ECOPHYSIOLOGICAL ADAPTATION OF *SEDUM CAUCASICUM*: IMPLICATIONS FOR PLANT BIODIVERSITY CONSERVATION IN THE CAUCASUS

Taliyya Orujova [1], Ulduza Gurbanova [1*], Novruz Guliyev [1]

[1]Institute of Molecular Biology, Ministry of Science and Education of the Republic of Azerbaijan, AZ1073, Baku, Azerbaijan

Abstract: *Sedum caucasicum* is a succulent species native to the Caucasus whose ecophysiological responses to environmental variation remain insufficiently understood. This study evaluated the adaptive responses of *S. caucasicum* to changes in temperature, irradiance, and water availability, with particular emphasis on CAM-related acid metabolism, leaf water status, and Photosystem II photochemical efficiency. Plants were investigated under natural conditions in the Lesser Caucasus and on the Absheron Peninsula, as well as under controlled-environment conditions. Diurnal changes in leaf titratable acidity, relative water content, and chlorophyll fluorescence were assessed. The study investigates how variations in these environmental conditions affect PSII efficiency and, in turn, the overall photosynthetic capacity of *S. caucasicum*. By integrating the study of titratable acidity with evaluations of Photosystem II efficiency, this research seeks to offer meaningful insights into how *S. caucasicum* manages CAM photosynthesis in relation to the distinctive environmental factors of the Caucasus region. Grasping these mechanisms may have wider implications for understanding drought resilience and photosynthetic adaptability in other CAM plants, especially in areas with extreme climatic conditions.
Keywords: CAM photosynthesis, chlorophyll fluorescence, drought stress, plant conservation, relative water content, titratable acidity.

***Corresponding author:** ulduzagurbanova@gmail.com

Received: 4 May 2026 **Accepted: 18 May 2026** **Published: 30 June 2026**

DOI: 10.54414/DDZQ5975

Introduction

Crassulacean Acid Metabolism (CAM) photosynthesis represents a specialized adaptive mechanism observed in various vascular plant species, which has likely evolved independently between 68 and 104-times during plant evolution. Currently, CAM is identified in approximately 5% of vascular plant species, comprising around 320 genera across 38 distinct families (Gilman & Edwards 2020; Gilman et al., 2023). Most CAM-utilizing plants are angiosperms, which exhibit a range of adaptations conducive to survival in arid environments.

CAM taxa are distributed across nearly all continents, apart from Antarctica, where environmental conditions are unsuitable for their growth. These plants encompass a variety of large and small succulents characterized by thickened stems and fleshy leaves, predominantly thriving in seasonally dry and semi-arid environments found in tropical and subtropical regions. Interestingly, although most CAM plants are associated with arid areas, numerous species also originate from humid tropical environments. These adaptations enable them to successfully inhabit a range of ecological settings, from the consistently wet conditions of tropical rainforests to the variable climates of temperate biomes, where they experience both wet and dry seasons. This adaptability underscores the ecological versatility of CAM plants and their capacity to thrive in diverse environmental conditions (Holtum 2023).

Species within the genus Sedum (Crassulaceae) are characterized by a high degree of variation, ranging from the C3 photosynthetic pathway to facultative and obligate CAM (Fioretto et al. 1999).

To better understand the plant's adaptability, we explored how *S. caucasicum* responds to various environmental factors, specifically temperature variations, light intensity levels, and the effects of drought stress.

The objective of these experiments was to enhance the understanding of this plant species and its potential resilience in the face of climate change.

Materials and Methods

Field studies were conducted in the Lesser Caucasus region, with a specific focus on the Tovuz district, (40°44′43″N 45°28′11″E / 40.74528°N 45.46972°E / 40.74528; 45.46972) as well as on the Absheron Peninsula (40°27'49.20" N 49°57' 27.29" E). These studies were undertaken under natural environmental conditions to facilitate the observation of *S. caucasicum* in its native habitat. *S. caucasicum* were collected from various rocky habitats, providing valuable information regarding their natural growth conditions and ecological adaptations (**Figure 1**).

Figure 1. *Sedum caucasicum* plant growing in natural conditions (Lesser Caucasus, Tovuz region).

Furthermore, controlled environment experiments were implemented in growth chambers, which are designed with adjustable temperature and light settings. This approach allowed researchers to create and manipulate specific environmental conditions to assess the physiological responses of *S. caucasicum* to various stresses.

Titratable acidity

For the analysis of titratable acidity in leaf disc samples, the samples were frozen to preserve their chemical integrity and then extracted using 4 mL of boiling water. A 1-mL aliquot of the solution was taken for titration, during which a 10 mM sodium hydroxide solution was added. Phenolphthalein served as the indicator to accurately determine the endpoint, indicated by a color change from acidic to neutral. This method allows for precise quantification of titratable acidity in the leaf samples (Maxwell et al. 1999).

Photosystem II activity

PSII photochemical efficiency was assessed using chlorophyll fluorescence measurements. Parameters such as Fv/Fm (maximum quantum yield) were recorded to evaluate PSII performance under varying light intensities and temperatures.

Determination of RWC

To measure the leaf RWC fresh mass, the mass after rehydration (equilibrated at 100% RH, at 4°C in distilled water for 24 h in the dark), and dry mass were determined. RWC was measured using the following formula:

$$\mathbf{RWC = [(FW-DW)/(SW-DW) \times 100}$$

where FW is the fresh weight, DW is the dry weight, and SW is the weight after rehydration (Castillo 1996).

Results

S. caucasicum is an obligate CAM plant found in temperate zones. In July-August 2016–2017, we studied the variation in the value of titratable acidity during the early and late parts of the day in species of the genus *Sedum* distributed under natural conditions in the northern part of the Lesser Caucasus. Among the species studied, only *S. caucasicum* exhibited high titratable acidity in the early morning, which decreased during the evening hours. *S. caucasicum* is a perennial plant that stores nutrient reserves in its dense roots, a characteristic that suggests it has a geophytic root system.

The intensive growing season of *S. caucasicum* is late spring and the first half of summer. The fact that *S. caucasicum* stops growing in the same year in response to unfavorable conditions in the growing environment suggests that it is an interesting indicator plant. Observations of *S. caucasicum* in its natural environment show that the arrangement of leaves on the stem prevents the upper leaves from shading the lower ones. This arrangement enables the plant to optimize its use of solar energy. To evaluate the titratable acidity in the plant, leaf samples were collected every four hours throughout the day, and the changes in titratable acidity over time were analyzed (**Table 1**).

The table data illustrates the patterns of titratable acidity in plant leaves during the day, from morning to evening. Titratable acidity is significantly high in the early morning, indicating substantial metabolic activity, and decreases to its lowest levels between 16:00 and 18:00 hours.

Additionally, leaves situated in the middle tier of the plant consistently show higher acidity compared to those in the upper and lower tiers, suggesting that leaf position may influence metabolic processes.

The dynamics of daily fluctuations in titratable acidity of *S. caucasicum* plants were investigated in a comparative manner, analyzing both natural conditions and conditions specific to plants introduced to the Absheron Peninsula. The titratable acidity levels in these plants exhibited significant variation throughout the day, influenced by various environmental parameters.

Table 1. Dynamics of changes in titratable acidity in various organs and layers of leaves of *Sedum caucasicum* plants growing in natural conditions, depending on the time of day.

Plant organs	8^{00}	12^{00}	16^{00}	20^{00}	24^{00}
Newly formed leaf	44±4.2	42±4,1	38±3,3	40±4,1	44±4,3
First leaf	114±11,3	46±4,2	26±2,5	20±2,2	60±5,9
Second leaf	122±12,1	76±7,2	26±2,5	22±2,2	70±7,3
Third leaf	126±12,3	88±8,1	24±2,4	20±2,1	70±7,3
Fourth leaf	130±13,1	96±9,3	24±2,4	20±2,2	65±5,9
Fifth leaf	124±12,2	88±8,1	24±2,4	20±2,2	60±5,8
Sixth leaf	124±12,2	60±6,2	22±2,0	18±1,6	60±5,9

In natural conditions observed in the Lesser Caucasus, specifically within the Tovuz district during June and July, ambient temperatures ranged from 20 to 24°C. A marked decrease in titratable acidity in the leaves of *S. caucasicum* was recorded from 11:00 AM to 12:00 PM. Conversely, temperatures exceeding 30°C were noted in open-air conditions on the Absheron Peninsula. Under these circumstances, the decline in total acidity of the plant leaves initiated early in the morning, beginning at approximately 7:00 AM (**Table 2**).

Table 2. Dynamics of changes in titratable acidity in the leaves of *Sedum caucasicum* grown in natural growing conditions and on the Absheron peninsula in June and July depending on different hours of the day.

Temperature	Times of the day						
	07^{00}	11^{00}	15^{00}	19^{00}	23^{00}	03^{00}	07^{00}
20-24^{0}C	124±11,2	90±7,2	20±2,1	18±0,8	60±6,1	116±11,2	120±11,2
30-32^{0}C	100±9,4	50±4,1	16±0,8	16±0,8	65±6,3	128±12,3	110±10,2

Note: The titratable acidity is expressed in μmol g-1 fresh weight.

To explore the effect of temperature, an essential environmental parameter, on the dynamics of titratable acidity changes, an investigation was conducted on *S. caucasicum* plants under varying temperatures in an artificial climate setting. The study focused on the variations in titratable acidity at different times throughout the day. The findings indicate that elevated temperatures significantly enhance the rate of stored malate consumption, as illustrated in **Figure 2**. The titratable acidity present in the leaves of plants positioned within an artificial climate chamber at a temperature of 30°C at 8:00 AM was quantified as H^+ = 140 ± 12.3 μmol·g^{-1} of crude weight. This measurement serves as a baseline for understanding the physiological responses of the plants under specific climate conditions. However, upon assessment at 14:00 hours, a notable decline in titratable acidity was observed in the leaves of *S. caucasicum* plants subjected to a cooler temperature of 12°C. This reduction amounted to a 1.75-fold decrease, resulting in a titratable acidity measurement of H^+ = 80 ± 6.2 μmol-g^{-1}. Conversely, the plants that were maintained in the warmer environment of 30°C demonstrated a more significant decline in acidity levels, exhibiting a 4-fold reduction, with a recorded value of H+ = 35±2 μmol·g^{-1} equating to a crude weight of 0.8 μmol·g^{-1}.

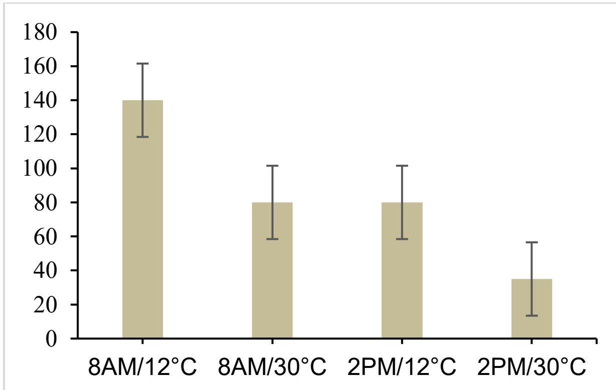

Figure 2. Changes in titratable acidity in *Sedum caucasicum* plants over time and in relation to temperature.

These findings substantiate the hypothesis that temperature acts as a critical environmental parameter that profoundly influences titratable acidity within plant tissues. Specifically, enhanced temperatures appear to accelerate the metabolic processes that lead to a more rapid consumption of organic acids in the leaves. This phenomenon likely signifies an increase in the decarboxylation processes, which are catalyzed by enzymatic activities that are temperature-dependent.

In summary, the research substantiates the assertion that the photosynthetic phases of *S. caucasicum* plants, specifically CAM-photosynthesis, exhibit periodic variations that are closely linked to alterations in titratable acidity throughout different time intervals within the diurnal cycle. This relationship underscores the importance of temperature as a regulating factor in the physiological and biochemical processes occurring within these plants.

The dynamics of change in the relative amounts of water and titratable acidity in the leaves of *S. caucasicum* plants were examined under two different conditions: regular irrigation over a period of 33 days and exposure to drought followed by re-irrigation. For this experiment, the plants were divided into two distinct groups. The first group received regular watering, while the second group, which experienced drought stress, was not irrigated for 24 days. Subsequently, this drought-stressed group was re-watered every other day for a duration of 9 days, with measurements taken on the third day. As illustrated in **Figure 3A**, both indices demonstrated elevated values throughout the experiment, with minimal variability observed in the dynamics of change in the relative amounts of water and titratable acidity in the leaves of the regularly irrigated plants.

The study examined the dynamics of changes in the relative amounts of water and titratable acidity in the leaves of *S. caucasicum* plants that were subjected to regular irrigation for a duration of 33 days, followed by a period of drought stress and subsequent re-watering. The experimental subjects were divided into two groups. The first group received consistent irrigation, while the drought-stressed plants were deprived of water for 24 days. Subsequently, for an additional 9 days, these plants were watered every other day, akin to the irrigated plants, with measurements taken on alternate days. The results, as illustrated in **Figure 3**, indicate that both indices exhibited high values throughout the experiment, demonstrating minimal variation in the dynamics of the relative amounts of water and titratable acidity in the leaves of the regularly irrigated plants. During the course of the experiment, the relative water content of irrigated plant samples was observed to range between 85% and 95%. The titratable acid content exhibited variability, measuring approximately ΔH+ = 140 ± 13.2 μmol·g^{-1}, which was contingent upon the crude leaf weight. Conversely, the plant samples subjected to drought stress, as illustrated in Fig. 3B, exhibited a relative water content of approximately 80% to 90% on the initial day of the experiment, with a corresponding titratable acidity of ΔH^{+}= 136 ± 13.2 μmol·g^{-1}.

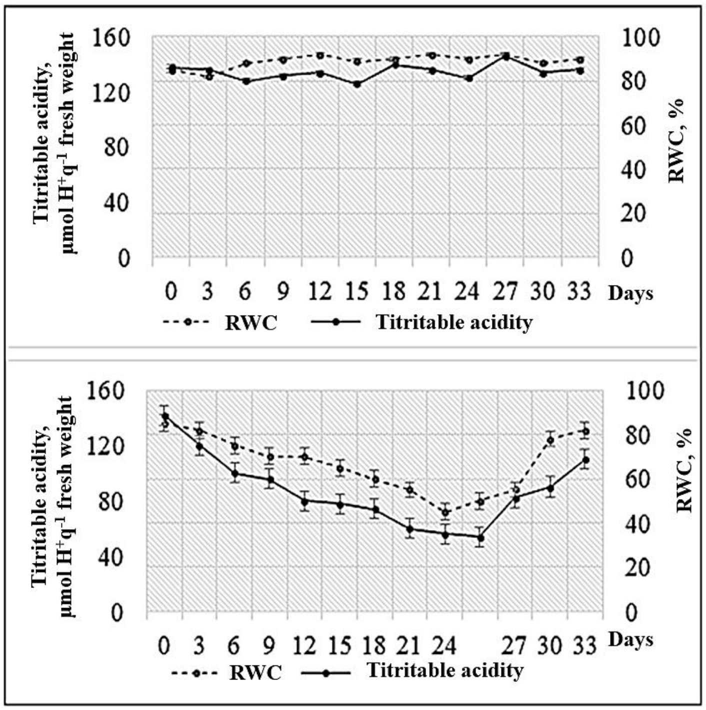

Figure 3. Dynamics of changes in relative humidity and titratable acidity in irrigated and drought-stressed *Sedum caucasicum* plants. (a) regular irrigation; (b) drought-stressed plants.

As demonstrated in **Figure 3B**, there was a notable and gradual decline in both the titratable acidity and relative water content as the duration of exposure to drought increased. On day 18, the titratable acidity had decreased by approximately 50% compared to day 1, reaching a level of ΔH+ = 50 ± 4 μmol · g-1 per leaf wet weight. By day 24, the titratable acidity further diminished to ΔH+ = 30 ± 2.7 μmol·g^{-1}, while the relative water content had reduced to between 45% and 50%.

Following the rewatering of drought-stressed plants on day 24, a marked increase in both the relative water content and titratable acidity of the leaves was observed. On day 33, the leaves of the rewatered *S. caucasicum* plants demonstrated a relative water content of 85%, with titratable acidity measured at ΔH^{+} = 130 ± 12.2 μmol·g^{-1}, based on crude weight.

This data illustrates a clear correlation between the relative water content in the leaves of the plants and the dynamic changes in titratable acidity. As a result of these experiments, a positive correlation was established between titratable acidity and metabolic activity in *S. caucasicum* leaves. The findings suggest that, with an increase in the duration of drought stress, the stomata exhibit delayed opening as a mechanism to minimize water loss from the plant. This response subsequently limits the amount of CO_2 assimilated from the atmosphere, thereby leading to a reduction in overall metabolic activity, including titratable acidity levels. The positive correlation observed between titratable acidity and relative water content in *S. caucasicum* leaves aligns with findings from experiments conducted on *P. marrubioides* (Herppich 1998) and various Crassulacean Acid Metabolism (CAM) species cultivated in field conditions (Herppich et al. 1996). These correlations appear to be influenced by fluctuations in environmental parameters. Research on certain CAM plants, such as *Kalanchoe blossfeldiana* (Pagter et al. 2013) and *Clusia uvitana* (Winter et al. 2008), has demonstrated that water deficit conditions can trigger the induction of CAM photosynthesis, resulting in an enhancement of this metabolic pathway. This phenomenon is evident in CAM species characterized by limited succulence (Davis et al. 2014; Winter & Holtum 2014), which possess the capability to perform both C_3 and CAM photosynthesis in response to varying environmental conditions. Conversely, certain CAM species experience a decline in CAM metabolism due to extended periods of drought stress, resulting in reduced concentrations of malate and diminished reserves of water within the leaves. These species are categorized as obligate CAM plants (Herppich et al. 1996). Our research indicates that the trends observed in obligate CAM species are anticipated to manifest similarly in the *S. caucasicum* species examined in our study.

Table 3. Dynamics of changes in titratable acidity of *Sedum caucasicum* plants grown outdoors on the Absheron Peninsula in July, analyzed based on time of day, relative humidity, and light intensity.

Time, hour	Temperature, $^{\circ}C$	Relative humidity, %	Light intensity, µmol m^{-2} s^{-1}	Titratable acidity, µmol $H^{+}q^{-1}$ fresh weight
07:00	30	45	37	150 ± 13,7
09:00	31	54	74	128 ± 11,6
11:00	33	41	1480	100 ± 9,4
13:00	36	44	1757.5	36 ± 3,3
15:00	32	48	1480	24 ± 0,8
17:00	30	56	111	20 ± 0,6
19:00	27	63	83.25	36 ± 1,2
21:00	27	65	dark	56 ± 2,3
23:00	26	70	dark	64 ± 2,9
01:00	25	73	dark	84 ± 3,7
03:00	24	72	dark	98 ± 6,6
05:00	24	61	166.5	170±14,9

The study focused on the dynamics of titratable acidity changes in *S. caucasicum* plants cultivated on the Absheron Peninsula, examining the relationship between these changes and various environmental parameters (**Table 3**). Under outdoor conditions in July, leaf samples were collected from the plants at two-hour intervals to assess the titratable acidity. The findings are presented in Table 3. During the experimental period, the air temperature ranged between 30 and 33 °C, with observations recorded during the morning hours from 7:00 to 11:00. Light intensity values were noted to be low, specifically at 200 × 10 µmol m^{-2} s^{-1}. The titratable acidity measured in plant leaves at 7:00 AM was quantified at H^+ = 150 ± 13.7 µmol g^{-1} of crude mass, declining to 100 ± 9.4 µmol g^{-1} of crude mass by 11:00 AM. It was observed that acidity continued to decrease throughout the afternoon, reaching a minimum value of 20 ± 0.6 µmol g^{-1} by 5:00 PM. As indicated in the accompanying table, this period corresponds approximately to phase III of CAM. During this phase, both temperature and light intensity exhibited an increase, concomitantly with a decrease in the titratable acidity.

The partial elevation in temperature contributed to an accelerated progression of phase III, resulting in a rapid utilization of the acid that was previously stored in the vacuoles. Photosystem II (PS II) is recognized for its high sensitivity to environmental damage. Chlorophyll fluorescence serves as an optimal method for assessing the photochemical efficiency of PS II and has been effectively utilized to identify stress-induced impairments. In investigations involving the leaves of *S. cacuasicum*, the dynamics of photochemical efficiency changes in PS II were examined in plants exposed to both light and shade throughout the day, considering variables such as ambient temperature and sunlight intensity (**Table 4**).

Table 4. Changes in photochemical efficiency of photosystem II in *Sedum caucasicum* leaves under light and shade conditions.

Time	Light				Shade			
	T,^{0}C	F_0	F_m	Y	T,^{0}C	F_0	F_m	Y
8^{00}	27,6	621	2422	740	27,0	666	3104	785
9^{00}	32,0	623	2429	743	32,0	710	3277	783
10^{00}	34,5	566	1574	640	30,0	591	3056	806
11^{00}	35,2	697	1597	563	31,1	645	3113	792
12^{00}	34,1	604	1973	600	31,9	597	3332	820
13^{00}	37,1	502	1168	570	31,2	522	2629	801
14^{00}	38,4	483	1130	572	31,4	354	1721	794
15^{00}	34,8	407	962	570	30,5	301	1509	800
16^{00}	34,0	590	1372	569	30,5	517	2645	804
17^{00}	33,4	505	2293	744	30,0	637	3365	810
18^{00}	30,3	584	2695	783	29,6	644	3282	803

Note: Y, $(F_m-F_o)/F_m$ - Maximum photochemical efficiency of photosystem II.

The results obtained indicate that the photochemical efficiency of Photosystem II (PS II) in the leaves of *S. caucasicum* plants maintained under shade conditions exhibited minimal variability. In contrast, for plants exposed to natural daylight, a notable decline in photochemical efficiency was observed as midday approached, correlating with an increase in light intensity. During the subsequent hours, specifically between 2:00 and 4:00 PM, the efficiency stabilized. As ambient light intensity diminished in the evening, the photochemical efficiency regained its prior levels. These findings suggest that elevated ambient light intensity can result in damage to PS II, thereby leading to photoinhibition in plant leaves. However, the efficiency of PS II demonstrates a capacity for recovery to morning levels as light intensity and ambient temperature decrease in the evening. This observation further substantiates the rapid recovery mechanisms and significant plasticity associated with the Crassulacean acid metabolism (CAM) plants in *S. caucasicum.*

Conclusion

This study underscores the remarkable adaptability of *S.caucasicum* in managing Crassulacean Acid Metabolism (CAM) photosynthesis under various environmental stresses. Our findings highlight that *S. caucasicum* demonstrates significant plasticity in its CAM photosynthesis mechanisms, enabling it to thrive under changing environmental conditions. This adaptability has significant implications for understanding the strategies that enable plants to survive and flourish in semi-arid climates. By investigating the physiological responses of *S. caucasicum,* we gain insights into the broader ecological and evolutionary processes that affect plant adaptation in challenging environments.

It would be interesting to study changes in the upper and lower limits of the plant's distribution in mountain belts, mineral requirements in the soil, changes in local relative humidity, and response to light intensity in relation to climate variability. On the other hand, a more detailed study of the dynamics of changes in the rate of acid consumption in the light phase and its influence on the activity of photosystem II as a function of temperature and environmental illumination may be important in the study of ecophysiological characteristics.

Conflict of Interest

The authors declare that there is no conflict of interest.

References

Castillo F.J. (1996) Antioxidative protection in the inducible CAM plant *Sedum album* L. following the imposition of severe water stress and recovery. *Oecologia*, 107:469–477.

Davis S.C., LeBauer D.S., Long S.P. (2014) Light to liquid fuel: theoretical and realized energy conversion efficiency of plants using Crassulacean Acid Metabolism (CAM) in arid conditions. *Journal of Experimental Botany*, 65(13):3471–3478.

Fioretto A., Fuggi A., Papa S., de Santo A.V. (1999) Crassulacean acid metabolism in *Sedum dasyphyllum* L. *Plant Biosystem*, 133(3):239–250.

Gilman I.S., Edwards E.J. (2020) Crassulacean acid metabolism. *Current Biology*, 30(2):R57–R62.

Gilman I.S., Smith J.A.C., Holtum J.A., Sage R.F., Silvera K., Winter K., Edwards E.J. (2023) The CAM lineages of planet Earth. *Annals of Botany*, 132(4): 627–654.

Herppich W.B. (1998) Photosynthesis but not CAM responded flexibly to changes in irradiance in *Plectranthus marrubioides* (Lamiaceae). *Photosynthetica*, 34(1):1–12.

Herppich W.B., Flach B.M.T., von Willert D.J., Herppich M. (1996) Field investigations of photosynthetic activity, gas exchange and water potential at different leaf ages in *Welwitschia mirabilis* during a severe drought. *Flora*, 191(1):59–66.

Holtum J.A. (2023) The diverse diaspora of CAM: a pole-to-pole sketch. *Annals of Botany*, 132(4): 597–625.

Maxwell K., Borland A.M., Haslam R.P., Helliker B.R., Roberts A., Griffiths H. (1999) Modulation of Rubisco activity during the diurnal phases of the crassulacean acid metabolism plant *Kalanchoe daigremontiana*. *Plant*

Physiology, 121(3):849–856.

Pagter M., Petersen K.K., Kjaer K.H. (2013) Direct and indirect effects of shoot- and/or root-chilling stress on growth, photosynthesis, and osmotic root water uptake in *Kalanchoe blossfeldiana* Poelln. ‘Molly’. *The Journal of Horticultural Science and Biotechnology*, 88(5):571–579.

Winter K., Holtum J.A. (2014) Facultative crassulacean acid metabolism (CAM) plants: powerful tools for unravelling the functional elements of CAM photosynthesis. *Journal of Experimental Botany*, 65(13):3425–3441.

Winter K., Smith J.A.C. (2022) CAM photosynthesis: the acid test. *New Phytologist*, 233(2):599–609.

Winter K., Garcia M., Holtum J.A. (2008) On the nature of facultative and constitutive CAM: environmental and developmental control of CAM expression during early growth of *Clusia*, *Kalanchoë*, and *Opuntia*. *Journal of Experimental Botany*, 59(7):1829–1840.