Modified atmosphere packaging and dark/light refrigerated storage in green leafy vegetables have an impact on nutritional value.

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Abbreviations: A, antheraxanthin; AMD, age-related macular degeneration; β-Car, β-carotene; Chl, chlorophylls; DLE, dim light-exposed; DW, dry weight; FW, fresh weight; MA, modified atmosphere; Neo, neoxanthin; Lut, lutein; LE, light-exposed; PPFD, photosynthetic photon flux density; t-Car, total carotenoids; V, violaxanthin; VAZ, the xanthophyll cycle involving the carotenoids violaxanthin, antheraxanthin and zeaxanthin; VDE, violaxanthin de-epoxidase; Z, zeaxanthin.

Abstract

The consumption of zeaxanthin (Z) through a vegetable-rich diet is recommended to reduce the progression of age-related macular degeneration. Due to Z's intrinsic dynamic character that results from its participation in the photoprotective xanthophyll cycle involving the carotenoids violaxanthin, antheraxanthin and zeaxanthin (VAZ), post-harvest handling practices and storage usually retain low amounts of this bioactive compound (compared to the rest of phytochemicals that are, in general, more stable). Thus, the aim of this work was to investigate in important consumed leafy vegetables the effects of different storage conditions on carotenoids (mainly Z) including i) packaging under three modified atmospheres (MAs), ii) light refrigerated supermarket storage and iii) dark refrigerated domestic storage. The results showed that an MA with low O_2 and high CO_2 enhanced the Z content under light. Moreover, both light and dark refrigerated storage showed dynamic and circadian pigment changes that enhanced the total VAZ pool. These results can contribute to generating practical recommendations for industries, supermarkets, and consumers when high Z content is a nutritional target.

Introduction

The consumption of carotenoids, their precursors, and their derivatives have been associated with many health benefits such as reduced risk of cancer [1], cognitive improvements, or reduced development of degenerative illnesses like age-related macular degeneration (AMD), which is the main cause of blindness in developed countries [2]. Three carotenoids are responsible for healthy vision, β-carotene (β-Car) (the precursor for vitamin A), lutein (Lut), and zeaxanthin (Z). Because our bodies cannot produce these bioactive compounds, their intake from a carotenoid-rich diet with fruits and vegetables is desirable. From the field to the consumers' table, vegetables remain biologically active during processing (e.g. packaging in a modified atmosphere, MA), transport, and/or storage (both on supermarket shelves and/or in consumers' refrigerators). Under these scenarios, the bioactive compounds of these vegetables are active and respond to environmental stimuli [3]. This is particularly the case for Z, whose pool is extremely dynamic and variable (usually present at low amounts). Other compounds such as Lut or β-Car are usually present at high and stable concentrations. In plants, Z is involved in the photoprotective xanthophyll (VAZ) cycle that dissipates excess light energy and protects plant tissues (for a review, please see [4]). When the plant is under intense light or/and environmental stress, violaxanthin (V) is de-epoxidised to Z by the enzyme violaxanthin de-epoxidase (VDE). This dissipates energy from the light harvesting complex. The Z pool is high under this dissipative state. Under non-stressed situations or in darkness, it is rapidly reconverted to violaxanthin (V). The VAZ-cycle is then completed within minutes to hours. Consequently, processing along the commercial chain usually causes leafy greens to have high Lut content, but only traces of Z at the time of consumption. The strategy by which plants protect their photosynthetic machinery via the VAZ-cycle offers a benefit and a challenge from a nutritional point of view: the opportunity to improve the nutritional quality of vegetables [5].

Carotenoid responses are much more dynamic in green tissues than in non-photosynthetic tissues, as is the case of Z formation through the VAZ cycle. However, the ability to enhance carotenoid contents in edible green leafy plants is limited by the fact that carotenoids are bound to the proteins that constitute the photosynthetic apparatus [6]. This is in contrast to fruits or seeds, where pools are not constrained by chloroplast structure and function [6]. As a result, the strategy is to identify approaches that can increase the total VAZ pool or can maintain the

cycle in the dissipated state (Z), or that can even enhance the Z content at the time of consumption to improve the overall nutritional quality of food.

Information available on changes to carotenoids during processing after harvest is limited. While there is a growing interest in MA packaging combined with low temperatures to extend the freshness of fresh-cut vegetables until consumption [7], few studies have focused on Z and Lut changes during this storage. It has been reported that the MA technique delays senescence, preserves quality, conserves phytonutrients, and extends the life of the vegetables [8]. Furthermore, some studies have analysed what occurs to carotenoids in green leafy vegetables during usual postharvest storage under a regime of light/dark cycles [9] at ambient or temperate refrigerated conditions [10]. Therefore, our study sought to investigate the effects on the carotenoid profiles—especially Z—of several common storage conditions including (i) MAs of varying CO_2 and O_2 (with and without light), (ii) storage in refrigerated supermarket shelves (with and without light), and (iii) storage in a domestic refrigerator (without light). We hypothesise that the different storage conditions applied here have an important effect on the carotenoid dynamics. As plant models, we studied two of the most popular leafy species for consumption: lettuce and spinach. These have a worldwide production of approximately 25 and 22 million tonnes, respectively. We also included parsley, a popular fresh aromatic herb, and corn salad, which has recently attracted considerable interest as a culinary vegetable for salads.

Material and methods

Biological material, experimental design, and sampling

Three different experimental designs were carried out to address the effect of storage on carotenoid contents: (i) MA packaging of CO_2 and O_2 (with and without light), (ii) dark/light refrigerated storage and (iii) storage in a domestic refrigerator (without light):

(i) **Storage in MA packaging:** Here, mature and visually good quality lettuce (*Lactuca sativa* L. var. oak leaf, Lactuceae), spinach (*Spinacia oleraceae* L. var. baby, Chenopodioideae), and corn salad (*Valerianella locusta* L. var. Verte de Louviers, Valerianaceae) obtained from a local market were stored at 4ºC for 12 h prior to processing. To avoid microbial interference, the leafy greens were cut, washed with 100 mg l^{-1} chlorinated water, and the excess surface water was removed with a handheld salad spinner for 40 s. Subsequently, a mixture of the three leafy greens (approximately 100 g per species) was packaged by the Tecnova Technology Centre (Almeria, Spain) in polymeric film plastic containers (400 ml) with three modified atmospheres based on the usual gas

concentrations employed commercially: low O_2 (1–5 %) and high CO_2 (5–10 %) [7, 11]. The treatments were as follows: (i) high CO_2, MA_1: 5% O_2 + 15% CO_2 + 80% N_2, (ii) low CO_2, MA_2: 5% O_2 + 5% CO_2+ 90% N_2, and (iii) atmospheric air, MA_3: 21% O_2 + 79% N_2 (container with holes). The plastic containers were kept at 4ºC for 48 hours, and half of them (30 containers) were exposed throughout the storage time to a light treatment of 300 μmol photons m^{-2} s^{-1} photosynthetic photon flux density (PPFD). A 500 W metal halogen projector (model 906609, Massive, Barcelona, Spain) supplied the illumination and the infrared radiation was reduced using thick glass covered with water. The other 30 containers were stored in the dark (controls). Samples were collected at the beginning of the experiment (time=0 hours) and then 3, 6, 24 and 48 hours later. Results are shown in Fig. 1.

(ii) Storage under supermarket conditions (illuminated and refrigerated): Two different experiments were performed: *(a) Parsley pigment composition in supermarket shelves*: 10 parsley (*Petroselinum crispum* (Mill.) Fuss. var. neapolitanum, Apiaceae) bunches (≈125 g, mature and visually good quality) comprising light-exposed (LE) and dim light-exposed (DLE; PPFD<4 μmol photons m^{-2} s^{-1}; shaded with other bunches) bunches from cooled supermarket shelves (4ºC) monitored *in situ* in the supermarket at local time 14:00 (6 hours after the shelving lights were switched on). Five different parsley bunches were selected for each condition (LE and DLE) based on the quality and exposure criteria. *(b) Daily course of pigment change under supermarket conditions:* Changes to pigments were followed across a day in spinach, parsley, and corn salad: 09:00 (1 hour before shelving lights were switched on) to 20:00 (two hours before shelving lights were switched off). Only LE-tissues were selected this time. Ten replicates from each condition/species were directly collected from the cooled supermarket shelves (4ºC) at 0 hours, and subsequently at 5 and 11 hours. The PPFD at the shelf level was provided by the supermarket system of illumination for both experiments and was ≈ 16 ± 2.6 μmol photons m^{-2} s^{-1}. It was measured with a quantum sensor (LI-189B, Li-Cor, Lincoln, NE, USA). Results are shown in Figs. 2, 3 and 4

(iii) Storage under household refrigerated conditions: The same commercial mixture of vegetables used for the MA packaging experiments (lettuce, spinach and corn salad) were kept at 4ºC and 90% relative humidity for 48 hours without any modified atmosphere (under air) to imitate household refrigerator conditions. Results are shown in Fig. 5. In the three experimental designs, leaves with defects such as yellowing or decay were discarded. In each of the sampling periods, five replicates per treatment/condition/species were collected. Each replicate was obtained by a random sampling and uniformly mixing twenty different leaves from one

independent container for each replicate. When sampling, approximately 200 mg of fresh mass was collected, weighed, frozen in liquid nitrogen, and stored at -80ºC until biochemical analyses were performed.

Pigment analysis

Pigments were extracted and homogenised as explained in Esteban et al. [12] and pigment composition was analysed by HPLC as described previously [13] using a photodiode array detector.

Data processing and statistics

Total carotenoids were calculated by the sum of neoxanthin (N), violaxanthin (V), antheraxanthin (A), Lut, Z and β-Car and the total VAZ pool was calculated by the sum of V, A, and Z. This work focuses on the nutritional quality so the results are also expressed via the Z/VAZ ratio, which indicates the proportion of Z within the VAZ pool. This index indicates the kinetic changes in the de-epoxidation state of the xanthophyll cycle, which directly correlates with the Z content. The *S*tudent t test was performed in order to assess significant differences between times (0h and 48h) in Fig. 1. One-way Analyses of Variance (ANOVA) were performed to evaluate the pigment contents in figures 1-6, considering the treatment as a fixed factor. All data were tested for normality and homogeneity of variances and log-transformed if necessary. Student–Newman–Keuls tests were used to discriminate between different treatments. All of the effects discussed in this paper were significant at $\alpha = 0.05$. All statistical analyses were performed using the SPSS 24.0 statistical package (IBM Corp., NY, USA).

Results and discussion

Plants photoprotect their tissues via operation of the VAZ-cycle, which regulates the thermal dissipation of excess light energy [4]. Over the last three decades, a huge effort has been employed to characterise this cycle through a comprehensive and integrated view of the photoprotection in plants. This has been a requisite to explore the way that Z or Lut could be enhanced in vegetables and fruits for foodstuffs with high contents of these phytochemicals. An interesting fact is that vegetables are active even after being detached and harvested. They remain responsive to environmental stimuli [9]. Indeed, the environment in which these vegetables are confined can be extremely important to the final content of phytochemicals [3, 6]. This fact is usually underestimated by postharvest techniques, which usually focus on the maintenance of good-looking fruits and vegetables, but not on the nutritional content. Although carotenoid responses to endogenous and environmental

factors are quite complex [3], here we report different storage methods applied as common post-harvest techniques with the central goal of monitoring phytochemical dynamics, and especially Z.

1. Storage in MA packaging: The effect on pigments of the three types of MA (differential proportion of CO_2 and O_2) was analysed in three important leafy vegetables: lettuce, spinach, and corn salad (Fig. 1). The interactive effect of illumination was studied by exposing the plastic containers with each of the atmospheres to light (300 μmol photons m^{-2} s^{-1}) for 48 h, while the controls were maintained in the dark. It has been reported that modified atmospheres delay senescence, extend the life of different crops, and maintain phytochemicals [14]. This is in accordance with our results showing that modified atmospheres resulted in a substantial maintenance of chlorophyll content in the three species under both light and dark conditions (Figs. 1C, G, K) and also enhancement of total Lut content (Figs. 1D, H, L). Regarding Z within the total VAZ pool, it was constant in spinach and corn salad during the darkness storage under the three treatments (MA_1, MA_2, MA_3) (Figs. 1A, E, I). A slight and transient Z/VAZ enhancement occurred in lettuce under MA_1 (Fig. 1A). Conversely, there was a significant augmentation of the Z/VAZ pool under illumination (Figs. 1A, E, I) for all the species (reflected also in the Z content; Figs. 1B, F, J). However, this increase was dependent on the atmosphere type. The highest increase was achieved in all the species under MA_1 (which has lower O_2 and higher CO_2). Indeed, the maximum Z content for all vegetables was accomplished after the first 24 hours under light storage. After this period of light exposure, the content dramatically decreased in all the treatments in all three species—probably in relation to the continuous light exposure. Interestingly, the total VAZ pool was enhanced in all atmospheres for the three species (Figs. 1B, F, J) especially in lettuce and corn salad. Several works have reported the effect of controlled atmospheres on β-Car, vitamin C, phenols, or Chls [15, 16], however, this is the first report of an increase in Z under MA (low O_2 and CO_2) (Fig. 1). As explained before, light induces the enzyme VDE to catalyse the epoxidation of the V pool to Z via A. This enzyme requires ascorbate and low pH, which is generated by the pumping of protons from the stroma to the lumen [17]. The high CO_2 content inside the plastic containers may provoke an internal acidification, which probably enhances the activity of the VDE enzyme. However, the beneficial effect of the MA under high light (300 μmoles m^{-2} s^{-1}) is limited. Thus, after 24 h of exposure, there was a net decrease in Z content suggesting that constant light during post-harvest storage may deteriorate plant

tissues and lead to browning [9]. Indeed, cycles of light/darkness slow down the decline in plant tissues compared to storage under constant light or darkness [9, 18].

2. Storage under supermarket conditions (illuminated and refrigerated): The phytochemical changes due to storage under the environmental conditions that prevailed on the supermarket shelves (4ºC and low light), and this was studied *in situ* in parsley (Fig. 2). This means that some of the tissues of the selected parsley bunches were fully light exposed (LE) or not (DLE) depending on their locations on the shelves. Despite the low PPFD reaching the shelves (<16 μmol photons m^{-2} s^{-1}), the Z within the VAZ pool (Fig. 2A) and the total VAZ pool (Fig. 2B) was significantly higher in LE tissues than in DLE tissues. The Z pool was, therefore, also higher in LE than in DLE (Fig. 2B). To further investigate the effect of the refrigerated/light storage in the profile of Chls and carotenoids under usual environmental conditions of the market shelves, we studied daily changes in parsley, corn salad, and spinach (from 8:00 to 20:00) (Figs. 3 and 4). Only LE-tissues were selected this time to assure maximal changes. The ratio of Z/VAZ, the total VAZ pool, and the total Z varied throughout the day in the three species (Figs. 3A, B). However, the patterns were different. In corn salad, the Z varied during the day, peaking significantly in the afternoon (14:00) with the same pattern in spinach (although it was not significant). In parsley, Z/VAZ and Z remained constant throughout the day (Fig. 3A, B). Regarding Chls *a*+*b*, spinach showed a significant decrease during the day versus the initial tracing (Fig. 3C). In parsley, Chl *a*+*b* also showed a dynamic response to local time (Fig. 3C). On the other hand, the Chl a/b ratio was not affected in any of the species (Fig. 3D). Neoxanthin did not change from the beginning to the end of the experiment; however, it showed dynamic behaviour (Fig. 4A), as did Lut and β-Car (Figs. 4 B, C). Total carotenoids in parsley (t-Car) increased between 8:00 and 14:00 and afterward decreased (Fig. 4D). The t-Car showed no significant changes during the day in corn salad and spinach (Fig. 4D). We report here the dynamic and circadian behaviour of carotenoids and Chls under light/dark cycles (14/10 hours) in cooled supermarket shelves. Parsley is the leafy vegetable with the most dynamic pigment composition (Figs. 2, 3 and 4). Photosynthetic pigments including the Chl *a*/*b* ratio has been described as having circadian regulation during constant darkness or light [19]. These oscillations could be attributed to the synthesis and/or degradation of trimeric light-harvesting complex II (reflected by the rhythmic changes in Chl *a*/*b*), with the antenna size minimal at night and maximal around subjective noon. The intensity of the light is also important in the pigment responsiveness. Indeed, low growth light supplemented with several short daily light pulses of higher intensity enhances the Z content in *Arabidopsis*

[20]. Here, we show that even under low light intensity (≈16 μmoles m^{-2} s^{-1}), the light responsiveness of carotenoids in parsley resulted in higher Z content in illuminated tissues than those in dim-light (Fig. 2).

3. Storage under household refrigerated conditions: Refrigerated conditions have also been demonstrated to be essential to avoid the deterioration of the fruits and vegetables, while also offering better phytochemical retention [10]. The effect of the refrigerator/dark storage of salads (including lettuce, spinach, and corn salad) was analysed over 48 h (Fig. 5). During this time, no weight loss was recorded due to refrigeration (data not shown), and the proportion of Z in the VAZ pool (Fig. 5A) and the total Z (Fig. 5B) did not show any significant changes. However, the light-harvesting apparatus that binds all the Z maintained dynamic changes as seen by the sustained decrease in the Chl *a/b* ratio for spinach and lettuce during storage in the first 10 hours (Fig. 5C). Interestingly, the total Lut pool showed a slight increase in spinach and corn salad (Fig. 5D) and the total VAZ in the three species (Fig. 5B) during refrigerator storage. This suggests that more V would be available for interconversion to Z if the vegetables were exposed to light. The dynamic behaviour of the Chl *a/b* ratio might be attributed to degradation of the trimeric light harvesting complex III due to constant dark conditions [19]. Therefore, during storage, the maintenance of a photoperiod (cycles of 12 hours of light followed by 12 hours of darkness), together with refrigerated conditions are beneficial for the vegetables' appearance and integrity, but also for the phytochemical content and the nutritional content. Regarding the total xanthophyll pool (VAZ), illumination also had a positive effect and indeed the effect took longer (up to 48 h) under all treatments (MA_1, MA_2, MA_3) (Fig. 1). In addition, refrigerated storage also increases the total VAZ (Fig. 5). This effect has been achieved in other approaches including applying stress while the plants are growing [12]. This enhancement could be due to a more long-term modulation of the xanthophyll cycle (probably new synthesis of V) [12]. This new pool may be inter-converted again to Z in the next illumination cycle if the plant tissues are again illuminated to enhance the nutritional value. Besides, some of the treatments also have a beneficial effect, slightly increasing the Lut content (Figs. 1, 5), probably due to *de novo* synthesis after long exposure (more than 24 hours).

4. Conclusions

This study demonstrates that some common habits, such as exposure to light or storage for a short time can be exploited to optimise plant nutritional quality (mainly Z for human eye health) without any disadvantages to

visual quality (based on chlorophyll content). Indeed, with low light intensities (the most common in supermarkets), the desirable effect on phytochemicals is achieved along with energy savings. The results suggest that refrigerated light intensity/cycles in the supermarket shelves and vegetable drawer can be modulated to increase carotenoid (Z) contents. This research therefore provides easy storage guidelines for supermarket management and consumers.

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Figure captions

Figure 1. Zeaxanthin kinetic changes within the total xanthophyll cycle pigments (Z/VAZ; panels A, C, E) and the total xanthophyll cycle pigments (VAZ) (μg/g DW) and the total Z content (bars; μg/g DW) (panels B, D, F), Total Chls *a*+*b* (mg/g DW) (panels C, G, K) and total Lut content (μg/g DW) (panels D, H, L) of lettuce, spinach, and corn salad during 48 h storage under modified atmospheres as follows: MA_1: 5% O_2 + 15% CO_2 + 80% N_2 (triangles), MA_2: 5% O_2 + 5% CO_2 + 90% N_2 (circles), and MA_3: 21% O_2 + 79% N_2 (diamonds) exposed to 300 μmoles $m^{-2}s^{-1}$ light (open symbols) or dark (closed symbols) conditions. Data are means ± SEM of five replicates. Error bars are shown when they are larger than the symbols. An asterisk indicates significant differences at α=0.05 between treatments (MA_{1light}, MA_{2light}, MA_{3light}, MA_{1dark}, MA_{2dark}, MA_{3dark}).

Figure 2. The Z content within the total xanthophyll cycle pigments (Z/VAZ, white bars) (panels A), the total xanthophyll cycle pigments (VAZ; dark grey bars), and the Z (black bars) (μg/g DW) (panels B) and the lutein (Lut) (light grey bars) (μg/g DW) (panels C) content of light-exposed (LE; ≈ 16 μmol $m^{-2}s^{-1}$) and dim light-exposed (DLE; <4 μmol $m^{-2}s^{-1}$) parsley in refrigerated shelves *in situ* in the supermarket. Data are means ± SEM of 5 replicates. Different lower case letters indicate statistically significant differences at $p<0.05$ after the Student-Neuman-Keul test. No letters indicates no significant difference ($p>0.05$).

Figure 3. Z content within the total xanthophyll cycle pigments (Z/VAZ; panel A), the total xanthophyll cycle pigments (VAZ) and the Z content (μg/g DW; panel B), the total Chls *a*+*b* (mg/g DW; panel C), and the ratio of chlorophyll *a* to *b* (Chl *a*/*b*; panel D) in parsley, corn salad, and spinach during a daily cycle under supermarket shelf conditions (light and refrigerated). The PPFD that reached the shelves was 16 μmol $m^{-2}s^{-1}$. The grey panels inside the graphs indicate the end of the dark period (from 22:00 to 08:00). The data are means ± SEM of 10 replicates. Error bars are shown when they are larger than the symbols. Different lower case letters denote statistically significant differences at $p<0.05$ after Student-Neuman-Keul tests. No letters indicates no significant difference ($p>0.05$).

Figure 4. Neoxanthin (Neo; panel A), lutein (Lut; panel B), β-carotene (β-Car; panel C), (μg g^{-1} DW), and total carotenoids (t-Car; panel D) (mg g^{-1} DW) in parsley, corn salad, and spinach during a day course under the supermarket shelf conditions (light and refrigerated). The grey panels inside the graphs indicate the end of the dark period (22:00-08:00). Data are means ± SEM of 10 replicates. Error bars are shown when they are larger than the symbols. Different lower case letters indicate statistically significant differences at $p<0.05$ after the Student-Neuman-Keul test. No letters indicates no significant difference.

Figure 5. The Z content within the total xanthophyll cycle pigments (Z/VAZ; panel A), the total xanthophyll cycle pigments and Z content (VAZ; panel B) (μg/g DW), the chlorophyll *a* to *b* ratio (Chl *a*/*b*; panel C) and the Lut content (panel D) (μg/g DW) in lettuce, spinach, and corn salad after 48 h under refrigerator conditions (4ºC and darkness). Data are means ± SEM of 5 replicates. Different lower case letters denote statistically significant differences at $p<0.05$ after Student-Neuman-Keul tests (black, dark grey, and white colours denote lettuce, spinach, and corn salad, respectively). No letters indicates no significant difference ($p>0.05$).

Fig 1

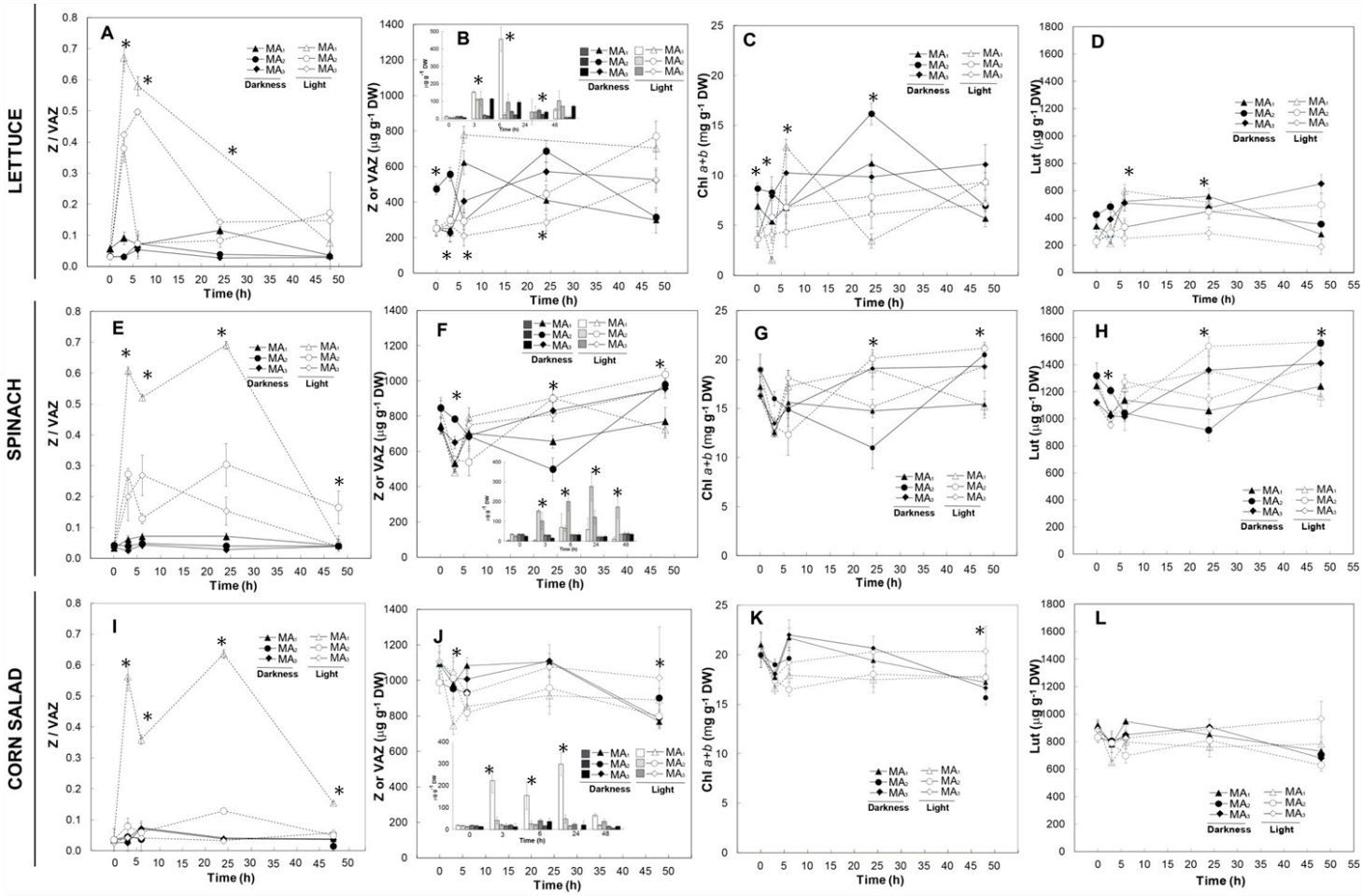

LETTUCE
SPINACH
CORN SALAD
A
B
C
D
E
F
G
H
I
J
K
L
Z / VAZ
Z or VAZ (µg g-1 DW)
Chl a+b (mg g-1 DW)
Lut (µg g-1 DW)
Time (h)
Darkness
Light
MA1
MA2
MA3

Fig 2

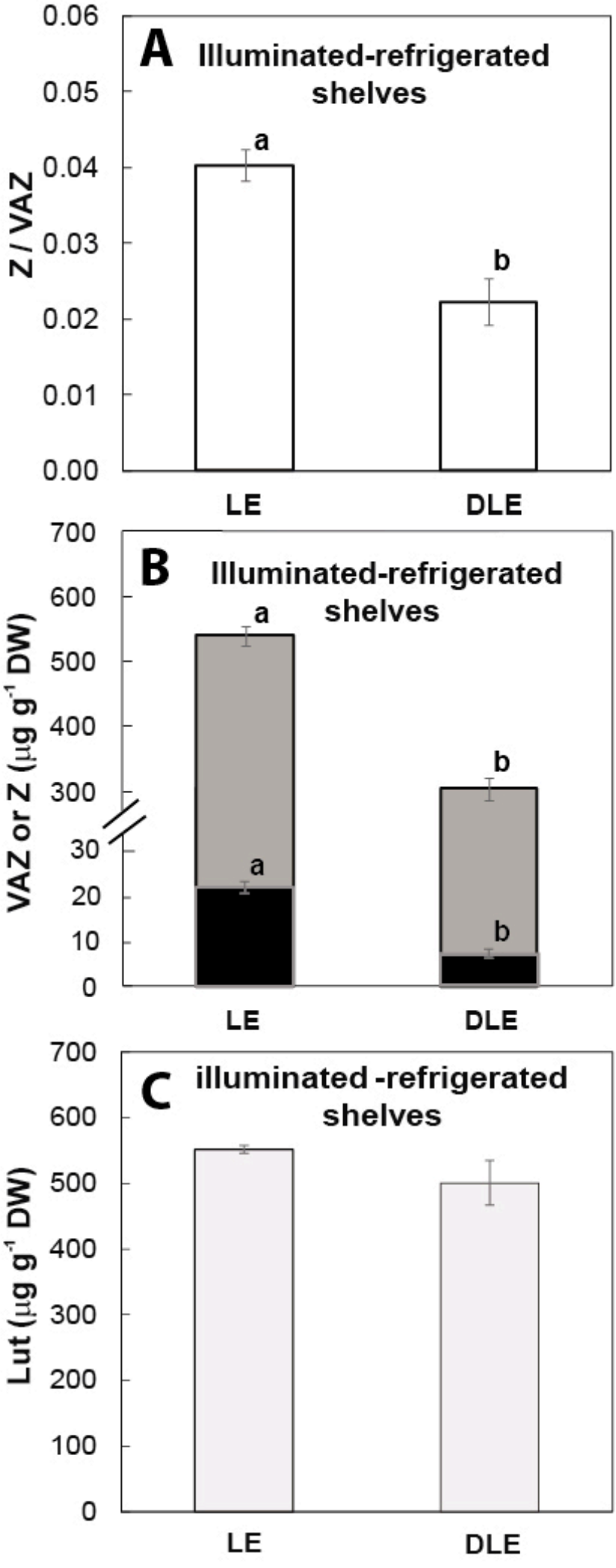

A
Illuminated-refrigerated shelves
Z / VAZ
0.06
0.05
0.04
0.03
0.02
0.01
0.00
a
b
LE
DLE
B
Illuminated-refrigerated shelves
VAZ or Z (μg g-1 DW)
700
600
500
400
300
30
20
10
0
a
a
b
b
LE
DLE
C
illuminated -refrigerated shelves
Lut (μg g-1 DW)
700
600
500
400
300
200
100
0
LE
DLE

Fig3

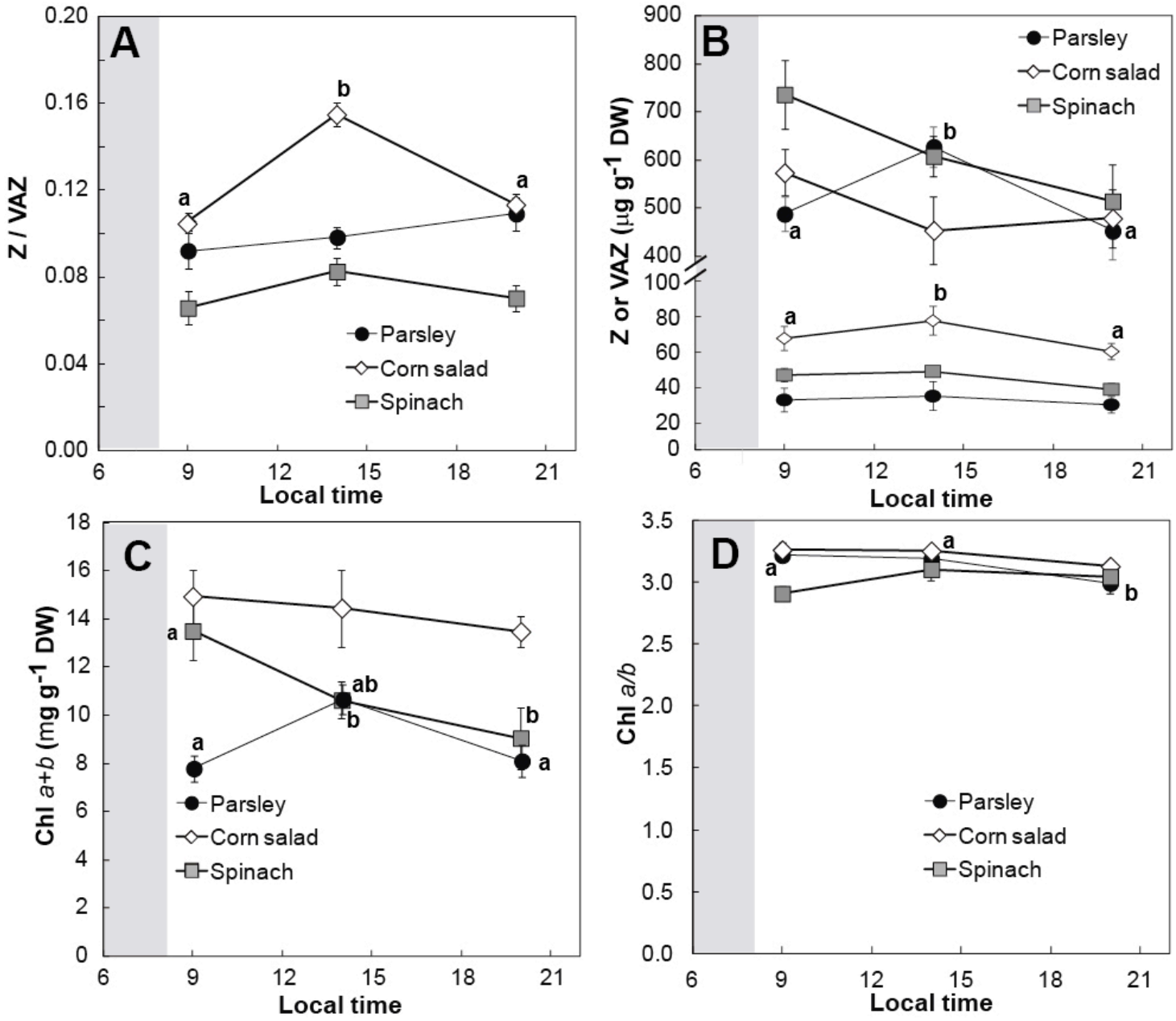

A
Z / VAZ
Local time
Parsley
Corn salad
Spinach
B
Z or VAZ (μg g-1 DW)
C
Chl a+b (mg g-1 DW)
D
Chl a/b

Fig 4

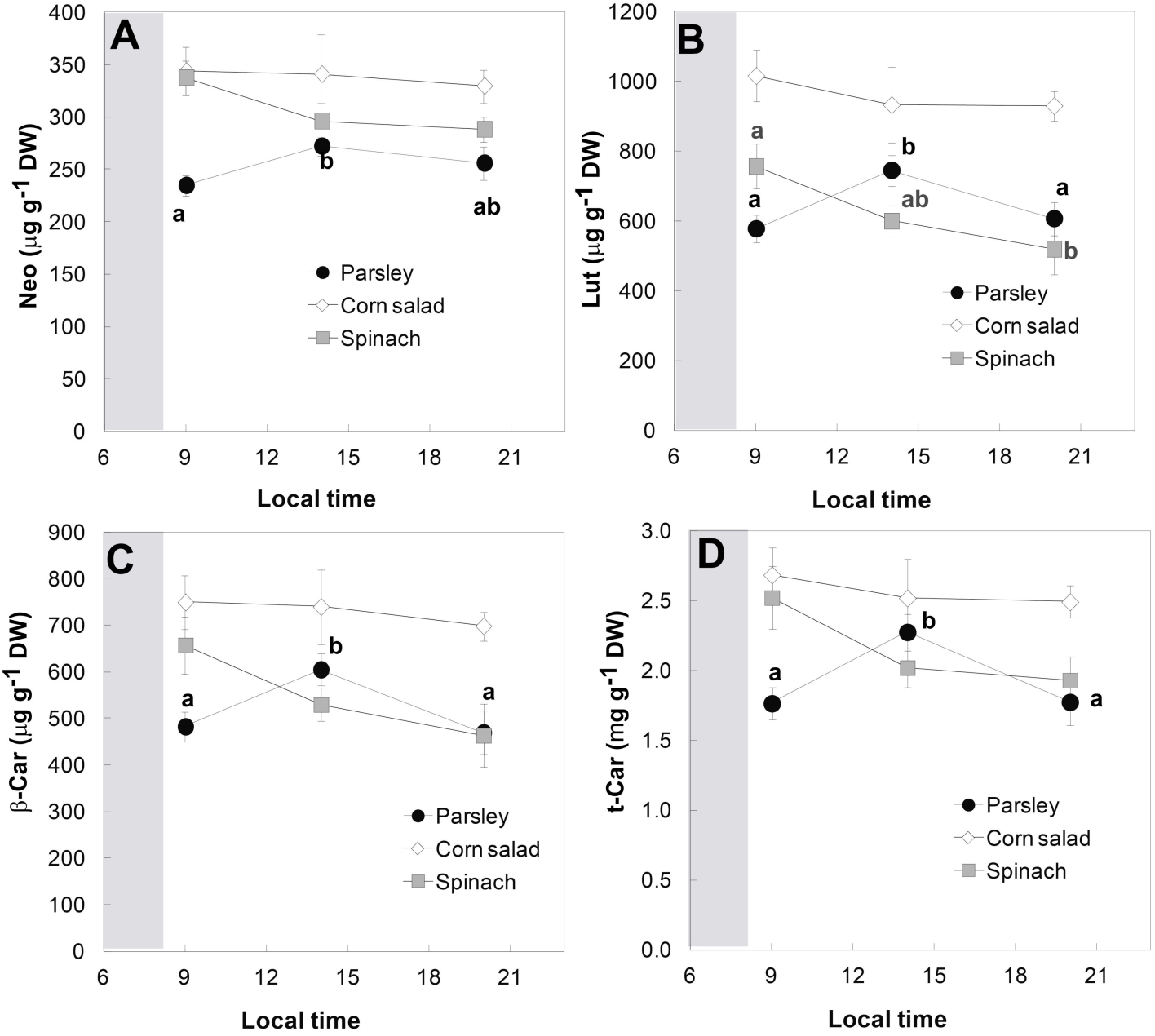
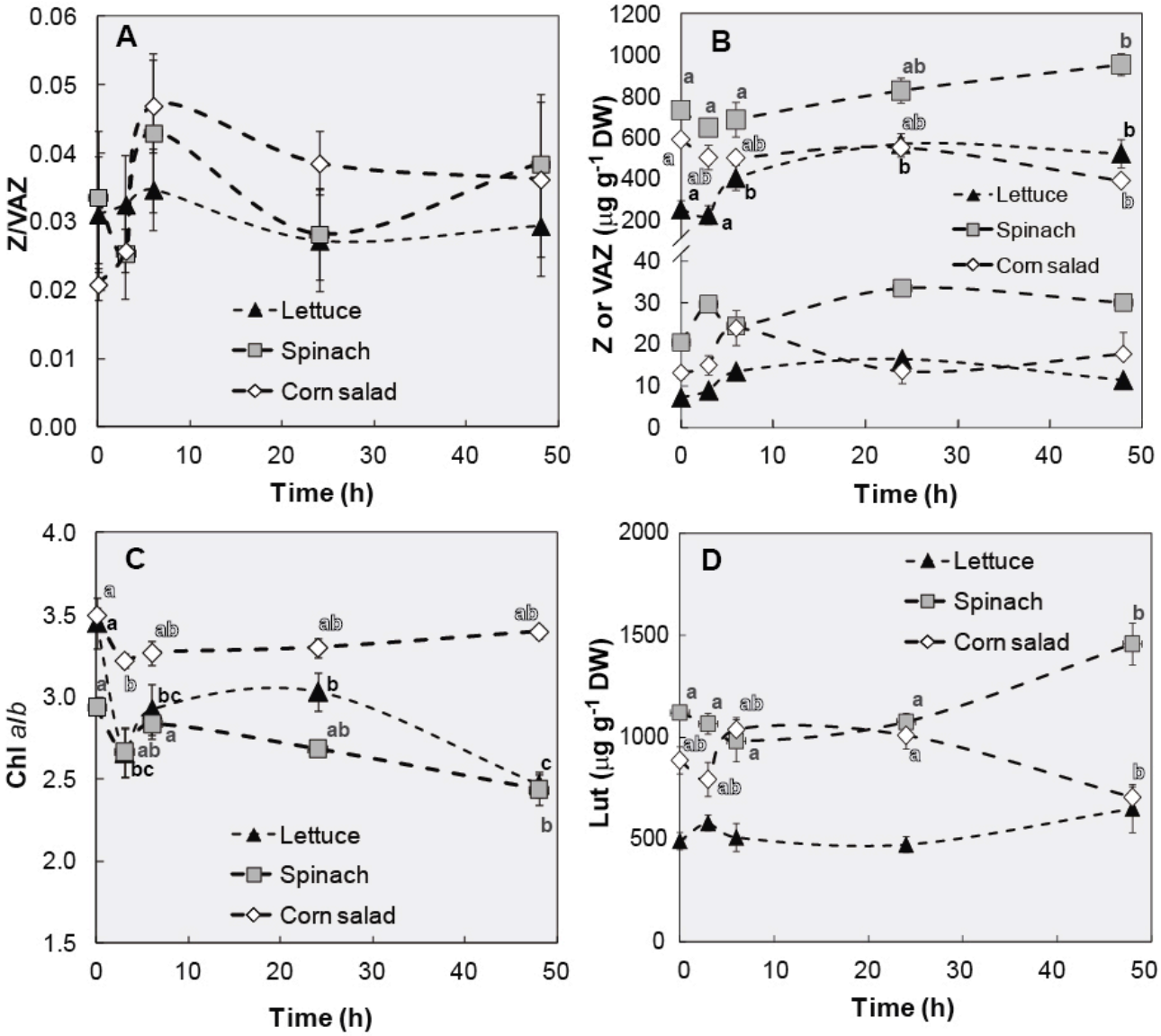

A
Neo (μg g-1 DW)
400
350
300
250
200
150
100
50
0
a
b
ab
Parsley
Corn salad
Spinach
6
9
12
15
18
21
Local time
B
Lut (μg g-1 DW)
1200
1000
800
600
400
200
0
a
b
a
a
ab
b
Parsley
Corn salad
Spinach
Local time
C
β-Car (μg g-1 DW)
900
800
700
600
500
400
300
200
100
0
a
b
a
Parsley
Corn salad
Spinach
Local time
D
t-Car (mg g-1 DW)
3.0
2.5
2.0
1.5
1.0
0.5
0.0
a
b
a
Parsley
Corn salad
Spinach
Local time

Fig 5


A
Z/VAZ
Time (h)
Lettuce
Spinach
Corn salad
B
Z or VAZ (µg g-1 DW)
Time (h)
Lettuce
Spinach
Corn salad
C
Chl a/b
Time (h)
Lettuce
Spinach
Corn salad
D
Lut (µg g-1 DW)
Time (h)
Lettuce
Spinach
Corn salad