICALS

One event that can damage chloroplasts is the development of free radicals. Typically, free radicals are stable molecules that contain pairs of electrons. When a chemical reaction breaks the bonds that hold the paired electrons together, free radicals are

produced. They contain an odd number of electrons, which make them unstable, short-lived and highly reactive. As they combine with other atoms that contain unpaired electrons, new radicals are created, and a chain reaction begins.

This chain reaction or accumulation of reactive oxygen species, in turf plants is generally ascribed to several possible sources: cell-wall-bound peroxidases, membrane-located NADPH oxidases, amine oxidases, xanthine oxidase, chloroplastic electron transport chains, mitochondrial electron transport chains, and peroxisomal fatty acid β -oxidation, which includes the H_2O_2 -generating argyl-coenzyme A oxidase steps. These sources can be attributed to environmental causes such as drought, heat, and ultraviolet light, or chemicals such as herbicides.

Accumulation of reactive oxygen species is central to plant response to several pathogens. One of the sources of reactive oxygen species is the chloroplast because of the photoactive nature of the chlorophylls. The free radicals, or reactive oxygen species, are singlet, hydroxyl, superoxide and hydrogen peroxide.

LIGHT

When photosynthetic organisms, such as turf, are exposed to ultraviolet radiation, significant, irreversible damage to important metabolic processes within the cell might occur (such as lesions in DNA and inhibition of photosynthesis). Through these reactions and others, radical forms of oxygen are often created. Many reports suggest this damage is because of oxidative stress resulting from UV-A exposure.

Photosynthetic light absorption and energy usage must be kept in balance to prevent formation of reactive oxygen species in the chloroplasts. Drought causes stomatal closure, which limits the diffusion of carbon dioxide to chloroplasts and thereby causes a decrease of carbon dioxide assimilation in favor of photorespiration that produces large amounts of hydrogen peroxide. Under these conditions, the probability of singlet oxygen production at photosystem II and superoxide production of photosystem I is increased. These can cause direct damage or induce a cell suicide program.

It has been known for a long time wavelengths in the ultraviolet-B region of the spectrum are effective in inactivating photosynthesis, and the molecular target is photosystem II. An excess of light brings about the inactivation of oxygenic photosynthesis, a phenomenon known as photoinhibition, and the molecular target of photoinhibition is photosystem II, a thylakoid multisubunit pigment-protein complex. The major effect of ultraviolet-B light on the thylakoid proteins is the breakdown of the reaction centre D1 protein.

SENESCENCE

Senescence results in massive levels of cell death, but the purpose of senescence isn't cell death; rather death only occurs when senescence has been completed. Senescence occurs in two stages. The first stage is reversible, and the cells remain viable throughout. The second stage results in cell death.

The key enzyme in the pathway to chlorophyll degradation during senescence appears to be pheophorbide-a-oxygenase. The activity of pheophorbide-a-oxygenase increases dramatically during senescence, implicating this enzyme as a control point in the process. Light absorption by pheophorbide-a-oxygenase also is believed to cause the production of singlet oxygen, which is a free radical.

Because senescence is reversible, it suggests that fully developed chloroplasts retain enough genetic information to support re-greening and chloroplast reassembly.

CALCIUM AND POTASSIUM

From a nutritional standpoint, there are various nutrients and compounds that can be applied in the process of strengthening and defending chloroplast damage.

Because the chloroplasts and thylakoid membrane are located inside the plant cell, the first line of defense would seem to be to strengthen the plant cell by keeping calcium and potassium at optimal levels. Calcium plays a key role in strengthening the cell walls of the turf plant, while potassium helps strengthen cell walls inside the turf plant, which makes it harder for physiological problems to occur inside the cell wall.

AMINO ACIDS

Amino acids are the building blocks of proteins. Under optimal conditions, proteins are able to perform the normal physiological function to synthesize amino acids, but intensively manicured turfgrass, such as golf courses and athletic fields, are rarely operating under optimal conditions because of stress caused by low mowing heights and traffic.

To date, 154 proteins in the turfgrass plant have been identified – 76 (49 percent) are integral membrane proteins. Twenty-seven new proteins without known functions, but with predicted chloroplast transit peptides, have been identified – 17 (63 percent) are integral membrane proteins. These new proteins are likely to play an important part in thylakoid biogenesis.


The application of amino acids plays an extremely important part in developing the proteins specifically designed to help chloroplasts, thylakoid membranes, photosystem I and photosystem II to function properly. These proteins are known as D1, D2 CP43, CP47 and cytochrome b559. Of special



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importance is the D1 protein because it exhibits the highest turnover rate of all the thylakoid proteins, and is highly vulnerable to singlet oxygen, a free radical.

ANTIOXIDANTS

The antioxidants a-tocopherol (vitamin E), ascorbic acid (vitamin C), carotenoids (B-carotene), vitamin B6 and mannitol in some biostimulants play a vital role in scavenging free radicals and helping protect chloroplasts, thylakoid membranes inside the chloroplasts, photosystem I and photosystem II.

In terms of its antioxidant properties, carotenoids can protect photosystem I and photosystem II in one of four ways: by reacting with lipid peroxidation products to terminate chain reactions; by scavenging singlet oxygen and dissipating the energy as heat; by reacting with triplet or excited chlorophyll molecules to prevent formation of singlet oxygen; or by dissipation of excess excitation energy through the xanthophyll cycle.

Xanthophylls function as accessory pigments for harvesting light at wavelengths that chlorophyll can't and transfer the light energy to chlorophyll. But, they also absorb excess light energy and dissipate it to avoid damage in the xanthophyll cycle.

A-TOCOPHEROL (VITAMIN E)

A-tocopherol (vitamin E) is considered a major antioxidant in chloroplasts in at least two different but related roles. It protects photosystem II from photoinhibition and thylakoid membranes from photooxidative damage. The antioxidant properties of vitamin E are the result of its ability to quench singlet oxygen and peroxides.

ASCORBIC ACID (VITAMIN C)

It's generally believed maintaining a high ratio of ascorbic acid is essential for the scavenging of free radicals and is needed in high concentrations in the chloroplasts to be effective in defending the turfgrass against oxidative stress. Although ascorbic acid can directly scavenge the free radicals superoxide and singlet oxygen, the main benefit ascorbic acid plays in the prevention of free radicals is that it's an excellent scavenger of the hydroxyl radical. The hydroxyl radical is dangerous to turfgrass because it can inhibit carbon dioxide assimilation by inhibiting several Calvin cycle enzymes.

VITAMIN B6

Apart from its function as a cofactor, vitamin B6 is also thought to act as a protective agent against reactive oxygen species,

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such as singlet oxygen. Vitamin B6 is also the master vitamin in processing amino acids and plays an important role in developing proteins specifically designed to help chloroplasts, thylakoid membranes, photosystem I, and photosystem II to function properly.

MANNITOL

The antioxidant mannitol has the ability to protect and quench two damaging free radicals: singlet oxygen and hydroxyl. Singlet oxygen is damaging because it can react with proteins, pigments and lipids and is thought to be the most important species for light-induced loss of photosystem II activity, as well as the degradation of the D1 protein. It has been demonstrated that when mannitol is present in the chloroplasts, it can protect plants against oxidative damage by the hydroxyl radicals.

MANGANESE AND MAGNESIUM

Both of these nutrients are attached to the chlorophyll molecule that's located inside the chloroplasts. These two nutrients play a part in making turfgrass greener by helping develop chlorophyll. They also transport other vital nutrients and are responsible for many enzymatic functions and help prevent chlorophyll degradation in the cells.

CARBON

There's new evidence carbon plays a role in the development of the turfgrass plant leaf, and that a reduction in carbon reduces photosynthetic activity, which reduces carbohydrate availability to the turfgrass plant. There's also new evidence to suggest proper development of the turfgrass plant can't occur without proper amounts of carbon in the chloroplast. There's more evidence to

suggest that, if there's an abundant source of carbon in the thylakoid membranes inside the chloroplasts, it can be mobilized for use as an energy source during senescence.

HUMIC ACIDS

Humic acids are another compound that contain antioxidant properties that promote the scavenging of free radicals. The added benefits of humic acid are that they increase the availability of micronutrients, phosphate and potassium to the plant and enhance the chlorophyll content of turfgrass.

Humic acids also stimulates root initiation because of the auxin-like activity they contain, which is most likely because of their ability to inhibit indoleacetic acid oxidase breakdown. ■

Jeff Haag is sports turf specialist at John Carroll University, University Heights, OH.

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