Diurnal variations of chlorophyll and dry matter contents of *Senna occidentalis* in response to zinc and soil moisture

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Abstract

The effect of concentration of zinc ions on chlorophyll (Chl) and dry matter contents in *Senna occidentalis* (L.) Link plants was studied under various availability of soil water. Diurnally, the internal Zn^{2+} content was predominantly affected by supplied Zn^{2+} and by daytime (T_m). Chl content, Chl *a/b* ratio, and Chl stability to heat were mainly affected by soil water content, and the dry matter content depended mostly on T_m.

Additional key words: Chl stability index, dry matter accumulation, relative water content.

Introduction

Deficiency in Zn^{2+} may reduce metabolic activity because Zn^{2+} is required for physiological processes as it is a component of some essential enzymes (Welch and Norvell 1993). Zn^{2+} deficiency can also reduce transpiration rate of individual leaves (Pearson and Rengel 1995) or enhance sensitivity of photosynthesis to low temperature (Marschner and Cakmak 1989).

The aim of the present study was to assess the effects of supplied Zn^{2+} on the daily fluctuations in Chl and internal Zn^{2+} concentrations, dry matter content and relative water content (RWC) in *Senna occidentalis* plants grown in soil under different water supply.

Materials and methods

The seeds of wild *Senna occidentalis* (L.) Link (grown naturally in the Egyptian desert) were germinated and grown in plastic containers (four plants per one container) with 1.5 kg air-dry sand-silty soil (3:1). A perforated plastic tube was introduced through the soil in order to regulate the distribution of irrigation solutions. The plants were watered with half strength Hoagland nutrient solution.

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Abbreviations: Chl - chlorophyll; CSI - chlorophyll stability index; RWC - relative water content; S_m - soil moisture; T_m - daytime.

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Soil water content was adjusted to 120, 90 or 40 g(H_2O) g^{-1}(dry soil). Soil water potential which correspond to maximal field capacity was -0.033 MPa and that to permanent wilting was -1.5 MPa (Kramer 1969). The available soil water contens were calculated from the difference between water content at field capacity and at permanent wilting.

The plants received Zn^{2+} in concentrations 0, 1, 3 and 6 g m^{-3} (Zn0, Zn1, Zn3 and Zn6 plants, respectively). After 9 d of treatment, the measurements were carried out at 08:00, 13:00, 16:00 and 20:00 h (after sunset). During plant testing in laboratory, air temperature was 25, 36 and 28 °C, relative humidity of air 43, 27 and 40 % and the soil temperature 24, 33 and 30 °C, in the morning, midday and evening, respectively. For each treatment three pots were chosen at random. The mass of leaf samples was determined and then they were immediately blended with 10 cm^3 of ice-cold distilled water and the supernatant was kept in deep freeze until the time of analysis.

In sap solutions, the concentration of Zn was measured by using atomic absorption spectrometer (model *AA-630-02, Shimadzu Corporation,* Kyoto, Japan) as described by Williams and Twine (1960).

The Chl contents (*a* and *b*) were determined in 85 % acetone according to Todd and Basler (1965), whereas the stability to heat (CSI) was calculated according to Murty and Majumder (1962) as the ratio between Chl content in heated leaves (56 ± 1 °C) and Chl content in fresh leaves and expressed in %.

The leaf relative water content (RWC) was determined as described by Weatherley (1950). Dry matter accumulation corresponding to increased stem or root elongation [cm] or leaf expansion [cm^2] (accumulation/elongation ratio) was estimated as ratio between dry matter [mg] and length [cm] or area [cm^2].

The effects of single factors or their interactions were evaluated by analysis of variance (*F* values), as coefficient of determination (η^2) and simple linear correlation coefficient, *r* (Ostle 1963).

Results

***In vivo* Zn concentration:** In Zn3 plants the internal Zn^{2+} content increased at low S_m (Fig.1). A considerable differences in internal Zn^{2+} content existed among various Zn-treatments and daytimes (T_m); *e.g.*, at evening, whereas in the Zn3 plants the internal Zn^{2+} content decreased, in the Zn6 plants it increases. Hence, the both T_m and Zn^{2+} supply determined the Zn^{2+} content of *Senna* leaves. The highly significant effect of S_m and T_m as single factors and ($S_m \times Zn$) and ($S_m \times T_m \times Zn$) interactions on the internal Zn^{2+} concentration, was found.

Relative water content: Near to sunrise and sunset, RWC of the plants markedly increased, especially at high S_m (Fig. 1). A depressed RWC was found in the afternoon. Slightly lower RWC was found in Zn6 plants. Therefore, Zn^{2+}, S_m and T_m singly and both ($S_m \times Zn$) and ($S_m \times T_m$) interactions had a significant effect on the RWC.

Chl content, ratio, and stability: *Senna* leaves exposed to different Zn^{2+} concentrations showed parallel changes of both Chl *a* and *b* at the tested S_m (Fig. 2). Only minor changes in Chl *a* content were found in water stressed plants with sub-optimal Zn^{2+} concentration (Zn0). Maximum Chl *a* and *b* contents were found in water stressed Zn3 plants at times of maximum irradiance. Total Chl (*a*+*b*) content increased with reduced S_m (Fig. 3).

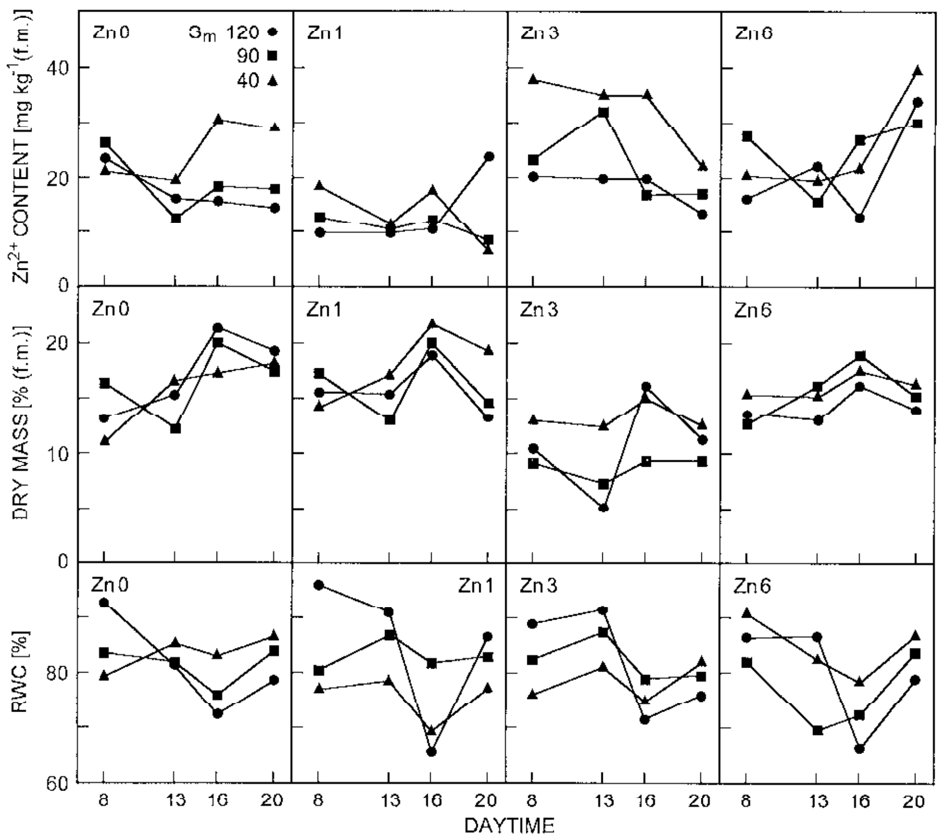

Fig. 1. Diurnal fluctuations of Zn^{2+}, dry matter content, and relative water content (RWC) in *Senna occidentalis* leaves under various external Zn supplies (0, 1, 3, 6 mg kg^{-1}) and soil moisture contents of 120 (*circles*), 90 (*squares*) or 40 (*triangles*) g(H_2O) kg^{-1}(d.m.). Dark started at 18:00 h.

The Chl *a*/*b* ratios ranged between 1.75 to 2.48. The maximum ratio was found in Zn6 water stressed plants (Fig. 2). Heat stability of both Chl *a* and *b* greatly depended on the external Zn^{2+} concentration and S_m. A high Chl stability (Fig. 3) existed at sub-optimal Zn^{2+} supply in water stressed plants, while in the Zn3 plants maximum Chl stability (CSI) was found in unstressed plants.

The statistical analysis showed that Chl contents were significantly affected by S_m, T_m and ($S_m \times Zn$) interaction. The same effect of ($S_m \times Zn$) interaction was found for Chl stability. However, Zn^{2+} concentration and ($S_m \times T_m$) interaction had a significant effect on the Chl *a*/*b* ratio. Moreover, the predominant role (judged by η^2) differed among these parameters. The Chl content was greatly affected by S_m, the ($S_m \times Zn$) and ($S_m \times T_m$) interactions had a major role on the Chl stability and Chl *a*/*b* ratio, respectively.

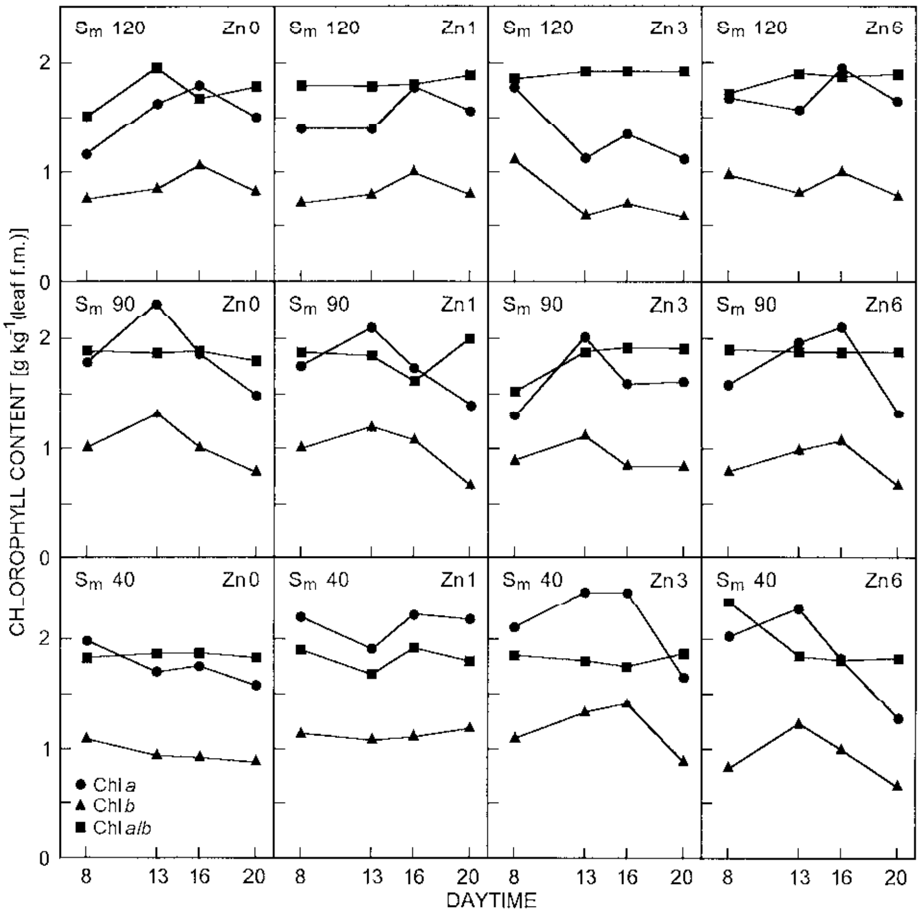

Fig. 2. Diurnal fluctuations of chlorophyll (Chl) *a* (*circles*) and *b* (*triangles*) contents as well as Chl *a*/*b* ratio (*squares*) in *Senna occidentalis* leaves at different soil moisture and Zn^{2+} concentration.

Dry matter content: At 16:00, higher dry matter contents were found in plants compared to other daytimes (Fig. 1) in both control and water stressed plants. This was in agreement with lowest RWC.

Discussion

Many assimilatory processes in leaves have a great demand for Zn^{2+} ions transported from roots to leaves *via* the xylem tissue. Leaves with the high rate of transpiration are actively growing and this may be affected by the internal Zn^{2+} concentration

(Pearson and Rengel 1995). In the investigated *Senna* plants which received moderate Zn^{2+} concentration, the internal Zn^{2+} content was elevated under water stress. The unstressed plants, had a low Zn^{2+} concentration at midday. This indicated a predominant role of the interaction between T_m and external Zn concentration. In plants that received moderate Zn^{2+} concentration, RWC markedly decreased during hot day period characterized by high evaporative demand.

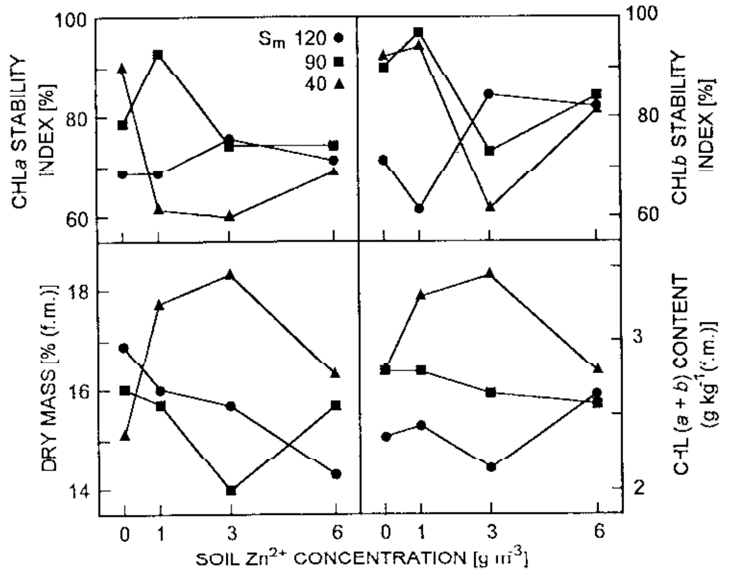

Fig. 3. Stability indexes of chlorophyll (Chl) *a*, Chl *b* and Chl (*a*+*b*), and dry matter contents in *Senna occidentalis* leaves at different Zn^{2+} concentrations and soil moistures.

The photosynthetic system adapts to increasing water stress (Aggarwal *et al.* 1984) by either an increase in Chl content or in activity of the photosynthetic apparatus (Farghali and El-Sharkawi 1990). The Chl *a* and *b* contents per fresh mass unit increased in the Zn3 plants and in plants exposed to low available water. In darkness, the Chl decreased in stressed Zn6 plants. The availability of soil water was the main factor controlling Chl contents, while the Chl *a*/*b* ratio was mainly affected by the ($S_m \times T_m$) interaction. This ratio and (CSI) help in indicating adaptations in plants (El-Sharkawi *et al.* 1986) inhabiting semi-arid areas. At low Zn^{2+} supply, the CSI was high in water-stressed plants and *vice versa*. The CSI in *Senna* leaves was influenced strongly by the interaction between external Zn^{2+} and S_m.

Good yields of leaf dry matter pointed to the improvement of plant photosynthetic systems avoiding the adverse environmental conditions (*e.g.* at mild Zn^{2+} supply). However, the excessive Zn^{2+} concentration in root medium lowered the biomass formation, despite a high content of available water. The ratio of dry matter accumulation to elongation (or expansion) in different plant organs depended on Zn^{2+} addition (Table 1). High ratio of dry matter accumulation to leaf length or area was found in leaves at sub-optimal Zn^{2+} supply, while the roots grown at high Zn gained more biomass.

Table 1. Leaf area [cm^2], stem or root length [cm], relative distribution of dry matter [%], and relative accumulation of dry matter [g cm^{-2}] or [g cm^{-1}] in *Senna occidentalis* organs at different Zn^{2+} concentrations (Zn 0 to Zn 6) and soil moisture 90 g(H_2O) kg^{-1}(soil d.m.).

	Area/length				Dry mass distribution				Relative accumulation			
	Zn0	Zn1	Zn3	Zn6	Zn0	Zn1	Zn3	Zn6	Zn0	Zn1	Zn3	Zn6
Leaf	7.0	17.6	27.3	29.8	24	22	25	23	2.3	2.2	0.6	0.5
Stem	7.3	7.2	9.7	9.5	38	30	30	29	4.5	9.0	7.7	7.4
Root	20.0	23.7	21.3	17.7	38	48	45	48	1.9	7.3	7.3	12.0

The low internal Zn^{2+} concentration enhanced the dry matter accumulation under changing S_m. This was mostly accompanied by an increase in Chl contents and photosynthetic activities. Conversely, a high concentration of internal Zn^{2+} due to excessive Zn^{2+} supply inhibited photosynthetic productivity and the Chl content decreased.

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