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Effects of day and night temperature and lighting conditions on growth, nutritional status and tipburn of lettuce *(Lactuca Sativa L.* `Frillice´*)* cultivated in controlled environment.

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Plant Sciences- Plant production systems

Acknowledgment

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Abstract

Growth chamber experiments were conducted with *Lactuca sativa* crisphead lettuce:`Frillice´ growing in nutrient film technique (NFT). The main objective was to study energy-efficient cultivation strategies without reductions in growth and quality. To do this, the effects of temperature, including aerial day temperatures (DT) and night temperatures (NT) as well as leaf temperatures, on the growth, mineral content, and tipburn of lettuce cultivated with different lamp types were investigated. High-pressure sodium lamps (HPS) and two different light emitting diodes (LEDs) suitable for lettuce production (LED I and LED II) were combined with standard (DT/NT: 20/18°C) and varied temperature (DT/NT: 20/13°C) conditions at two different light intensities of 150 µmol/m^2/s and 300 µmol/m^2/s. The growth and morphology of the plants were recorded together with the tipburn data and leaf temperatures. Mineral analysis was also performed for Ca, Mg, K, Fe, and Mn to see if the changes in content could be linked to tipburn. Higher light intensity was more conducive for tipburn establishment. There was no difference in biomass in lettuces grown under HPS at standard or varied temperatures, but higher tipburn was seen in varied temperatures. Under varied temperatures, LED I (41% red, 13% far-red, and 10% blue) gave better plant growth than HPS but induced more tipburn. Further, the same LED I gave better growth and more tipburn in comparison with LED II (containing 17.23% red, 3.2% far-red, and 21.95% blue) under both standard and varied temperature conditions. The leaf temperature of plants exposed to LED was always lower than plants exposed to HPS (1.53-2.57°C) but were similar between the two LEDs.

In conclusion, the results showed that LED I improved growth compared with HPS and LED II for the growth of plants, but the risk for tipburn is higher to the level that the entire product can be discarded. Lamp types and light intensity are more important in the development of tipburn than air/leaf temperature but did not affect mineral composition very much. Furthermore, the best subset regression did not show high correlations between any of the cations and tipburn. Thus, overall LED I can replace HPS and potentially reduce the energy costs for lighting, but the light intensity should not be high to avoid light stress and tipburn. Though growth was not much hampered, and condensation was not observed, the lower night temperature (13°C) can still cause condensation in the shoot apical meristem and/or guttation due to a higher root pressure and then induce tipburn in greenhouses. Thus, a night temperature >13°C is recommended, together with good control of the air humidity during day and night.

Keywords: *lettuce, tipburn, leaf temperature, day/night temperature, LED, mineral*

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Abbreviations

ADT	Average daily temperature
ANOVA	Analysis of variance
Ca	Calcium
CEC	Cation exchange capacity
DLI	Daily light integral
DT	Day temperature
EC	Electrical conductivity
Fe	Iron
HPS	High-pressure sodium lamp
K	Potassium
LED	Light-emitting diode
LUR	Leaf unfolding rate
Mg	Magnesium
Mn	Manganese
N	Nitrogen
NFT	Nutrient film technique
NT	Night temperature
P	Phosphorus
PAR	Photosynthetic active radiation
Pers. com	Personal communication
PPFD	Photosynthetic photon flux density
PSI	Photosystem I
PSII	Photosystem II
QTL	Quantitative trait loci
RH	Relative humidity
ROS	Reactive oxygen species
SE	Standard error
St. Dev	Standard deviation
Temp	Temperature
UV	Ultraviolet

1. Introduction

Lettuces (*Lactuca sativa* L.) are the most common leafy vegetables worldwide, belonging to the family *Asteraceae*. Originated anciently as a salad crop from wild winter annuals in the Mediterranean basins of southern and eastern Europe, now the crop is cosmopolitan with wide morphological and physiological variations in due course of cultivation by human civilization (Subbarao, 1998; Thompson et al., 1979). Based on their head and leaf types mostly, lettuces are characterized into seven different types, viz. butterhead lettuce, crisphead/iceberg lettuce, cos/romaine lettuce, bunching/loose-leaf lettuce, stem/stalk lettuce, Latin/loosehead lettuce, and oilseed group (De Vries, 1997). It is a source of minerals like calcium (Ca), phosphorus (P), magnesium (Mg), potassium (K), iron (Fe), and vitamins like A, C, E, folates, fiber, and other bioactive compounds. It is an anti-diabetic, anti-inflammatory, cholesterol-lowering, and anti-cancerous food (Kim et al., 2016). Such nutritional value and awareness of its high health benefits contribute to increased consumption. It is commonly used in salads, sandwiches, and wraps. Among all, crisphead is the most popular lettuce. Unfortunately, it is the one with the lowest nutrient contents mentioned above due to its enclosed leaves in the head that limits the light penetration (Kim et al., 2016; Mou, 2009).

Among 106 countries producing lettuces, China, India, and the USA are the three leading countries in terms of yield and the gross area used for the production (Shatilov et al., 2019). A total of 27.3 million tons of lettuce and chicory were produced globally in more than 1.27 million hectares of land in 2020 and showed an increase in gross area and production from 2010-2020. Of this total production, 12.5% belongs to Europe (FAOSTAT, 2020). From 2010-2017, both the area and production increased in Norway. After that, a fluctuation occurred, plummeting the area to 880 hectares from 1000 hectares in 2017 and production to 21 thousand tons from 28 thousand tons in 2017.

Tipburn, a physiological disorder occurring in hydroponic lettuce production, is a common problem worldwide, including in Norway. Tipburn appears as necrosis on the outer leaves (outer tipburn) or on the young inner leaves (inner tipburn). This disorder has been a problem for many years, and it is still a problem with no definitive understanding that could help formulate a proper solution to it (Becker, 1971; Collier & Tibbitts, 1982). The necrosis of leaf edges renders the product unmarketable, causing substantial economic losses to commercial farmers. According to Benoit and Ceustermans (1986), tipburn could cause a loss of 50% of

the total production. Annually, 15%-20% estimated loss has been recorded in the greenhouse-produced crisphead salad in Norway (personal communication; Per Osmund Espedal). Based on the result of 2016, on average, this means out of 7,617,000 crisphead salads produced, 1,500,000 plants were lost, which accounts for a loss of NOK 19 million at an estimated average price of NOK 12.90 per salad (Grofondet, n.a.). With just more than 5% of tipburn, the entire product becomes unacceptable and prone to microbial rotting (Jenni & Hayes, 2010). This vast loss incurred by hard-working farmers could be improved by avoiding this tipburn damage. Compared to the lettuces growing in natural sunlight, those grown under supplementary lighting produce more biomass under a reduced production cycle while raising more susceptibility to tipburn incidence (Gaudreau et al., 1994). In Norway, where lettuce is produced the year round in greenhouses, supplementary lighting is needed throughout the winter, and tipburn can be a serious problem.

To cope with the tipburn problem, different solutions have been discussed in different literatures, like breeding robust cultivars, harvesting early before the symptoms are visible, and cultivating under an optimal climate. Hence, understanding how climate factors like temperature, light, and air humidity affect tipburn and controlling the climate accordingly is essential. The main objective of this thesis work was to test how varied air temperature treatments combined with different lamp types and light levels affect growth and incidence of tipburn. Saving energy is important in a controlled environment. In the winter months, heating costs are a limitation for greenhouse production in Northern latitudes. As such, lowering the air temperature at night can be an alternative to reduce heating costs (Morgan et al., 1979). Further novel lighting technology, like light-emitting diodes (LEDs), is more energy efficient than the traditional high-pressure sodium lamp (HPS). There is a need for more information on how LEDs with different spectral distributions in combination with different temperature regimes will affect growth, mineral status, and tipburn. Overall, the results from the experiments in this thesis will give indications if the altered temperature by lowering the temperature at night has any positive effect on the overall growth, tipburn, and mineral status of the crisphead lettuce. Furthermore, knowledge about the interaction between temperature regimes and lamp type will provide important information for future energy-saving strategies in the production of crisphead lettuce.

1.1 Lettuce cultivation: `Frillice´

Lettuces are cool temperate crops, and as stated by Tindall (1983), they can grow very well under a temperature range of 15-20°C. According to Jones Jr (2016), the optimal temperature for lettuces ranges from 17 to 28°C day temperature (DT) and 3 to 12°C night temperature (NT), while the pH, as given by Resh (2022), ranges from 5.8 to 6.5 and electrical conductivity (EC) of 1.5. In later years they have become the most common vegetable plant grown in hydroponics because of their higher growth capacity and nutrient uptake capacity (Kaiser & Ernst, 2016; Sharma et al., 2018).

`Frillice´ lettuce is a cross between crisphead/iceberg lettuce and curly endive (Seeds, 2022). It possesses crispy, curly, green-shiny leaves with a sweet taste that makes it popular among people. It is known to have a good tolerance to bolting and tipburn. As it can be grown both indoors and outdoors, it can be sown and cultivated all year-round indoors, while sowing can be done from the end of March to the end of August, and harvesting can be done around the end of May to the end of October outdoors (Bayer, 2022; Seeds, 2022). They can be grown in soil or hydroponics.

For growing lettuce in soil/outdoors, soil temperature between 7°C and 18°C and air temperature of 20°C is considered ideal. In a well-prepared seed bed, seeds are seeded at a depth of 1 cm, covered with soil, and then watered lightly. Sowing can be either broadcasting and thinning out later or row-row planting where 30-40 cm spacing is desired for crisphead lettuces. Fertilized and well-watered soil gives better yield. Crisphead lettuce can be harvested when the center becomes firm. Late harvesting makes them bitter and woody. Higher temperature initiates the bolting (Almanac, 2022).

1.1.1 Hydroponics

Derived from the Greek words `hydro´ means water and `ponos´ means labor, the term meaning water work in hydroponics, plants are grown in nutrient solutions together with the use of inert medium like gravel, rock wool, peat moss, vermiculite, coconut fiber, coir dust, sawdust, etc. to provide mechanical support. Nutrient film techniques (NFT), deep water culture, ebb and flow, wick, and drip are different types of hydroponics, and NFT is the most popular on-field (Sharma et al., 2018).

NFT consists of plastic or metal pipes ranging from 1-20 m in length with troughs of varying diameter depending on the crops grown (from 4-8 cm for lettuces to 15 cm for tomatoes or sweet peppers) slanted at a slope of 0.3-2%. In the troughs, a thin film of nutrient solution is supplied from the high end, which flows continuously downward due to the slope, and a drain is kept at the end. When seedlings are ready for transplanting, the small pots are directly placed in the troughs where their roots meet the nutrient film below. In a closed system, the drained nutrient solution is recycled and reused; in an open system, the drained solution is not reused (Van Os et al., 2019).

The technique is beneficial because, under fully automated conditions, it reduces labor otherwise required for field preparation or intercultural operations like weeding and fertilization. It saves 70 to 80% of water, fertilizers are readily available to the plant's roots, and climatic conditions are all controlled, so it has less risk of diseases and pests and has less production time yielding high-quality harvest (Sharma et al., 2018). Despite these benefits, there is a problem that the more suitable growing conditions are, the faster the plant grows, and the higher the chance of getting physiological disorders like tipburn (Uno et al., 2016). Siomos et al. (2001) found more tipburn disorders in hydroponics compared to traditional soil-based cultivation systems. Likewise, lettuce grown on perlite had higher Ca level in their leaves and lesser tipburn than lettuce grown in the deep flow technique (Assimakopoulou et al., 2013).

For hydroponics cultivation inside greenhouses in Norway, effective shading in summer months and heating in winter is required. Seedlings are first prepared in a separate germination room under very low lighting provided by HPS or fluorescent lamps (Fig 1). Seeds are placed in peat, perlite, or rock wool in small pots in seed trays. A germination temperature of 18-20°C, high humidity, and low light are maintained. Fertigation is done by the ebb and flow bench. Germination occurs within a week, and after that, light level and temperature are slightly increased. After another two to three weeks, they become ready for transplanting. In a system where NFT is used, transplanting is done by placing pots with seedlings directly in the holes of plastic pipes where their roots meet the nutrient solution flowing in the holes. For lettuces, the nutrient flow rate recommended is between 0.26 to 0.53 gallons per minute. Nutrients supplied are a mix of calcium nitrate with other nutrients coming from two different tanks that mix directly at the supplying line. The pH needs to be monitored continuously because hydroponics lacks buffering capacity of the soil. When plants mature, harvesting is done together with the roots or roots cut at the base (Kaiser & Ernst, 2016). In commercial

productions, moveable hydroponic structures are used so that the planting, spacing, and harvesting become easier to manage.

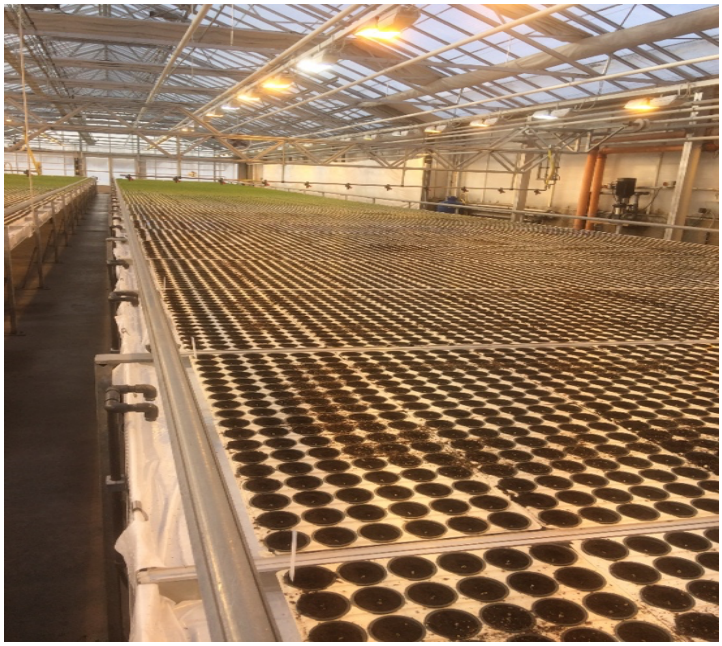


Figure 1: Pre-cultivation of lettuce from germination to seedling stage under HPS lamp with low light intensity. (Pic credit: Sissel Torre)

1.1.2 Growth stages of lettuce

The overall growth period of lettuce can be divided into three stages: germination, where soil temperature, moisture, and aeration are controlling factors; soil coverage, where light and air temperature play a significant role, and after that, the period till harvest, where total radiation becomes the most primary factor affecting yield (Bierhuizen et al., 1973).

1.2 Tipburn

Tipburn is a physiological disorder common in vegetable crops like lettuces, cabbage, brussels sprouts, chervil, chicory, Chinese cabbage, fennel, and potatoes. It is known to be caused by localized Ca deficiency, leading to necrosis in the edges of young, rapidly developing leaves (Kuo et al., 1981; Olle & Bender, 2009; Saure, 1998). Low Ca levels have been registered in lettuce leaves with necrosis (Barta & Tibbitts, 2000; Sago, 2016). The Ca deficiency seen in these leaves may not necessarily be because of low Ca content in their growing system or nutrient medium. Despite being sufficient in the soil or nutrient medium, localized Ca inadequacy has been observed in tipburn leaves (Kirkby & Pilbeam, 1984). Under such conditions, adding Ca salts to the tipburn leaves had shown positive results that prove the deficiency seen in the leaves is due to the imbalanced distribution of Ca within the plant cells rather than a deficiency in the growing medium (Mason & Guttridge, 1974).

Calcium is a phloem immobile mineral and gets transported only via the transpiration route from roots to the shoots with water through the xylem vessel (Kirkby & Pilbeam, 1984; Ziegler, 1975). Within the plant's system, transpiration pull and the cation exchange capacity (CEC) of the rhizosphere, root, stem, and shoot together are responsible for the absorption and transportation of Ca (Kumar et al., 2015). As a direct effect, the part that transpires more gets more Ca while the part that transpires less gets less Ca, creating an unequal Ca distribution (Busse & Palta, 2006).

The young inner leaves transpire less than the more mature exterior leaves in lettuces because young inner leaves are mostly enclosed, and they develop a humid microclimate. As a result, they accumulate less Ca and become more susceptible to tipburn. In an experiment where enclosed and exposed lettuce heads were compared, 53% inner tipburn was reported in enclosed heads and 1% in exposed heads (Barta & Tibbitts, 1986). In addition, xylem vessels are poorly developed in the young leaves, incapable of supplying enough Ca, exacerbating the situation (Barta, 1989). Such tipburn occurring in the inner leaves is called inner tipburn; necrosis in the outer leaves is called outer tipburn (Dimsey, 2010 in Knoop 2019).

Ca has a vital role in cell structure and function. Lack of Ca disturbs cell wall integrity and membrane structure, making cells weak and prone to damage (Sanders et al., 2002; White & Broadley, 2003). According to Cox et al. (1976), the tipburn event is affected by the growth rate of plants. The faster the plants grow, the more the requirement for Ca, and the slower the growth rate, the less the requirement, which creates unbalanced Ca distribution. Barta and Tibbitts (2000) found lesser Ca levels in the enclosed head and higher tipburn as the head enlarged in butterhead lettuce.

While inner tipburn is more related to the Ca disbalance, the causes for outer tipburn are not so clear. Outer tipburn is seen first as necrosis in leaf margins where vessels end in hydathodes. Under low humidity, much water is transpired and may lead to salt accumulation in the tips and cause cellular damage. These conditions are more severe when the nutrient solution’s EC is high. Since butterhead lettuce growing in outer rows of NFT exposed to higher airflow easily got an outer tipburn excessive airflow can also be one of the causes of outer tipburn (Mattson, 2015).

In all cases, the tip becomes necrotic first, and then the entire leaf rots with rotting of all interior leaves in severe cases. The necrosis results from the collapse of the epidermal and mesophyll cells between the vascular bundles exacerbated by the crushing of the adjacent cells due to hypertrophy of parenchyma cells and blockage of xylem vessels as observed in Ca deficit cells (Struckmeyer & Tibbitts, 1965). The collapses are from the increased latex pressure in laticifers walls, which then ruptures and releases latex in the surrounding cells, as observed in tipburn-affected lettuce leaves (Misaghi & Grogan, 1978). Such ruptures are more likely to occur during the early phase of rapid leaf development, making young leaves susceptible to tipburn (Tibitts et al., 1965).

Saure (1998) adds that tipburn could also be a stress-associated disorder when stress tolerance is exceeded. Exposure to mild stress from an early stage could increase their stress tolerance to tipburn. Tipburn, however, is not only limited to stressed plants. Stress-free plants are luxuriantly growing. They have a high content of active gibberellins, which interferes with the Ca^{++} transport to actively growing tissues, making their cell membranes more permeable and prone to damage, causing tipburn.

A positive correlation has been established between the tipburn severity and the irradiation sum, head fresh weight, temperature, day length, and maximum sum of daily irradiation. Among all, irradiation sum was the main factor in the tipburn severity when all other factors were statistically excluded. This means that light stress alone has a significant effect which indicates that other factors only have magnifying roles (Wissemeier & Zühlke, 2002). Higher temperature and higher light photosynthetic photon flux density (PPFD) are well-known factors that increase the tipburn of lettuce and their growth rate and yield. But blowing air directly into their growing meristem has eliminated tipburn in light as high as 1000 $\mu mol/m^2/s$ and temperatures of 25°C to 30°C giving four times the yield than under standard greenhouse conditions (Frantz et al., 2004). Multi fan system could be used inside lettuce cultivation to improve the horizontal airflow in the plant canopy and produce tipburn-free plants (Ahmed et al., 2020).

Relative humidity (RH) is another crucial factor affecting tipburn. Higher humidity of 82% gave more production but also more tipburn in cabbage, while the plants with 52% showed no tipburn at all (Palzkill et al., 1980). In young tomato leaves, 95% RH also gave more growth but had less Ca than those grown at 50% RH (Armstrong & Kirkby, 1979). However, lowering

the daytime humidity and increasing the night-time humidity delay or reduce tipburn. Such conditions create root pressure, which drives the uptake of water and Ca and gives an equal distribution to the newly expanding leaves (Collier & Tibbitts, 1984; Wiebe et al., 1977). High RH (95%-100%) combined with lower EC (1.5 dS/m) during night-time caused zero tipburn in Chinese cabbage; transpiration stopped, and positive root pressure was created that supplied the uptake evenly to the less transpiring parts (Van Berkel, 1988). Bradfield and Guttridge (1979) also found similar results in strawberries. Moreover, even at a normal humidity range, less tipburn without any effect on the plant's growth rate could be obtained by supplying air all day into the inner leaves of lettuce. There was increased transpiration along with associated Ca uptake from the root (Goto & Takakura, 1992).

The severity of tipburn varies with the cultivars used, the environmental conditions they are grown on, and their interactions. This creates a possibility of breeding lettuce cultivars' resistance to tipburn based on their morphological features and natural selection against the tipburn (Jenni & Hayes, 2010). Morphologically different cultivars have different patterns of internal uptake of Ca that may have resulted in this cultivar's difference in tipburn symptoms (Olle & Bender, 2009).

In research on crisphead lettuce, three significant quantitative trait loci (QTL) were found for tipburn incidence and severity associated with pleiotropic effects on head type, stem length, head firmness, and ribbiness. One QTL, qTPB5.2, was highlighted to bring about 38-70% variation in tipburn incidence rendering it a candidate gene in marker-assisted selection for tipburn resistance breeding (Jenni et al., 2013). Similarly, two significant QTLs were identified in the cv. Salinas in linkage groups 1 and 5 which also have pleiotropic effects with leaf crinkliness. Still, they could be a potential gene in tipburn resistance breeding via a marker-assisted selection field (Macias-Gonzalez et al., 2019).

Altogether, this Ca deficiency-led disorder is exacerbated by environmental factors like high temperature, high light intensity and duration, higher EC, reduced transpiration due to less air movement inside the enclosed head with growing leaves, and high relative humidity (RH) during the day. Further, when all factors are suitable, a faster growth rate also leads to tipburn. As explained above, tipburn is connected to the transpiration process and Ca distribution and any environmental factor that changes the transpiration rate of plants.

1.3 Role of Temperature in plant growth

Temperature is a major factor affecting the growth and development of the plants, having more influence on the latter. The transition of the plant from seed to seedling, to mature young vegetative stage, and from vegetative to reproductive stage, all depends on an optimum temperature range. There always exists an optimal, sub-optimal, and supra-optimal temperature for any physiological processes occurring inside the plant. The optimal temperature range always gives higher quality and quantity of produce. Both the sub-optimal and supra-optimal temperatures bring negative impacts to the plants. Any production inside the greenhouse provides an efficient way of managing this temperature at the optimal range along with other climatic factors as per the need of a specific crop.

All the temperature-dependent changes plants undergo in their life cycle are either from altered biochemical reactions in various metabolic processes or from developing water stress in plants by altering physical processes like transpiration rates. Biochemical reactions include the enzymatic reactions that get doubled with every 10°C rise in temperature. Bringing about the change in biochemical reactions, temperature changes plant photosynthesis which is mostly a light-dependent phenomenon (Downs, 2012). The temperature range between 10°C to 35°C is mostly favorable for most crops, while the temperature outside this limit damages the photosynthetic apparatus of the plants and disturbs their system cohesion. The plants adapted to cool climatic conditions can give a higher photosynthetic rate at low leaf temperature, while the plants grown in deserts can maintain so at higher leaf temperatures which can be seen in figure 2 below, taken directly from (Berry & Bjorkman, 1980).

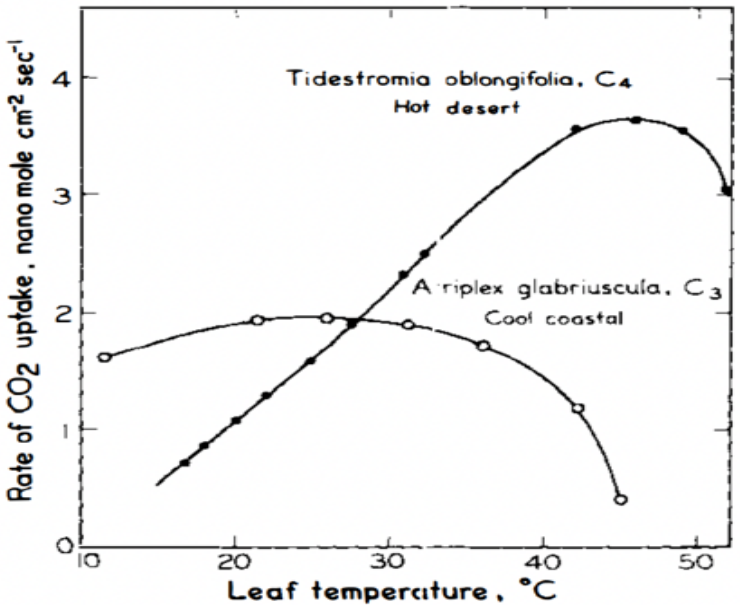


Figure 2: Comparison of the temperature dependency of photosynthesis by whole plants of Tidestromia oblongifolia during the summer in Death Valley, California, and Atriplex glabriuscula, grown under a temperature regime simulating that of its native coastal habitat (Berry & Bjorkman, 1980).

Temperature also influences the morphology of plants. Plants grown under higher temperatures have elongated stems, thinner leaves, and less dry matter (Downs, 2012). Friend and Pomeroy (1970) found the increase in leaf length, epidermal cell number, and length with an increase in temperature in spring wheat grown from 10°C to 30°C. The difference in the day and night temperature is called DIF, and this has a strong morphological effect on plants. Positive DIF is when DT is higher than NT, and its opposite is called negative DIF, while no difference in DT and NT is called zero DIF. Positive DIF is known to induce increased stem elongation and plant height, leaf expansion, development of more upright leaves, and high chlorophyll content resulting in greener plants. The stronger the positive DIF during the rapid growth phase, the more increased plant height in determinant crops (Myster & Moe, 1995). In sweet pepper, plant height and leaf area were also affected by DIF. However, the 24 hr. mean temperature showed relatively greater effects than any DIFs considered in its vegetative growth. The number of leaves and fresh weight of plants were affected only by mean temperature. Moreover, no effects of light on the optimum vegetative growth of sweet pepper were found (Bakker & Van Uffelen, 1988). Similarly, Moe and Heins (1989) reported that it is the average daily temperature (ADT) that affects the leaf unfolding rates (LURs) and not the DIF. Similar results have been found in the case of the tomato (De Koning, 1988) and the cucumber (Slack & Hand, 1983). Light quality, light intensity, and photoperiod interact with this DIF.

In terms of energy used in greenhouses, the lowered night temperature below the optimal level is further beneficial in saving the greenhouse energy without compromising much of the growth and development of plants; the low-temperature effect during the cold night gets compensated by the accelerated physiological processes during the day (Gent et al., 1979). Also, the symptoms of low night temperature in peppers' vegetative and reproductive growth were successfully prevented by exposing them to higher day temperatures (Pressman et al., 2006). These studies confirm that the average 24-hour daily temperature plays a significant role in plant growth and development rather than the specific day and night temperatures.

1.3.1 Dynamic climate control in greenhouses

The term dynamic temperature control further justifies the 24-hour ADT concept to be more accurate and effective inside the greenhouse to save the greenhouse energy and optimize plant growth and yield. It is a temperature integration procedure based on natural irradiance levels. Here, the temperature is allowed to fluctuate within a specific range in relation to the irradiance level. Inside the greenhouse, higher temperatures can be maintained at higher light irradiance, and CO_2 can be added to enhance their benefits further and lower the temperature during the night, which altogether balances the daily temperature requirement of the crop and saves energy from 20% to 30% (Aaslyng et al., 2003). It is the ADT that is of utmost importance than the fluctuations in temperature within a given range for the growth and development of plants (Hurd & Graves, 1983). Using this strategy has saved a significant amount of energy in the greenhouse production of ornamentals, bell peppers, and *Hibiscus rosa-sinensis* with no production differences or increased dry matter accumulation with appropriate lower night temperatures (Lund et al., 2006; Ottosen et al., 2003; Ottosen et al., 2004). However, there is a need to devise a crop photosynthesis model specific to each crop to exploit every benefit of dynamic climate control (Körner, 2003). The dynamic climate control based on the crop temperature instead of greenhouse air temperature can further save more energy and is more effective as crop temperature is the one that influences the crops (Körner et al., 2007).

1.3.2 Leaf temperature

Leaf temperature is not the same as air temperature; all the effects seen on plants are from the changes in their leaf temperature rather than from the ambient air temperature. Further, the whole plant does not have a uniform temperature; parts exposed to light or shade have different temperatures; angles from the light source also alter its temperature (Downs, 2012). Even the

temperature within a single leaf at different points differs. Lambers et al. (1998) explained the leaf's heat energy balance that affects the leaf temperature, as shown in figure 3 below.

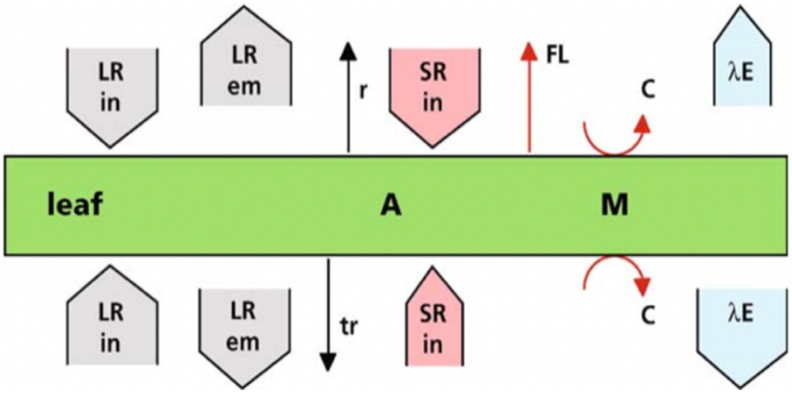


Figure 3: Diagrammatic representation of all the components in the energy balance of a leaf. It consists of short-wave radiation (SR), long wave radiation (LR), both incident(in) and emitted (em), convective heat transfer(C), and evaporative heat loss(λE). Reflection(r), transmission(tr), and fluorescent emission (FL) are only given for SR incident on the upper side of the leaf. A and M are CO_2 assimilation and heat heat-producing metabolic processes, respectively.

The light energy that falls on the plant is absorbed, reflected, or transmitted. Some of the absorbed light is lost in the form of heat. It absorbs the short-wave radiation and reflects the long wave while absorbing some long waves from the surrounding objects and sky. Convective heat loss occurs when the air temperature is less than the leaf temperature. Transpiration also causes heat losses. And then some metabolic processes release some heat and other such consumes it. This heat gain and loss process remains balanced when the air temperature rises under normal conditions. Leaf temperature is altered if any changes occur in the above heat balance processes. Plant heat loss processes can make the leaf temperature vary by 10°C more or less than the ambient air temperature. So, the air temperature of the whole environment only estimates the plant temperature, which in fact, is more dependent on the microclimate. In the Amazon, 10°C higher leaf temperatures than air temperature was obtained which supports that the microclimate of the plants is more critical, as that deviates leaf temperature from the air temperature (Doughty & Goulden, 2008). Maes and Steppe (2012) also showed that apart from the air temperature, radiation, wind speed, relative humidity, soil conditions like its type and water content, and canopy features like morphology, height, density, etc., change the plant and leaf temperature.

In a closed room or in greenhouses, the leaf temperature further matters more because of the use of artificial lamps and a closed, controlled environment. The amount of heat energy the plant receives differs based on the lamp type used. This is mainly because of the portion of the non-effective spectrum emitted by the lamps. The part of the spectrum that is absorbed by the

plants, but is not utilized, only heats them up. The leaf temperature will exceed the ideal limit if the air temperature is not kept cool. In addition, there are infrared radiations absorbed by the surrounding objects and trapped inside, warming the room even after the lamp is turned off till all the energy is lost. Thus, under lamps with non-effective spectrums, these things must be kept in mind before fixing the ambient air temperature. LED lights with the most effective photosynthetic active radiation (PAR) always heat the leaf much less than the HPS lamps. Even the LED with a non-efficient light spectrum like that in white LED with more green and yellow spectrum heat the leaves more than the LED with an effective spectrum like blue and red (Dannehl et al., 2021; Fender, 2017). As leaf temperature varies at different parts and points, this is further exacerbated by the distance to lamp type and air movement inside the closed chambers. Under higher leaf temperatures, maintenance respiration exceeds the growth respiration that consumes much plant energy and hampers leaf expansion as the plant spends more energy on cooling itself by transpiration, reducing the cell pressure required for cell expansion. Altogether these reduce plant growth and yield (Batts & Burgner, 2021).

1.3.3 Temperature and lettuce

Lettuces, well-known as cool temperate crops, have a high photosynthetic capacity under lower temperatures and grow well around 20°C. In lettuce, it has been found that temperatures lower than 13°C drastically reduce plant growth and nitrogen (N) uptake (Manrique, 1993). Jie and Kong (1998) found 50% lower photosynthetic capacity and productivity under higher ambient air temperature due to photo-inhibition compared to shoots maintained at higher air temperature with roots at a lower temperature of 20°C; this indicates that along with the aerial temperature, root temperature is equally important for the quantity and quality of lettuces. They also found that higher aerial temperature and higher irradiances maintain higher leaf temperature that happens to be at midday than in the morning and evening. Moreover, temperature also affects the composition of pigments in plants. In lettuce, low temperature increases the number of antioxidants, phenylalanine, peroxidase, and polyphenol oxidase, which play significant roles in plant defense against abiotic stresses (Boo et al., 2011). Higher temperature increases the nitrate content in lettuces, a disadvantage to lettuce growers (Richardson & Hardgrave, 1992).

1.3.4 Temperature and tipburn

Given that many modern lettuce cultivars are primarily adapted to cultivation in cool climates, their exposure to high temperatures during their vegetative phase can cause tipburn. Although genetic differences in cultivars exist, the incidence of tipburn increases with increasing temperature and light intensities, along with the improvement in growth characteristics. Nonetheless, a higher temperature decreases the head formation in crisphead lettuce. In contrast, even in higher light intensity conditions, lower temperatures than standard improves the quality of the lettuce in greenhouses (Lee et al., 2019).

If it is a tipburn-sensitive cultivar, then even the lower temperatures (18°C) can cause tipburn equivalent to that of higher temperature (25°C). Instead, a supply of stable air flow for 24 hours effectively reduced tipburn than the lower air temperatures (Lee et al., 2013). This would create a lower difference in Ca content between plants' inner and outer leaves by causing increased transpiration in the inner leaves. Knoop (2019) found no effect of temperature in tipburn, not even when the effect was compounded with elevated RH.

In Chinese cabbage and chervil plants, higher temperatures gave higher tipburn and Ca deficiencies (Borkowski & Szwonek, 1993; Kleemann, 2001). In addition to tipburn, other physiological disorders, like bolting and ribbiness in the lettuce, are also associated with higher temperature conditions (Al-Said et al., 2018; Jenni, 2005). Ribbiness is more related to the genotype, while tipburn is influenced by both genotype and environmental interactions (Jenni & Yan, 2009). Air temperature always seems more important in influencing tipburn than the root temperature. Studies performed to see if root temperature plays any role in altering Ca uptake and subsequent tipburn reduction gave no positive outcome (Collier & Tibbitts, 1984; Wiebe et al., 1977).

As per the data from Sand (2022), the average maximum and minimum temperature for lettuce cultivation differ among the Norwegian greenhouses (Fig 4). This study was done to see if the temperature strategies differed among growers. Clearly, the average maximum temperature recorded in the greenhouses was in the range between 20°C and 23°C, while the minimum temperature was between 14°C to 18°C, as shown in Fig 4. Hence, different growers use different temperature strategies in their production, making it difficult to find recommendations on what to choose.

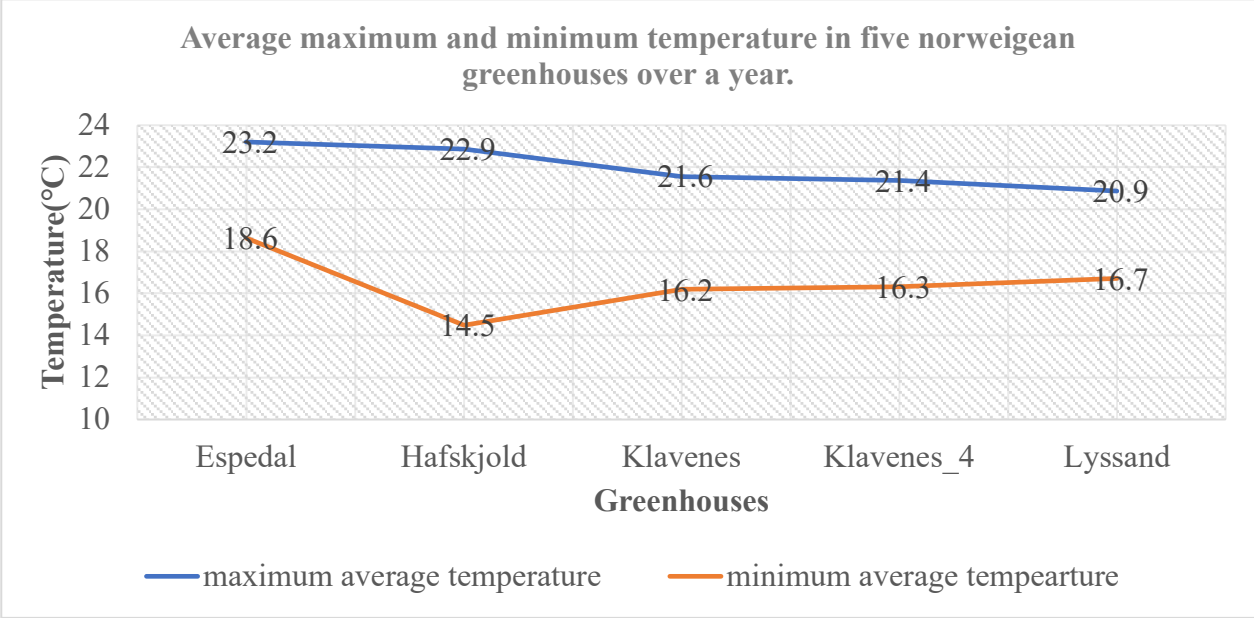


Figure 4: Average maximum and minimum temperature in five Norwegian greenhouses over a year (From A. Sand, unpublished).

1.4 Role of light on plant growth

Light is the ultimate energy source for plants, and the sun is its only natural source. It is known to govern various physiological processes in plants, being photosynthesis and photomorphogenesis the major ones (Wang et al., 2022). Plants get affected by light quality, quantity, direction, and periodicity (Hart, 2012). There are photoreceptors in plants that receive specific wavelengths of light and then bring about change in their growth and development processes (Franklin et al., 2004).

1.4.1 Light intensity

The plant can only use light for photosynthesis that falls under a visible spectrum of 400-700 nm, called photosynthetically active radiation (PAR) (Fig 5).

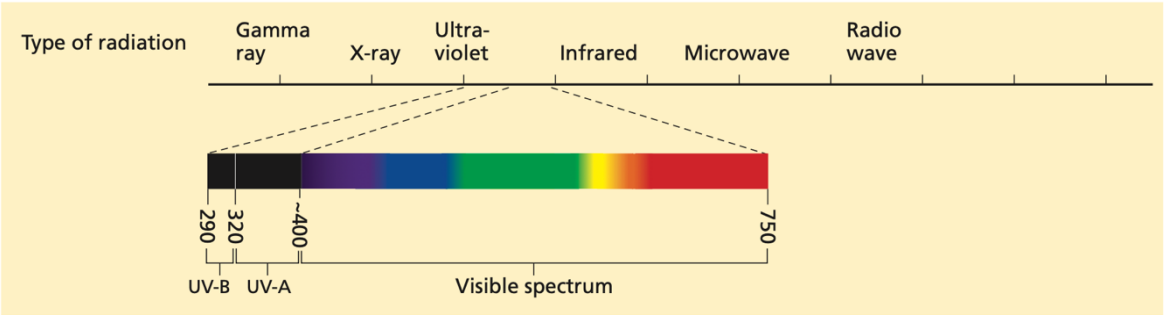


Figure 5:Visible spectrum of light (Taiz et al., 2015)

This PAR is expressed in terms of PPFD for easy quantification of its role in photosynthesis, as it is the number of photons that matters. There are different light compensation points for plants adapted to sun and shade; for sun plants, it is 10 to 20 μmol/m^2/s, and for shade plants, it is 1 to 5 μmol/m^2/s. Above this point, the photosynthesis is linear with the increased PPFD till it reaches the light saturation point, which is higher for the sun and lower for shade plants, with most plants saturating between 500 to 1000 μmol/m^2/s. The higher the light level within the limit, the higher the biomass production (Taiz et al., 2015). The lettuce plant falls under the crops having the ability to perform well under shaded conditions (Glenn et al., 1984; Marrou et al., 2013).

When plants receive too much light, their photosynthetic apparatus gets damaged; both the photosystem II (PSII) and photosystem I (PSI) complex gets damaged, but the frequency of PSI damage is less. The higher the light intensities, the lesser the capacity of the plants to repair their damage; as such, photosynthesis is reduced, which is called photoinhibition (Willey, 2018). This damage is caused by the production of highly reactive oxygen species (ROS) as a by-product of photosynthesis for which plants have developed photoprotective processes like adjusting their electron flow via cyclic electron flow or water-water flow, thermal dissipation of heat, scavenging of reactive oxygen species by antioxidants and scavenging enzymes and by chloroplasts movement (Niyogi, 1999; Willey, 2018).

However, optimum light is always necessary for plants to survive and function well. A reduction of 1% of the light will reduce production by 1% (Stanghellini et al., 2019). According to Knight and Mitchell (1983), growth chambers with higher light intensities give higher plant growth rates. It is not just the PPFD that has a role in plant growth. Any effects that the PPFD brings about depend on the amount of time the PPFD has been given. The amount of time any plant is exposed to the day and night period in 24 hours is called a photoperiod. Photoperiod works together with the light intensity and quality to drive plants' physiological and morphological processes. It is because PPFD and photoperiod combined give daily light integral (DLI), which is actually the one that affects plant yield and quality (Marcelis et al., 2005). Zhang et al. (2018) reported an increase in lettuce biomass with an increment in the DLI. Longer photoperiods with low intensity can give a similar yield as shorter photoperiods in higher intensity (Kang et al., 2013; Kelly et al., 2020). Loose-leaf lettuce doubled its yield under an increased photoperiod from 16 to 24 hours. Transplants grown at 50% increased radiation under lower PPFD for longer photoperiod has higher dry weight than transplants

grown at lower radiation from higher PPFD for short photoperiod in lettuce (Craker & Seibert, 1982; Koontz & Prince, 1986).

1.4.2 Light quality

Light quality deals with the spectral composition of the light and the spectral energy that supports the entire plant's growth and development. Plants have photoreceptors to sense this quality of light: phytochromes sense the red (R) to far-red (FR) ratio, cryptochromes and phototropins sense the ultraviolet (UV)-A/blue light, UVR8 sense the UV-B, and there are also ZTL/FKF1/LKP2 receptors. Altogether they govern photosynthesis, where blue and red wavelengths are the most effective, and photomorphogenesis, where blue, red, and far-red have major roles (Mawphlang & Kharshiing, 2017). Blue light reduces plant height, plant dry weight, and leaf area but gives darker green leaves and more lateral branches (Mortensen & Strømme, 1987). It gives more compact plants. However, the effect is reversed at 100% blue light (Hernández & Kubota, 2016). Also, the photosynthetic capacity increases with blue light from increased stomatal conductance, nitrogen content per area, chlorophyll per area, and leaf mass per unit area (Hogewoning et al., 2010). However, plant malformation occurs with only red light. Far-red triggers the shade avoidance response and increases biomass accumulation through more leaf expansion and increased shoot mass but decreases chlorophyll concentrations (Meng, Q. et al., 2019). It is actually the ratio of R: FR that triggers such a response in plants (Morgan et al., 1980). Low R: FR promotes stem elongation from internode elongation, petiole elongation, leaf expansion, hyponasty, accelerated flowering, increased allocation of nutrients to aerial parts, less root hair density, etc. (Demotes-Mainard et al., 2016).

1.4.3 Lamp types

Production inside the greenhouses depends on artificial lighting, mainly in the Nordic countries in winter when the natural light becomes limited. Different lamp types emit different spectra. However, including artificial lighting has increased the production of many crops. For example, Gaudreau et al. (1994) found increased lettuce biomass by 1.4 to 2.7 times and decreased production cycle by 25% when they were given additional supplementary lighting of 50-100 $\mu mol/m^2/s$ over a photoperiod of 16 to 24 hours in winter months as compared to natural light only. With the development of growth chambers and plant factories, artificial lamps have turned from additional lighting to the only source of lighting for plants. There also have been

advancements in the lamp types so that the high production demand of vegetable crops could be met with as low incurring electrical costs as possible.

The most common type of auxiliary light used in vegetable production is HPS. The low cost, high life span, high light emission, and electrical efficiency have made it popular. It converts 35-37% of electrical energy into PAR, and the rest is converted into heat that raises the greenhouse temperature and thus alters the climate. The crop temperature also rises, making it unsuitable for placing near the crop canopy (Dorais, 2003). For example, leaf temperatures of plants growing under HPS are higher (0.9-1.3°C) than in LED (Bergstrand et al., 2016). Its spectral composition has a significant amount of infrared, more green and yellow light, less blue, and low red to the far-red ratio (Dorais, 2003).

The recent lamp type drawing attention is LED. The introduction of LEDs has made it possible to incorporate just the necessary wavelengths, and the lack of infrared and UV radiation makes it possible to place it near plants and provide sufficient light without increasing crop temperature. Previously, high price and less efficiency had become a problem in its use in greenhouses; however, the current decline in price due to mass production and its increased brightness and quantum efficiency has increased its prospect of being a suitable and economical light source for plant cultivation inside greenhouses and plant factories (Watanabe, 2009). Although much is needed to be understood on the morphological and physiological effects of different LEDs on different crops, the most popular LEDs are red (660nm), blue (450nm), and far-red (730nm). LEDs are known to be 60% more efficient than HPS, have a narrow spectrum of monochromatic light with the feasibility of choice, low heat emission, very long-life span, less light pollution, and good safety characteristics, which altogether makes it an alternative to HPS (Morrow, 2008).

If correct wavelengths of light are combined at a proper light intensity, then the production under LED could be comparable to that of HPS (Currey & Lopez, 2013; Singh et al., 2015). Experiments have shown that red and blue light are the most effective in photosynthesis and promoting plant growth, while yellow and green suppress growth (Yang et al., 2017). Strawberry growth was best when 70% red was combined with 30% blue light (Nhut et al., 2003). Tomatoes grown under LED developed higher photosynthetic capacity than under HPS, with less production and quality differences between them (Dueck et al., 2011). The combination of HPS and LED gave a higher fruit yield in cucumber than under two lights alone

(Särkkä et al., 2017). Studies by Yanagi et al. (1996) showed that lettuce plants grown under red LEDs alone had more leaves and longer stems than plants grown under blue LEDs. Lettuce grown under additional far-red light gave more dry matter than those under additional red light due to an increase in leaf area (Li & Kubota, 2009). Similarly, a combination of blue, red, and far-red gave the highest dry matter in cucumber and tomato, while under blue and red only, plants were more compact, and dry matter was similar to that from HPS lighting (Meinen et al., 2012).

1.4.4 Light and tipburn

Higher light intensities and longer photoperiods are known to increase the incidence of tipburn (Gaudreau et al., 1994; Pressman et al., 1993). According to Sago (2016), with the increase in light intensities from 150 μmol/m^2/s to 300 μmol/m^2/s, there is an increase in tipburn. The outer leaves transpire more with an increase in light intensity and accumulate more Ca, while no such increase in inner leaves causes tipburn in them. Thibodeau and Minotti (1969) found no tipburn under low light intensity. Islam et al. (2004) found contrasting results in *Eustoma grandiflorum,* where high PPFD and longer photoperiods do not always cause tipburn, and lower intensity always does not necessarily prevent tipburn when using PPFDs of 60, 120, 180, 240 μmol/m^2/s. Under the same daylight intensities, repeating a shorter day-night cycle reduces the rate of tipburn (Goto & Takakura, 2003). Less tipburn was obtained when the 14-hour day and 10-hour night cycle was changed to 105 minutes of light and 75 minutes of the dark period. Under shorter cycles, the pressure in the laticifers is less, preventing cell rupturing and subsequent tipburn. Continuous light with higher light intensity increases the dry matter with the corresponding tipburn while the same continuous light at a lower light intensity (250 μmol/m^2/s) reduces its severity without affecting the growth (Oda et al., 1989). Compared to the HPS, white LED gives more outer tipburn while adding far-red reduces the tipburn (Knoop, 2019). He also found the presence of ROS in tipburn leaves and the amount being more under high light intensities.

1.5 Combined role of light and temperature in plant growth and tipburn

Among others, light and temperature are considered the major factors that provide radiant and thermal energy respectively to plants. Though they have their own way of affecting plants, the ultimate effect seen in the plants is always from the interaction between these two factors. It is hard to separate these and conclude the effect from only one. The thermal energy is responsible

for the developmental rate, while the radiant energy drives the photosynthesis obtained as plant dry weight at each developmental stage. Together they affect the plant quality. The higher the ratio between these two energies, the higher stem dry weight, specific leaf weight, and stem area have been found in *Euphorbia pulcherrima* (Liu & Heins, 1996).

At constant air temperatures under longer photoperiods, necrotic and chlorotic disorders have been seen in tomato leaves (Arthur et al., 1930). In contrast, low air temperature (Withrow & Withrow, 1949) or daily alteration of air temperature effectively prevented such disorders (Omura, 2001). Under alternating air temperature (DT/NT: 28/16°C), even under 24-hour photoperiod combined with lower PPFD (200 μmol/m^2/s), no physiological disorders like chlorosis and necrosis were seen in tomatoes (Ohyama et al., 2005a; Ohyama et al., 2005b).

Only under high light intensity and high CO_2 can gross photosynthesis be significantly affected by temperature. At higher temperatures, the rate of plant development, i.e., increase in the number of new leaves, is higher. In tomatoes, the low pulse temperature of 12°C and 15°C for 2 hours before the end of the photoperiod inhibits the incidence of chlorosis in the 20-hours photoperiod condition. High light with high temperature increases leaf widths while also increasing the incidence of tipburn, bolting, and puffy heads (Dorais, 2003).

Tipburn was reduced in the treatment of QRSL (where the light temperature was increased rapidly to 23°C and set as such in the first half up to 2 pm and then decreased by 1°C every two hours. in the second half till dark in 16 hr. photoperiod) and growth rate was increased. While it was increased in QDSL (where the light temperature was slowly increased by 1°C every two hours during the first half and then set as 23°C during the second half till dark) (Kumazaki, 2022). This gives some possibility of tipburn reduction by fluctuating temperatures in the light and dark periods.

1.6 Mineral ions and tipburn

Ca, Mg, and K are the essential macronutrients required by plants that remain in the ionic form. Fe and Manganese (Mn) are the micronutrients mainly involved in redox reactions (Taiz et al., 2015). Ca and tipburn have been studied extensively, but studies relating tipburn to other ions are scarce. The other ions may or may not have a direct role in tipburn, but the interaction among them can alter the uptake and distribution of Ca in the plant cells. According to Wallace

and Mueller (1980), Ca uptake and distribution is affected by the cations K^+, Mg^{++}, Na^+, $NH4^+$, and H^+. Cations have antagonistic relation to Ca, while anions decrease the severity of tipburn (Olle & Bender, 2009). Increased N concentration to its optimum level increased tipburn in field-grown lettuce, while the reduction of N reduced tipburn in NFT-grown chervil and parsley (Brumm & Schenk, 1992; Kleemann, 2018).

Higher Mg increases tipburn incidence. Lower K levels corresponded to lesser tipburn; with the increase in K levels, leaf Ca, Mg, and Mn decreased. Likewise, when Ca level was increased in the nutrient solution Mg and K levels were reduced in the tomato fruit (Paiva et al., 1998). Uptake and translocation of Ca and Mg are affected negatively by the higher levels of K in the soil (Jakobsen, 1993). More tipburn was on strawberry leaves grown under less Ca and high potassium soils (Chiu & Bould, 1976). A different result was obtained by Bres and Weston (1992), where different concentrations of K had no significant effect on the incidence of lettuce tipburn.

Mn is mainly transported via xylem (Pearson et al., 1996), and under increased Mn concentration, uptake of Ca was decreased (Juice et al., 2006). Mn can easily replace other divalent metal ions like Ca, Mg, Fe, Cu, and Zn and alter the uptake, distribution, enzyme activities, and use of other nutrients (Lavres Junior et al., 2010). An antagonistic effect on Ca, Mg, K, Fe, and Cu with an increased supply of Mn was found in the outer leaves of the Chinese cabbage (Lee et al., 2011). However, no effect on Ca was seen in hydroponic lettuce with increased Mn concentration, while there was a decrease in K, Mg, and Fe (Kleiber, 2014). Excess Mn in the tissues will cause a deficiency of Fe, leading to chlorosis, and later necrosis occurs under further accumulation and oxidation into manganese oxides (Fernando & Lynch, 2015). Mn toxicity affects the leaves based on maturity, causing chlorosis in young leaves and necrosis in mature leaves (Li et al., 2019). Concentrations higher than 100 mg/kg in plant tissues are considered toxic depending on the plants, whereas lettuce is considered to have higher resistance with tolerance above 1500 mg/kg dry matter (Howe et al., 2004). Fe and Mn have antioxidation roles, and they detoxify ROS through redox reactions (Sebastian & Prasad, 2015), while the excess can cause excessive production of ROS (Li et al., 2010). It also decreases stomatal conductance. Nonetheless, increased concentration of Mn in inner leaves has been found to reduce inner tipburn in lettuce (Kodua, 2022).

2. Objectives of the study

2.1 Main objective

The main objective of this thesis was to study energy-efficient cultivation strategies without reductions in the growth and quality of crisphead lettuce. To do this, the effects of temperature, including aerial day (DT) and night temperatures (NT) as well as leaf temperatures, on the growth, mineral content, and tipburn of lettuce cultivated with different lamp types were investigated.

2.2 Specific objectives

Exp I: To compare standard (DT/NT: 20/18°C) air temperature and low NT (DT/NT: 20/13°C) on the morphology, leaf temperature, tipburn, and mineral status of lettuces cultivated with HPS lamps.

Exp II: To compare HPS and LED in a temperature regime with low NT (DT/NT: 20/13°C) to study differences in morphology, leaf temperatures, tipburn, and mineral status of lettuces.

Exp III: To compare two different LEDs suitable for lettuce production at standard temperature (DT/NT: 20/18°C) on the morphology, leaf temperature, tipburn, and mineral status.

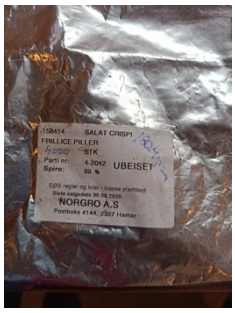
Exp IV: To compare two different LEDs suitable for lettuce production in a temperature regime with low NT (DT/NT: 20/13°C) to study differences in morphology, leaf temperatures, tipburn, and mineral status of lettuces.

3. Methods

3.1 Plant material preparation

Seeds (Norgro, Norway, Fig 6A) of Frillice lettuce (*Lactuca Sativa L. 'Frillice')* were seeded in biodegradable pots and kept in a seed tray (Fig 6C&D). The pots were filled with peat (Vekesttorv, Degernes Torvstrøfabrikk Norway, Fig 6B). Then seeds were sown at a depth of 1 cm, covered by peat, and finally irrigated with tap water. The seeds were clay coated to maintain a uniform shape so that machine seeding would be easier if it were to be done so. The peat contained mixed leca beads, which increases aeration in the potting substrates.

After seeding, the trays were kept in the dark room covered with a black cloth at about 15°C for three days and then transported to a greenhouse with supplementary lighting under HPS lamps supplied from Gavita (Gavita AS, Norway) for 16 hrs. daily at a PPFD of 150 µmol/m^2/s. The seedlings were allowed to grow until the four leaves stage (Fig 6G). In the beginning, the seed trays were covered by agryl white cloth to prevent them from excessive drying out (Fig 6E). They were produced at a temperature of 20°C and 70% RH, and irrigation was done using a nutrient solution of 1.5 mS/cm EC (Fig 6F).



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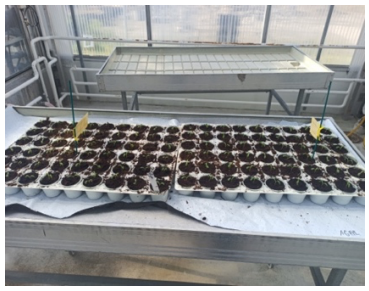

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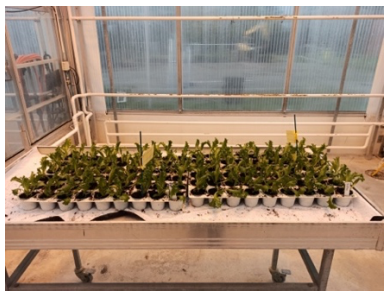

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Figure 6: A) Seeds of frillice lettuce; B) Peat sack; C) Peat compacted in biodegradable pots in seed tray; D) Seeds seeded; E) Trays covered by agryl in the greenhouse after bringing from the dark; F) Seeds germinated in the greenhouse; G) 4 leaf stage seedlings ready to be planted in the growth chamber.

3.2 Growth chambers and hydroponic setup

The seedlings were pre-cultivated in a greenhouse for two weeks. Then the seedlings were moved to enclosed growth chambers without any natural daylight. In the greenhouse and the chambers, a hanging climate sensor was connected to a Priva climate computer (Priva, Zijlweg, The Netherlands) containing a wet and dry thermometer to measure the air temperature and RH (Fig 7B&C). The humidity in the chambers was maintained at ±70%. The lighting in each chamber was set as per the experimental requirement, and the amount of light was measured by a Li-cor quantum sensor (LI-250, Light meter, LI-COR, USA, Fig 7A), which gives PPFD in µmol/m^2/s. A complete hydroponic system setup was placed inside each chamber (Fig 7D), and NFT was used in all the experiments. On a table, four gutters (Vefi AS, Norway, Fig 7E) were placed 25 cm apart, each 1.5 m long and 10 cm wide, with ten holes at 15 cm spacing in between. At one end of each gutter was a small hole on top to insert the hose pipe for irrigation, and the other end was open to let the drained water out. The gutters were kept slightly inclined towards the outlet for easy flow of drainage water. Two plastic trays were kept at the bottom to collect this drained water. Water for irrigation was kept below the gutters in a plastic bucket. A pump (Aquarium Systems Maxi-Jet 500, France) was fitted with a timer (müeller SC 28 11 pro, Germany) to force the water up to the gutters at a definitive time through the drip pipe (9A&B). Also, the drip pipe had a control knob to let the fixed amount of water into the gutters through the small hose pipes (Fig 9C).

A B C

D E

Figure 7: A) Li-Cor quantum sensor; B) Hanging PRIVA climate sensor; C) Inside of PRIVA climate sensor: dry thermometer above and wet thermometer below; D) Growth chamber setup showing gutters, irrigation water bucket to bottom right, drained water collecting trays to bottom left, hose pipes fitted in gutter holes at top; E) Gutters with seedlings.

3.3 Nutrient solution preparation and irrigation

A nutrient solution with electrical conductivity (EC) of 2 mS/cm and pH of 5 was used throughout every experiment. To prepare the nutrient solution, two 75-80 liters tanks were filled with 50 liters of tap water (Fig 8B) and then kept at room temperature. Two different stock solutions were used (Fig 8C), whose recipes are shown in Table 1. Both the stock solutions were stirred thoroughly with a bamboo stick before using them.

150 ml of each stock solution was poured into the water tank and stirred thoroughly (Fig 8D). The EC was measured using an EC meter (ScanGrow Conductivity meter, Denmark, Fig 8A). The process was then repeated until the EC of the water reached 2 mS/cm. Altogether approximately 750-800 ml of each stock solution was added to each tank. Every 2-3 days, the black plastic buckets inside the growth chamber were filled with this nutrient solution, then pumped up into the gutters via the water pump and hose pipes (Fig 9A & C).

Table 1: Stock solutions 1 and 2 and their content of fertilizers in 50 liters of water.

Stock solution 1		Stock solution 2	
Nutrients	Amount	Nutrients	Amount
Water	50 liters	Water	50 liters
Calcium nitrate	2.5 kg	Pioneer basic cucumber	3.125 kg
Potassium nitrate	0.625 kg	Pioneer Iron chelate, 6% EDDHA	0.025 kg
Calcium chloride	0.15 kg		

Also, a sample from the final solution with 2 EC was taken and sent to Eurofins Agro Testing Norway AS for analysis of nutrient content. The result obtained can be seen in Table 2.

Table 2: Actual amount of nutrients and micronutrients in the nutrient solution supplied to the lettuce with pH 5.

Cation's ppm (mg/l)						
NH_4	NH_4-N	K	Na	Ca	Mg	
1.8	1.4	282	32	148	29	
Anions ppm (mg/l)						
NO_3	NO_3-N	Cl	S	HCO3	P	
750	169	64	48	6.1	37	
Micronutrients ppb (µg/l)						
Fe	Mn	Zn	B	Cu	Mo	Si (ppm-mg/l)
1843	483	275	292	133	86	2.8

A) B) C) D)

Figure 8: A) EC meter; B) 75-80 liters water tank; C) Two different stock solutions; D) Stock solutions in measuring cups.

At moderate light intensity (150 µmol/m^2/s), the nutrient solution was supplied nine times during the day at the rate of 1 minute every 2 hrs. When the light level was increased to 300 µmol/m^2/s, instead of 1 minute 9 times, it was alternated 2 min every 2 hrs. The water frequency was the same for all experiments. The amount of water coming out of the hose pipes was set to 130 ml per watering for all gutters by adjusting their control knobs (Fig 9C). The entire irrigation schedule can be seen in Table 3.

Table 3: The irrigation schedule at two different light intensities: the water was given nine times a day throughout the experiment, where at 150 µmol/m^2/s, it was for 1 min every 2 hours, while at 300 µmol/m^2/s, it was 1 min for four times and 2 min for five times alternately.

Light intensity	Time of watering	8:55/56	11:05/06	13:05/06	15:05/06	17:05/06	19:05/06	21:05/06	23:05/06	12:55/56	No water period
150 µmol/m^2/s	Time period	1 min	1 min	1 min	1 min	1 min	1 min	1 min	1 min	1 min	1-8:54
300 µmol/m^2/s	Time period	2 min	1 min	2 min	1 min	2 min	1 min	2 min	1 min	2 min	1-8:54

A) B) C)

Figure 9: A) Water pump; B) Water timer; C) Water hose pipes inserted into gutter holes.

3.4 Light treatments

Inside the growth chambers, the plants were exposed to artificial lighting from 9:00 AM till 1:00 AM. There were 16 hours of daylight (9:00 AM to 1:00 AM) and eight hours of darkness (1:00 AM to 9:00 AM). Four different experiments were run, and the following light conditions were given:

Experiment I: Four hundred watts of HPS lamps (Gavita AS, Norway) were used in both chambers (Fig 11A). The initial PPFD was 150 µmol/m^2/s for about three weeks, and in the fourth week, the light level was increased to 300 µmol/m^2/s.

Experiment II: The same HPS lamps were used in one of the chambers; in the other, LED I (Evolys, Norway) was used. LED I had a combination of white LED and additional FR in it (Fig. 11B&C). The condition of PPFD was the same as in experiment I.

Experiment III and IV: Two different LEDs (Evolys, Norway) were used. One was the same LED I used in experiment II. The other used LED II (Fig 11D), which used only white LED. The condition of PPFD was the same as earlier. The R/FR sensor (Skye Instruments, The UK, Fig 10) measured the R/FR ratio, which was 2.89 for LED I and 8.3 for LED II.

Figure 10: R/FR sensor

A B C D

Figure 11: Figures showing experimental chambers under different lights: A) Chamber with HPS I; B) LED I, ceiling view; C) Chamber with LED I; D) Chamber with LED II.

Spectral composition of light

The spectral composition for HPS was taken from Knoop (2019) as the same HPS lamps were used in both experimental works, which had a red-far ratio of 3.8 stated by Gavita, Norway and can be seen in Fig 14. For LED I and LED II, the spectral composition was measured with Spectra Pen mini (https://handheld.psi.cz/products/spectrapen-mini/), and the spectrum can be seen in Fig 12 and Fig 13. The blue, red, and far-red percentages were calculated for two LEDs with wavelengths from 400-780 nm to incorporate FR under the same calculation (Table 4).

LED I contained 10% blue, 41% red, and 13% far-red. LED II included 22% blue, 17% red, and 3% far-red and had a higher percentage of green and yellow than LED I.

Table 4: Percentage of different wavelengths of lights in LED I and LED II.

Light quality	Blue	Green	Yellow	Orange	Red	Far-red
LED I	10%	20%	6.3%	9.1%	41%	13%
LED II	21.95%	37%	10.92%	9.4%	17.23%	3.2%

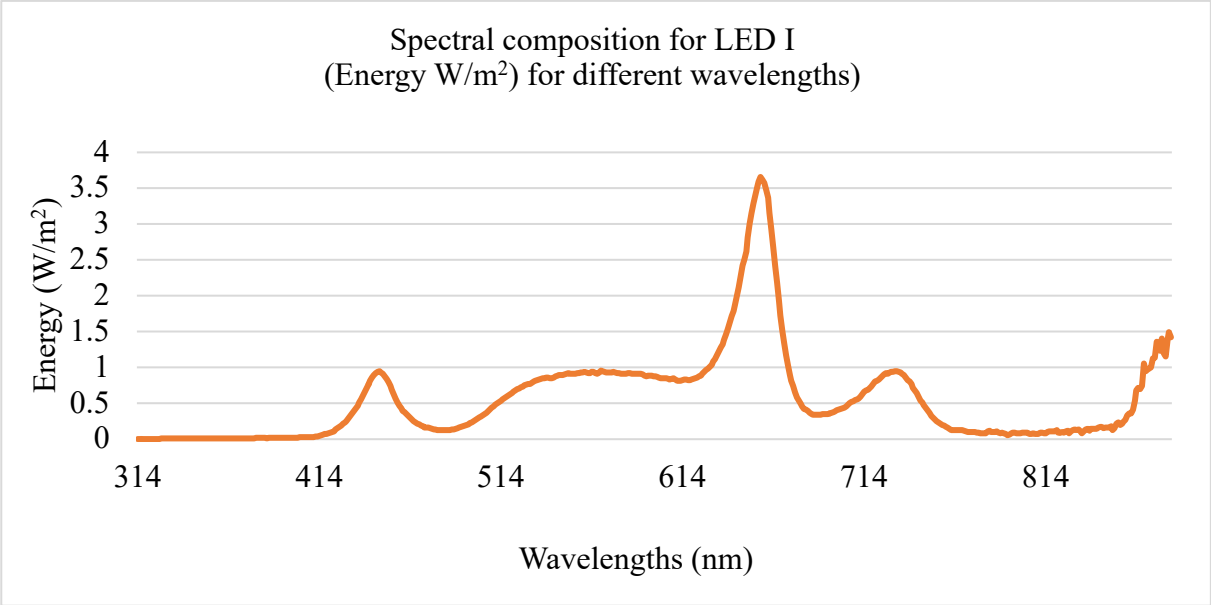


Figure 12: Light spectral composition under LED I (Evolys, Norway) used in growth chambers in experiments II, III, and IV.

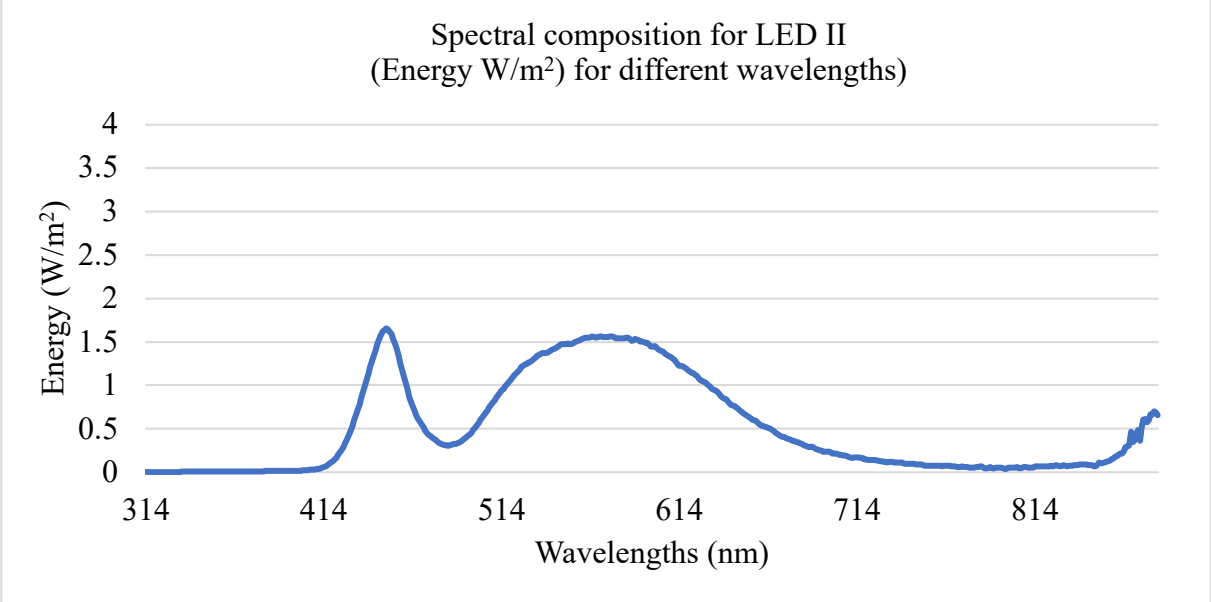


Figure 13: Light Spectral composition under LED II (Evolys, Norway) used in growth chambers in experiments III and IV.

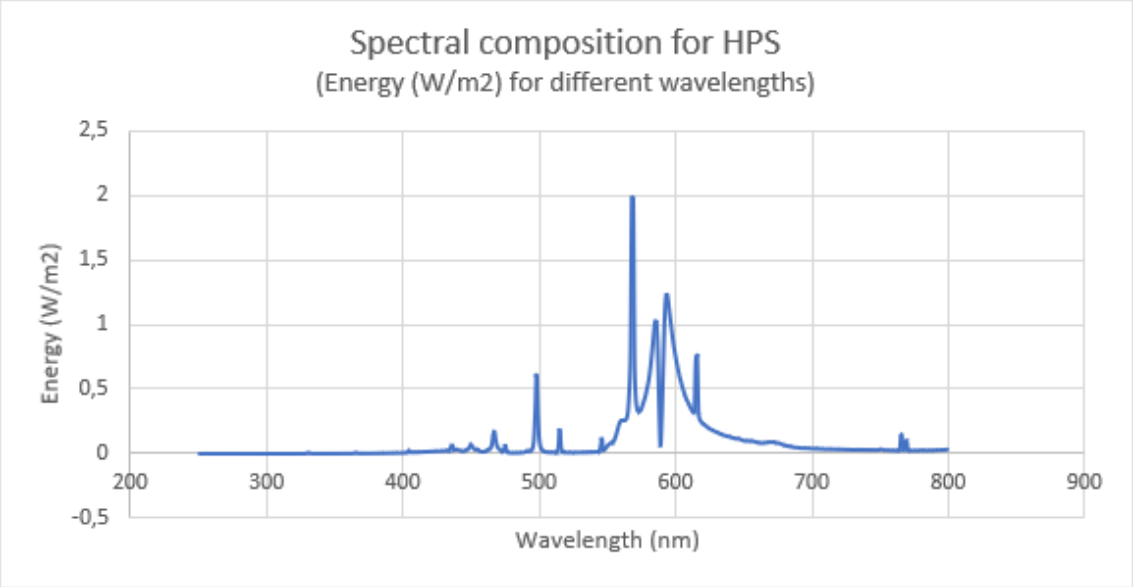


Figure 14: Light Spectral composition for 400 W HPS (Gavita Norway) used in the greenhouse compartment and the growth chambers in experiments I and II (M Knoop, 2019).

3.5 Temperature treatments

The temperature of the control treatment in all experiments was 20°C during the photoperiod and 18°C during darkness which is the "Standard temperature" used by most of the commercial lettuce growers (pers. com. P.O. Espedal). Night (darkness) was from 1:00 am to 9:00 am, and the temperature in the growth chamber was lowered to 18°C. Then as the light period started from 9:00 am, the temperature was directly increased to 20°C and then stabilized at 20°C throughout the whole day period till 1:00 am again in the night (Fig 15).

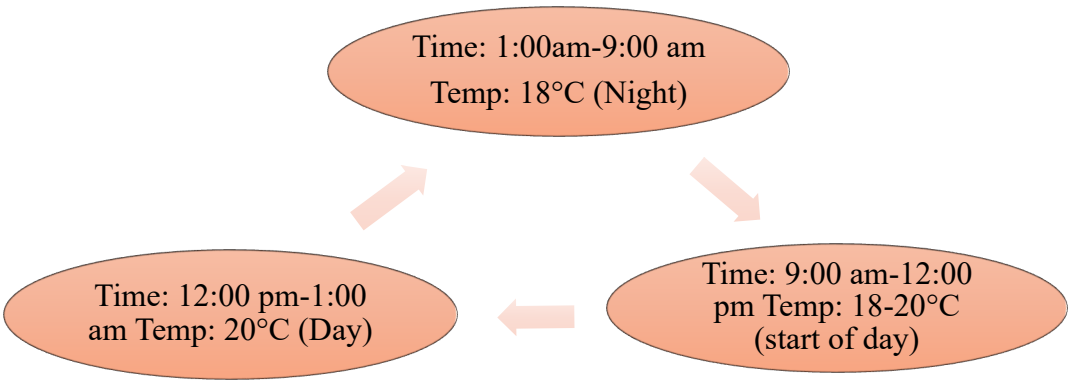


Figure 15: Standard temperature treatment (Temp: temperature)

For the temperature variation treatment called as varied temperature, the NT was controlled at 12-14°C. Then, as the light was switched on, the temperature was increased to 16°C during the

first hour. Then the temperature was increased to 18°C during the second hour, and during the third hour, it was increased to 20°C, then it was stabilized at the same temperature throughout the whole day until 1:00 am. Then the light was turned off, and the temperature was dropped again to 12-14°C. Before temperature variation started, the plants were all grown at 20°C DT and 18°C NT for two weeks. The temperature setup is illustrated in Fig 16.

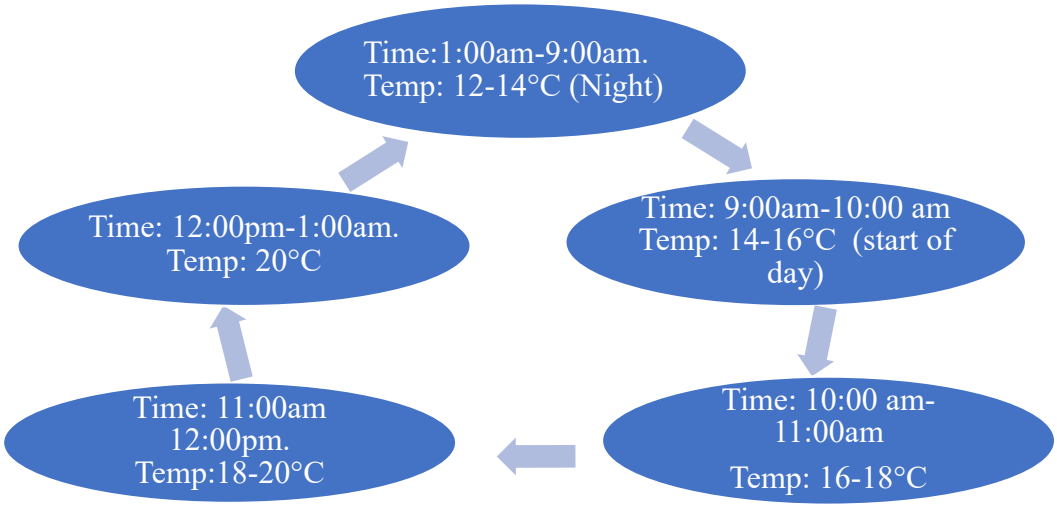


Figure 16: Varied temperature treatment (Temp: temperature)

Average daily temperature (ADT)

The average daily temperature was calculated by multiplying the maintained temperature by the number of hours it was kept so, and then adding all such values over 24-hour period and finally dividing by 24 hours (Runkle, 2008) (Table 5).

Table 5: ADT for the two temperature treatments used in all experiments. All plants in both chambers were exposed to "Standard temperature" during the first two weeks of cultivation in the chambers. Later, one chamber was kept at standard temperature, and one was changed to "Varied temperature."

Treatments	Calculations	ADT
"Standard temperature"	(20*16+18*8) / 24	19.33°C
"Varied temperature"	(13*8+16*1+18*1+20*1+ 20*13) / 24	17.41°C (approx. 13°C taken as night temperature is between 12-14°C)

3.6 Measurement of leaf temperature:

Figure 17: Infrared thermometer

The leaf temperature was measured using an infrared thermometer (Fluke 62 Max IR thermometer, the USA, Fig 17). It was held firmly and then pointed at each plant's outer and inner leaf, and the temperature it displayed was noted down. The innermost six leaves were considered the inner leaves, while the outer ones were all the remaining ones. For ease, while pointing the thermometer, it was usually 5th, 6th, or 7th leaves taken as outer while the innermost region was taken as inner leaves. The first leaf temperature was taken at 8:50 am, just before the light was turned on to get an overview of the leaf temperature at night, and after that every half an hour till 12:00 pm.

Layout for collection of leaf temperatures

Five plants were randomly selected from the first two gutters only in each growth chamber because of the limited space to reach the plants in the last two gutters (Table 6).

Table 6: A sample demonstration for the layout of plants used to follow leaf temperature.

Gutter 4										
Gutter 3										
Gutter 2	1				3			4		
Gutter 1			2							5

3.7 Experimental designs and description of the schedules:

Table 7: Table showing the schedule of all the activities performed in experiments I, II, and IV from start to the end: when the temperature treatments started in the chambers, when the light level was increased, and when the data were collected.

Days	**Activities**
Day 1	Seedlings transferred to chambers.
Day 14	1st morphological data and tipburn score taken.
Day 15	Temperature treatment started.
Day 16+17	Leaf temperature data followed with air temperature.
Day 23	The light level increased to 300 µmol/m^2/s.
Day 24+25	Leaf temperature data followed with air temperature.
Day 30	Experiment ended: last morphological + tipburn score data were taken

Table 7 gives all the activities performed in experiments I, II, and IV from start to end. Before day 23 plants were grown under 150 µmol/m^{2}/s. Also, before temperature treatment started the lettuces were grown under DT/NT of 20/18°C (Table 7).

3.7.1 Experiment I

Table 8 gives the summarization of experiment I after temperature treatment started in the chambers.

Table 8: Treatments used in experiment I after two weeks and how the temperature was maintained at different time points. The yellow and black colour shows the light and dark period inside the chambers respectively.

Treatments / Time	9:00am-10:00am	10:00am-11:00am	11:00am-12:00pm	12:00pm-1:00am	1:00am-9:00am
Treatment I: HPS + varied temperature	16°C	18°C	20°C	20°C	12-14°C
Treatment II: HPS + standard temperature	20°C	20°C	20°C	20°C	18°C

3.7.2 Experiment II

Table 9 gives the summarization of experiment II after temperature treatment started in the chambers.

Table 9: Treatments used in experiment II after two weeks with how the temperature in them was maintained at different time points. The yellow and black colour shows the light and dark period inside the chambers respectively.

Treatments / Time	9:00am-10:00am	10:00am-11:00am	11:00am-12:00pm	12:00pm-1:00am	1:00am-9:00am
Treatment I: LED I + varied temperature	16°C	18°C	20°C	20°C	12-14°C
Treatment II: HPS + varied temperature	16°C	18°C	20°C	20°C	12-14°C

3.7.3 Experiment III

Table 10 gives the summarization of experiment III with temperature treatments and Table 11 gives all the activities performed in experiments III from start to end. Standard temperature was used in this experiment from start to end in both chambers. Due to some technical problems in the chambers, experiment had to be ended sooner than expected.

Table 10: Treatments used in experiment III with how the temperature in them was maintained at different time points. The yellow and black colour shows the light and dark period inside the chambers respectively.

Treatments / Time	9:00am-10:00am	10:00am-11:00am	11:00am-12:00pm	12:00pm-1:00am	1:00am-9:00am
Treatment I: LED I + standard temperature	20°C	20°C	20°C	20°C	18 °C
Treatment II: LED II + standard temperature	20°C	20°C	20°C	20°C	18 °C

Table 11: Diagram showing schedule of data collection activities performed in experiment III at different days. DT/NT was the same throughout the experiment.

Days	**Activities**
Day 1	Seedlings transferred to chambers.
Day 14	1st morphological data and tipburn score taken.
Day 15	Light level was increased to 300 µmol/m^2/s.
Day 19	Experiment ended: Last morphological+tipburn score data taken.

3.7.4 Experiment IV

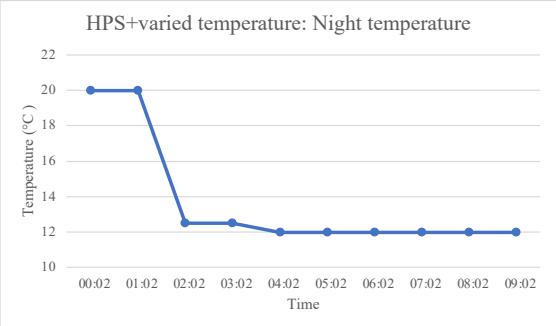
Table 12 gives the summarization of experiment IV after temperature treatment started in the chambers.

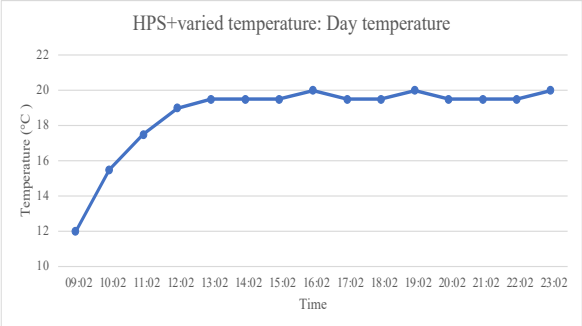
Table 12: Treatments used in experiment IV after two weeks with how the temperature in them was maintained at different time points. The yellow and black colour shows the light and dark period inside the chambers respectively.

Treatments/time	9:00am-10:00am	10:00am-11:00am	11:00am-12:00pm	12:00pm-1:00am	1:00am-9:00am
Treatment I: LED I + varied temperature	16°C	18°C	20°C	20°C	12-14°C
Treatment II: LED II + varied temperature	16°C	18°C	20°C	20°C	12-14°C

3.7.5 Exact air temperature scenario in chambers (19.10.2021)

Chamber 1





Chamber 2

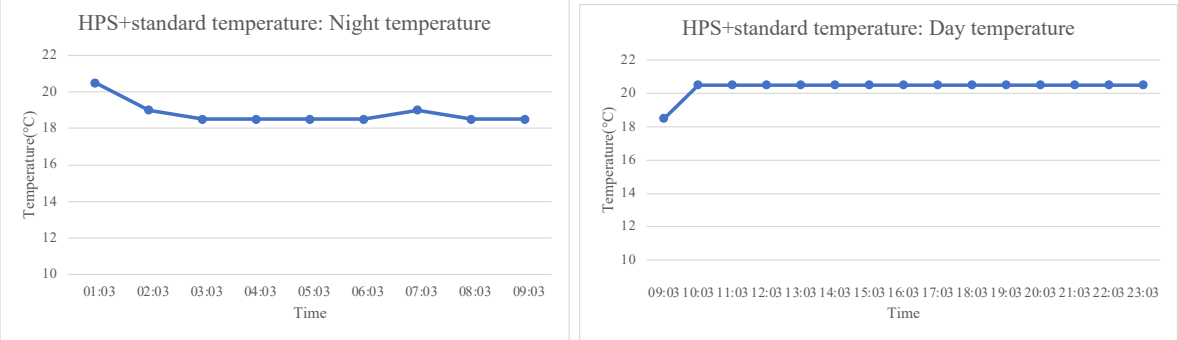


Figure 18: How the exact air temperature scenarios turned out to be in two chambers with HPS + varied temperature and HPS + standard temperature during the day(light) and night(dark) periods.

The day and night temperature data were obtained from the Priva climate computer for all experiments. A random date (19.10.2021) was picked during the first experimental period and plotted to show how the exact air temperature scenarios looked like in all the experiments with standard and varied temperature treatments during day and night (Fig 18).

3.7.6 Summary of lamp types and temperature

Table 13 below summarizes the all the experiments performed.

Table 13: *Summary of experimental set-up with different lamp types and temperatures in experiment I-IV.*

Experiments	Lamp types and Treatments: (150 µmol/m^2/s for the first three weeks and then 300 µmol/m^2/s afterwards. Temperature treatments were started after 2 weeks.)	
	Treatment I	Treatment II
Experiment I	HPS + Varied temperature	HPS + Standard temperature
Experiment II	LED I + Varied temperature	HPS + Varied temperature
Experiment III	LED I + Standard temperature	LED II + Standard temperature
Experiment IV	LED I + Varied temperature	LED II + Varied temperature

3.8 Data registrations

3.8.1 Growth and morphology

Five random plants from each chamber were selected for the first growth and morphological data set before the temperature treatment started. Growth and morphological data taken were:

1) The number of leaves: Each plant was first separated into individual leaves and placed in respective order from outer to inner leaves (Fig 19A). The smallest leaves, up to 1 cm, were counted. The first two cotyledon leaves were always excluded in any of the data taken.

2) Length of longest leaf: As the lettuce leaves are curled and unproportionate in shape, for measuring the length of leaves, they were first folded about the midrib and somewhat straightened to hold correctly. Then with a measuring scale (Fig 19B), the length was measured from the base to the tip of the leaf. The leaf number with the longest leaf was noted down together with its size.

3) Fresh weight (wt.) of the plant: All the leaves were collected and put into paper bags, and their fresh wt. was measured with a weighing device which was kept the same for all experiments. Then, the paper bags were labeled with their plant number, chamber number, experiment number, date, fresh weight, and name.

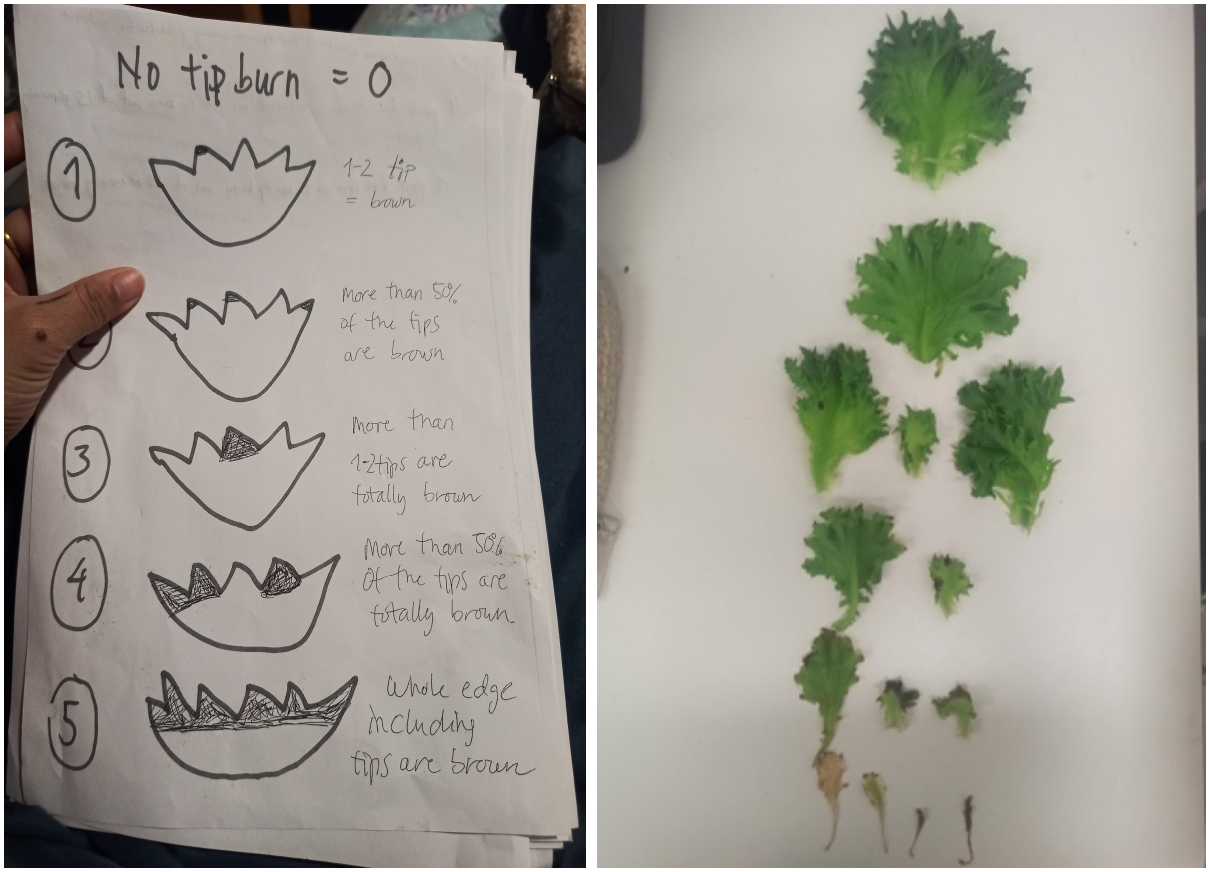
4) Dry weight (wt.) of the plant: The labeled paper bags were then kept in a drier at 60°C for a week (Fig 22C). After this, each plant's dry weight was measured using the same weighing device.

A) B)

Figure 19: A) Plant separated into individual leaves with tip burn scoring reference sheet; B) Showing scales, scissors, and vials.

3.8.2 Tipburn scoring format

Before collecting the leaves for fresh and dry weight, a tipburn assessment on each of the leaves was done separately. A scoring format was drafted with a scale of 0-5 based on the severity of tipburn and followed in scoring the tipburn as shown in Fig 20 & 21.



A) B)

Figure 20: A) Reference scoring format and B) Scores taken on its basis: dead, 5,4,3,2,1 from bottom to top.

A B C

D E F

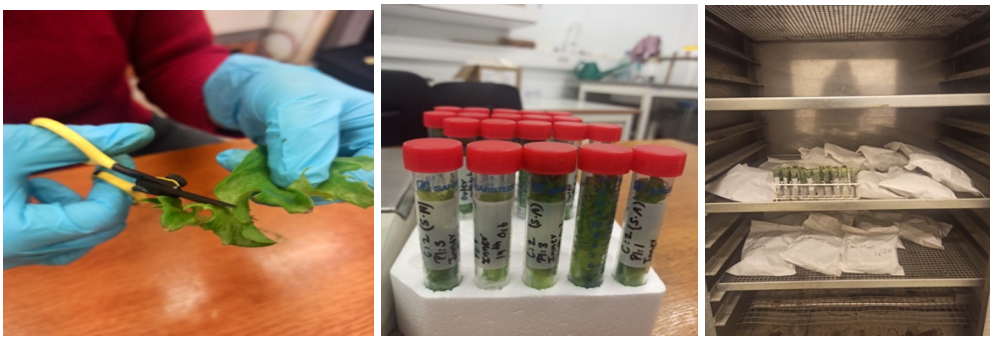
Figure 21: Tipburn scores given to the leaves: A) Dead leaves; B) Score=5; C) Score=4; D) Score=3; E) Score=2, and F) Score=1.

The same procedure for both morphological and tipburn assessment was repeated at the end of the experiment, but ten plants were randomly taken from each chamber this time. Then, tipburn on each leaf was observed again according to a scale from 0 to 5 based on the severity, as shown in Fig 20 and 21.

3.8.3 Mineral analysis

On the same day as the first morphological data was taken, another five plants from each chamber were randomly selected for mineral analysis. Samples were taken separately for each plant from the outer and inner leaves. All the plant leaves were separated individually like earlier. Then, the third and fourth leaves were taken, and their edges with tips (1-2 cm), were cut with a scissor (Fig 22A). The leaf edges were put in labeled test vials (Fig 22B). For inner leaves, the 3-4 innermost were taken and kept in vials. Twenty labeled vials were prepared from both chambers and kept in a drier with an open lid at 60°C for a week (Fig 22C). Each vial was labeled with plant number, chamber number, experiment number, inner or outer leaf, date, and name. Afterward, they were ground to fine particles in mortar and pestle, stored in 15 ml small vials, and later sent to the Lab-Tek (NMBU) for mineral analysis (Fig 23A&B). The study was performed for Ca, Mg, K, Fe, and Mn. The analysis was performed with ICP-AES (Inductively Coupled Plasma-Atomic Emission Spectroscopy) method (Greenfield, 1983).

Similarly, the same procedure was followed at the end of the experiment. The edge from leaves 3 and 4 and the 3-4 inner leaves were taken from 5 random plants selected from both chambers and put in 20 vials.



A) B) C)

Figure 22: A) Cutting of outer edge of leaf; B) Leaf edges kept in vials to dry for mineral analysis; C) Vials and paper bags kept in drier with open lid.

A) B)

Figure 23: A) The dried parts were grounded in mortar and pestle for mineral analysis; B) After grinding, all the dry powder was kept in 15ml vials and labeled to be sent to the lab for mineral analysis.

3.9 Data analysis

Data analysis was performed by using Microsoft Excel, and Minitab software version 21.1. 0 2022, USA. Means, standard deviations (St. Dev) and standard errors (SE)were calculated. For p-values, on growth and morphology data, a one-way analysis of variance (ANOVA) was performed while for mineral data two-way ANOVA analysis was performed and the Tukey test was done to compare means. For tipburn data, the Mann-Whitney test was used. The significant level was set at $p < 0.05$. Basic subsets regression was performed to evaluate the best mineral effect on the inner and outer tipburn.

4. Results

4.1 Experiment I: Temperature treatments with HPS as a light source

4.1.1 Growth and morphology

Table 14: Average mean values (± St. Dev) for growth and morphology of lettuces grown with high-pressure sodium lamps (HPS) as light source and standard temperature of DT/NT: 20/18°C (ADT:19.33°C) at a PPFD of 150 µmol/m²/s for two weeks. N= 5, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
HPS+standard temperature	19.82 (±2.35)	0.92 (±0.16)	9.6 (±0.89)	14.08 (±0.16)
HPS+standard temperature	21.57 (±2.87)	1.03 (±0.18)	10.2 (±0.84)	14.02 (±0.71)
p-values	0.322	0.342	0.305	0.859

Table 15: Average mean values (± St. Dev) for growth and morphology of lettuces when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C (ADT:19.33°C) compared to a varied temperature of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
HPS+varied temperature	170 (±39.3)	11.09 (±1.20)	23.3 (±1.06)	17.76 (±1.02)
HPS+standard temperature	159.87 (±19.01)	11.01 (±0.73)	25.2 (±1.03)	16.75 (±0.72)
p-values	0.471	0.866	0.001	0.020

Tables 14 and 15 show the growth and morphological data obtained from experiment I for moderate (150 µmol/m²/s) and high light intensity (300 µmol/m²/s) respectively. The results did not show any significant difference after two weeks of cultivation ($p>0.05$) as the temperature treatments were the same here (Table 14). When the plants were grown for two more weeks and the light was increased to 300 µmol/m²/s in the last week, a significant difference between the two temperature treatments was found in the number of leaves and length of the longest leaf with similar fresh and dry weights. The temperature variation induced longer leaves but a fewer leaf number compared with plants exposed to standard temperature (Table 15).

4.1.2 Tipburn

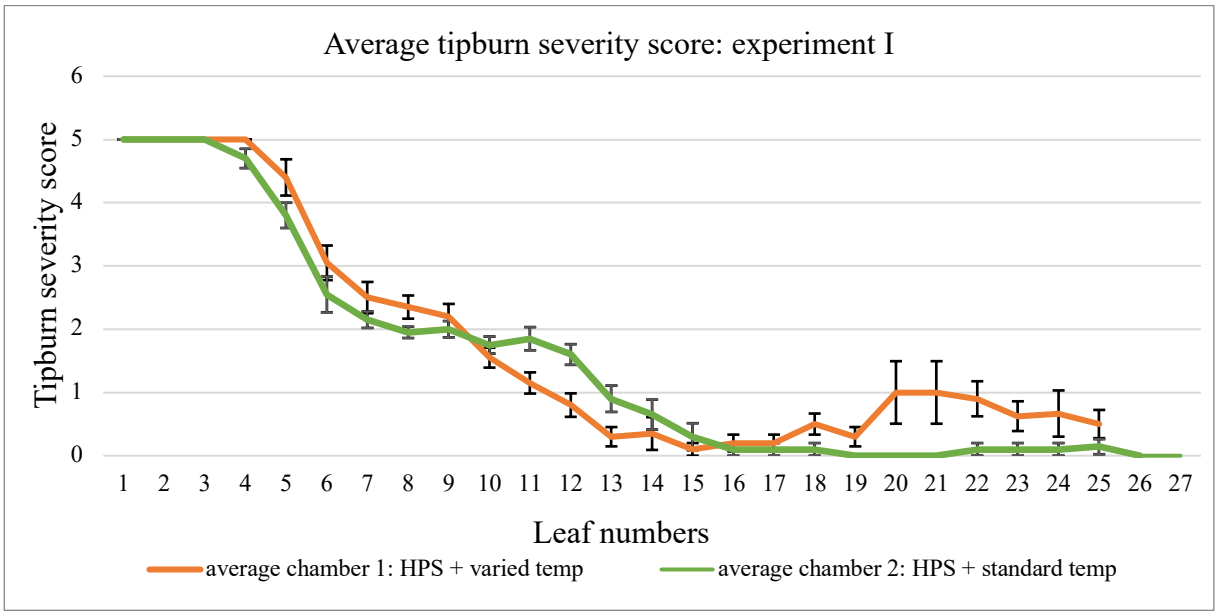


Figure 24: Average tipburn severity score (0-5) on individual leaves when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C compared to a varied temperature of DT/NT: 20/13°C after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, bars=standard error (SE).

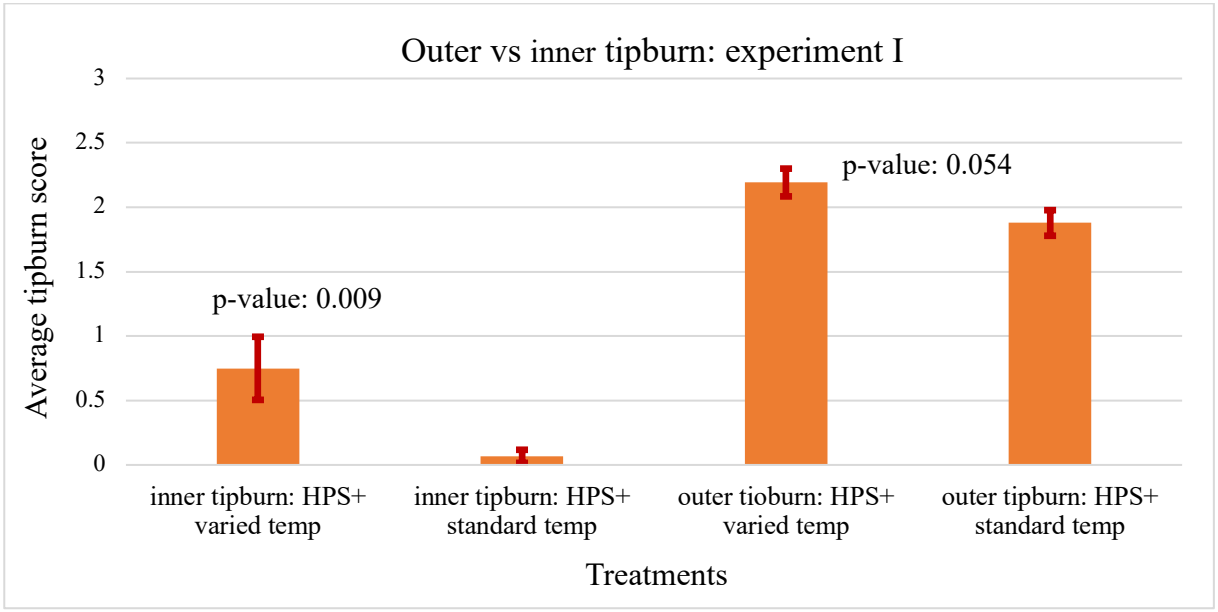


Figure 25: Average outer vs. inner tipburn (0-5) when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C compared to a varied temperature of DT/NT: 20/13°C after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N=10, bars=standard error (SE). Mann-Whitney test: significance level was set at p<0.05.

Figures 24 and 25 show the incidence of tipburn development in experiment I. The outer leaves showed severe outer tipburn, but the differences between standard and varied temperatures were small and insignificant ($p > 0.05$). However, plants exposed to varied temperatures

showed more inner tipburn compared with standard temperature ($p < 0.05$) (Fig 24 & 25). The inner tipburn was almost negligible for HPS+ standard temperature.

4.1.3 Mineral analysis

Table 16: Average content of selected minerals (± St. Dev) of lettuce leaves when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C (ADT:19.33°C) compared to a varied temperature of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different mean within each mineral.

		Treatments(T)		p-values		
Minerals	**Leaf type (L)**	**HPS + varied temperature**	**HPS + standard temperature**	**T**	**L**	**T*L**
Ca (%)	outer	1.44 ± 0.49 b	2.14 ± 0.21 a	0.01	< 0.001	0.014
	inner	0.22 ± 0.04 c	0.24 ± 0.05 c			
Mg (%)	outer	1.04 ± 0.15 a	1.18 ± 0.16 a	< 0.001	< 0.001	0.006
	inner	0.2 ± 0 c	0.68 ± 0.08 b			
K (%)	outer	5.7 ± 1.57 b	7.36 ± 0.39 a	0.089	< 0.001	0.031
	inner	4.32 ± 0.60 b	4.10 ± 0.40 b			
Fe	outer	200.2 ± 58.8 a	173.4 ± 15.61 ab	0.129	< 0.003	0.946
(mg/kg)	inner	143.8 ± 36.0 ab	119.20 ± 12.64 b			
Mn	outer	285.6 ± 53.7 a	323.2 ± 26 a	0.254	< 0.001	0.126
(mg/kg)	inner	32.80 ± 3.11 b	27 ± 6.28 b			

When morphological data were taken, temperature treatments had not yet started at 150 µmol/m²/s, so both the chambers were under the same climatic conditions. Mineral contents of selected cations were measured when the plants were grown for two weeks at 20/18°C (results not shown), after which, in one chamber temperature was varied.

Table 16 shows the mineral result from experiment I with two different temperature treatments under higher light intensity. A significant interaction between the treatments and leaf types was found for Ca, Mg, and K. Ca and K were highest in outer leaves of HPS+ standard temperature and Mg was equal in outer leaves in both treatments. In inner leaves, there was no difference in Ca and K content between the two treatments, while Mg was higher in HPS+ standard temperature. The content of Fe and Mn were not statistically affected by the different temperature treatments but were affected by leaf type. The outer leaves had higher amounts than the inner leaves.

4.1.4 Leaf temperatures

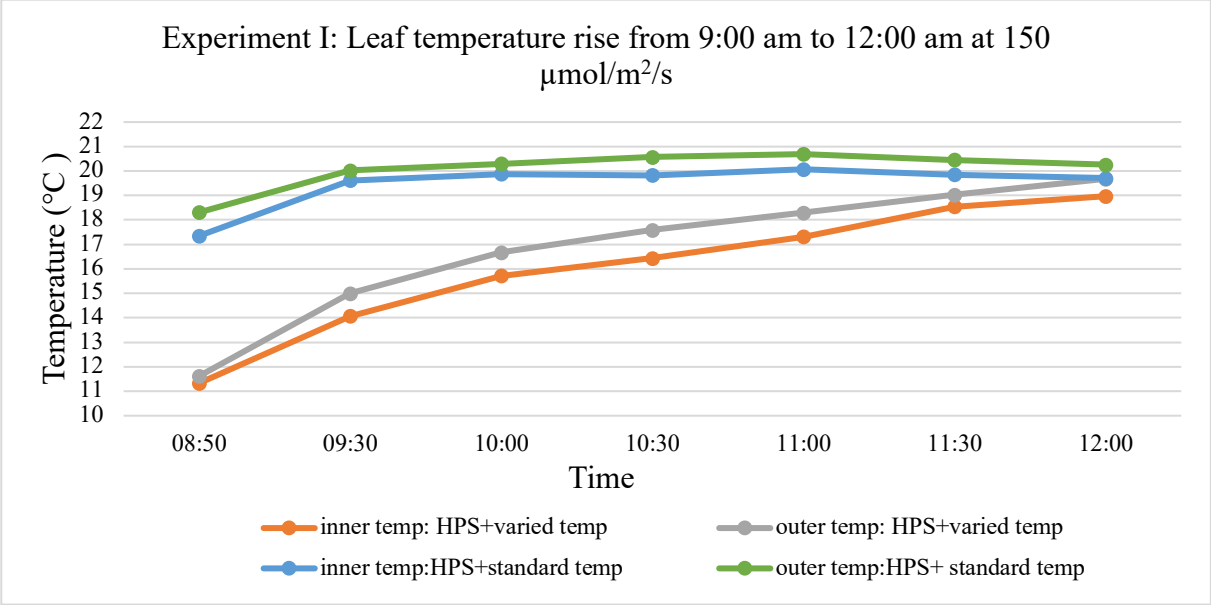


Figure 26: Average inner and outer leaf temperature rise from 9:00 am to 12:00 am at 150 µmol/m²/s when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C (ADT: 19.33°C) and varied temperature of DT/NT: 20/13°C (ADT: 17.41°C). Lamps were turned on at 9:00 am. N=5

As seen in Fig 26, inner leaves' temperatures were always lower than outer leaf temperature at all time points in both temperature treatments, but the temperature gap was a bit higher between them in varied temperatures (0.7-1°C) than in standard temperatures (0.4-0.6°C). The inner leaf temperature was approximately a degree lower than the air temperature, while the outer temperature was proportionate to the air temperature at each point in HPS+ varied temperature. At noon, inner leaf temperature also did not reach 20°C. However, in HPS+ standard temperature, both the inner and outer leaf temperatures were around 20°C.

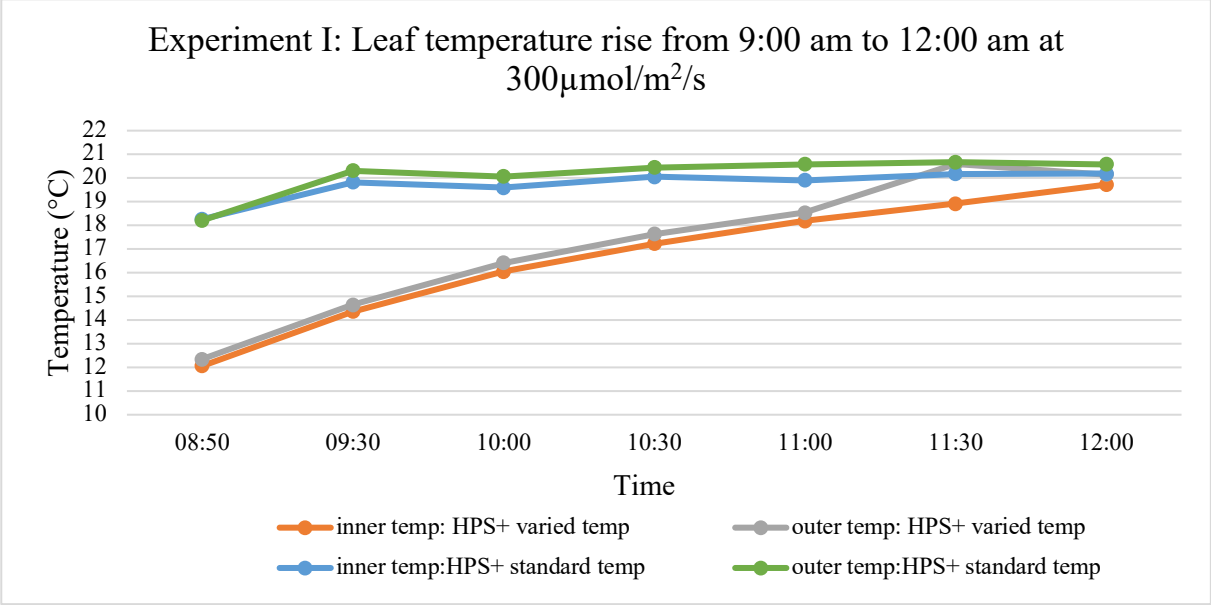


Figure 27: Average inner and outer leaf temperature rise from 9:00 am to 12:00 am at 300 µmol/m²/s when lettuces were grown under HPS lights with a standard temperature of DT/NT: 20/18°C (ADT: 19.33°C) and varied temperature of DT/NT: 20/13°C (ADT: 17.41°C). Lamps were turned on at 9:00 am. N=5.

When the light was turned on, an immediate leaf temperature rise in both treatments according to their air temperatures could be seen at 300 µmol/m²/s (Fig 27). However, the gap between inner and outer leaves narrowed here, than at 150 µmol/m²/s. The narrowing resulted from the rising of inner leaf temperatures along with the air temperatures. At 11:30 am, an increase in the outer leaf temperature beyond the trend could be seen in HPS+ varied temperature which at noon, again stabilized at 20°C. All the leaves in both treatments reached 20°C at noon.

4.2 Experiment II: Varied temperature treatments with HPS and LED I as light sources

4.2.1 Growth and morphology

Table 17: Average mean values (± St. Dev) for growth and morphology of lettuces grown with HPS lamps compared with LED I as light sources and standard temperature treatment of DT/NT: 20/18°C (ADT:19.33°C) at a PPFD of 150 µmol/m²/s for two weeks. N= 5, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I + standard temperature	12.09 ± 1.54	0.66 ± 0.09	9.2 ± 0.45	11.30 ± 0.837
HPS + standard temperature	11.24±11.16	0.62 ± 0.02	10.40 ± 0.55	11.82 ± 0.54
p-values	0.294	0.486	0.005	0.277

Table 18 Average mean values (± St. Dev) for growth and morphology of lettuces when they were grown with the HPS lamps compared with LED I as light sources and varied temperature treatment of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N=10, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I + varied temperature	136.59 ± 20.25	8.45 ± 1.24	21.33 ± 1.8	16.67 ± 0.94
HPS + varied temperature	106.02 ± 22.02	6.62 ± 1.58	22.7 ± 1.25	15.32 ± 1.09
p-values	0.005	0.010	0.070	0.009

Tables 17 and 18 show the growth and morphological data obtained from experiment II at moderate (150 µmol/m²/s) and high light intensity (300 µmol/m²/s), respectively. After two weeks of cultivation, HPS + standard temperature had a significantly higher number of leaves than LED + standard temperature. When the temperature variations were introduced and the light was increased to 300 µmol/m²/s, LED + varied temperature gave bigger fresh, and dry wt, and longer leaves than HPS + varied temperature. The treatments however brought no significant difference in the number of leaves.

4.2.2 Tipburn

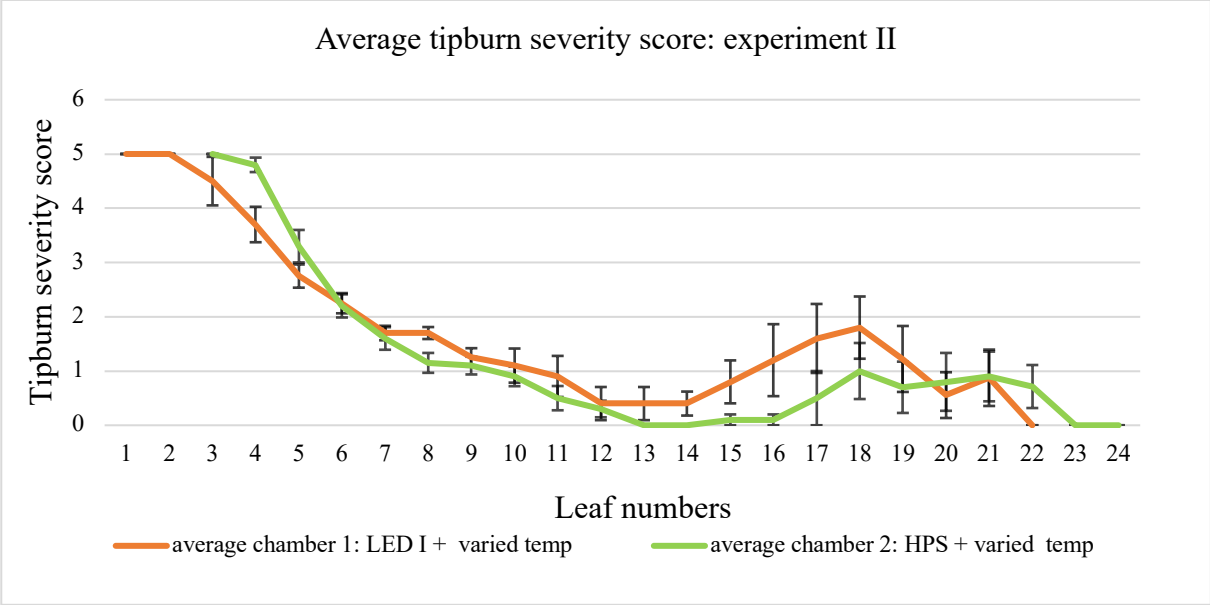


Figure 28: Average tipburn severity score (0-5) on individual leaves when lettuces were grown with the HPS lamps compared with LED I as light sources and varied temperature of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, bars = SE.

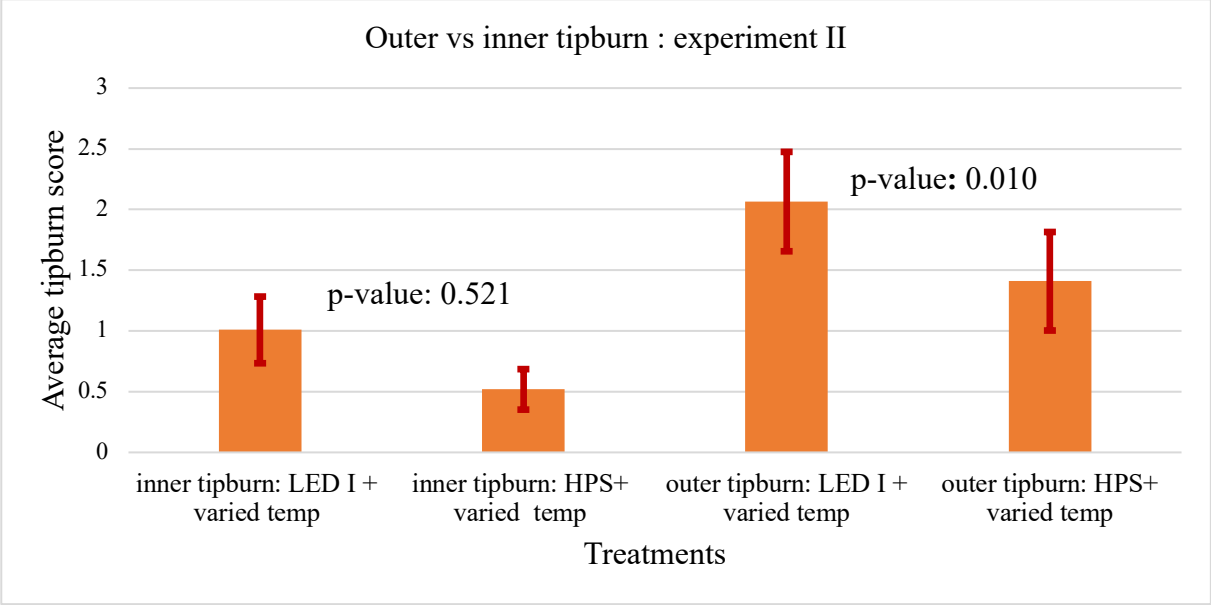


Figure 29: Average outer vs, inner tipburn (0-5) when lettuces were grown with HPS lamps compared with LED I as light sources and varied temperature of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, bars = SE, Mann-Whitney test: significance level was set at p<0.05.

Figures 28 and 29 depict the incidence of tipburn development in experiment II as affected by the LED I+ varied temperature and HPS+ varied temperature. There was significantly higher outer tipburn in LED I+ varied temperature. Inner tipburn was severe on both treatments with LED I+ varied temperature giving more tipburn; however, the difference between the two treatments was insignificant.

4.2.3 Mineral analysis

Table 19: Average content of selected minerals (± St, Dev) of lettuces leaves when lettuces were grown with HPS lamps compared with LED I as light sources at a PPFD of 150 µmol/m²/s and standard temperature of DT/NT: 20/18°C (ADT:19.33°C) for two weeks. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		p-values		
Minerals	**Leaf type (L)**	**LED I + standard temperature**	**HPS+ standard temperature**	***T***	***L***	***T*L***
Ca (%)	outer	1.22 ± 0.11 a	1.04 ± 0.05 b	*0.042*	*<0.001*	0.003
	inner	0.34 ± 0.06 c	0.38 ± 0.05 c			
Mg (%)	outer	1.4 ± 0.34 a	0.74 ± 0.11 b	*0.001*	*<0.001*	0.001
	inner	0.30 ± 0.00 c	0.28 ± 0.05 c			
K (%)	outer	9.32 ± 0.26 a	9.20 ± 0.62 a	*0.656*	*<0.001*	0.823

	inner	6.26 ± 0.37 b	6.22 ± 0.19 b			
Fe (mg/kg)	outer	142.20 ± 17.74 a	159.8 ± 38.0 a	*0.698*	*<0.001*	0.239
	inner	143.20 ± 18.09 a	134.20 ± 16.69 a			
Mn (mg/kg)	outer	180.8 ± 25.0 a	161.20 ± 16.72 a	*0.397*	*<0.001*	0.092
	inner	49.60 ± 9.84 b	56.40 ± 9.07 b			

Table 19 shows the mineral result for experiment II with two different lights at standard temperature treatments under moderate light intensity (150 µmol/m^2/s). There was a clear interaction between treatments and leaf type for Ca and Mg; both were highest in outer leaves of LED I+ standard temperature while inner leaves had the lowest of them and were similar in amounts under both treatments. K and Mn content between the two treatments were similar for both outer and inner leaves but, within each treatment, the content was lower in the inner leaves than in the outer. Fe content was similar for all leaves on both treatments.

Table 20: Average content of selected minerals (± St, Dev) in leaves of lettuces when lettuces were grown with HPS lamps compared with LED I as light sources and varied temperature of DT/NT: 20/13°C (ADT:17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m^2/s from 150 µmol/m^2/s during the last week. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		**p-values**		
Minerals	**Leaf type (L)**	**LED I + varied temperature**	**HPS+ varied temperature**	**T**	**L**	**T*L**
Ca (%)	outer	2.22 ± 0.24 a	2.28 ± 0.13 a	*0.438*	*<0.001*	0.876
	inner	0.14 ± 0.06 b	0.18 ± 0.05 b			
Mg (%)	outer	1.14 ± 0.06 b	1.3 ± 0.16 a	*0.048*	*<0.001*	0.048
	inner	0.20 ± 0.00 c	0.20 ± 0 c			
K (%)	outer	7.66 ± 0.66 a	7.18 ± 0.37 a	*0.375*	*<0.001*	0.115
	inner	3.7 ± 0.30 b	3.84 ± 0.17 b			
Fe (mg/kg)	outer	117.8 ± 15.09 a	111.2 ± 22.4 a	*0.497*	*0.303*	0.122
	inner	98.80 ± 7.29 a	115.2 ± 14.45 a			
Mn (mg/kg)	outer	242.8 ± 16.8 a	247 ± 18.07 a	*0.466*	*<0.001*	0.986
	inner	22.20 ± 5.97 b	26.60 ± 4.16 b			

Table 20 shows the mineral result from experiment II with two different lights at varied temperature treatments under higher light intensity (300 µmol/m^2/s). There was an interaction between treatments and leaf type only for Mg which was highest in outer leaves of HPS+ varied temperature and inner leaves have similar doses under both treatments being the lowest. All

the other minerals were higher in outer leaves than in inner leaves with no difference between the treatments except Fe which was similar in all irrespective of treatments and leaf types.

4.2.4 Leaf temperature

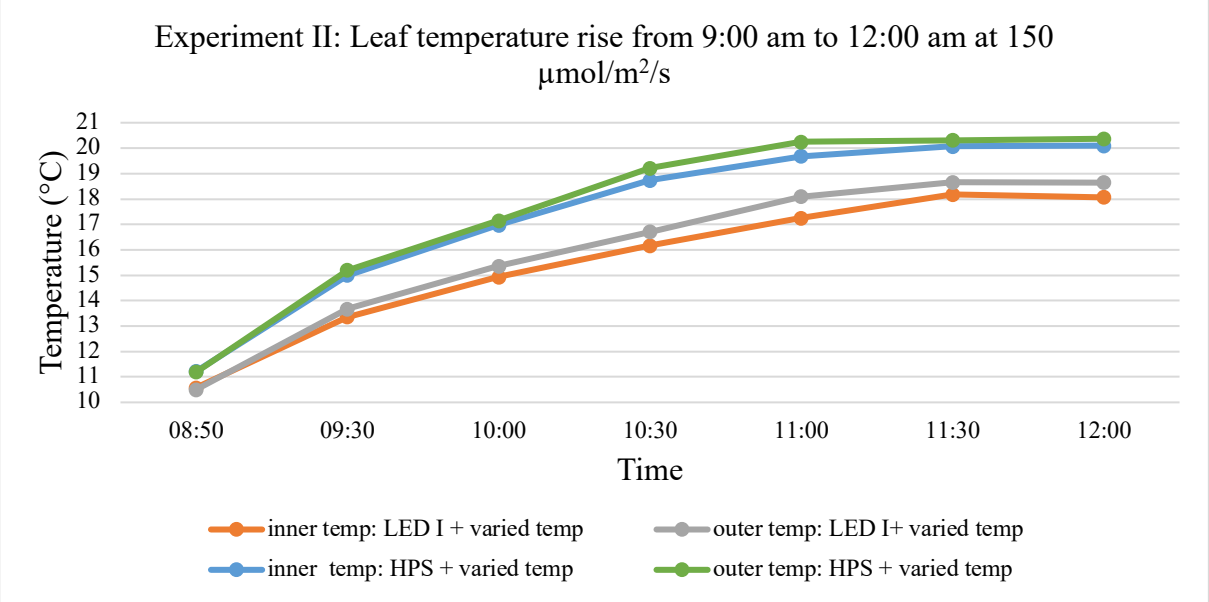


Figure 30: Average inner and outer leaf temperature rise from 9:00 am to 12:00 am on lettuces grown with HPS lamps compared with LED I as light sources at a PPFD of 150 µmol/m²/s and varied temperature of DT/NT: 20/13°C (ADT:17.41°C). Lamps were turned on at 9:00 am, N=5.

Figure 30 gives the inner and outer leaf temperature rise from 9:00 AM to 12:00 PM for experiment II at moderate light intensity (150 µmol/m²/s). Both the inner and outer leaf temperature of lettuces under HPS+ varied temperature rose faster than the lettuces under LED I+ varied temperature at all time points being inner leaf temperature in LED I+ varied temperature always lower than outer. At all-time points, the inner and outer leaf temperatures were lower than the air temperature in LED I but were higher in HPS light.

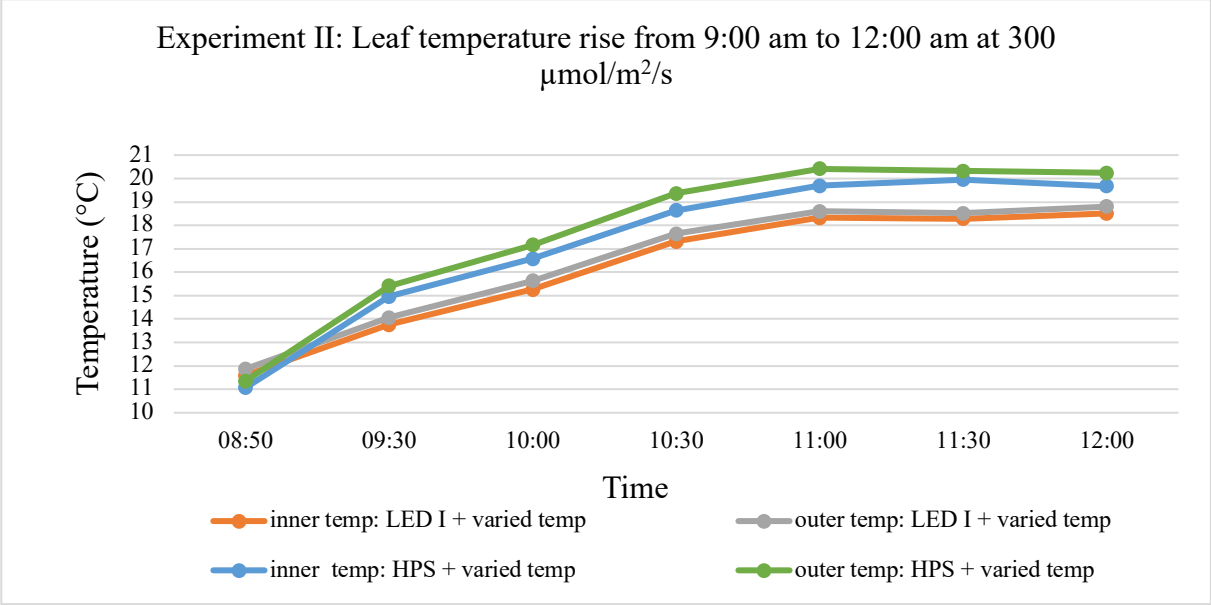


Figure 31: Average inner and outer leaf temperature rise from 9:00 am to 12:00 am on lettuces grown with HPS lamps compared with LED I as light sources at a PPFD of 300 µmol/m²/s and varied temperature of DT/NT: 20/13°C (ADT:17.41°C). Lamps were turned on at 9:00 am. N=5.

Figure 31 gives the inner and outer leaf temperature rise from 9:00 AM to 12:00 PM for experiment II at the higher light intensity (300 µmol/m²/s). The leaf temperature gap between outer and inner leaves increased a bit in HPS treatment while the temperature was almost equal under LED I light. Moreover, inner and outer leaf temperatures under HPS were always higher than air temperatures but under LED I, they were always lower at all time points.

4.3 Experiment III: Standard temperature treatments with LED I and LED II as light sources

4.3.1 Growth and morphology

Table 21: Average mean values (± St, Dev) for growth and morphology of lettuces grown with LED I compared with LED II as light sources and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at a PPFD of 150 µmol/m²/s for two weeks. N= 5, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I + standard temperature	15.61 ± 1.53	1.76 ± 0.02	10.80 ± 0.45	13.26 ± 1.37
LED II + standard temperature	10.96 ± 1.59	1.55 ± 0.11	10.60 ± 0.55	11.48 ± 0.91
p-values	0.002	0.003	0.545	0.041

Table 21 shows the growth and morphological results at moderate light intensity (150 µmol/m^2/s) in experiment III. The results showed significantly higher fresh and dry weight and longest leaf in LED I+ standard temperatures than in LED II+ standard temperatures.

Table 22: Average mean values (± St, Dev) for growth and morphology of lettuces grown with LED I compared with LED II as light sources and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at an increased PPFD of 300 µmol/m^2/s from 150 µmol/m^2/s after first two weeks. N=10, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I + standard temperature	94.74 ± 10.74	5.45 ± 1.13	20.20 ± 1.32	15.92 ± 0.68
LED II+ standard temperature	60.82 ± 15.53	3.57 ± 0.98	17.40 ± 2.12	12.78 ± 0.60
p-values	*<0.001*	0.001	0.002	*<0.001*

Table 22 shows the growth and morphological results at the higher light intensity (300 µmol/m^2/s) in experiment III. LED I+ standard temp, brought significantly higher fresh and dry weight, with longer and a greater number of leaves than under LED II + standard temperature.

4.3.2 Tipburn

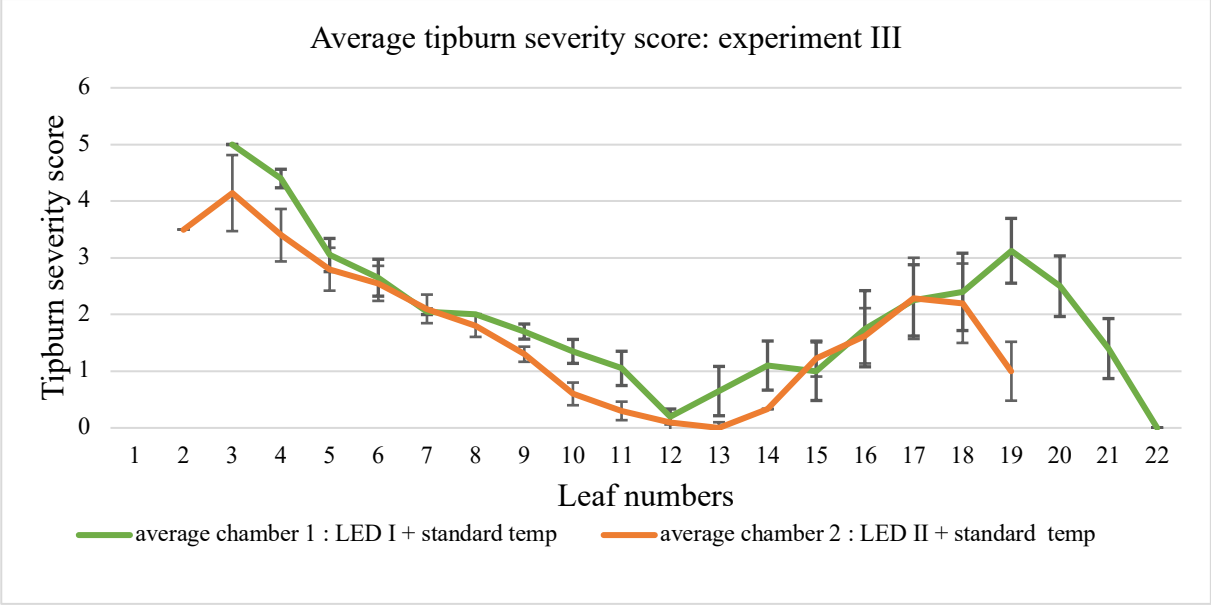


Figure 32: Average tip burn severity score (0-5) on individual leaves when lettuces were grown with LED I compared with LED II as light sources and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at an increased PPFD of 300 µmol/m^2/s from 150 µmol/m^2/s after first two weeks. N= 10, bars = SE.

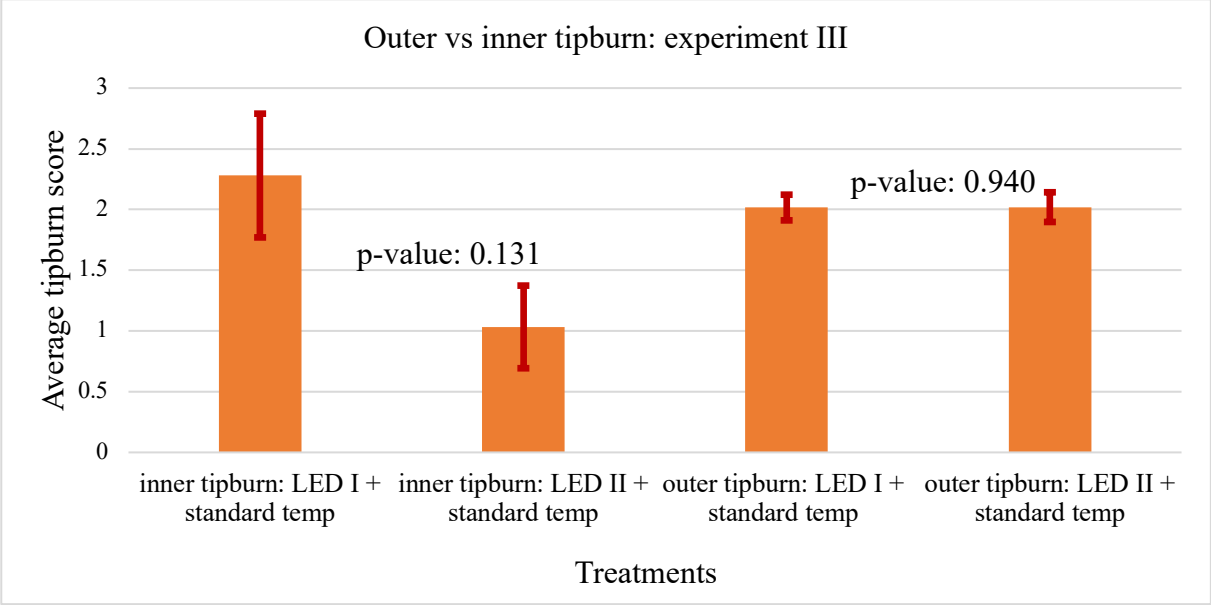


*Figure 33: Average outer vs, inner tipburn (0-5) when lettuces were grown with **LED** I compared with LED II as light sources and standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s after first two weeks. N= 10, bars = SE, Mann-Whitney test: significance level was set at p<0.05.*

Figures 32 and 33 show the tipburn severity score as affected by the two treatments in experiment III. There were extreme outer and inner tipburn under both treatments, but the results were not significantly different between the two. Outer tipburn is at a similar level but, inner tipburn was more severe under LED I + standard temperature.

4.3.3 Mineral analysis

Table 23: Average content of selected minerals (± St, Dev) of lettuce leaves when lettuces were grown with LED I compared with LED II as light sources, and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at a PPFD of 150 µmol/m²/s for two weeks. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		p-values		
Minerals	**Leaf type (L)**	**LED I + standard temperature**	**LED II + standard temperature**	**T**	**L**	**T*L**
Ca (%)	outer	1.2 ± 0.07 a	1.16 ± 0.05 a	*0.150*	*<0.001*	0.008
	inner	0.34 ± 0.05 c	0.46 ± 0.05 b			
Mg (%)	outer	0.9 ± 0.07 a	0.92 ± 0.08 a	*0.077*	*<0.001*	0.274
	inner	0.32 ± 0.05 b	0.4 ± 00 b			
K (%)	outer	8.38 ± 0.24 a	7.98 ± 0.58 a	*0.318*	*<0.001*	0.007
	inner	4.90 ± 0.56 c	5.7 ± 0.21 b			

Fe (mg/kg)	outer	211.6 ± 70.9 a	201 ± 10.72 a	*0.813*	*0.251*	0.393
	inner	177.2 ± 17.08 a	195.8 ± 10.01 a			
Mn (mg/kg)	outer	189.0 ± 7.33 a	186.0 ± 7.33 a	*0.054*	*<0.001*	0.024
	inner	59.4 ± 14.43 c	84 ± 7.62 b			

Table 23 gives the mineral results for experiment III at moderate light intensity (150 µmol/m^2/s). There was a clear interaction between treatments and leaf type for Ca, K, and Mn; outer leaves had the highest of all three with no difference among the two treatments, However, in inner leaves, they were higher in LED II+ standard temperature and lower in LED I+ standard temperature. The amount of Fe was significantly similar in all the leaves irrespective of treatments and leaf type.

Table 24: Average content of selected minerals (± St, Dev) of lettuces grown with the LED I compared with LED II as light sources and standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at an increased PPFD of 300 µmol/m^2/s from 150 µmol/m^2/s after first two weeks. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		**p-values**		
Minerals	**Leaf type (L)**	**LED I + standard temperature**	**LED II + standard temperature**	**T**	**L**	**T*L**
Ca (%)	outer	1.76 ± 0.29 a	1.92 ± 0.16 a	0.257	<0.001	0.374
	inner	0.20 ± 0.0 b	0.22 ± 0.04 b			
Mg (%)	outer	1.06 ± 0.11 a	1.12 ± 0.13 a	0.332	<0.001	0.624
	inner	0.20 ± 0.00 b	0.22 ± 0.04 b			
K (%)	outer	7.9 ± 1.08 a	7.6 ± 0.26 a	0.478	<0.001	0.079
	inner	3.46± 0.19 b	4.14 ± 0.30 b			
Fe (mg/kg)	outer	129 ± 13.44 b	152.6 ± 9.61 a	<0.001	0.955	0.127
	inner	120.2 ± 6.18 b	160.8 ± 15.75 a			
Mn (mg/kg)	outer	211.6 ± 26.2 a	218.8 ± 3.66 a	0.528	<0.001	0.918
	inner	28.80 ± 2.95 b	34 ± 8.19 b			

Table 24 gives the mineral results for experiment III at higher light intensity (300 µmol/m^2/s), There was no interaction between treatments and leaf type. The treatment effect was only seen for Fe where LED II +standard temperature had higher amounts in both inner and outer leaves than LED I+ standard temperature. In contrast, Ca, Mg, K, and Mn were significantly higher in outer leaves than inner leaves with no difference between the treatments.

4.4 Experiment IV: Varied temperature treatments with LED I and LED II as light sources

4.4.1 Growth and morphology

Table 25: Average mean values (± St, Dev) for growth and morphology of lettuces grown with LED I compared with LED II as light sources and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at a PPFD of 150 µmol/m²/s, for two weeks. N= 5, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I + standard temperature	19.67 ± 1.62	1.02 ± 0.10	9.80 ± 0.45	12.28 ± 0.31
LED II+ standard temperature	14.72 ± 1.45	0.86 ± 0.09	9.20 ± 0.45	10 ± 0.39
p- values	0.001	0.028	0.067	*<0.001*

Table 25 shows the treatments' effect on the growth and morphology for experiment IV at moderate light intensity (150 µmol/m^2/s). LED I+ standard temperature gave significantly higher fresh and dry weight and longer leaves than LED II+ standard temperature.

Table 26: Average mean values (± St. Dev) for growth and morphology of lettuces grown with LED I compared with LED II as light sources and the varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) after the first two weeks with an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, ANOVA: significance level was set at p<0.05.

Treatments	Fresh weight (gm)	Dry weight (gm)	No of leaves	Length of longest leaf (cm)
LED I +varied temperature	124.55 ± 12.12	8.12 ± 0.72	22.7 ± 1.89	16.18 ± 0.91
LED II +varied temperature	88.69 ± 16.37	5.66 ± 0.97	20.30 ± 1.06	13.62 ± 0.66
p- values	*<0.001*	*<0.001*	0.003	*<0.001*

Table 26 shows the treatments' effect on the growth and morphological parameters for experiment IV at higher light intensity (300 µmol/m^2/s). Fresh and dry weight were significantly higher in LED I+ varied temperature together with longer, and a greater number of leaves than in LED II+ varied temperature.

4.4.2 Tipburn

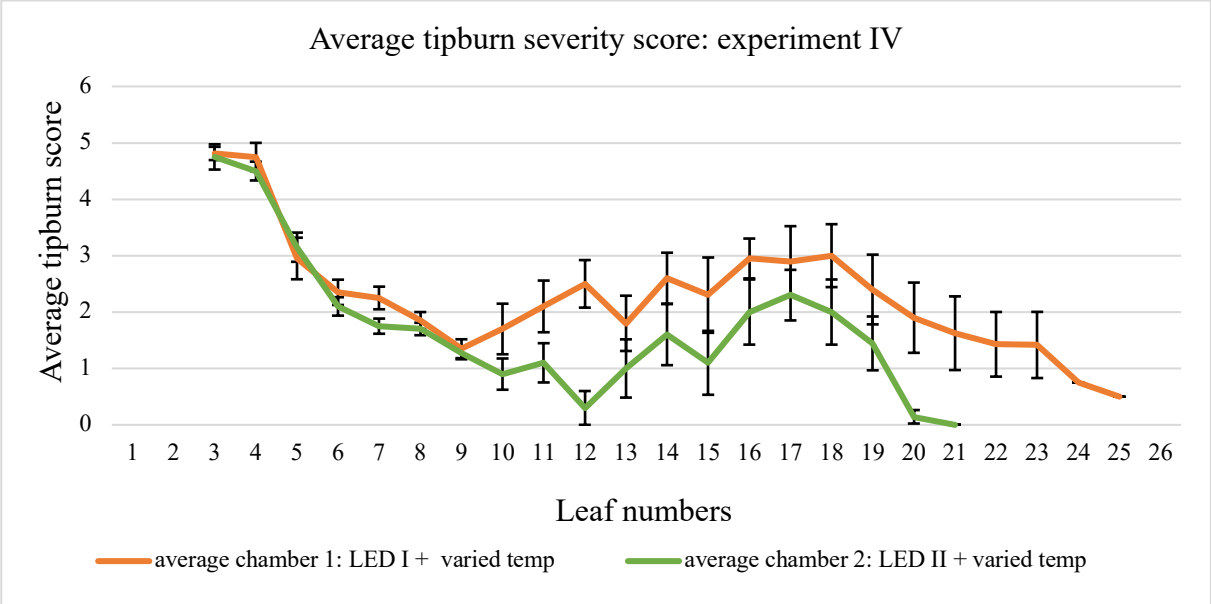


Figure 34: Average tip burn severity score (0-5) on individual leaves when lettuces were grown with LED I compared with LED II as light sources and varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, bars = SE.

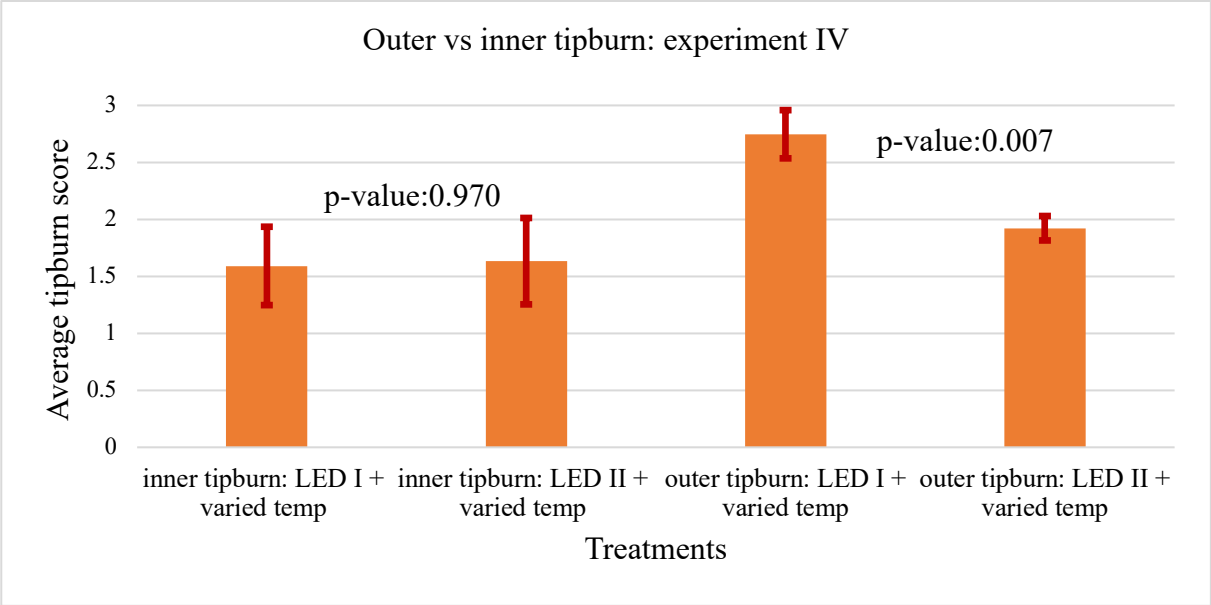


Figure 35: Average outer vs inner tipburn (0-5) when lettuces were grown with LED I compared with LED II as light sources and varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. N= 10, bars = SE, Mann-Whitney test: significance level was set at p<0.05.

Figures 34 and 35 show the tipburn severity score as affected by the two treatments in experiment IV. The outer and inner tipburn were severe under both treatments; however, only the outer tipburn was significantly higher with LED I+ varied temperature, and the inner tipburn had an insignificant difference between the two treatments.

4.4.3 Mineral analysis

Table 27: Average content of selected minerals (± St. Dev) of lettuce lettuces when lettuces were grown with LED I compared with LED II as light sources and the standard temperature treatments of DT/NT: 20/18°C (ADT: 19.33°C) at a PPDF of 150 µmol/m²/s for two weeks, Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		p-values		
Minerals	**Leaf type (L)**	**LED I + standard temperature**	**LED II+ standard temperature**	**T**	**L**	**T*L**
Ca (%)	outer	1.3 ± 0.28 a	1.36 ± 0.15 a	*0.589*	*<0.001*	0.786
	inner	0.4 ± 0.07 b	0.42 ± 0.04 b			
Mg (%)	outer	0.54 ± 0.05 a	0.5 ± 0.0 a	*0.301*	*<0.001*	0.301
	inner	0.22 ± 0.04 b	0.22 ± 0.04 b			
K (%)	outer	8.80 ± 0.96 a	8.4 ± 0.20 a	*0.509*	*<0.001*	0.038
	inner	5.36 ± 0.31b	6.10 ± 0.46 b			
Fe (mg/kg)	outer	139 ± 2.55 a	147.40 ± 11.97 a	*0.242*	*<0.001*	0.709
	inner	152.40 ± 7.30 a	156.80 ± 18.78 a			
Mn (mg/kg)	outer	144 ± 22.8 b	174.00 ± 18.18 a	*0.008*	*<0.001*	0.284
	inner	51 ± 9 .82 c	65 ± 9.90 c			

Table 27 gives the mineral results at moderate light intensity (150 µmol/m²/s) in two different treatments for experiment IV. A clear interaction between treatments and leaf type was there for K; it is highest in the outer leaves of both treatments and lowest in their inner leaves. All other minerals were higher in outer leaves except for Fe, which was statistically similar in all.

Table 28: Average content of selected minerals (± St. Dev) of lettuces grown with LED I compared with LED II as light sources and the varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) after the first two weeks and an increased PPFD of 300 µmol/m²/s from 150 µmol/m²/s during the last week. Samples were taken from the edge of outer leaves and young unexpanded inner leaves. N=5, Two-way ANOVA: significance level was set at p<0.05, abcd= Tukey pairwise comparison where different letters indicate significantly different means within each mineral.

		Treatments (T)		p-values		
Minerals	**Leaf type (L)**	**LED I + varied temperature**	**LED II+ varied temperature**	**T**	**L**	**T*L**
Ca (%)	outer	1.96 ± 0.29 b	2.36 ± 0.23 a	*0.045*	*<0.001*	0.029
	inner	0.20 ± 0.10 c	0.18 ± 0.08 c			
Mg (%)	outer	0.70 ± 0.10 a	0.78 ± 0.08 a	*0.189*	*<0.001*	0.189
	inner	0.20 ± 0.0 b	0.20 ± 0.00 b			

K (%)	outer	8.34 ± 0.96 a	8.80 ± 0.18 a	*0.234*	*<0.001*	0.519
	inner	3.62 ± 0.444 b	3.76 ± 0.15 b			
Fe (mg/kg)	outer	112.4 ± 15.18 a	128.20 ± 12.03 a	*0.069*	*0.978*	0.829
	inner	114.2 ± 19.01 a	126.80 ± 17.96 a			
Mn (mg/kg)	outer	194.2 ± 35 a	231.2 ± 26.2 a	*0.063*	*<0.001*	0.123
	inner	26.00 ± 9.67 b	29.80 ± 6.34 b			

Table 28 gives the mineral results at the higher light intensity (300 µmol/m^2/s) in two different treatments for experiment IV. Here, a clear interaction effect of treatments and leaf type was seen in only Ca; it was highest in outer leaves of LED II+ varied temperature and lowest in inner leaves that were statistically similar for both the treatments. For Mg, K, and Mn content, outer leaves had significantly higher doses than inner leaves except for Fe content which was similar in all leaves in all cases.

4.4.4 Leaf temperature

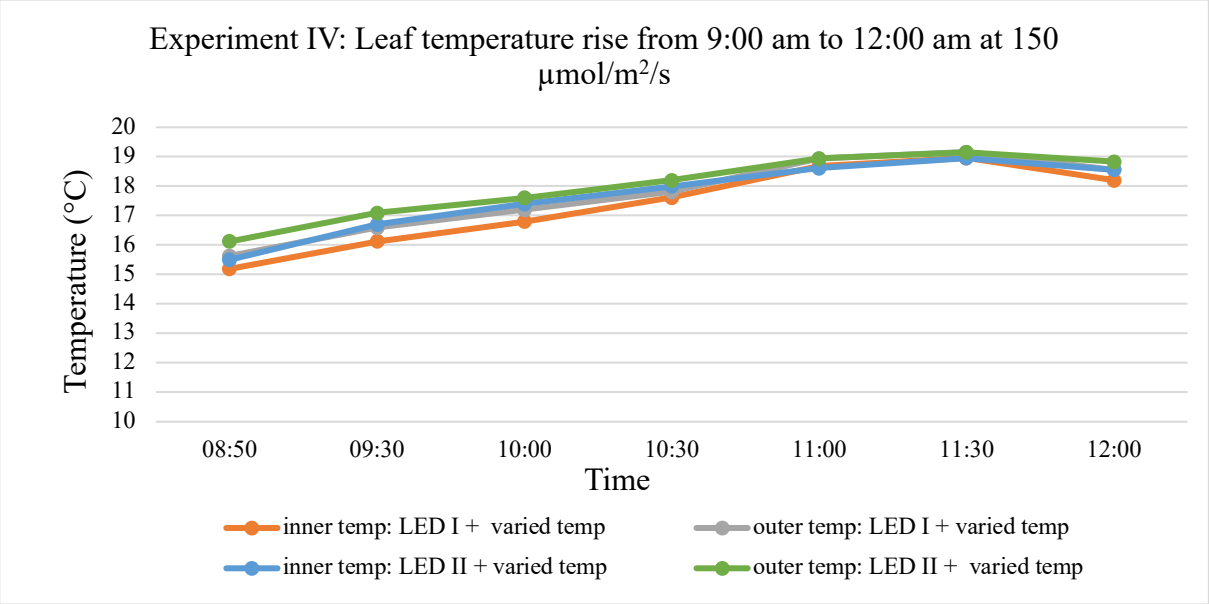


Figure 36:Average inner and outer leaf temperature rise from 9:00 am to 12:00 am on lettuces grown with LED I compared with LED II as light sources, and the varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) at a PPFD of 150 µmol/m^2/s. Lamps were turned on at 9:00 am. N=5.

Figure 36 shows the inner and outer leaf temperature rise from 9:00 am to 12:00 am at 150 µmol/m^2/s in the two treatments for experiment IV. Not much gap (0.2-0.47°C) existed between inner and outer temperatures under both treatments. Under both treatments, both the outer and inner leaf temperatures followed the air temperature till 11:00 am, while at noon all

of them were below the air temperature of 20°C. At the same time, inner and outer leaf temperatures under LED I were just a bit lower than under LED II (0-0.5°C).

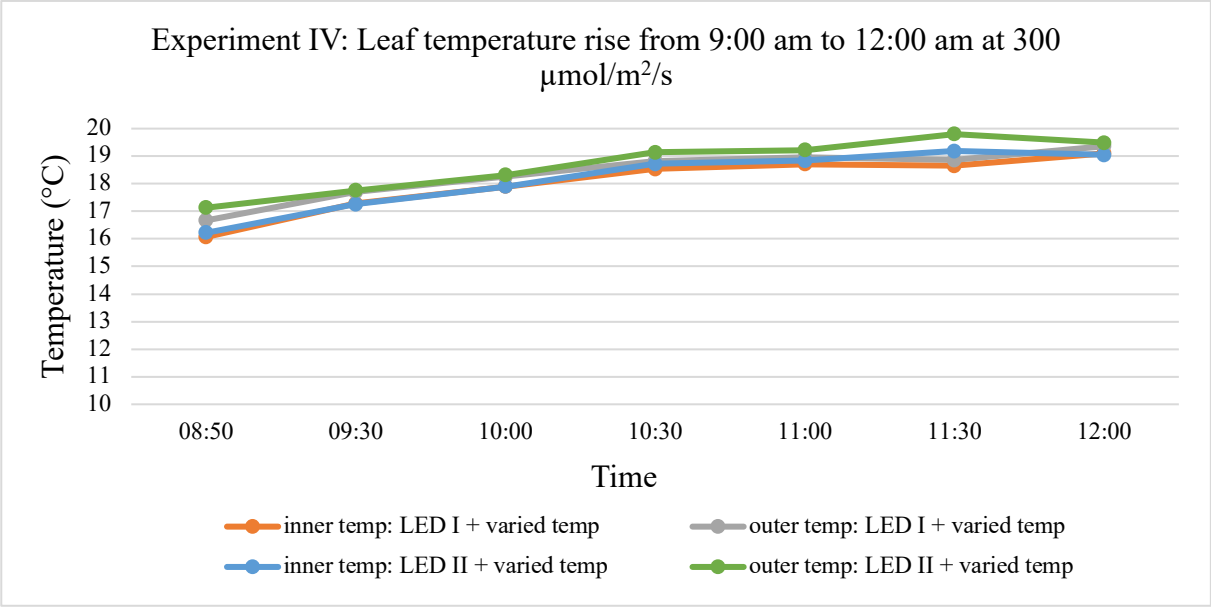


Figure 37: Average inner and outer leaf temperature rise from 9:00 am to 12:00 pm on lettuces grown with LED I compared with LED II as light sources, and the varied temperature treatments of DT/NT: 20/13°C (ADT: 17.41°C) at a PPFD of 300 µmol/m²/s. Lamps were turned on at 9:00 am. N=5.

Figure 37 shows the inner and outer leaf temperature rise from 9:00 am to 12:00 am at higher light intensity for experiment IV. Not much gap (0.2-0.6°C) existed between inner and outer temperature under both treatments which further lowered with time. Outer leaf temperature at 11:30 am increased above the trend for LED II+ varied temperature, which at noon again decreased a bit. All the leaves were at 19°C at noon. Further, the inner and outer leaf temperatures in LED II were just higher by 0.13°C and 0.31°C on average than that of LED I (data not shown).

5. Discussion

5.1 Growth and morphology

The experiments were conducted to gain insight into climate optimization for crisphead lettuce by testing three different lamp types (traditional HPS and two types of LEDs) at moderate (150 µmol/m^2/s) and high light intensities (300 µmol/m^2/s) with standard (DT/NT 20/18°C) and varied temperature conditions (DT/NT 20/13°C). The aim was to study the growth and morphology of plants together with the tipburn occurrences to come up with relevant advice for commercial lettuce growers in Norway. To save energy, it is of interest to replace HPS with LEDs, but the optimal spectral content for crisphead lettuce is not known. Furthermore, reducing the NT will lead to energy savings because less heating is required in periods when the outdoor temperature is low. Hence, one sub-goal was to investigate if the LED lamps could replace the HPS lamps in the greenhouse and if the NT could be reduced without any adverse effects on growth and tipburn.

In experiment I, there was no significant difference ($p>0.05$) at the moderate light intensity (150 µmol/m^2/s) when lettuces were grown for two weeks under HPS+ standard temperatures (ADT: 19.33°C) (Table 14). This was expected since the seedlings were grown under the same lamp type with a similar standard temperature of DT/NT: 20/18°C. The leaf unfolding rates (Leaves/day) and dry weight/leaf were all identical during the two first weeks (Table 29). When the temperature in one of the chambers was changed to varied (DT/NT 20/13°) and compared with standard temperature (DT/NT 20/18°C), and later the light intensity was increased in both chambers, a significantly higher number of leaves from higher leaf unfolding rates was found in plants cultivated with standard temperature than in varied temperature, but more elongated leaves were found in varied temperature regime. This shows that the number of leaves and length of the longest leaf are affected by temperature. Higher ADT in standard temperature is probably responsible for the higher number of leaves. This result corresponds to the fact that the number of leaves produced per day depends on ADT. The leaf production rate is usually higher under a higher ADT and lower under a lower ADT and is not determined by DT or NT (Bensink, 1971; Moe & Heins, 1989; Walters & Lopez, 2021). As for leaf elongation, a lower ADT induced longer leaves at a higher light intensity which is opposite to what is usually found: more elongated leaves are obtained for treatment having higher ADT compared with lower ADT at high light intensities (Bensink, 1971). However, it could be noted that the varied temperature (DIF ≥ + 6°C) results in a stronger positive DIF than in standard temperature (DIF

= + 2°C). Positive DIF induces more elongation in the leaf length (Myster & Moe, 1995). Further, LURs were higher at higher light intensities than at lower light intensities, which are similar to what was found by (Bensink, 1971). No significant changes were observed for fresh and dry weight, and not much difference was in dry weight/leaf (Table 15 & 29). Similar results were obtained by Lund et al. (2006), and Ottosen et al. (2004), where dry matter accumulation was almost similar under dynamic climate control with lower night temperature compared to higher night temperature in ornamentals. Ottosen et al. (2003) found a similar case in sweet pepper where lowering night temperature did not affect the dry matter accumulation of fruits in bell peppers.

Likewise, when LED I was compared with HPS at a PPFD of 150 µmol/m^2/s for the first two weeks with the same standard temperature (DT/NT 20/18°C), more leaves were obtained with HPS. This shows that HPS is inducing more leaves than LED I at standard temperature. As already explained, ADT plays a significant role in LURs. However, leaf temperature may have played a role here. Bergstrand et al. (2016) found a higher leaf temperature of plants exposed to HPS by (0.9-1.3) than in LED. The same could have happened in the present experiment. Fig 30 shows that the leaf temperature in plants exposed to HPS was always higher than in LED in a range from 1.53-2.57°C for both inner and outer leaves after the temperature was varied at 150 µmol/m^2/s during the time it was noted. No difference in biomass accumulation and leaf elongation was observed during the two first weeks of cultivation. But when temperatures were changed to varied temperatures and later when light PPFD was also increased to 300 µmol/m^2/s (DT/NT 20/13°C) in both chambers, 28.8% higher fresh weight with higher dry weight and longer leaves was obtained in LED I than in HPS (Table 18). Similarly, the leaves exposed to LED showed 37.93% higher dry weight accumulation/leaf than with HPS. The lower R/FR ratio of 2.89 in LED I with more blue and red than in HPS lamps (R/FR ratio: 3.8) could have brought this result. Red and blue increases plant photosynthetic capacity and biomass accumulation and improve the growth (Hogewoning et al., 2010; Yang et al., 2017). Low R/FR ratio causes shoot elongation, leaf expansion, increased biomass accumulation, and nutrient allocation to the shoots (Demotes-Mainard et al., 2016; Morgan et al., 1980). Meinen et al. (2012) also found higher dry matter in cucumber and tomato under a good combination of blue, red, and far-red LED than in HPS. Further, there was no difference in leaf number because the LURs were equal in both treatments (Table 29). Since even under lower ADT, LED I performed better than HPS, the results obtained show that LED I can replace HPS as an effective source for cultivation in growth chambers even under varied

temperature conditions. The results clearly show that the growth potential is higher for LED I compared with HPS, even if the leaf temperature is almost 1.5°C higher for plants exposed to HPS due to the high infrared radiation. There is also a possibility of increasing dry matter accumulation per leaf with LED I. In conclusion, the lettuce is very responsive to light quality as a parameter to improve growth, and the production potential is improved with the use of LED I.

LED I had a better combination of blue, red, and far-red wavelengths with more red and far-red wavelengths and less blue as compared to LED II (Fig 12 & Fig 13). More blue light suppresses the plant growth and gives compact plants with lesser dry weight (Mortensen & Strømme, 1987). The higher effectiveness of LED I could be reflected by higher fresh and dry weights and longer leaves under both lower and higher light intensities compared with LED II. However, an equal number of leaves were obtained on both lamps at moderate light intensities with similar leaf unfolding rates (Table 21 & 22 & 29). But a greater number of leaves with higher LURs were there in LED I than in LED II at higher light intensity. This shows that more leaves are formed in higher light intensities, which are similar to the finding of Kang et al. (2013). The effect to be seen more under LED I could be from the more red spectrum in it. But the effect of light quality on the number of leaves is unclear and seems to be species dependent as red light gave more number of leaves in tomato, blue did so in *Alternanthera* but reduced the number in cucumber (Cao et al., 2016; Hernández & Kubota, 2016; Macedo et al., 2011). Further, red LED gave more leaves in strawberries than in the combination of blue and red LED (Meng, L. et al., 2019). The dry weight accumulation/ leaf was also higher with LED I lamps, especially at higher light intensities than at moderate light intensities which were same for all treatments. Because of some technical problems in the chambers in experiment III, the plants were only grown for five days under higher light intensities, so the results obtained from it are incomplete, and the plants were not fully developed. But it still shows LED I is better than LED II.

Exact similar results were obtained when the temperature was varied in experiment IV under high light intensity with the same LED I and LED II lamps (Table 25&26). Despite the lower ADTs in varied temperature conditions, growth was not much hampered. At both moderate and high light intensities, neither was there much difference between the inner and outer leaf temperatures in either of the treatments, nor was there much difference between inner-inner and outer-outer leaves in between the two treatments. In all cases, the difference was less than

1°C (Fig 36&37). The more ineffective the light spectrum, the more the leaf gets heated. Even among LEDs, the one with the most effective spectrum heats the leaves less (Dannehl et al., 2021; Fender, 2017). However, based on the leaf temperature rise, since there were similar leaf temperatures under both lamps, one cannot be considered to have a less effective spectrum than the other.

Nevertheless, as the growth and morphology of plants are considered, lettuces will give better growth and yield under LED I lamps compared with the other two lamp types used even under lowered night temperatures. Taken together, the growers will have the potential to save energy by choosing LED I and varied temperature.

Table 29: Summary of leaf unfolding rates (LUR) and dry weight/leaf from experiments I to IV. LUR was calculated by dividing the total number of leaves by the total number of days the plants were grown under the respective treatment. Dry weight/leaf was calculated by dividing the dry weight of the plant by the total number of leaves.

		Leaf unfolding rates (no. of leaves/day)		**Dry weight/leaf (gm/leaf)**	
		150 µmol/m^2/s	300 µmol/m^2/s	150 µmol/m^2/s	300 µmol/m^2/s
Experiment I	**HPS+varied temperature**	0.69	0.86	0.09	0.48
	HPS+standard temperature	0.73	0.94	0.09	0.44
Experiment II	**LED I + varied temperature**	0.66	0.76	0.07	0.40
	HPS + varied temperature	0.74	0.77	0.06	0.29
Experiment III	**LED I + standard temperature**	0.77	1.88	0.16	0.27
	LED II + standard temperature	0.76	1.36	0.15	0.21
Experiment IV	**LED I + varied temperature**	0.7	0.81	0.10	0.36
	LED II + varied temperature	0.66	0.69	0.09	0.28

5.2 Tipburn

At moderate light intensity, the tipburn data was taken after two weeks of growth, but very little outer tipburn was recorded (tipburn score ≈ 2), and no inner tipburn was observed (results not shown). Knoop (2019) confirmed the increase in severity of inner tipburn with the higher light intensity (300 µmol/m^2/s) in `Frillice´. At higher light intensity, severe inner tipburn was found along with more outer tipburn in all experiments in this thesis. Sago (2016) also saw an increase in tipburn severity with an increase in light intensity with HPS from 150 to 300 µmol/m^2/s. Islam et al. (2004) further stated that lower intensity does not always prevent tipburn while using four different light intensities from 60 to 240 µmol/m^2/s with HPS.

Lee et al. (2013) reported lower tipburn under lower DT at first but an equivalent level of tipburn to that of higher temperature later till harvest while using 18, 22, and 25°C as constant DTs. Instead, horizontal airflow significantly reduced tipburn compared to lower temperature but decreased the fresh weight and leaf area. However, Lee et al. (2019) reported that lower temperatures (18/14 °C DT/NT) decreased the tipburn even under high light intensity (250 µmol/m^2/s) compared to the higher temperature. Depending on the temperature regime, no effect of temperature on tipburn was also reported by Knoop (2019). In his study, the high temperature was tested (>25°C), but this did not influence the tipburn incidence. In the first experiment of this thesis, the leaf temperatures during the morning were higher under standard temperatures than under varied temperatures with HPS lamps. The more inner tipburn *(<0.05)* and more outer tipburn *(>0.05)* with varied temperatures than with standard temperatures under HPS lamps in experiment I is therefore difficult to explain, but more inner tipburn can occur due to guttation in the inner leaves at the shoot apical meristem, but it was not visible during cultivation (Curtis, 1943). Low NT can lead to a higher root pressure and, consequently, more guttation (Singh, 2016). In the study of Frantz et al. (2004), air movements on the shoot apical meristem were found to reduce tipburn. According to Aizarani (2021), there was a lesser tipburn in high light intensity of 300 µmol/m^2/s under DT/NT of 20/14°C than at 18/17°C.

Several studies compare LED and HPS to investigate production potential for lettuces, and some studies focused on tipburn development. But the studies on temperature variation and tipburn are missing. A proper combination of irradiance and light spectrums can improve lettuce's growth and morphology as shown by the results. LED I induced better growth compared to both HPS and LED II in the present experiment. However, it is well known that

an increase in growth rate also increases the tipburn occurrence in lettuce (Cox et al., 1976). The present experiment is in line with this as higher inner and outer tipburn were found under LED I+ varied temperatures than HPS+ varied temperatures though a significant difference was only found in the outer tipburn between the two treatments (Fig 28). Further, Kleemann (2002) found decreased tipburn incidence in leaf lettuce when far-red light was filtered from the light source using far-red absorbing plastics. Comparatively, the LED I had higher far-red than HPS, which can also be one of the causes of higher tipburn in it. However, a striking difference in HPS and LED I was the peak in red (660-670 nm). Red light together with blue will lead to very efficient photosynthesis. On the other hand, lettuce is a shade plant, and too much light will lead to photoinhibition and ROS formation (Ruangrak & Khummueng, 2019; Willey, 2018). One point to note is also that LED I showed lower leaf temperatures than HPS. The light quality of LED I is probably inducing more tipburn because of light stress and ROS accumulation and the lower temperature seems not to be much effective on reducing tipburn severity.

As of the third experiment, the lettuces were only grown for twenty days with standard temperature in both LED I and LED II, but severe inner and outer tipburn was observed on both with insignificant differences between them (Fig 32&33). Though insignificant, the inner tipburn under LED I was higher, which could be because of a greater number of leaves in LED I, and the more red and far-red in it could also be the reason. As of experiment IV, there was severe outer and inner tipburn, but outer tipburn was significantly higher in LED I than in LED II (Fig 34&35). Higher growth rate and more red in LED I than LED II can be the reason behind the higher tipburn in LED I, as mentioned earlier. The amount of tipburn seen in these last two experiments was so much that the entire product could be discarded. Also, the leaf temperatures under two LEDs did not show much difference. Though the temperatures were varied in the fourth experiment and no difference in leaf temperatures between the two were noticed, the high tipburn scores obtained show that light intensity and light quality are more critical factors in tipburn severity than the temperature (leaf/air).

Growers sometimes observe condensation on the inner leaves in the morning (pers.com P.O. Espedal), and it was hypothesized that too low NT would lead to condensation of the inner leaves and, therefore, more inner tipburn. However, no condensation was observed on the inner leaves in any of the experiments. According to calculations using the Mollier diagram, the dew point was found to be between 8 and 10°C in the early morning after the light was turned on.

A leaf temperature above 12°C was measured in all conditions with varied temperature treatments, which are far from the expected dew point. In a commercial greenhouse, the air volume is larger than in the small chambers used in these experiments, and it takes a longer time to increase the air and leaf temperature in a greenhouse than in a growth chamber. It is likely that condensation can occur in a commercial greenhouse if the NT goes too low, especially when the growers are not using energy curtains and the sensible heat loss is too high (Stanghellini et al., 2019).

In conclusion, varied temperatures could not reduce the tipburn in combination with high light intensity. Instead, inner tipburn was seen in all experiments, and it was more was under the LED I lamp. Based on the previous literature and findings and the current results from this thesis, temperature regimes do not affect the tipburn severity much, but the light intensity and quality are the more critical factors. Varied temperatures under moderate or low light intensity were not tested in these experiments, but this could be something that will further confirm if varied temperatures will be beneficial or not. From these results, it can be concluded that the temperature should not be too low, and probably not lower than 13-14°C depending on the season and climate control in the greenhouse and this can be done together with moderate light intensity. Further, LED I can be tested so that growth can also be improved.

5.3 Minerals

The relation between Ca and tipburn has been a topic of interest for a long time. Tipburn leaves have a lower concentration of Ca as compared to leaves with no tipburn. It is the result of localized Ca deficiency in the edges of the young growing leaves (Barta & Tibbitts, 2000; Saure, 1998). In the present study in experiment I, a higher Ca level ($p<0.05$) was seen in the outer leaves of HPS and standard temperatures which also had lesser outer tipburn ($p > 0.05$) than in HPS and varied temperatures (Table 16 & Fig 25). However, the inner leaves had significantly higher inner tipburn in HPS + varied temperature but had similar levels of Ca in both the treatments. The results of K were similar to that of Ca, but the content of Mg was significantly equal in the outer leaves of both treatments. Cations compete with each other to be taken up and their high content in the nutrient solution can lead to lower uptake and changed Ca distribution and can cause Ca deficiency (Olle & Bender, 2009). However, our result shows no such increment or decrement of Ca with respect to Mg and K. In inner leaves, also Mg was higher in standard and lower in varied; as for K, they were in equal amounts.

Though the outer tipburn under LED I and the varied temperature was higher than in HPS and varied temperature, in outer leaves, no difference in Ca between them but lower Mg in LED I and the varied temperature was found, which is contrasting to the fact that tipburn leaves have lower Ca and higher Mg (Paiva et al., 1998). Equal levels of K here show no role of K in tipburn, which is similar to the finding of Bres and Weston (1992), who used different K concentrations but found no effect on lettuce tipburn (Table 20 & Fig 29). In inner leaves, all the minerals were significantly similar, and so was the tipburn.

In experiment III, when LED I was compared with LED II under standard temperatures, no significant difference was observed in tipburns, and similar was the case for minerals. Outer leaves always had higher mineral contents than inner leaves, probably because the transpiration rate is higher for outer than inner leaves (Table 24 & Fig 33).

However, in experiment IV, when LED I was compared to LED II under varied temperatures significantly higher outer tipburn was found under LED I with no difference in the case of inner tipburn. Here, the Ca level was lower in LED I and varied temperature than in LED II and varied temperatures, which was according to the earlier findings mentioned above. As for Mg, K and Mn, they were all higher in outer leaves with equal amounts under both treatments and lower in inner leaves, which were also equal in both treatments. Fe was in equal amounts in all inner and outer leaves which is surprising (Table 28 & Fig 35). A higher level of Fe in the outer leaves than in inner leaves was found by Mou and Ryder (2002) in crispy and romaine lettuce while investigating the relationship between nutritional value and head structure of lettuce. Baslam et al. (2013) also found higher Fe in outer leaves than in inner leaves in green and red pigmented lettuces. In all experiments, Mn was always higher in outer leaves and lower in inner leaves with no difference between the treatments.

Since non-conclusive relations were observed between the minerals and tipburn occurrences in the experiments, to understand better which mineral plays a significant role, best subsets regression was performed separately for inner and outer tipburn for all four experiments (Table 30&31). The inner tipburn could not be explained clearly from the effect of a single cation; no role of Ca was surprising to see. The highest R^2 (62.9) was obtained when all five cations were included in the model. Similar was the case for outer tipburn. The R^2 value was quite low for all cations combinations, and even when all the five cations were included in the model, R^2

was still lower than 60. Therefore, the minerals seem to have no definitive role in the tipburn occurrences in these experiments altogether. Similar results were found by Su et al. (2016) where no correlation between tipburn severity and endogenous Ca, and with other cations like Mg, Fe, Zn, Mn, Na, and K were found in Chinese cabbage grown in Hoagland´s medium.

Table 30: Best subsets regression: inner tipburn score versus selected minerals from experiments I to IV.

Vars	R^2	R^2-adjusted	R^2-predicted	Mallows Cp	S	Ca	Mg	K	Fe	Mn
1	48.3	39.6	10.4	-1.2	0.40302		X			
1	25.4	13	0	0	0.48381			X		
2	56.5	39.1	0	0.3	0.40496		X	X		
2	48.4	27.8	0	0.8	0.44071		X		X	
3	62.6	34.5	0	2	0.4197		X	X	X	
3	61.4	32.4	0	2.1	0.42649		X	X		X
4	62.9	13.4	0	4	0.48275	X	X	X	X	
4	62.7	13	0	4	0.48377		X	X	X	X
5	62.9	0	0	6	0.59108	X	X	X	X	X

Table 31: Best subsets regression: outer tipburn score versus selected minerals from experiments I to IV.

Vars	R^2	R^2-adjusted	R^2-predicted	Mallows Cp	S	Ca	Mg	K	Fe	Mn
1	15.1	0.9	0	0.2	0.36099		X			
1	12.8	0	0	0.3	0.3658	X				
2	48.4	27.8	0	0.6	0.30816	X		X		
2	26.1	0	0	1.7	0.36895	X				X
3	49.7	11.9	0	2.5	0.34043	X		X		X
3	48.7	10.1	0	2.6	0.34381	X		X	X	
4	50.3	0	0	4.5	0.3905	X		X	X	X
4	50.1	0	0	4.5	0.39129	X	X	X		X
5	59.9	0	0	6	0.42961	X	X	X	X	X

5.4 Leaf temperature

In all the three experiments in which leaf temperatures were taken, the outer leaf always had higher leaf temperatures than the inner leaves. This means inner leaves take a slightly longer time to warm up than the fully exposed outer open leaves. In experiment I, the gap between

inner and outer leaves was higher in HPS+ varied temperature than in HPS+ standard temperature which further narrowed in higher light intensity. Experiment two with HPS+ varied temperature and LED I+ varied temperature confirms that leaf temperature under HPS always is higher than that under LEDs. Higher light intensity narrowed the gap between inner and outer leaf temperatures than at moderate light intensity; however, for the fourth experiment, there is not much difference in the gap between the two light intensities. This makes it hard to quote a concrete conclusion of how the leaves' temperatures are affected by the treatments at two different light intensities.

5.5 Practical implications

LED I can replace HPS because its spectral compositions improve the growth potential of lettuce. However, it is important to grow the lettuces at moderate light intensity. In the present study, 300 µmol/m^2/s induced both inner and outer tipburn. The optimal light intensity with LED I needs to be tested and will probably vary with the season and age of the plants. In all experiments, 150 µmol/m^2/s was used when the plants were young, and very little tipburn was observed at this stage. As of lower night temperature, it is clear that growth would not be much hampered, but condensation can still be a problem. Though no condensation was seen in the growth chambers, it’s likely that there will be some in greenhouses because the air volume in a greenhouse is huge, and it takes more time to get heated up than the growth chambers. Further, the test needs to be done in a commercial greenhouse. But for now, night temperatures not lower than 13-14°C are recommended.

6. Conclusions

- The varied temperature did not reduce the severity of tipburn in combination with high light intensity.
- Temperature variation with a lower night temperature (DT/NT 20/13°C) caused more tipburn than standard temperature (DT/NT 20/18°C) under HPS lamps.
- Higher irradiance caused a higher tipburn score, and more outer and inner tipburn were observed under higher light intensity. Lamp type had a stronger effect on tipburn than the temperature regimes.
- Leaf temperature was higher with HPS than with LED, and inner leaves' temperature was lower than outer and increased more slowly when the light was turned on.
- LED I with 10% blue, 41% red, and 13% far-red resulted in higher biomass compared with other lamp types tested but induced more tipburn.
- Neither Ca, nor other cations in the outer edge of old leaves or young inner leaves were found to correlate with outer or inner tipburn score.

7. References

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