TURFGRASS TRENDS

AGRONOMY

Thermal Tolerance: The Role of Heat Shock Proteins

By Karl Danneberger, Ph.D.

Plants live within a narrow temperature range of 32 to 122 F (0 to 50 C) with metabolic processes restricted to 50 to 104 F (10 to 40 C). Optimum shoot growth for cool season turfgrasses occurs between 60 to 75 F (10 to 24 C). When temperatures are outside the optimum range, turfgrass growth is hindered or reduced. High temperatures result in morphological changes including reduced shoot and root growth, decreased stand density and leaf width.

Physiological changes for cool season turfgrasses include reduction in photosynthesis, and an increase in respiration and photorespiration, resulting in decreased carbohydrate levels. As temperatures approach lethal levels, degradation of proteins, and membrane disruption occur resulting in overall shutdown of cellular functions. As cellular functions are disrupted or destroyed, cell death occurs leading to plant death.

The killing or lethal temperature for cool season turfgrasses is dependent on the temperature and exposure time. Upper limit for cool season turfgrasses is considered to range from 113 to 131 F (45 to 55 C), with killing temperatures for Kentucky bluegrass, perennial ryegrass, and annual bluegrass occurring around 117 F (47 C).

Indirect temperature stress where temperatures are above the optimum but below lethal, are common on cool season turfgrasses. Decrease in shoot and root growth along

with a reduction in stand density are common symptoms observed by golf course superintendents and turfgrass managers during summer stress times.

On the other hand, direct high temperature kill of turfgrass plants from a historical perspective is considered a rare event. However, as we expand the use of On the other hand, direct high temperature kill of turfgrass plants from a historical perspective is considered a rare event.

cool season turfgrasses, especially creeping bentgrass, into climactic regions that are considerably less adaptable due to higher temperatures, and as management practices become more "on the edge" (low height of cut, wear, etc.), the probability of direct temperature kill increases.

Heat shock response

We have studied how turfgrass plants respond to thermal stress. Although not a common event, turfgrasses — especially cool season turfgrasses, in the transition zone can reach canopy temperatures considered lethal, yet the plants survive without any apparent damage.

IN THIS ISSUE

Heat shock response

HSP in turfgrasses

Measuring Sod Strength of Kentucky Bluegrass and Supina Bluegrass.....6

Early sod testers

Materials and methods

Building sod strength

Consistency of measurements

 Management Forum: Control of Bluegrass in Bentgrass Fairways....10

How efficient is foliar feeding?

Growth Regulator May Help in Crabgrass Control

Bermuda and crabgrass

Sneak Preview of the Olympics 15

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Figure 1. Perennial ryegrass seedlings were exposed to various heat treatments. On the left, these seedlings were grown for 7 days at 68 F. The seedlings on the right were grown at 68 F but exposed for 1 hour at 131 F on the fourth day then returned to 68 F. The seedlings in the middle received 1 hour of 99-104 F followed by 2 hours at 131 F.

For example, we did a laboratory study where we took perennial ryegrass seedlings and grew them at an optimum temperature of 68 F (20 C) for seven days. A second group of seedlings we grew at the same temperature of 68 F but on the fourth day these seedlings received 1 hour at 131 F (55 C) then returned to 68 F for the remaining days. The third group of seedlings on the fourth day received 1 hour at 99-104 F (37-40 C) followed by 2 hours at 131 F then returned to 68 F for the remainder of the experiment.

AGRONOMY

The results from this type of experiment are shown in Figure 1. The seedlings grown at 68 F were well developed and healthy. The seedlings on the far right that were

The question that arises is how does a turfgrass plant, or any plant, survive a lethal temperature?

exposed to the 1-hour of 131 F were killed. The seedlings in the middle, which received a 2-hour acclimation temperature before the 131 F lethal temperature, survived.

The question that arises is how does a turfgrass plant, or any plant, survive a lethal temperature?

Over the last 20+ years considerable research on how eukaryotic organisms respond to "heat shock." If we look at what happens during elevated temperatures, normal protein synthesis slows or stops. At the same time, as temperatures rise the production of specific proteins called heat shock proteins occur (Parsell and Linguist, 19993; Vierling, 1991). Heat shock proteins (HSP) are synthesized when supraoptimal temperatures (~98-104 F) are reached but below lethal temperatures.

The correlation between HSP formation and cellular resistance to thermal stress has led to the hypothesis that the accumulation of HSP's increases the thermal tolerance of organisms. It believed that HSP work as "molecular chaperones."

Molecular chaperones are proteins that bind to partially folded proteins promoting correct folding or preventing unfolding (Hendrick and Hartl, 1993; Waters et al., 1996). The unfolding or aggregation of pro-



Figure 2. Heat shock proteins are believed to provide thermal protection to plants by helping prevent normal proteins from unfolding during high temperature periods.

teins is a direct result of elevated temperatures, which leads to cellular death. It is hypothesized that these HSP proteins work like matrices to prevent the unfolding of normal proteins during periods of high temperature (Figure 2). The HSP are highly conserved among the eukaryotes.

The major classes of HSP are based on their molecular weight and are known as HSP100¹, HSP90, HSP70, HSP60 and small HSPs (17 to 30 kDa). Regarding plants in general, the small HSPs are the most important in thermal tolerance and the ones produced in the greatest abundance.

The small HSPs are divided into five classes partially based on cellular location:

- chloroplast,
- endoplasmic reticulum,
- two classes in the cytosol, and
- mitochondria.

HSP in turfgrasses

Over the years, there has been considerable research on turfgrass responses to high temperature stress. The majority of these studies have looked at whole plant morphological and physiological responses. Little research has been conducted into understanding the thermal tolerance response at the molecular level.



Figure 3. In vitro translation products of total RNA from "Accolade" (A) and "Caravelle" (C) perennial ryegrass seedlings. The arrows point to the presence of mRNA translating for heat proteins.

3

AGRONOMY

Little research has been conducted into understanding the thermal tolerance response at the molecular level. During a 10 year period, we studied the heat shock response and production of heat shock proteins in turgrasses (DiMascio et al., 1994;

Dimascio and Danneberger, 1990: DiMascio et al., 1989; Danneberger, et al., 1987; Sweeney et al., 1997).

In one study (Diemascio et al., 1994) we compared a heat-tolerant perennial ryegrass cultivar (Accolade) with a heat sensitive perennial ryegrass cultivar (Caravelle). We found that a broad range of HSP was induced in both cultivars at elevated temperatures (Figure 3). We did find that a detectable difference in the levels of heatinduced HSP 26 messenger RNA (mRNA)² occurred between the two cultivars. The heat tolerant cultivar produced more HSP26 mRNA than the sensitive cultivar. If a greater amount of mRNA is produced, then more protein is also produced.

The ramification from a breeding perspective is that if heat tolerant cultivars produce specific HSP in greater abundance than sensitive cultivars, then screening for these proteins may be beneficial.

Researchers at Mississippi State University have studied HSP response in creeping

TURFGRASS MANAGEMENT STRATEGIES

In summary, the heat shock response occurs in the following steps:

As temperatures rise above the optimum, normal protein synthesis begins to slow.

■ Once temperatures rise to a supraoptimal range (acclimation phase) a special group of proteins called heat shock proteins (HSP) form in the plant. The formation of HSP is associated with thermal tolerance.

Heat-tolerant cultivars tend to produce more messenger RNA (mRNA) for certain HSP than nontolerant cultivars.

■ Once the heat shock period passes, the HSP mRNA degrades and normal protein synthesis resumes. It maybe that heat-tolerant species or cultivars resume normal protein synthesis quicker after the heat shock period. This is believed to give a competitive advantage to heat-tolerant cultivars over nontolerant cultivars.

The insight gained from understanding the heat shock response is directly related to management strategies. Survival at high temperatures is dependent on the turfgrass plant successfully going through an acclimation phase.

If this phase is disrupted by a rapid rise in temperature, thermal tolerance will not be achieved. Thus, management practices that could interfere with the acclimation phase should be avoided. Some of these practices are:

■ Topdressings – should be avoided during midday. The application of topdressing during high temperatures can cause a rapid increase in plant temperatures by acting as a cover or blanket, in effect, heat shocking the plant.

■ Clippings – should be removed during periods of high temperature. Clipping left on a green or more likely a shortcut fairway turf, can act as a blanket or topdressing causing a rapid rise in temperature. Collecting or blowing the clippings off the turf can help reduce temperature buildup.

Mowing – height should be raised to help reduce the heat load of the turf.

■ Mechanical – practices such as brushing and verticutting should be done (if at all) will care during high temperature periods. The mechanical damage that can occur can cause rapid increase in the plant temperature.



bentgrass (Park et al. 1996; Park et al. 1997).

In their first study, they found the heat tolerant creeping bentgrass variants produced HSP25 while the nontolerant vari-

The ramification from a breeding perspective is that if heat tolerant cultivars produce specific HSP in greater abundance than sensitive cultivars, then screening for these proteins may be beneficial.

ants did not (Park et al. 1996). In a followup study looking at the recovery from heat shock, both the heat-tolerant and nontolerant variants produced the HSP25 but greater production of HSP25 mRNA in the heat-tolerant variants of creeping bentgrass was observed (Park et. al. 1997).

They also found that once the heat shock period passed, normal protein synthesis occurred sooner in the heat-tolerant variants than the nontolerant.

Park et al. (199) proposed that in the field HSP would be produced during midday during temperature stress, and then when temperatures decreased later in the day, the heat-tolerant creeping bentgrass would be able to resume normal protein synthesis sooner, and capture the remaining sunlight more efficiently.

NOTES

¹ 100 is expressed in kiloDalton (kDa) which is a molecular weight measurement. A Dalton is the unit of mass equivalent to the mass of a hydrogen atom $(1.66 \times 10 - 24 \text{ gram})$. Kilo-is the metric prefix meaning 10^3 .

²mRNA is a linear sequence of nucleotides (transcribed from DNA) that carries the protein-building instructions.

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5