

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Root Growth, Morphological and Physiological Characteristics of Subtropical and Temperate Vegetable Crops Grown in the Tropics Under Different Root-Zone Temperature

Jie He

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/64824>

Abstract

Root growth and morphology are important for maximizing water uptake and mineral absorption. Similar to the plants grown in the soil, in a soilless culture such as an aeroponic system, the amount of water and nutrient available to a plant is determined by the root surface area and volume of nutrient solution with which its roots are in contact. Furthermore, plant roots can alter their nutrient acquisition capacity by adjusting their morphological and/or physiological characteristics to meet changes in shoot nutrient demand in response to environmental stress. Subtropical and temperate vegetables have successfully been grown aeroponically in the tropics by simply cooling the root zone (RZ) while their aerial portions are subjected to fluctuating atmospheric temperatures. This paper focused on RZ temperature (RZT) on root and shoot growth, and root morphology of subtropical and temperate vegetable crops grown in the tropics. The impacts of RZT on water relations as well as nitrate (NO₃⁻) uptake and assimilation of these vegetable crops were also discussed.

Keywords: nitrate uptake and assimilation, root morphology, root-zone temperature, water relations

1. Introduction

Plants are photoautotrophic and manufacture their own food through the process of photosynthesis. However, they must acquire water and minerals from the environment for photo-

synthesis to occur. While CO_2 comes from the atmosphere for photosynthesis, plants obtained water and minerals from the soil or other growth media via their roots. Plants are sessile and adapt their morphological structures to the encountered environmental conditions. The morphology of the root system such as root length, number of root tip, root diameter, root surface area, and root volume varies greatly depending on the plant species, soil composition, and water and mineral nutrients availability [1, 2] and root-zone temperature (RZT) [2, 3]. Root plasticity provides the sessile plants to adjust their structure to environmental conditions as they change [1]. Changes in root morphology are important for maximizing water uptake [1, 3, 4] and mineral absorption to alleviate stress of nutrient deficiency [4–7]. Furthermore, plant roots alter their nutrient acquisition capacity not only by adjusting the morphological characteristics but also the physiological performance such as nitrate (NO_3^-) uptake and assimilation to meet changes in shoot nutrient demand in response to environmental stress [2, 4].

Root morphology and physiology are closely associated with the growth of the aerial parts and shoot productivity [2, 3, 8]. For instance, plant growth and productivity are often limited by high temperature and thus restrict the growth of subtropical and temperate crops in the tropics. However, since 1997, our research team has successfully grown certain subtropical and temperate crops in Singapore by simply cooling their roots while their aerial portions are subjected to hot fluctuating atmospheric conditions [9, 10]. Working on subtropical and temperate vegetable crops grown in the tropics, in this paper, the author first focused on effects of RZT on root morphology. The impacts of RZT on water uptake and water relations as well as NO_3^- uptake and assimilation were also discussed.

2. Effects of root-zone temperature on root morphology

Root systems have higher ratios of surface area to volume that effectively explore a larger volume of soil [11]. Similar to the plants grown in the soil, in aeroponic systems, root surface area determines the amount of water and mineral uptake [2, 12, 13]. As root systems are responsible for acquisition of water and mineral nutrients, it is not surprising that root morphology is highly influenced by rhizosphere environments such as RZT [2].

When the plant is exposed to high temperatures, root development is adversely affected [14]. However, manipulating RZT alone has a great influence on root development, perhaps even greater than shoot temperature manipulation [15–18]. Because of the changes of RZT, there is great variation of root morphology, especially root length which is a more sensitive indicator of RZT impact compared to root biomass. For example, grown under optimum RZT of 30°C , root length of rape (*Brassica napus* cv. “Emeralk”) was fivefold longer compared to those grown under superoptimum RZT of 35°C . However, there was only twofold difference in root biomass, which was found between the plants grown under two different RZTs. Higher root length/weight biomass ratio was due to the smaller diameter of the successive orders of lateral roots [15]. It was reported that in both heat tolerant and sensitive clones of potato (*Solanum tuberosum* L), smaller number and shorter length of lateral roots were observed at 30°C RZT than 20°C RZT resulting from a reduction of cell division rate and followed by cessation of cell

elongation in roots [16]. It was found that maximum seminal root elongation and first-order lateral root initiation and elongation of Sorghum (*Sorghum bicolo*), occurred at 25°C but they were severely inhibited at 40°C [17].

We have found that subtropical and temperate vegetables grown in the tropical greenhouse under cool RZTs (C-RZTs) established much bigger root systems compared to those subjected to hot ambient RZT (A-RZT) [2]. For instance, the total root length of capsicum plants (*Capsicum annuum* Indra F1-hybrid) at C-RZT increased about 400 cm in 2 weeks during the experimental period (**Figure 1A**). When plants were transferred from C-RZT to A-RZT (C → A-RZT), increases in their total root length seemed to stop compared to C-RZT plants [19]. Capsicum plants grown at A-RZT did not show any increases and had the lowest total root length during the entire experimental period. However, after transferring capsicum plants from A-RZT to C-RZT (A → C-RZT), linear increase in total root length was observed.

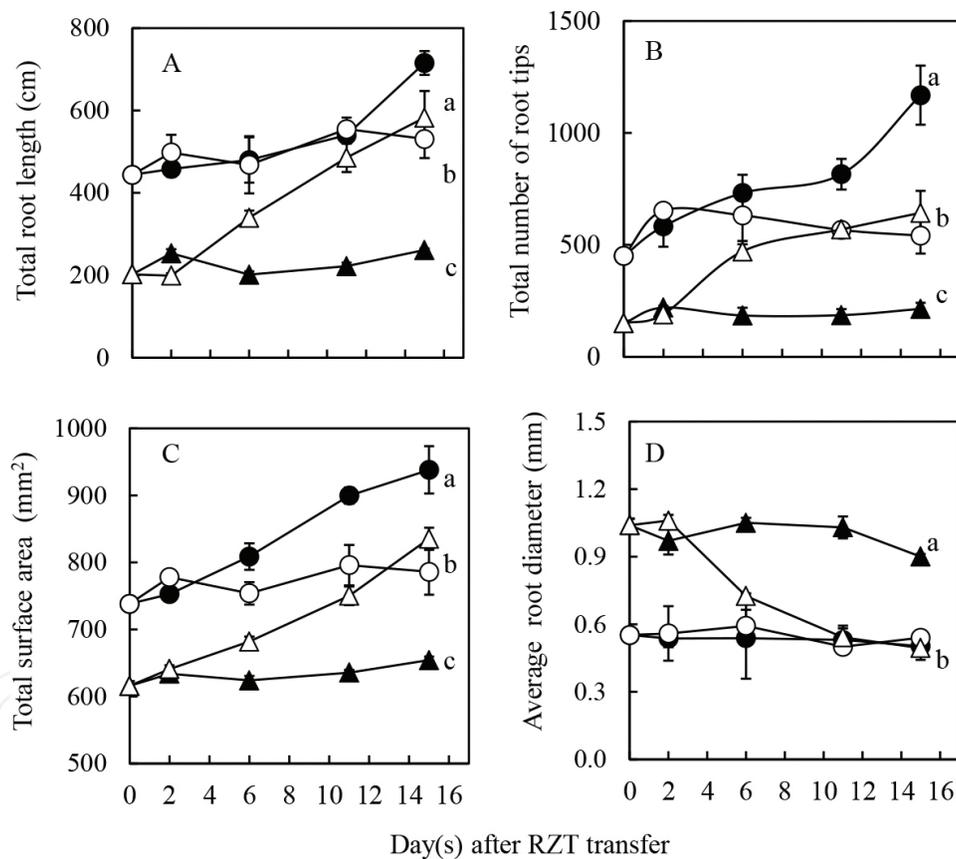


Figure 1. Total root length (A), total number of root tips (B), total surface area (C), and average root diameter (D) of capsicum (*Capsicum annuum* Indra F1-hybrid) during different RZT treatments: C-RZT (●), A-RZT (▲), C → A-RZT (○), and A → C-RZT (Δ). RZT treatments were started 3 weeks after transplanting. Means with different letters are statistically different ($P < 0.05$; $n = 6$) as determined by Tukey's multiple comparison test. Redrawn from Tan [19].

Differences in total number of root tip (**Figure 1B**) and total surface area (**Figure 1C**) among the four different RZT treatments were similar to those of total root length (**Figure 1A**). Compared to plants grown at A-RZT, the average root diameter of C-RZT plants was much

smaller (**Figure 1D**). It is surprising to note that there were no increases in root diameter after transferring plants from C-RZT to A-RZT (C → A-RZT) as capsicum plants were similar to those of C-RZT plants throughout the experiment. However, A → C-RZT transfer resulted in the development of new and finer roots in A → C-RZT plants. Thus, the average root diameter of A → C-RZT capsicum was significantly smaller than A-RZT by day 6 after RZT transfer (**Figure 1D**). Similar to capsicum plants, lettuce grown at C-RZT showed linear increase in total root length over the 2-week period (**Figure 2A**). Unlike the capsicum plants (**Figure 1A**), increase in total root length was also observed in C → A-RZT lettuce plant and the increase rate was similar to C-RZT lettuce plants from day 0 to day 8 after RZT transfer. However, C → A-RZT lettuce plants maintained their total root length of about 600 cm from day 10 to day 14 after RZT transfer. There was no significant increase in total root length of A-RZT lettuce over the 2-week experimental period. Again, unlike the capsicum plants (**Figure 1A**), A → C-RZT lettuce exhibited increase in total root length only after 8 days of RZT transfer (**Figure 2A**). The total root tip number of C-RZT lettuce was consistently higher than lettuce grown at other RZTs (**Figure 2B**). Unlike the capsicum plants (**Figure 1B**), there was an increase in total root tip number for C → A-RZT lettuce although the net increase decreased significantly

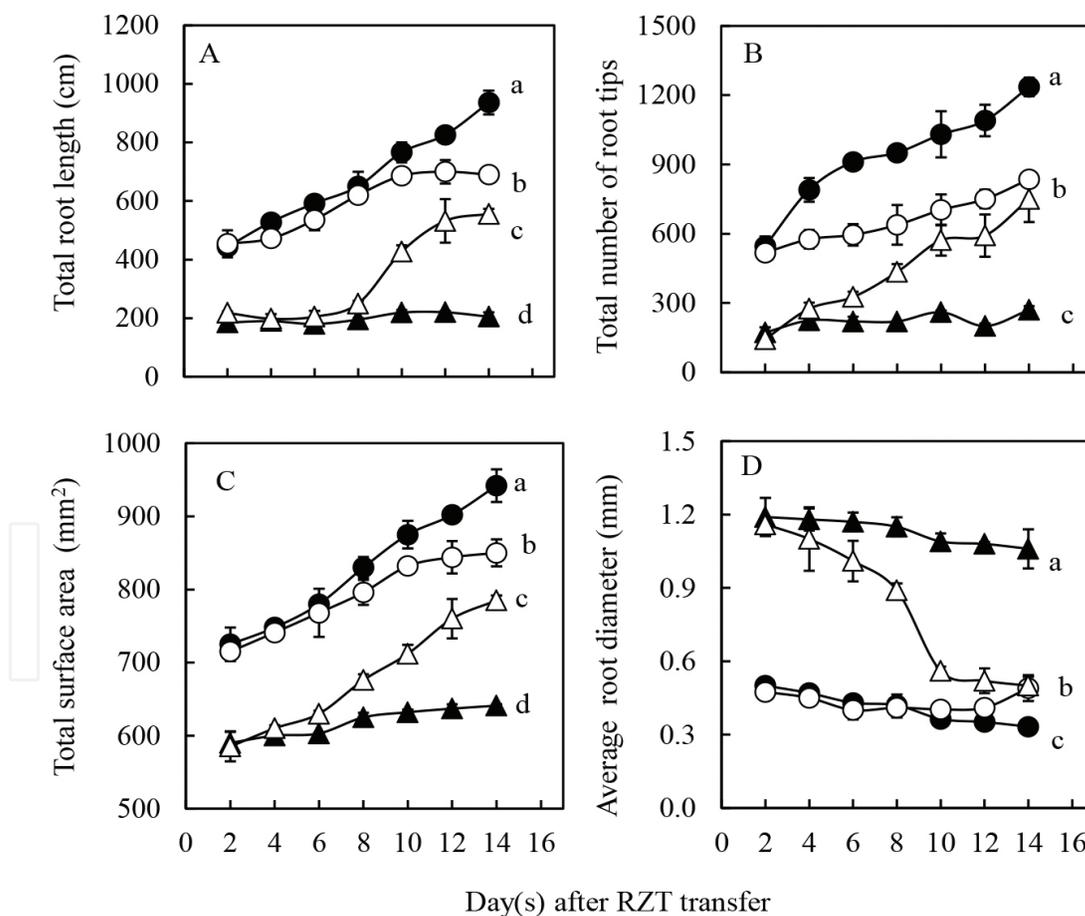


Figure 2. Total root length (A), total number of root tips (B), total surface area (C), and average root diameter (D) of lettuce (*Lactuca sativa* L. “Panama”) during different RZT treatments: C-RZT (●), A-RZT (▲), C → A-RZT (○), and A → C-RZT (△). RZT treatments were started 3 weeks after transplanting. Means with different letters are statistically different ($P < 0.05$; $n = 6$) as determined by Tukey’s multiple comparison test. Redrawn from Tan [19].

after C → A-RZT transfer (**Figure 2B**). The total root tip number of A-RZT lettuce did not change significantly throughout the whole experiment. When lettuce was transferred from A → C-RZT, there was rapid increase in the total root tip number. The root tip number of A → C-RZT lettuce became significantly higher than A-RZT lettuce by day 6 after RZT transfer. At day 14 after RZT transfer, total root tip number of A → C-RZT lettuce was 2.8 times higher than A-RZT plants. Responses of total surface area of lettuce plants to different RZTs (**Figure 2C**) were similar to those of total root length (**Figure 2B**). Similar to those of capsicum plants (**Figure 1D**), lettuce grown at A-RZT had the highest average root diameter while C-RZT plants had the least (**Figure 2D**). The average root diameter of C → A-RZT lettuce remained similar to C-RZT plants during the first 10 days of RZT transition. However, unlike capsicum plants (**Figure 1D**), there was slight root thickening in C → A-RZT lettuce roots after 2 weeks of RZT transfer. By day 14 after RZT transfer, the average root diameter of C → A-RZT lettuce was much thicker than C-RZT plants. Similar to those of capsicum plants, A → C-RZT lettuce, developed new and finer root (**Figure 2D**).

Root morphological analysis of both subtropical and temperate vegetables revealed that high RZT inhibited root elongation, branching, and hair formation but increased root diameter. These were also observed in other plant species [16, 17]. However, effects of RZT transfer on morphology were different between capsicum and lettuce plants (**Figures 1 and 2**). These finding suggest that effects of RZT on root morphology is species-dependent. It was also reported that root length and diameter appeared to be inversely related to a study using *Secale cereale* seedlings [20]. Root thickening, or an increase in diameter, was controlled through signals emanating from shoot apices and root tips [21]. Root thickening may also be accompanied by associated changes in microfibril angles within expanding cell walls [21]. The chemical signals involved in root thickening may be ethylene [22]. The role of ethylene in inhibition of root elongation and root thickening was further confirmed by our team [13]. Our recent study with a recombinant inbred line (RIL) of lettuce and its parental lines (*Lactuca serriola* × *Lactuca sativa* “Salinas”) that were grown in a tropical greenhouse under 24°C-RZT and hot A-RZT showed that higher RZ ethylene concentrations accumulated in A-RZT plants compared to that of 24°C-RZT plants. Lowest RZ ethylene concentration corresponded with highest shoot fresh weight [23]. Our results indicated that the presence of an ethylene inhibitor promoted root elongation at high RZT of 38°C. Without ethylene inhibitor, root elongation at high RZT was significantly inhibited. Our previous ¹⁴C feeding experiments implied that both capsicum and lettuce grown under C-RZT indeed had higher assimilation rates of ¹⁴C and their younger developing leaves exhibited greater sink strength [24]. Further studies concluded that plant growth was the result of interaction between source leaves and carbon partitioning among competitive sinks [25–27]. It was interesting to note that A-RZT lettuce had higher fresh weight and dry weight root/shoot ratio than that of C-RZT lettuce [19, 23]. This suggested that more photoassimilate may be distributed to the lettuce roots under hot A-RZT conditions. The RZT transfer experiments confirmed that A-RZT induced greater levels of ¹⁴C delivered to the lettuce root system. However, it was surprising that the high ¹⁴C translocated to the roots of A-RZT lettuce which was not accompanied by a greater root development [19, 24]. This may be attributed to the higher respiration rates in the roots which may require energy for the active uptake of water and nutrients in a poorly developed root system [26, 28]. The high rate of

respiration may have taken place at the expense of root development. It is highly likely that there exists a negative feedback mechanism among root respiration, water uptake, nutrient absorption, root morphology, and high RZT. Our results showed that capsicum grown at C-RZT had lower root/shoot ratios than A-RZT plants [19]. C-RZT resulted in longer total root length, greater total root tip number, larger root surface area, and smaller root diameter in both lettuce and capsicum plants. However, the trend of the capsicum root/shoot ratios was different from that of lettuce because the capsicum grown at C-RZT had higher root/shoot ratio than A-RZT capsicum [19]. C → A-RZT transfer also caused a decline in root/shoot ratio while A → C-RZT resulted in an increase in root/shoot ratios of capsicum. These suggested more photo-assimilate partitioning to roots of C-RZT capsicum and less so for A-RZT capsicum. This was further supported by the ^{14}C feeding experiments which showed that more ^{14}C was found in the roots of C-RZT capsicum compared to the roots of A-RZT capsicum. The transfer from C → A-RZT also resulted in decreased ^{14}C found in C → A-RZT capsicum roots. The reverse was demonstrated in A → C-RZT capsicum which had higher root ^{14}C than A-RZT plants. Therefore, lettuce grown at A-RZT distributed more ^{14}C to their roots while capsicum grown at C-RZT distributed more ^{14}C to the roots [24].

3. Effects of root-zone temperature on water relations

Changes of root morphology are important for maximizing water and dissolve mineral uptake [1, 11, 29–31]. Plant roots can alter not only their morphological but also their physiological characteristics to meet changes in shoot water and nutrient demand in response to environmental stress [29]. Manipulation of RZT resulted in changes of root morphology, water, and mineral uptake and transport [12, 32–35]. For plants grown in soil, high RZT causes not only poor root growth and development [36] but also results in spatial water and mineral nutrient availability in soil [37] and reduces uptake efficiency per unit root length [36, 37]. In our aeroponic system, plant roots are continuously sprayed with nutrient mist, and there is no spatial variation in water and mineral nutrient availability. However, water deficits and mineral deficiency occurred at high A-RZT due to poor root system development and lower rate of water uptake [9, 10, 12].

We have previously reported that supra-optimal A-RZT caused shoot water deficit by altering the balance between water uptake by the root system and water loss from the shoot [38, 39]. Water deficits resulting in stomata closure measured by stomatal conductance are due to the direct effect of reduced shoot water potential (ψ_{shoot}) and leaf relative water content (RWC) [38–42]. **Table 1** summaries ψ_{shoot} of subtropical vegetable crops of capsicum (*C. annuum* Indra F1-hybrid) and Chinese broccoli (*Brassica alboglabra* Bailey) [38] grown at 25°C-RZT and A-RZT while their shoots were maintained at fluctuating ambient temperatures. **Table 1** shows predawn and midday ψ_{shoot} were higher in 25°C-RZT than A-RZT plants in both vegetable species. Leaf RWC were further determined in both subtropical (capsicum and Chinese broccoli) and temperate vegetables (lettuce). Similar to the results of ψ_{shoot} RWC was significantly lower in all plants grown at A-RZT than at C-RZT measured predawn and midday (**Table 2**).

Vegetable species	25°C-RZT	A-RZT
Capsicum		
Predawn ψ_{Leaf} (Mpa) ($n = 6$) *	-0.51 ± 0.070	-2.95 ± 0.18
Midday ψ_{Leaf} (Mpa) ($n = 6$) *	-1.45 ± 0.092	-4.87 ± 0.24
Chinese broccoli		
Predawn ψ_{Leaf} (Mpa) ($n = 6$) *	-0.21 ± 0.03	-1.07 ± 0.06
Midday ψ_{Leaf} (Mpa) ($n = 6$) *	-0.85 ± 0.07	-2.03 ± 0.12

* Significant interaction between the two RZTs at $P < 0.01$.

Table 1. ψ_{Leaf} of subtropical vegetable crops of capsicum [19] and Chinese broccoli (*Brassica alboglabra* Bailey) [38] grown at two different RZTs while their shoots were maintained at fluctuating ambient temperatures under 100% prevailing solar radiation. All the measurements were done after 30 days of transplanting. Values shown are means \pm standard deviation.

Vegetable species	Cool-RZT*	A-RZT
Capsicum		
Predawn RWC (%) ($n = 6$) [#]	93.2 ± 0.22	80.3 ± 0.28
Midday RWC (%) ($n = 6$) [#]	81.3 ± 0.31	51.7 ± 0.29
Chinese broccoli		
Predawn RWC (%) ($n = 6$) [#]	97.6 ± 0.23	85.4 ± 0.16
Midday RWC (%) ($n = 6$) [#]	85.5 ± 0.27	64.9 ± 0.12
Lettuce		
Predawn RWC (%) ($n = 6$) [#]	95.1 ± 0.17	88.8 ± 0.35
Midday RWC (%) ($n = 6$) [#]	89.2 ± 0.29	60.1 ± 0.46

* Cool-RZT, 25°C-RZT for capsicum and Chinese broccoli; 20°C-RZT for lettuce.
[#] Significant interaction between the two RZTs at $P < 0.01$.

Table 2. Leaf RWC of subtropical capsicum (*Capsicum annuum* Indra F1-hybrid) [19] Chinese broccoli (*Brassica alboglabra* Bailey) [19], and temperate vegetable crops of lettuce [39] grown at two different RZTs while their shoots were maintained at fluctuating ambient temperatures under 100% prevailing solar radiation. All the measurements were done after 30 days of transplanting. Values shown are means \pm standard deviation.

Based on the results shown in **Tables 1** and **2**, it is obviously that subtropical and temperate vegetables grown in the tropical greenhouse had experienced permanent water deficit (reduced predawn ψ_{shoot} or/and leaf RWC) and midday (lower midday ψ_{shoot} or/and leaf RWC) when they were grown at A-RZT. In another experiment with capsicum, leaf g_s , root hydraulic conductivity, and shoot ψ_{shoot} declined after transferring plants from 20°C-RZT to A-RZT [42]. It was explained that supraoptimal RZTs caused a reduction in root hydraulic conductivity and might lower ψ_{shoot} which in turn could cause stomatal closure [42]. However, water deficit in 20 or 25°C-RZT plants was alleviated due to the larger root system [19].

4. Effects of root-zone temperature on NO_3^- uptake and assimilation

NO_3^- is the major N source available in aerobic soils [43]. Once uptaken by root cells, NO_3^- can be redirected out of the root cell, either by extrusion into the external medium or by unloading into the xylem vessels to reach the aerial organs [29]. The third possible fate for NO_3^- , in roots as well as in leaves, is its uptake by the vacuole where it participates in the general osmoticum or serves as a reservoir to sustain the growth process when the external nitrogen supply becomes limiting [44].

The effects of RZT on NO_3^- and N contents in plants have been reported [45–47]. N contents were reduced in plants grown under high RZT [39, 48–50]. For instance, Du and Tachibana [48] grew cucumber (cv. “Sharp I”) plants hydroponically at several RZTs: 25 (control), 30, 35, or 38°C, with shoot’s temperature at $26/23 \pm 3^\circ\text{C}$ (day/night). Total N concentration in leaf was reduced as the RZT was raised to 35°C and to 38°C in particular. Similar results were obtained our research team with lettuce (cv. “Palma”) plants [39]. Leaf organic N content was 32% lower in A-RZT plants than 20°C-RZT plants. Transfer of plants between these two RZTs altered leaf N content after 6 days. Leaf N content increased in A \rightarrow 20°C-RZT plants, while decreased in 20°C \rightarrow A-RZT plants. After 10 days of reciprocal RZT transfer, A \rightarrow 20°C-RZT plants and 20°C \rightarrow A-RZT plants had a similar leaf N content [39]. The results also showed tight temporal coupling of leaf N content, and light- and CO_2 - saturated photosynthetic O_2 evolution rate throughout the reciprocal temperature transfers. He et al. [39] suggested that the decreased nutrient status of lettuce plants caused nonstomatal limitation of photosynthesis under high A-RZT conditions. In lettuce (cv. “Panama”), Tan et al., [12] reported that 20°C-RZT plants had higher leaf N concentrations on the basis of per unit dry weight compared with plants grown at A-RZT. Total shoot and root NO_3^- of 20°C-RZT plants were higher than A-RZT plants. 20°C \rightarrow A-RZT plants suffered from a reduction of total mineral accumulation, and A \rightarrow 20°C-RZT plants increased in total mineral accumulation [12]. In another study, Yeager et al. grew *Ilex crenata* Thunb. “Rotundifolia” plants in sand culture with the RZTs at 28, 34, or 40°C for 6 h daily. They found that root and shoot N accumulation (mg N/g dry weight) decreased when RZTs were increased from 28 to 40°C. These plants were fertilized twice daily with 500 mL of either 75, 150, or 225 mg N/L, to determine applied N rate on growth and N accumulation of “Rotundifolia” holly. The results showed that root and shoot N accumulation depended on RZT and the N rate. N accumulated by roots and shoots increased when the applied N concentration increased from 75 to 225 mg/L at each RZT. However, root and shoot N accumulation decreased with the root zone at 40°C compared with 28°C for 75 and 225 mg N/L applied. These data indicated that increased N fertilization rates would not alleviate growth reductions of holly caused by high RZT (40°C) [49].

In contrast, high RZT increased N content in plants has also been reported [49–51]. Johnson and Ingram exposed root systems of *Pittosporum tobira* Thunb plants to temperatures of 27, 30, or 40°C for 6 h daily for 7 months with air temperature around 30/24°C (day/night). They found that N level in leaf tissue (newly expanded leaves) was increased in plants at 40°C medium temperature [50]. This was supported by the work of Gosselin and Trudel who transferred the 10-week-old pepper (cv. “Bell Boy”) plants to five different RZTs (12, 18, 24, 30, or $36 \pm 2^\circ\text{C}$)

with air temperature maintained at 22–24/18–20°C (day/night), for a period of 8 weeks. Their results showed that leaf N concentration increased with the increase of RZT from 12 to 36°C [51]. Cruz et al. grew Carob (*Ceratonia siliqua* L.) seedlings at different RZTs (10–40°C) with shoot temperature at $24 \pm 1/20 \pm 1^\circ\text{C}$ (day/night). When nitrate (3 mM) was supplied as nitrogen source of nutrient solutions, they found that organic N concentration in roots increased with the increase of RZT from 10 to 40°C. Total N concentration in both shoot and root increased with the increased RZT from 10 to 25°C. Investigators explained that increasing RZTs induced increased in ion-uptake rates, mainly nitrogen, might be responsible for the higher N content in plant. This situation increased also the demand for carbon in the root of Carob plant [52]. As discussed earlier, the amount of mineral nutrient especially nitrogen available to a plant is determined by the root morphological structure [3, 12, 13]. The amount of mineral nutrient uptake and transport also depend on transpiration rate as long-distance movement of nutrient through plants is predominantly by bulk flow in the xylem [53].

For most higher plants, NO_3^- assimilation is the major pathway by which inorganic N is converted to an organic form [54]. The conversion of NO_3^- to NH_4^+ , which can be directly used to synthesize organic N containing compounds, is a reduction process that occurs in two steps. NO_3^- is first reduced to NO_2^- in the cytosol by nitrate reductase (NR). NO_2^- is then translocated to the chloroplast where it is reduced into NH_4^+ by nitrite reductase (NiR). NR is NO_3^- inducible enzyme. Transcription of NR genes is induced by NO_3^- [54–56]. The reduction of NO_3^- could take place either in roots or in leaves or both [54, 56, 59]. Significant translocation of NO_3^- to the shoot would occur only when the net NO_3^- uptake rate was fast enough to saturate the reduction process in the roots.

In our study of subtropical vegetable Nai Bai (*Brachyponera chinensis* L.) plant and Baby butterhead lettuce (*L. sativa* L.) plants, NO_3^- concentrations of leaf and root and maximum nitrate reductase activity (NRA) of leaf and roots were determined after 10, 20, and 30 days of transplanting (**Figures 3 and 4**).

For Nai Bai plants, the leaf NO_3^- concentrations of either 25°C-RZT or A-RZT plants were similar at 10, 20, and 30 days after transplanting (DAT). However, the leaf NO_3^- concentrations of A-RZT plants were significantly lower than those of 25°C-RZT plants (**Figure 3A**). Regardless of growth stage, 25°C-RZT plants had significantly higher root NO_3^- concentrations than those of A-RZT plants (**Figure 3C**). Root NO_3^- concentrations of A-RZT remained constant at all growth stages (**Figure 3C**). However, 25°C-RZT plants had lower root NO_3^- concentrations at 10 DAT compared to those at 20 and 30 DAT (**Figure 3C**). At 10 DAT, leaf NRA of 25°C-RZT plants was significantly lower than those of A-RZT plants (**Figure 3B**). However, it increased from 10 to 20 DAT, and remained constant from 20 to 30 DAT. Leaf maximum NRA of A-RZT plants decreased from 10 to 20 DAT, and remained constant from 20 to 30 DAT. Therefore, at 20 and 30 DAT, maximum leaf NRA of 25°C-RZT plants was much higher than those of A-RZT plants (**Figure 3B**). Root maximum NRA of 25°C-RZT and A-RZT plants decreased from 10 to 20 DAT, and then remained constant to the 30 DAT. At 10 DAT, no significant difference in maximum root NRA was measured between 25°C-RZT and A-RZT plants. However, at 20 and 30 DAT, maximum root NRA of 25°C-RZT plants was significantly higher than those of A-RZT plants (**Figure 3D**).

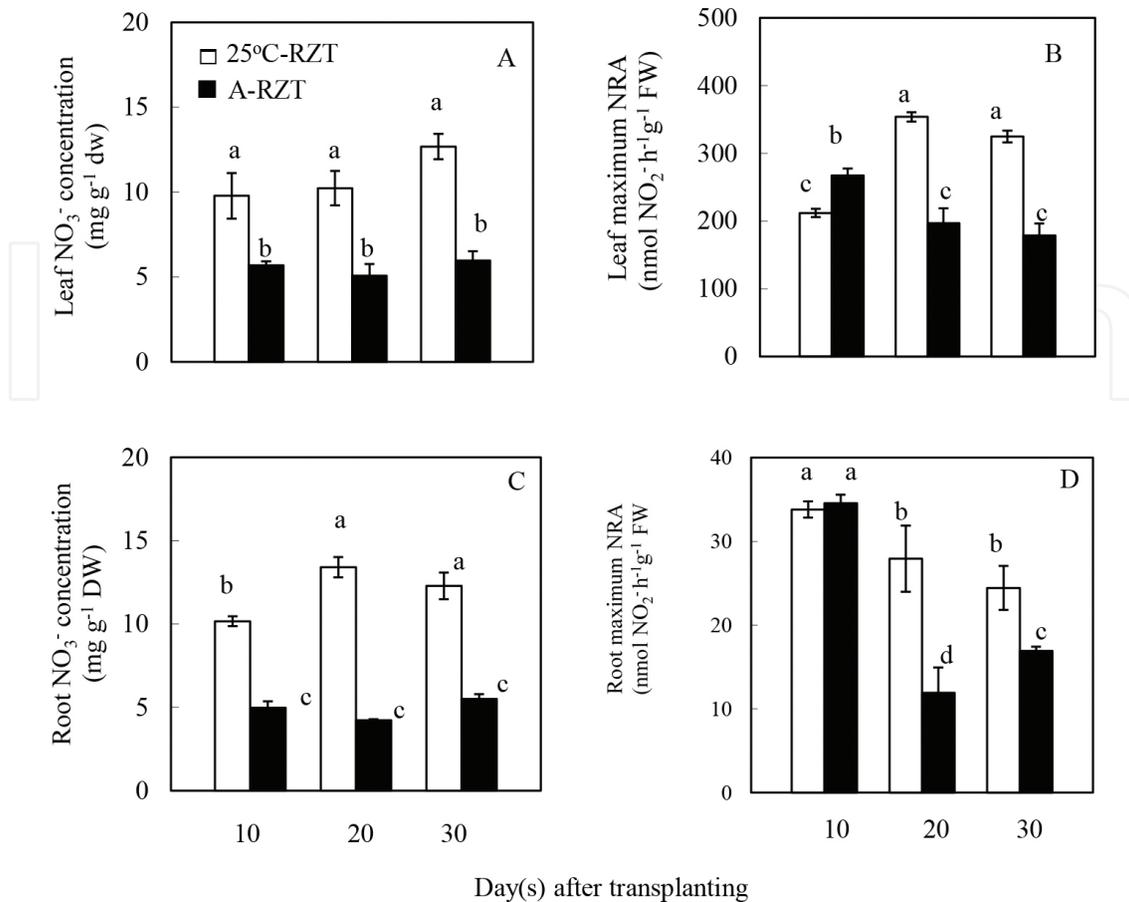


Figure 3. Leaf NO_3^- concentration (A) and maximum NRA (B), root NO_3^- concentration (C) and maximum NRA (D) of Nai Bai (*Brachypogon chinensis* L.) plant. Each point is the mean of five measurements of five different leaves from two different bins. Vertical bars represent the standard errors. Means with different letters above the columns are statistically different ($P < 0.001$) as determined by Tukey's multiple comparison test (unpublished data).

The leaf NO_3^- concentrations of 20°C-RZT were significantly higher compared to those of A-RZT plants, at different growth stages (**Figure 4A**). With prolonged growth to 30 DAT, leaf and root NO_3^- concentrations of 20°C-RZT plants increased (**Figure 3A**). However, there was no significant difference in leaf and root NO_3^- concentrations at A-RZT among the three growth stages (**Figure 4C**). Leaf maximum NRA in 20°C-RZT and A-RZT plants was highest at 10 DAT and decreased at 20 DAT and further decreased at 30 DAT (**Figure 4A**). Regardless of growth stage, leaf maximum NRA was significantly higher in 20°C-RZT than in A-RZT plants (**Figure 4B**). It was surprising to observe that root maximum NRA was many times higher in A-RZT plants than in 20°C-RZT plants, indicating that RZT altered the site of NO_3^- assimilation (**Figure 4D**).

The reduction of NO_3^- could take place either in roots, in leaves, or in both [57, 58]. For Baby butterhead plants, hot A-RZT treatment switched NO_3^- reduction from shoot to root, evidenced by the higher NRA in A-RZT roots than in leaves (**Figure 4D**) while NRA of 20°C-RZT leaves was much higher than that of roots (**Figure 4B**). However, this was not seen in Nai Bai plants (**Figure 3D**). High NO_3^- assimilation rate in the root may be due to low reduced N

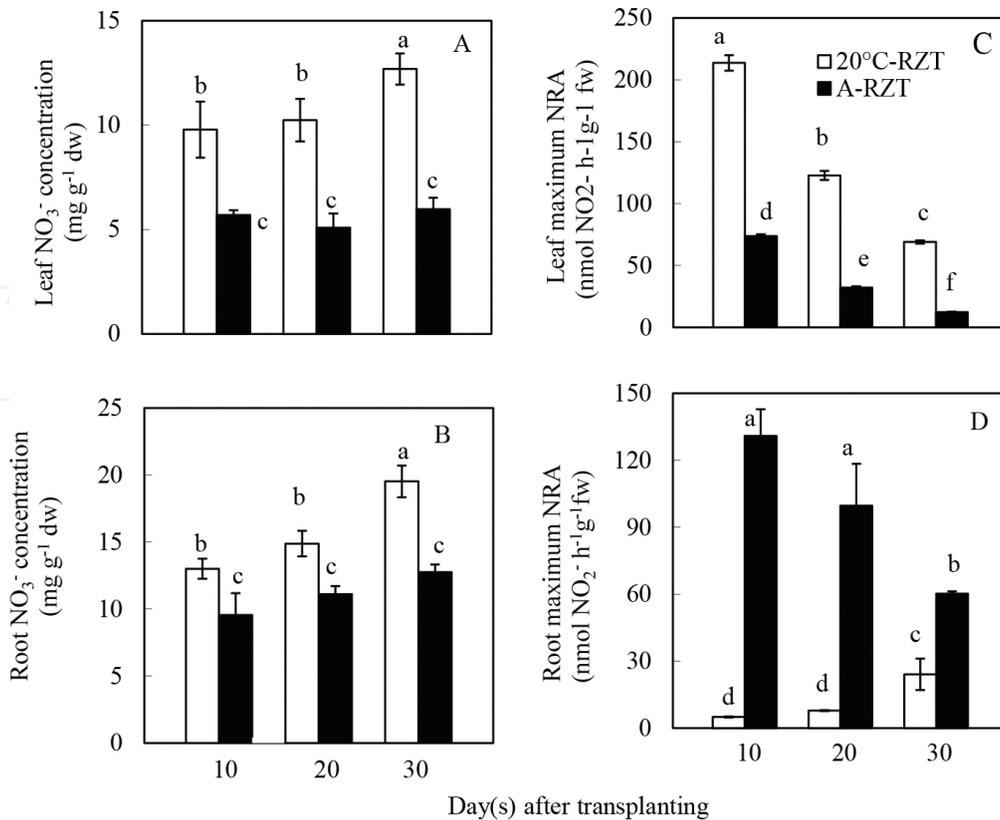


Figure 4. Leaf NO₃⁻ concentration (A) and maximum NRA (B), root NO₃⁻ concentration (C), and maximum NRA (D) of Baby butter head lettuce (*Lactuca sativa* L.) plant. Each point is the mean of five measurements of five different leaves from two different bins. Vertical bars represent the standard errors. Means with different letters above the columns are statistically different ($P < 0.001$) as determined by Tukey's multiple comparison test (unpublished data).

concentration in A-RZT roots (data not shown). Laurie and Stewart [60] grew cheakpea (*Cicer arietinum* L. ICARDA cultivar no. ILC 482) plants at high (40/25°C, day/night), and concentration in A-RZT roots, and moderate (25/17°C, day/night) temperature regimes. The results showed that high temperature (40/25°C) reduced shoot NRA (*in vivo*) but had little effect on root NRA [57].

On a total plant basis, high temperature growth shifted NRA from shoot to root, particularly in the young plant. Laurie and Stewart [60] also observed that there was a greater decline in leaf NRA with age [60]. This was in accordance with the present finding that leaf maximum NRA of 20°C-RZT Baby butterhead lettuce plants was highest in expanding young leaves (10 DAT) and lowest in fully matured leaves at 30 DAT (**Figure 4B**). Although leaf maximum NRA of A-RZT Baby butterhead lettuce plants decreased with the growth of plants, the decrease of NRA from 20 DAT to 30 DAT was not caused by the leaf age. Because the youngest fully expanded leaves were selected for the NRA analysis at both growth stages. Therefore, the repress of leaf maximum NRA in A-RZT plants at the late growth stage may be due to NR protein degradation caused by long-term supraoptimum RZT treatment. Although root maximum NRA was higher in A-RZT Baby butterhead lettuce plants compared to those of 20°C-RZT plants, the lower NO₃⁻ uptake and transport capacity (lower root and leaf NO₃⁻ concentration) still restricted NO₃⁻ reduction of A-RZT plants. This was evidenced by the lower

total reduced N concentration of leaf and root of A-RZT plants compared to 20°C-RZT plants, especially at 20 and 30 DAT (data not shown). For Nai Bai plants, NO₃⁻ reduction took place both in leaves and roots. However, the major proportion (more than 80%) of total plant NO₃⁻ reduction occurred in leaves for both 25°C-RZT and A-RZT plants (**Figure 3C and D**). At very early growth stage (10 DAT), supraoptimum A-RZT did not decrease maximum NRA in leaves and roots of Nai Bai plants. Because of very low growth rate of the plant at this very early growth stage, the lower NO₃⁻ content in A-RZT plants did not limit NO₃⁻ reduction. As a result, both of total reduced N concentrations in leaves and roots of A-RZT Nai Bai plants remained at the similar levels to those of 25°C-RZT plants at 10 DAT (data not shown). However, at 10 DAT, sugar concentrations (especially, glucose, fructose, and starch) both in leaves and roots of A-RZT Nai Bai plants increased significantly compared to 25°C-RZT plants (data not shown). Sugar accumulation in roots may be considered as a general consequence of impaired growth [61]. These indicate that, for Nai Bai plants, carbohydrate metabolism is more sensitive to supraoptimum RZT than NO₃⁻ metabolism. Controversial results of effect of high RZTs on N metabolism in shoots and roots have been reported [38, 39, 59–61]. In our study, it was confirmed that RZTs significantly affect N metabolism both in leaves and roots of subtropical vegetable Nai Bai (*B. chinensis* L (**Figure 3**) and temperate vegetable Baby butter head lettuce (**Figure 4**).

5. Conclusions

Our studies showed that high A-RZT inhibited root elongation, branching, and hair formation but increased root diameter of subtropical and temperate vegetables grown in the tropics. However, cooling the RZ promoted root growth and development as well as shoot productivities of aeroponically grown temperate and subtropical vegetables in the tropics. Manipulation of rhizosphere environment can alter not only root morphology but also physiological characteristics of roots to meet changes in shoot water and nutrient demand in response to atmospheric high temperature. Hot A-RZT caused shoot water deficit of aeroponically grown plants with continual spraying nutrient solution due to the negative water balance between water uptake by the root system and water loss from the shoot. Thus, subtropical and temperate vegetables grown in the tropics greenhouse had experienced mid-day and permanent water deficit when they were grown at A-RZT. However, water deficit of C-RZT plants was alleviated due to the larger root system. Compared to subtropical and temperate vegetables grown at hot A-RZT, cooling the RZ enhanced NO₃⁻ uptake and its assimilation of shoots. However, effects of RZT on NO₃⁻ assimilation of roots depend on species. Adequate levels of nutrient, especially N of C-RZT plants, alleviated nonstomatal limitation of photosynthesis.

Acknowledgements

All experiment work on subtropical and temperate vegetables grown aeroponically was supported by the Academic Research Fund (Grant reference numbers: RP10/95 HJ, RP 1/97 HJ,

RP 22/98 HJ, RP12/01, RI7/05 HJ, Ministry of Education, Singapore and Singapore Millennium Foundation (Project code: SMF-Farming System).

Author details

Jie He

Address all correspondence to: jie.he@nie.edu.sg

National Institute of Education, Nanyang Technological University, Singapore

References

- [1] Malamy JE. Intrinsic and environmental response pathways that regulate root system architecture. *Plant Cell, Environment*. 2005; 28: 67–77, DOI: 10.1111/j.1365-3040.2005.01306.x/full.
- [2] He J. Mineral nutrition of aeroponically grown subtropical and temperate crops in the tropics with manipulation of root-zone temperature at different growth irradiances. *Plant Stress*. 2010; 4: 14–30, http://www.globalsciencebooks.info/JournalsSup/10PS_4_SI2.html.
- [3] He J. Impact of root-zone temperature on photosynthetic efficiency of aeroponically grown temperate and subtropical vegetable crops in the tropics. In: Buchner Th. B. Ewingen NH, Editors. *Theory and Applications in Energy, Biotechnology and Nanotechnology*. Nova Science Publishers Inc.: New York; 2009. pp. 111–143, <http://trove.nla.gov.au/work/27991465?versionId=45343866>. Ch 4.
- [4] Hodge A, Berta G, Doussa C, Merchan F, Crespi M. Plant root growth, architecture and function. *Plant Soil*. 2009; 321: 153–187, DOI: 10.1007/s11104-009-9929-9.
- [5] Kirk GJD, Van DL. Changes in rice root architecture, porosity, and oxygen and proton release under phosphorus deficiency. *New Phytologist*. 1997; 135: 191–200, DOI: 10.1046/j.1469-8137.1997.00640.x
- [6] Neumann G, Massonneau A, Martinoia E, Römheld V. Physiological adaptations to phosphorus deficiency during proteoid root development in white lupin. *Planta*. 1999; 208: 373–382, <http://link.springer.com/article/10.1007%2Fs004250050572>.
- [7] Xie Y, An S, Yao X, Xiao K, Zhang C. Short-time response in root morphology of *Vallisneria spiralis* to sediment type and water column nutrient. *Aquatic Botany*. 2005; 81: 85–96, DOI: 10.1016/j.aquabot.2004.12.001.

- [8] Yang JC. Relationships of rice root morphology and physiology with the formation of grain yield and quality and the nutrient absorption and utilization. *Scientia Agriculture Sinica*. 2011; 44: 36–46, DOI: 10.3864/j.issn.0578-1752.2011.01.005.
- [9] He J, Lee SK. Growth and photosynthetic characteristics of lettuce (*Lactuca sativa* L.) grown under fluctuating hot ambient temperatures with the manipulation of cool rootzone temperature. *Journal of Plant Physiology*. 1998; 152: 387–391, DOI: 10.1016/S0176-1617(98)80252-6.
- [10] He J, Lee SK. Growth and photosynthetic responses of three aeroponically grown lettuce cultivars (*Lactuca sativa* L.) to different rootzone temperatures and growth irradiances under tropical aerial condition. *Journal of Horticulture Sciences and Biotechnology*. 1998; 73: 173–180, DOI: 10.1080/14620316.1998.11510961.
- [11] Lynch J. Root architecture and plant productivity. *Plant Physiology*. 1995; 109: 7–13, DOI: 10.1104/pp.109.1.7.
- [12] Tan LP, He J, Lee SK. Effects of root-zone temperature on the root development and nutrient uptake of *Lactuca sativa* L. cv 'Panama' grown in an aeroponic system in the tropics. *Journal of Plant Nutrition*. 2002; 25: 297–314, DOI: 10.1081/PLN-100108837.
- [13] Qin L, He J, Lee SK, Dodd IC. An assessment of ethylene mediation of lettuce (*Lactuca sativa*) root growth at high temperatures. *Journal of Experimental Botany*. 2007; 58: 3017–3024, DOI: 10.1093/jxb/erm156.
- [14] Kuroyanagi T, Paulsen GM. Mediation of high-temperature injury by roots and shoot during reproductive growth of wheat. *Plant, Cell and Environment*. 1988; 11: 517–523, DOI: 10.1111/j.1365-3040.1988.tb01790.x.
- [15] Cumbu IP, Nye PH. Root zone temperature effects on growth and nitrate absorption in rape (*Brassica napus* cv. Emeralk). *Journal of Experimental Botany*. 1982; 33: 1138–1446, <http://www.jstor.org/stable/23690485>.
- [16] Sattelmacher B, Marschner H, Kühne R. Effects of the temperature of the rooting zone on the growth and development of roots of potato (*Solanum tuberosum*). *Annals Botany*. 1990; 65: 27–36.
- [17] Pardales JR, Yamauchi A, Kono Y. Growth and development of sorghum roots after exposure to different periods of a hot root-zone temperature. *Environment Experimental Botany*. 1991; 31: 397–403, DOI: 10.1016/0098-8472(91)90037-O.
- [18] Menzel CM, Turner DW, Doogan VJ, Simpson PR. Root–shoot interactions in passionfruit (*Passiflora* sp.) under the influence of changing root volumes and soil temperatures. *Journal of Horticulture Science*. 1994; 69: 353–364, DOI: 10.1080/14620316.1994.11516487.
- [19] Tan LP. Root-zone temperature effects on vegetable physiology in a commercial aeroponic system in Singapore. [thesis]. Nanyang Technological University, Singapore; 2004.

- [20] Abbas Al-Ani MK, Hay RKM. The influence of growing temperature on the growth and morphology of cereal seedling root systems. *Journal of Experimental Botany*. 1983; 34: 1720–1730, DOI: 10.1093/jxb/34.12.1720.
- [21] Pierik R, Verkerke W, Voeselek RACJ, Blom KWPM, Visser EJW. Thick root syndrome in cucumber (*Cucumis sativus* L.): a description of the phenomenon and an investigation of the role of ethylene. *Annals Botany*. 1999; 84: 755–762, DOI: 10.1006/anbo.1999.0980.
- [22] Arshad M, Frankenberger WT Jr. Ethylene: Agricultural Sources and Applications. Kluwer Academic/Plenum publishers: New York. 2002; pp. 155–156.
- [23] Choong TW, He J, I. Dodd. Growing *Lactuca* in an aeroponic system within a tropical greenhouse – lower accumulated rootzone ethylene concentrations corresponded with higher shoot growth at cooler rootzone temperatures. *Frontier* (accepted).
- [24] He J, Tan LP, Lee SK. Root-zone temperature effects on photosynthesis, ¹⁴C-photo-assimilate partitioning and growth of temperate lettuce (*Lactuca sativa* cv. 'Panama') grown in the tropics. *Photosynthetica*. 2009; 47: 95–103, DOI: 10.1007/s11099-009-0015-6.
- [25] Minchin PEH, Thorpe, M.R. What determines carbon partitioning between competing sinks? *Journal of Experimental Botany*. 1996; 47: 1293–1296, DOI: 10.1093/jxb/47.
- [26] Minchin PEH, Farrar JF, Thorpe MR. Partitioning of carbon in split root system of barley: effect of temperature of the root. *Journal of Experimental Botany*. 1994; 45: 1103–1109, DOI: 10.1093/jxb/45.8.1103.
- [27] Schurr U, Heckenberger U, Herdel K, Walter K, Feil R. Leaf development in *Ricinus communis* during drought stress: dynamics of growth processes, of cellular structure and of sink-source transition. *Journal of Experimental Botany*. 2000; 51: 1515–1529, DOI: 10.1093/jexbot/51.350.1515.
- [28] Klock KA, Taber HG, Graves WR. Root respiration and phosphorus nutrition of tomato plants grown at a 36°C root-zone temperature. *Journal of the American Society for Horticultural Science*. 1997; 122: 175–178, <http://cat.inist.fr/?aModele=afficheN&cpsid=10736550>.
- [29] Forde BG, Clarkson DT. Nitrate and ammonium nutrition of plants: Physiological and molecular perspectives. *Advances in Botanical Research*. 1999; 30: 1–90, DOI: 10.1016/S0065-2296(08)60226-8.
- [30] McCully M. How do real roots work? Some new view of root structure. *Plant Physiology*. 1985; 109: 1–6, DOI: 10.1104/pp.109.1.1.
- [31] Grime JP, Crick JC, Rincon JE. The ecological significance of plasticity. *Symposia of the Society for Experimental Biology*. 1986; 40: 5–29, <http://www.ncbi.nlm.nih.gov/pubmed/3544310>.

- [32] Cooper AJ. Root temperature and plant growth: a review. In: Commonwealth Bureau of Horticulture and Plantation Crops. Commonwealth Agriculture Bureaux, Farnham Royal, Slough: England, 1973. p. 73, <http://trove.nla.gov.au/version/25843400>.
- [33] Delucia EH, Scott AH, Day TA. Effects of soil temperature on growth, biomass allocation and resource acquisition of *Andropogon gerardii* Vatman. *New Phytologist*. 1992; 10: 543–549, <http://www.jstor.org/stable/2557416>.
- [34] He J, Chua NYA, Qin, L. Interaction between iron stress and root-zone temperature on physiological aspects of aeroponically grown Chinese broccoli (*Brassica alboglabra*). *Journal of Plant Nutrition*. 2008; 31: 1–20, DOI: 10.1080/01904160701742121.
- [35] Luo HY. Mechanisms of Ca, P and K uptake and partitioning characteristics of certain temperate vegetables in relation to different irradiance and root-zone temperatures [thesis]. Nanyang Technological University, Singapore; 2008.
- [36] Engels C, Münkle L, Marschner H. Effect of root zone temperature and shoot demand on uptake and xylem transport of macronutrients in maize (*Zea mays* L.). *Journal of Experimental Botany*. 1992; 43: 537–547, DOI: 10.1093/jxb/43.4.537.
- [37] Macay AD, Barber SA. Soil temperature on root growth and phosphorus uptake by corn. *Soil Science Society of America*. 1984; 48: 818–823, DOI: 10.2136/sssaj1984.03615995004800040024x.
- [38] He J, Lee SK. Relationship among photosynthesis, Ribulose-1,5-Bisphosphate Carboxylase (Rubisco) and water relations of subtropical vegetable Chinese broccoli grown in the tropics by manipulation of root-zone temperature. *Environmental and Experimental Botany*. 2001; 46: 119–128, DOI: 10.1016/S0098-8472(01)00089-2.
- [39] He J, Lee SK, Dodd IC. Limitations to photosynthesis of lettuce grown under tropical conditions: alleviation by root-zone cooling. *Journal of Experimental Botany*. 2001; 52: 1323–1330, DOI: 10.1093/jexbot/52.359.1323.
- [40] Turner NC. Stomatal behavior and water status of maize, sorghum, and tobacco under field conditions II. At low soil water potential. *Plant Physiology*. 1974; 53: 360–365, <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC367352/>.
- [41] Turner NC, Jones MM. Turgor maintenance by osmotic adjustment. In: Turner NC, Kramer PJ. Editors. *A Review and Evaluation. Adaptation of Plants to Water and High Temperature Stress*. Wiley-Interscience: New York, USA; 1980. pp. 87–103, <http://www.cabdirect.org/search.html?q=turgor+maintenance+by+osmotic+adjustment.+a+review+and+evaluate>.
- [42] Dodd IC, He J, Turnbull, CGN, Lee SK, Critchley C. Influence of supra-optimal root temperatures on growth and stomatal conductance in *Capsicum annuum* L. *Journal of Experimental Botany*. 2000; 51: 239–248, DOI: 10.1093/jexbot/51.343.239.
- [43] Marschner H. *Mineral Nutrition of Higher Plants*. Academic: London, UK ; 1995. p. 889, <http://www.sciencedirect.com/science/book/9780124735422>

- [44] der Leij M, Smithe SJ, Miller AJ. Remobilization of vacuolar stored nitrate in barley root cells. *Planta*. 1998; 205: 64–72, <http://link.springer.com/article/10.1007/s004250050297>.
- [45] Atkin OK, Cummins WR. The effect of root temperature in the induction of nitrate reductase activities and nitrogen uptake rates in arctic plant species. *Plant and Soil*. 1994; 159: 187–197, DOI: 10.1007/BF00009280.
- [46] Engels C. Effect of root and shoot meristem temperature on shoot and root dry matter partitioning and internal concentrations of nitrogen and carbohydrates in maize and wheat. *Annals Botany*. 1994; 73: 211–219, DOI: 10.1006/anbo.1994.1025.
- [47] Ali IA, Kafkafi U, Yamaguchi I, Sugimoto Y, Inanaga S. Response of oilseed rape plant to low root temperature and nitrate:ammonium ratios. *Journal of Plant Nutrition*. 1998; 21: 1463–1481, DOI: 10.1080/01904169809365496.
- [48] Du YC, Tachibana S. Effect of supraoptimal root temperature on the growth, root respiration and sugar content of cucumber plants. *Scientia Horticulturae*. 1994; 58: 289–330, DOI: 10.1016/0304-4238(94)90099-X.
- [49] Yeager TH, Harrison RH, Ingram DL. ‘Rotundifolia’ holly growth and nitrogen accumulation influenced by supraoptimal root-zone temperatures. *HortScience*. 1991; 26: 1381–1388, <http://hortsci.ashspublications.org/content/26/11/1387.full.pdf>.
- [50] Johnson CR, Ingram DL. *Pittosporum tobira* response to container medium temperature. *HortScience*. 1984; 19: 524–525, <http://openagricola.nal.usda.gov/Record/GUA84131934>.
- [51] Gosselin A, Trudel MJ. Interaction between root-zone temperature and light levels on growth, development and photosynthesis of *Lycopersicon esculentum* Mill. cultivar ‘vondor’. *Scientia Horticulturae*. 1984; 23: 313–321. www.sciencedirect.com/science/article/pii/030442388490027X
- [52] Cruz C, Lips SH, Martinsloucao MA. Effect of root temperature on carob growth – nitrate versus ammonium nutrition. *Journal of Plant Nutrition*. 1993, 16: 1517–1530, DOI: 10.1080/01904169309364629.
- [53] Atkinson CJ, Ruiz LP, Mansfield TA. Calcium in xylem sap and the regulation of its delivery to the shoot. *Journal of Experiment Botany*. 1992; 43: 1315–1324, DOI: 10.1093/jxb/43.10.1315.
- [54] Solomonson LP, Barber MJ. Assimilatory nitrate reductase: functional properties and regulation. *Annual Review of Plant Physiology and Plant Molecular Biology*. 1990; 41: 225–253, DOI: 10.1146/annurev.pp.41.060190.001301.
- [55] Cheng CL, Dewdney J, Kleinhofs A, Goodman HM. Cloning and nitrate induction of nitrate reductase mRNA. *Proceedings of the National Academy of Sciences USA*. 1986; 83: 6825–6826, <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC386602>.
- [56] Savidov NA, Tokarev BI, Lips SH. Regulation of Mo-cofactor, NADH- and NAD(P)H-specific nitrate reductase activities in the wild type and two *nar*-mutant lines of barley

(*Hordeum vulgare* L.). *Journal of Experiment Botany*. 1997; 48: 847–855. DOI: 10.1093/jxb/48.4.847.

- [57] Cen YP, Layzell DB. *In vivo* gas exchange measurement of the site and dynamics of nitrate reduction in soybean. *Plant Physiology* 2003; 131: 1147–1156, <http://www.plant-physiol.org/content/131/3/1147.full.pdf>.
- [58] Scheurwater I, Koren M, Lambers H, Atkin OK. The contribution of roots and shoots to whole plant nitrate reduction in fast- and slow-growing grass species. *Journal of Experimental Botany*. 2002; 55: 1635–1642.
- [59] Gojon A, Passard C, Bussi C. Root/shoot distribution of NO_3^- assimilation in herbaceous and woody species. In: Roy R, Garnier E, editors. *A Whole Plant Perspective on Carbon-Nitrogen Interactions*. SPB Academic Publishing: The Hague; 1994. pp. 131–147, <http://link.springer.com/article/10.1023/A%3A1000903504144>
- [60] Laurie S, Stewart GR. Effects of nitrogen supply and high temperature on the growth and physiology of the chickpea. *Plant, Cell and Environment*. 1993; 16: 609–621, DOI: 10.1111/j.1365-3040.1993.tb00479.
- [61] Hansch R, Fessel DG, Witt C, Hesberg C, Hoffmann G, Walch-Liu P, Engels C, Kruse J, Rennenberg H, Kaiser W, Mendel RR. Tobacco plants that lack expression of functional nitrate reductase in roots show changes in growth rates and metabolite accumulation. *Journal of Experimental Botany*. 2001; 52: 1251–1258, DOI: 10.1093/jexbot/52.359.1251.