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DIVERSITY OF PHYSIOLOGICAL TRAITS IN *JERUSALEM ARTICHOKE* GENOTYPES UNDER NON-STRESS AND DROUGHT STRESS

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Abstract

Physiological traits such as SPAD Chlorophyll Meter Reading (SCMR), specific leaf area (SLA) and harvest index (HI) play an important role in crop yield. The objectives of this work were to study the effect of drought stress on HI, SCMR and SLA and explore genetic variability for these physiological traits in Jerusalem artichoke (JA) (*Helianthus tuberosus* L.). Field experiments were conducted in the dry period of 2010/11 and 2011/12 in the Northeast of Thailand using a strip plot design with four replications. A horizontal factor was three different water regimes (W1: 100% Crop water requirement (ETcrop), W2: 75% ETcrop and W3: 45% ETcrop) and a vertical factor was 40 JA genotypes. Measurements on HI, relative water content (RWC), SLA and SCMR were conducted at 40, 60 and 70 days after transplantation. Drought stress significantly reduced RWC and SLA but significantly increased SCMR. High variations in SCMR (32-59) and SLA (78-213 cm^2 g^{-1}) were found among genotypes. The correlations between HI and SCMR ($r = 0.56$ to 0.78, $p \leq 0.01$) were positive and significant, whereas the respective ones between HI and SLA ($r = -0.60$ to -0.76, $p \leq 0.01$) were negative and significant as those between SCMR and SLA ($r = -0.73$ to -0.90, $p \leq 0.01$). These findings suggested that SCMR was linked with SLA and HI in JA. SCMR could be used as a physiological trait for indirect selection for HI and productivity under various water regimes in JA.

Key words: Harvest index, SPAD chlorophyll meter readings, Specific leaf area, Inulin, Water regimes, Drought tolerance.

Introduction

Jerusalem artichoke (*Helianthus tuberosus* L.) is an important health food and also an alternative crop for bio-ethanol production (Kay & Nottingham, 2008). Drought is a major constraint of yield in Jerusalem artichoke (JA) (Ruttanaprasert *et al*., 2014). Improvement of drought tolerance in JA would be an alternative option to maintain the productivity and sustainability of the crop.

Any increase or decrease in yield of JA is caused by variable response of plant genotypes by means of physiological fluctuation. Thus, the development of cultivars for water limited environments would concern selection and incorporation of physiological traits related to drought resistance through breeding programs. The identification of drought resistant genotypes with good physiological traits is important for research and breeding of JA. The use of physiological traits which are easy to evaluate as selection criteria for drought tolerance should speed up the breeding programs. It is essential to access the germplasm with genetic diversity for physiological traits. An attempt has been made to understand genetic variations in physiological traits in JA germplasm to enable breeders to select parents for breeding programs.

Harvest index (HI) explains the partitioning of biomass into economic yield. Passioura (1977) proposed that grain yield of crops in water-limited environments could be analyzed in terms of 3 factors that are largely independent: grain yield = water transpired (T) × water-use efficiency (WUE) × HI. However, water transpired and water use efficiency was difficult to measure and take a long time to record. Maximizing HI is an important for sustaining yield under drought conditions (Lawn, 1988). In the tuber crop such as potato, the genotypes with high HI may contribute to achieve high and stable yields in drought prone environments (Deguchi *et al*., 2010). Thus, HI is an important trait determining yield under drought conditions.

Specific leaf area (SLA), an indicator of leaf thickness, is negatively related to SPAD chlorophyll meter reading (SCMR) and photosynthetic capacity. The thicker leaves usually have a higher density of chlorophyll and proteins per unit leaf area and have a greater photosynthetic capacity than thinner leaves. Therefore, the plant with high photosynthetic capacity could be inclinable a good varieties with a high yield. However, SLA has often been reduced under drought conditions (Marcelis *et al*., 1998; Monti *et al*., 2005). Decreases in SLA occur in response to drought in leaves as a result of a reduced transpiration leaf area, allowing increased resistance to drought conditions (Chaves *et al*., 2003).

The researches in peanut, Nageswara Rao *et al*. (2001) and Upadhyaya (2005) found a significant negative correlation between SLA and SPAD chlorophyll meter reading (SCMR), and suggested that this chlorophyll meter could be used as a rapid and reliable measure to identify genotypes with low SLA. HI was also found to be correlated with SLA and SCMR under both non stress and drought stress conditions (Songsri *et al*., 2008; Girdthai *et al*., 2012). Putative selection criteria that could be used as indirect selection to increase yield in JA have not been identified. In addition, the information on the effect of drought stress and the variation in physiological traits in a large number of JA genotypes under non stress and drought stress is rather limited has not been adequately researched.

The objectives of this study were to determine the effect of drought stress to HI, SCMR and SLA and to explore genetic variability for these three physiological traits in a large number of JA germplasm under non stress and drought stress conditions. The information would enable breeders to make decision about suitable JA parents with high SCMR and HI and low SLA in breeding program for drought resistance and to predict indirect responses for drought resistance selection.

Materials and Methods

Experimental design: Field experiment was conducted during the dry period (October to January) of two consecutive growing seasons (2010 - 2011 and 2011 - 2012) at the Field Crop Research Station of Khon Kaen University, Thailand (16°28' N, 102°48' E, 200 m above mean sea level). Soil type is Yasothon series (Yt: fine-loamy; siliceous, isohypothermic, Oxic Paleustults).

The experiment was set up in a strip plot design with four replications. Three water treatments were assigned as horizontal factor, and 40 JA genotypes with differences in morphological and physiological traits (harvest date, plant height and biomass) (Table 1) were arranged in vertical direction. A line source sprinkler system (Hanks *et al*., 1976) was installed at the center of the experiment field to supply water to JA at three water levels (W1, W2 and W3)**.** The water levels were placed horizontally along the line source sprinkler at the distances from the center of 1 - 5 (W1 = 100% ETcrop), 5 - 9 (W2 = 75% ETcrop) and 9 - 13 m (W3 = 45% ETcrop), respectively. Water amount applied to the crop of each level was measured by catch cans (24 cans for each water level treatment).

Preparation of plant materials and crop management: Seed tubers of all JA genotypes were cut into small pieces with 2 - 3 buds per piece. These tuber pieces were then pre-sprouted in coconut peat medium under ambient conditions for 4 - 7 days and later they were transferred to germinating plug trays with mixed medium containing burnt rice husk and soil for 7 days for complete sprouting. The healthy seedlings were selected for transplantation.

Conventional tillage was practiced for soil preparation, including primary plough, secondary plough, harrowing and leveling. Plot size was 2×4 m^2 with the spacing was 50×30 cm^2 of 4 rows per plot. Weed was manually hand controlled at 14 days after transplantation (DAT), and single dose fertilization of N- P_2O_5 -K_2O formula 15-15-15 at the rate of 156.25 kg ha^{-1} was spread over the trail at 30 DAT.

Water management: Water was supplied uniformly to the experimental field by drip irrigation system at water holding field capacity to facilitate uniform plant stand and crop establishment until 10 DAT. The line source sprinkler system supplied water gradients to the crop at 14 DAT until harvest. W1 level was a control treatment maintained at crop water requirement until harvest. The amount of crop water requirement was calculated as described by Doorenbos & Pruitt (1992), using the following equation;

$$ETcrop = kc \times ETo,$$

ETo is evapotranspiration of reference crop and kc is a coefficient of the crop at different growth stages. However, crop coefficient (kc) of the JA was not found in literature, and kc of sunflower was used (Monti *et al*., 2005).

Table 1. Forty genotypes of Jerusalem artichoke used in the experiment, their characteristics and sources of origin.

Genotypes	Characteristics	Sources of origin
JA 1 , JA 4, JA 6, JA 36, JA 70, JA 92, JA 114	early, short plant and low biomass genotype	PGRC[1], Canada
JA3, JA 16, JA 21, JA 37, JA 38, JA 97, JA 132	early, short plant and high biomass genotype	PGRC, Canada
JA 5, JA 122	early, high plant and low biomass genotype	PGRC, Canada
HEL 324	early, high plant and low biomass genotype	IPK[2], Germany
HEL 53, HEL 61, HEL 231, HEL 335	early, high plant and high biomass genotype	IPK, Germany
CN 52867	early, high plant and high biomass genotype	PGRC, Canada
KKUAc001	early, high plant and high biomass genotype	Jowaman Khajarern[3]
JA 61	early, high plant and high biomass genotype	PGRC, Canada
JA 46, JA 60, JA 109	late, short plant and low biomass genotype	PGRC, Canada
JA 76, JA 77	late, short plant and high biomass genotype	PGRC, Canada
HEL 62	late, short plant and high biomass genotype	IPK, Germany
HEL 246, HEL 257	late, high plant and low biomass genotype	IPK, Germany
JA 15, JA 67, JA 125	late, high plant and high biomass genotype	PGRC, Canada
JA 89	late, high plant and high biomass genotype	PGRC, Canada
HEL 65, HEL 253,HEL 256	late, high plant and high biomass genotype	IPK, Germany
JA102×JA89(8)	late, high plant and high biomass genotype	Jerusalem artichoke Research Project[4]

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Data collection

Soil and meteorological conditions: Soil moisture contents were detected by neutron probe at weekly intervals throughout the course of the experiment. The soil of both years is Yasothon soil series (loamy sand in 2010/11 and sand in 2011/12) with the subsequent chemical and physical properties (Table 2).

Weather condition of the experiment was shown in Fig. 1. Rainfall, humidity, evaporation (E_0), maximum and minimum temperature were recorded daily from transplantation until harvest by a weather station located 100 m away from the experimental field.

Table 2. Soil physical and chemical properties of experiment in 2010/11 and 2011/12.

Soil properties	2010/11	2011/12
Soil physical properties		
Sand	85%	90%
Silt	7%	8%
Clay	8%	2%
Texture class	Loamy sand	Sand
Soil chemical properties		
pH	6.08	6.12
EC (dS m^{-1})	0.03	0.03
CEC (cmol kg^{-1})	5.22	5.93
OM (%)	0.44	0.42
Total N (%)	0.02	0.01
Available P (mg kg^{-1})	23.95	37.97
K (mg kg^{-1})	33.09	37.83
Ca (mg kg^{-1})	418.33	448.75

SPAD chlorophyll meter reading (SCMR), Relative water content (RWC) and Specific leaf area (SLA): Data of SCMR, SLA and RWC were collected at 40, 60 and 70 DAT. Five plants were randomly selected in each plot to record SCMR following the procedure described by Ruttanaprasert *et al*. (2012), that SCMR was recorded from the third fully-expanded and intact leaf from the top of the main stem between 09:00 am to 11:00 am hours using a Minolta SPAD-502 meter (Tokyo, Japan).

RWC to estimate plant water status was measured following Kramer (1980), using the second leaves from the top of the main stems of 5 plants for each plot. The leaves were bored by a disc borer with 1 cm^2 in leaf area. RWC was calculated as:

$$RWC = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Saturated weight} - \text{Dry weight}} \times 100$$

Saturated weight was determined by putting the leaf sample in water for 8 hours, blot drying the outer surface, and then measuring leaf weight. The leaves samples were dried in an oven at 80°C for at least 48 hours or until the weight was constant to determine leaf dry weight. The leaves were weighted immediately after drying. SLA was derived as leaf area (the same leaves samples of RWC) per unit leaf dry weight (cm^2 g^{-1}).

Harvest index (HI): The plants were harvested at maturity. The mature plants determined by defoliation and stem browning of 50% were cut at soil surface and separated into shoots and tubers. The plants at 2 ends of the rows were discarded. As plants were bordered by adjacent plots, 14 plants in an area of 2.1 m^2 were harvested. The samples were oven-dried at 80°C for at least 72 hours or until the weight was constant. Tuber dry weight and biomass (including shoot dry weight and tuber dry weight) were recorded. HI was calculated from as tuber dry weight divides by biomass.

Statistical analysis: Analysis of variance was performed for each character followed a strip plot design (Gomez & Gomez, 1984). When the differences of main effects were significant ($p \leq 0.05$), Duncan's multiple rang test (DMRT) was used to compare means. Simple correlations were used to determine the relationships between HI, SLA and SCMR. All calculations were performed using MSTAT-C package (Fischer, 1990) and graphical presentation was conducted using Microsoft Excel.

Results

Climate, soil moisture and plant water status: Average air temperatures in 2010/11 and 2011/12 were 18.4 to 30.3 °C and 19.5 to 30.5°C, respectively (Fig. 1a, b). Daily pan evaporation ranged from 2.0 to 7.7 mm in 2010/11 and 2.2 to 9.8 mm in 2011/12 (Fig. 1c, d). The relative humidity values were 84.0% and 86.6% in 2010/11 and 2011/12, respectively.

Soil moisture contents measured by neutron method at the soil depth of 0 - 30 cm at W1 were slightly higher than those at W2, and they were generally higher than those at W3 in both years (data not shown). The plant water status for relative water content (RWC) showed clear differences among three water levels at 40, 60 and 70 DAT (Fig. 2). RWC values for W1 ranging from 62.9% to 93.6% were significantly higher than those for W2 ranging from 56.9% to 88.9%, whereas RWC values for W2 were significantly higher than those for W3 ranging from 49.7% to 82.6%.

Combined analysis of variance: Years and genotypes were significantly different for HI (Table 3). Genotypes contributed to a large portion of total variations for HI (38.8%). Years contributed to rather small portion of variation for HI (23.6%). The interaction between years and genotypes effects contributed to rather small portions (4.6%) of variations for HI. Combined analysis of variance exposed large portion and significant differences between genotypes for SCMR (39.4 - 68.3%) and SLA (35.3 - 49.0%) at 40, 60 and 70 DAT (Table 3.). The interaction effects of years × genotypes for SCMR and SLA under three water levels conditions at 40, 60 and 70 DAT were significant ($p \leq 0.01$ and $p \leq 0.05$), excepting for SLA at 60 DAT under W2 and W3 levels conditions and SLA at 70 DAT under W1 and W2 conditions. However, the portions of variances of years × genotypes interaction were smaller than genotype effect.

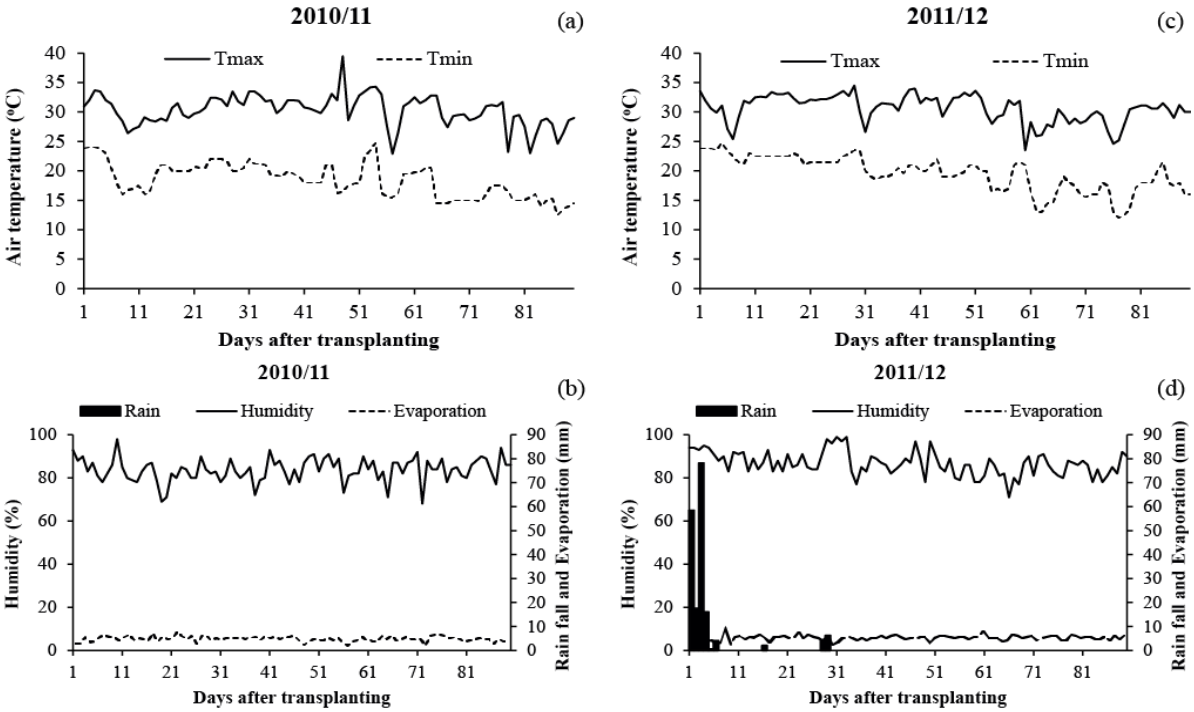

Fig. 1. The meteorological conditions as maximum air temperatures (Tmax ^{o}C), minimum air temperatures (Tmin °C), rainfall (mm), evaporation (mm) and humidity (%) in the dry period 2010/11 (a and b) and the dry season 2011/12 (c and d).

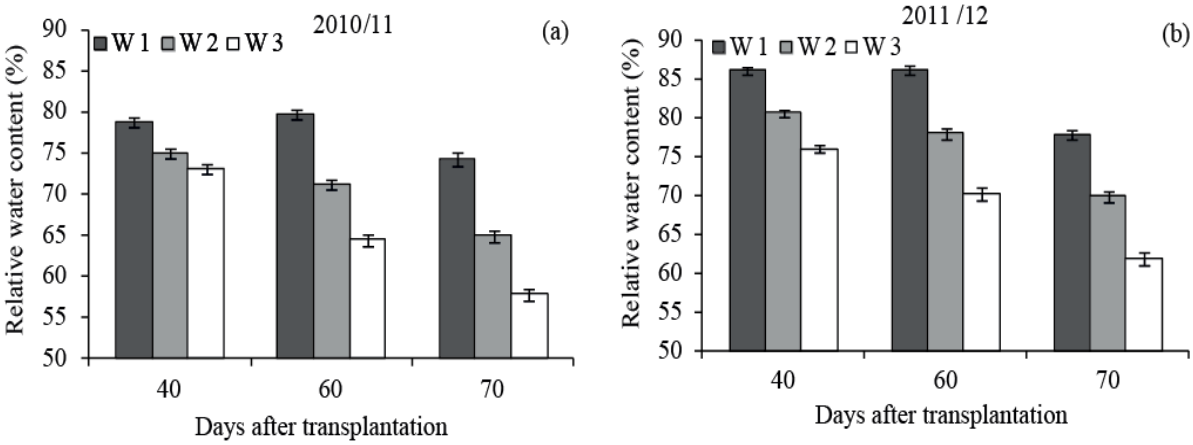

Fig. 2. Relative water content (%) at 40, 60 and 70 days after transplantation (DAT) of 40 Jerusalem artichoke genotypes grown under three water regimes 2010/2011 (a) and 2011/2012 years (b) Vertical bars indicate standard errors; (W1= 100% ET, W2= 75% ET and W3=45% ET).

Effect of drought stress on physiological traits and genetic variability: HI values for all JA genotypes varied from 0.69 to 0.89 in 2010/11 and 0.62 to 0.80 in 2011/12, and JA genotypes were significantly different under three water regimes (Table 4). SCMR were measured at 40, 60 and 70 DAT. The evaluation times at 70 DAT were not appropriate because the times were too late, based on CV and percent of sum squares values in analysis of variance. Evaluation at 40 DAT was the most appropriate sampling date to discriminate the differences among JA genotypes for SCMR as indicated by high percent of sum squares and low CV values. Therefore, only the results of 40 DAT are reported.

Drought stress significantly increased SCMR values. Genotypes and water levels were significantly different for SCMR in 2010/11 and 2011/12 (Fig. 3). In 2010/11, SCMR values at 40 DAT under W1 condition ranged from 33 to 52, under W2 condition ranged from 36 to 52 and under W3 condition ranged from 38 to 54. In 2011/12, SCMR values at 40 DAT under W1 condition ranged from 32 to 48, under W2 condition ranged from 33 to 52 and under W3 condition ranged from 35 to 53. However, the genotypes with high SCMR at 40 DAT could then be identified. JA 4, JA 36, JA 70, JA 92, JA 16, JA 21, JA 37, JA 38, JA 97, JA 5, HEL 324, JA 61, CN 52867, JA 46, JA 60, JA 77, HEL 246, HEL 257 and JA 125 had consistently high SCMR across water levels in both years.

Table 3. Mean squares from the combined analysis of variance for harvest index (HI) at final harvest, SPAD chlorophyll meter reading (SCMR) and specific leaf area (SLA) at 40 days after transplantation (DAT), 60 DAT and 70 DAT under 3 water management levels of 40 Jerusalem artichoke genotypes.

Source of variation	df	HI		SCMR						SLA					
		Harvest		40DAT		60DAT		70DAT		40DAT		60DAT		70DAT	
Years (Y)	1	1.310	(23.6)**	370	(1.6)**	1914	(8.1)**	2694	(10.9)**	15749	(2.9)**	590	(0.4)ns	1311	(1.0)**
Rep. within Y	6	0.006	(0.6)ns	155	(5.7)**	55	(8.2)**	122	(17.4)**	4000	(9.3)**	696	(12.8)*	672	(11.0)**
Waters (W)	2	0.008	(0.3)ns	672	(4.0)**	966	(1.4)**	2145	(3.0)**	24807	(4.5)**	9280	(2.9)**	6891	(3.2)**
Y x W	2	0.002	(0.1)ns	5	(0.0)ns	48	(0.4)*	311	(2.5)**	1138	(0.4)*	311	(0.4)ns	572	(0.9)**
Error (a)	12	0.005	(1.0)	11	(0.5)	10	(0.5)	8	(0.4)	196	(0.4)	202	(1.7)	69	(0.7)
Genotypes (G)	39	0.055	(38.8)**	411	(68.3)**	356	(58.6)**	250	(39.4)**	6733	(49.0)**	1511	(40.6)**	1138	(35.3)**
Y x G	39	0.007	(4.6)**	26	(4.3)**	31	(5.1)**	23	(3.6)**	771	(5.6)**	212	(5.7)**	199	(6.2)**
Error (b)	234	0.003	(14.0)	9	(8.7)	8	(7.7)	11	(10.7)	366	(16.0)	95	(16.0)	107	(19.9)
W x G	78	0.002	(2.3)ns	3	(1.0)*	6	(1.8)**	7	(2.3)**	166	(2.4)**	63	(3.4)*	63	(3.9)**
Y x W x G	78	0.002	(2.8)*	4	(1.3)**	7	(2.3)**	5	(1.7)ns	172	(2.5)**	49	(2.6)ns	41	(2.5)ns
Error (c)	468	0.001	(11.9)	2	(4.4)	3	(5.9)	4	(8.1)	83	(7.0)	44	(14.2)	41	(15.4)
Total	959														
CV%(a)		9		8		7		6		10		14		8	
CV%(b)		8		7		6		7		14		9		10	
CV%(c)		5		3		4		5		6		6		6	

Numbers within the parentheses are percent (%) of sum squares to total sum of squares

ns, *,** Non significant, significant and highly significant at p≤0.05 and ≤0.01 probability levels, respectively

SLA decreased as affected by drought stress. Means and ranges of SLA values were different among genotypes in both years (data not shown). In 2010/11, SLA at 40, 60 and 70 DAT under W1 condition ranged from 78 to 186 $cm^2 g^{-1}$, under W2 condition ranged from 83 to 168 $cm^2 g^{-1}$ and under W3 condition ranged from 82 to 165 $cm^2 g^{-1}$. In 2011/12, SLA values at 40, 60 and 70 DAT under W1 condition ranged from 92 to 213 $cm^2 g^{-1}$, under W2 condition ranged from 83 to 186 $cm^2 g^{-1}$ and under W3 condition ranged from 81 to 180 $cm^2 g^{-1}$.

Genotypic correlations for physiological traits: Positive and significant correlations were found between HI and tuber dry weight under non stress (W1) and drought stress (W2 and W3) in 2011/12 (r = 0.19, p≤0.05 to r = 0.31, p≤0.01), but the correlations were not significant in 2010/11 (data not shown). Significant correlations between HI, SCMR and SLA were observed (Table 5). Positive correlations were found between HI and SCMR at 40, 60 and 70 DAT under W1, W2 and W3 conditions (r = 0.56 to 0.78, p≤0.01 in 2010/11 and r = 0.61 to 0.71, p≤0.01 in 2011/12). The results indicated that selection for higher SCMR would result in higher HI in JA. But negative correlations between HI and SLA at 40, 60, and 70 DAT were also significant (r = 0.60 to 0.76, p≤0.01 in 2010/11 and r = 0.60 to 0.74, p≤0.01 in 2011/12). Thus, genotypes with low SLA tended to have high HI. Negative correlations were found between SCMR and SLA at 40, 60 and 70 DAT under W1, W2 and W3 conditions (r = 0.73 to 0.90, p≤0.01 in 2010/11 and r = 0.85 to 0.87, p≤0.01 in 2011/12).

Discussion

JA grown in 2010/11 had higher HI (0.80) than in 2011/12 (0.72). Lower temperature in 2010/11 might be the main cause of this difference. Lower daily minimum temperature during 7 to 14 DAT (16.0 to 19.5°C) in 2010/11 (Fig. 1a) reduced vegetative growth of JA, and biomass production in 2010/11 was lower than in 2011/12. Low temperature promoted partitioning of assimilates into harvestable tubers and therefore increased HI. In contrast, high temperature had negative correlation with HI (Ruttanaprasert *et al*., 2013), but it induced high shoot dry weight (Kocsis *et al*., 2007; Kocsis *et al*., 2008; Ruttanaprasert *et al*., 2013).

Differences in plant water status as indicated by relative water content were also similar to soil water status as indicated by soil moisture content. The plant and soil water status clearly separated water regimes and drought levels in plants and showed reasonable management of water treatments in the experiment.

The interactions between year and genotype for HI, thought significant, were very small compared to main effects (years, water levels and genotypes). The interactions between water regimes and genotypes and the interactions between years and water regimes were not significant for HI. This indicates that the genotypes perform rather consistently across years and water regimes. Low interaction favors selection of better genotypes under both of non-stress or drought stress conditions. Drought did not significantly reduced HI. Under long-term drought, HI was increased from 0.60 to 0.73 (Conde *et al*., 1991). However, HI could be reduced from 0.60 to 0.57 under terminal drought (Conde *et al*., 1991). In potato, terminal drought slightly reduced HI from 0.35 to 0.30 (Schafleitner *et al*., 2007).

The results suggested that drought has small effect on HI and in some cases could increase HI. Effect of drought on HI depends on timing of drought imposed to the crop and crop varieties. Early season drought, mid-season drought and long-term drought increased HI but terminal drought slightly reduced HI (Conde *et al*., 1991). The differences in the effects of drought at different growth stages on HI could possible due to the fact that terminal drought hindered the transport of sugar from stem to tuber and thus reduced HI, while early season drought, mid-season drought and long-term drought reduced shoot growth and increased more proportion of assimilate to the tuber (Deguchi *et al*., 2010).

Table 4. Harvest index (HI) of 40 Jerusalem artichoke genotypes in dry period 2010/11 and 2011/12.

Entry no.	Genotypes	HI			
		2010/11		2011/12	
1	JA 1	0.85	a-c	0.77	a-g
2	JA 4	0.83	c-g	0.73	f-j
3	JA 6	0.80	d-h	0.71	h-m
4	JA 36	0.84	b-d	0.75	b-h
5	JA 70	0.82	c-g	0.72	g-k
6	JA 92	0.79	f-h	0.74	b-h
7	JA 114	0.82	c-g	0.71	h-l
8	JA 3	0.85	a-c	0.74	e-i
9	JA 16	0.87	ab	0.74	c-i
10	JA 21	0.82	c-g	0.79	a-c
11	JA 37	0.84	b-d	0.78	a-e
12	JA 38	0.85	a-c	0.74	d-i
13	JA 97	0.80	d-g	0.78	a-e
14	JA 132	0.81	c-g	0.72	g-k
15	JA 5	0.83	b-e	0.74	e-i
16	JA 122	0.82	c-g	0.77	a-g
17	HEL 324	0.79	e-h	0.68	k-o
18	JA 61	0.89	a	0.78	a-e
19	CN 52867	0.83	b-f	0.75	a-h
20	KKUAc001	0.72	i-l	0.64	op
21	HEL 53	0.73	i-l	0.64	op
22	HEL 61	0.76	hi	0.66	m-p
23	HEL 231	0.73	i-k	0.66	l-p
24	HEL 335	0.70	kl	0.68	k-o
25	JA 46	0.80	d-g	0.79	a-d
26	JA 60	0.84	b-d	0.78	a-f
27	JA 109	0.80	d-h	0.68	k-o
28	JA 76	0.82	c-g	0.75	b-h
29	JA 77	0.83	b-f	0.79	ab
30	HEL 62	0.74	ij	0.69	i-n
31	HEL 246	0.83	b-e	0.72	g-k
32	HEL 257	0.82	c-g	0.80	a
33	JA 15	0.81	c-g	0.71	h-l
34	JA 67	0.70	kl	0.65	n-p
35	JA 89	0.76	hi	0.68	j-o
36	JA 125	0.82	c-g	0.80	a
37	HEL 65	0.79	gh	0.72	g-k
38	HEL 253	0.71	j-l	0.62	p
39	HEL 256	0.69	l	0.67	l-o
40	JA102 x JA 89 (8)	0.74	i-k	0.67	l-p
	Means	0.80		0.72	

Means in the same column followed by the same letter(s) are different at $p \leq 0.01$ probability levels by Duncan's multiple range test (DMRT)

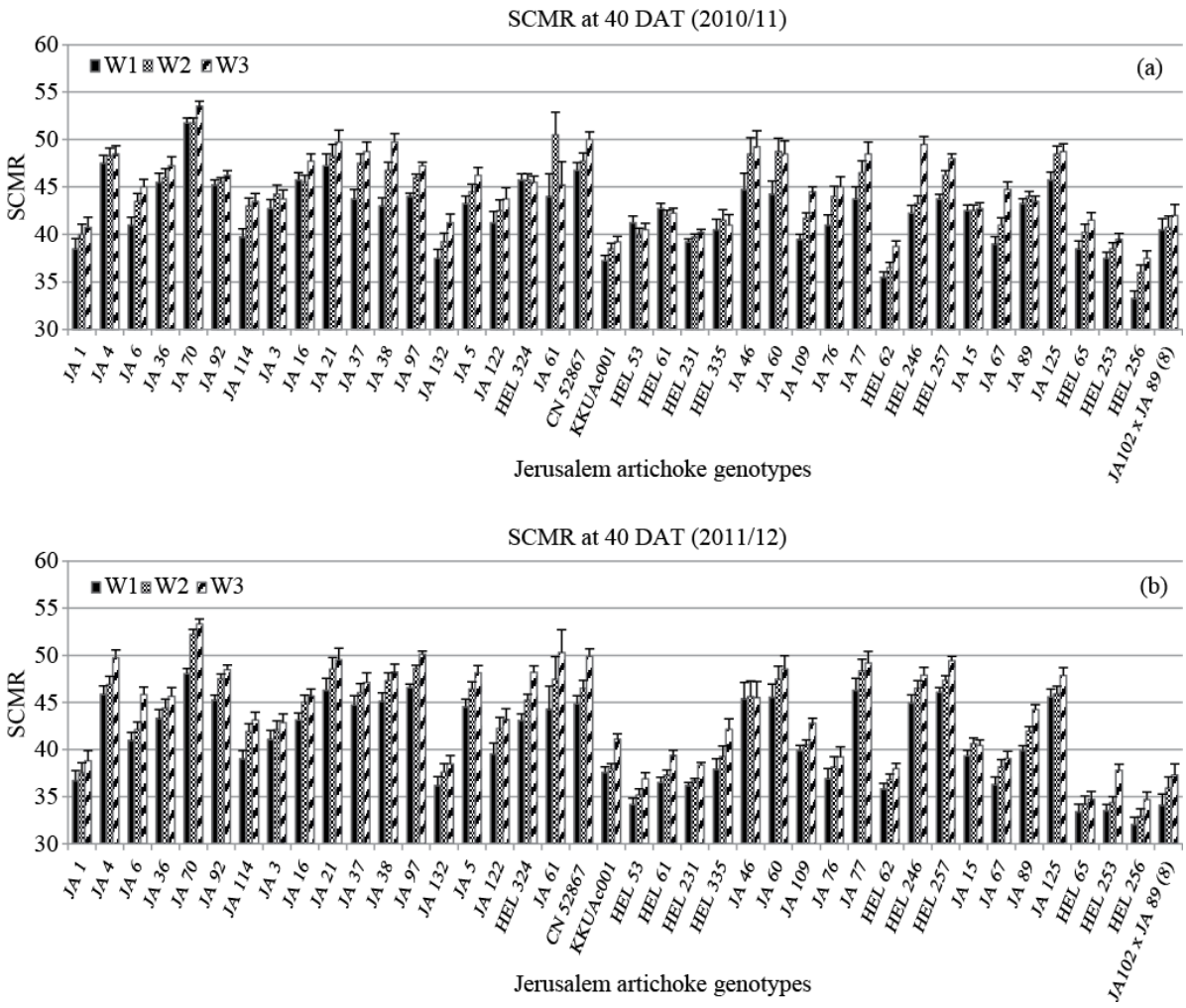

Fig. 3. SPAD chlorophyll meter riding (SCMR) of 40 Jerusalem artichoke genotypes (n = 160) for each water level, in three water levels applied at 40 days after transplantation (DAT) in 2010/11 (a) and 2011/12 (b). Bars represent the standard errors.

Table 5. Genotypic correlation among drought resistance traits (harvest index; HI, SPAD chlorophyll meter reading; SCMR and specific leaf area; SLA at 40 days after transplantation; DAT) of forty Jerusalem artichoke genotypes under W1 = 100% ETcrop, W2 = 75% ETcrop and W3 = 45% ETcrop in 2010/11 and 2011/12.

2010/11			2011/12		
Traits	**W1**		**Traits**	**W1**	
	SCMR	**SLA**		**SCMR**	**SLA**
HI	0.56**	-0.60**	HI	0.63**	-0.60**
SCMR		-0.73**	SCMR		-0.87**
Traits	**W2**		**Traits**	**W2**	
	SCMR	**SLA**		**SCMR**	**SLA**
HI	0.78**	-0.76**	HI	0.71**	-0.74**
SCMR		-0.90**	SCMR		-0.85**
Traits	**W3**		**Traits**	**W3**	
	SCMR	**SLA**		**SCMR**	**SLA**
HI	0.71**	-0.74**	HI	0.61**	-0.66**
SCMR		-0.85**	SCMR		-0.86**

** highly significant at $p \leq 0.01$ probability level

In this study, SCMR was increased as affected by drought stress, whereas SLA was decreased. The results were similar to those reported previously under drought stress (Castro-Dı´ez *et al*., 2000; Navas & Garnier, 2002; Li *et al*., 2009). Drought reduced SLA due to a significant decrease in individual leaf area and the leaves had higher tissue density under drought than under wetter conditions.

Variations in HI among JA genotypes averaged across the three water regimes in this study ranged from 0.62 to 0.89. HI values under early season drought had ranges from 0.60 to 0.63, HI values under mid-season drought had ranges from 0.61 to 0.69, HI values under terminal-drought had ranges from 0.53 to 0.57, and HI values under long-term drought had ranges from 0.72 to 0.73 (Conde *et al.*, 1991). In another tuber crop, the HI of potato under long-term drought had ranges from 0.28 to 0.39 and under terminal drought stress had ranges from 0.11 to 0.43 (Schafleitner *et al*., 2007). HI values in previous study had lower ranges than in this study because HI values in this study were observed from three water regimes whereas HI values in previous study (Conde *et al.*, 1991; Schafleitner *et al*., 2007) were determined from individual water regimes. Furthermore, the studies were also different in materials used and growing conditions. High range of HI values in this study indicated that there is a good possibility to use these genotypes for improving HI in JA.

SCMR values across water regimes in this study ranged from 32 to 59. Previous investigations reported the ranges of 32 to 46 under rainfed conditions, 33 to 44 under well-watered conditions (Monti *et al*., 2005) and 35 to 47 under well-watered conditions (Rodrigues *et al*., 2007). SCMR values were somewhat higher than those in previous studies. It is possibly due to the fact that this study included the ranges under both well-watered and drought conditions, whereas the ranges in other studies included either the range under well-watered or the range under drought. Thus, the range in this study was comparable to those in previous study. However, the range of variation in SCMR was wide enough for discrimination of JA genotypes. Therefore, SCMR can be used for some extent in screening of JA especially for germplasm with high variation for this trait.

Variation in SLA among JA genotypes across three water regimes in this study ranged from 78 to 213 $cm^2\ g^{-1}$. SLA values ranging from 30 to 196 $cm^2\ g^{-1}$ were reported for sunflower grown under non-stressed conditions (Nagarathna *et al*., 2012). Therefore, range of variation in SLA in this study was rather high, and selection for high SLA in this germplasm would be possible.

Significant and positive correlations between SCMR and chlorophyll density of JA has been reported (Ruttanaprasert *et al*., 2012). SCMR is an indicator of the photo-synthetically active light-transmittance characteristics of the leaf, which is dependent on the unit amount of chlorophyll per unit leaf area (chlorophyll density) (Richardson *et al*., 2002). Differences in levels of water supply had significant effects on SCMR and SLA in which JA genotypes subjected to severe drought had darker leaf color than did JA genotypes grown under well-watered conditions. Variations among JA genotypes were high for SCMR and SLA at 40, 60 and 70 DAT. The results indicated that it is possible to select JA genotypes for better performance for SCMR and SLA. Genetic variability of traits plays an important role on plant survival, adaptability, and can also be used to predict the genetic gain form selection in breeding programs.

The positive and close correlation between HI and SCMR under both non stress and drought stress found in this study are agreed well with the earlier finding report in peanut (Songsri *et al*., 2008; Girdthai *et al*., 2012). Therefore, SCMR can be used as indirect selection trait for improvement of HI in JA under non stress and long-term drought conditions. Close and negative correlations between SLA and SCMR were observed at all water levels. In previous studies, the close relationship between SLA and SCMR has been reported under non-stressed conditions (Nageswara Rao *et al*., 2001; Upadhyaya, 2005) and end of season drought conditions (Nigam & Aruna, 2008). In this study, the relationships between SLA and SCMR were consistent under different water regimes. JA genotypes having an ability to maintain higher SCMR and lower SLA under non stress and drought stress condition. Genotypic associations in present study demonstrated that lower SLA and higher SCMR were associated with increased HI under both of non stress and drought stress conditions. Hence, a breeding approach using SCMR traits with less expensive and rapid could be an effective tool to increase HI in JA.

Conclusions

Drought stress reduced RWC and SLA but increased SCMR in JA. HI was not significantly affected by drought stress. However, genotypes were significantly different for these traits and contributed to a large portion of total variations for these traits. High variations were found among JA genotypes for SCMR and SLA, and selection for theses character is possible. Correlation coefficients between HI and SCMR ($r = 0.56$ to $r = 0.78$, $p \leq 0.01$) were positive and significant but correlation coefficients between HI and SLA ($r = 0.60$ to $r = 0.76$, $p \leq 0.01$) were negative and significant. The correlation coefficient between SCMR and SLA ($r = 0.73$ to $r = 0.90$, $p \leq 0.01$) was negative and significant. SCMR could be used as a physiological trait for rapid assessment of relative chlorophyll status in JA genotypes as well as for indirect selection of HI and SLA under non stress and drought stress conditions in JA. JA 4, JA 36, JA 70, JA 92, JA 16, JA 21, JA 37, JA 38, JA 97, JA 5, HEL 324, JA 61, CN 52867, JA 46, JA 60, JA 77, HEL 246, HEL 257 and JA 125 had consistently high SCMR across different water levels in both years. These genotypes could be also used as parents in JA breeding for drought resistance.

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