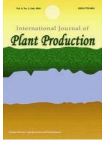

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Responses of inulin content and inulin yield of Jerusalem artichoke to seasonal environments

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Abstract

Seasonal variation (e.g. temperature and photoperiod) between growing seasons might affect inulin content and inulin yield of Jerusalem artichoke. However, there is limited information on genotypic response to seasons for inulin content and inulin yield. The objective of this study was to investigate the variability in genotypic response to seasons for inulin content and inulin yield of Jerusalem artichoke. Field experiments were conducted during the early-rainy season from June to September 2011 and the post-rainy season from September to December in 2011 and these 2 seasons were repeated in 2012 at the Field Crop Research Station of Khon Kaen University. A randomized complete block design (RCBD) with 5 replications was used. Four Jerusalem artichoke genotypes were studied during both seasons in each year. Data were recorded for brix value, inulin content, tuber yield, inulin yield, biomass and harvest index at harvest. The results revealed that seasonal variations had significant effects on inulin content, inulin yield, tuber yield, biomass and harvest index but not on brix value. The results indicated that growing Jerusalem artichoke in the early-rainy season with high temperature and long photoperiod resulted in greater inulin content and biomass. In contrast, growing Jerusalem artichoke in the post-rainy season with low temperature and short photoperiod resulted in greater tuber yield and inulin yield. The present study revealed that temperature and photoperiod were important for producing tuber yield and inulin yield. This information can be used to select the appropriate growing seasons for sustainable production of inulin content, inulin yield and tuber yield of Jerusalem artichoke in Thailand.

Keywords: Sunchoke; Growing season; Temperature; Photoperiod; Tuber yield; Fructan.

Introduction

Inulin, non-digestible oligosaccharides belong to a class of carbohydrates known as fructans. Inulin is a prebiotic that can be used to selectively stimulate growth of bifidobacteria and lactobacilli but limit growth of non-beneficial bacteria such as *Escherichia* in the intestine and thus improve host health (Coudray et al., 2005; Gibson et al., 2004). Additionally, inulin can increase intestinal absorption of minerals in humans and animals and help to prevent the development of deficiency diseases (Coudray et al., 2005). Inulin is considered as functional food that is beneficial to human health through reduction in the risk of some diseases like intestinal infections, constipation, non-insulin dependent diabetes, obesity and colon cancer (Roberfroid, 2002; Roberfroid, 2007). Several studies have shown that inulin reduces the risk of colon cancer and improve the management of inflammatory bowel diseases (Roberfroid, 2002; Pool-Zobel, 2005; Rafter et al., 2007). Clausen et al. (2012) indicated that inulin is suitable for a diabetic diet because it did not increase blood glucose levels. Moreover, inulin has low caloric value, thus, it is consumed as a dietary supplement for obese people (Kaur and Gupta, 2002). Therefore, inulin has been increasingly used in various foods due to its beneficial nutritional attributes.

Inulin a storage carbohydrate is found in many plants including leek, onion, garlic, wheat, banana, yacon, chicory and Jerusalem artichoke (Kaur and Gupta, 2002). However, only a limited number of species are suitable for non-food and industrial food applications. The main sources of inulin that are used in the food industry are chicory and Jerusalem artichoke. Jerusalem artichoke is a major source for inulin production in the tropics, where chicory cannot be grown commercially.

Jerusalem artichoke (*Helianthus tuberosus* L.) is native to North America (Kays and Nottingham, 2008) and is cultivated widely in many countries for different purposes such as for the food industry and also for the production of ethanol (Kleessen et al., 2007; Kerckhoffs and Renquist, 2013). Jerusalem artichoke can be grown successfully across a wide range of climatic environments including tropical climates in Thailand (Ruttanaprasert et al., 2013; Ruttanaprasert et al., 2014; Puangbut et al., 2012). Jerusalem artichoke can successfully be grown in two seasons (early-rainy and late-rainy) in Thailand due to its wide range of adaptability (Puttha et al., 2012). In the early-rainy season, Jerusalem artichoke is grown during May to September under high temperatures and long days while during the late-rainy season the crop is grown under medium temperatures and short days of September to December. Seasonal variations such as photoperiod, temperature and maturity, might affect inulin content and inulin yield of Jerusalem artichoke.

Previous studies indicated inulin content can be affected by photosynthesis, temperature and maturity (Ernst et al., 1995; Saengthongpinit and Sajjaanantakul, 2005; Kocsis et al., 2008; Puangbut et al., 2012). Legnani and Miller (2001) stated that inulin accumulation was increased by long photoperiod and high temperature conditions in *Dahli*a. Recent studies indicated that genotypes, days to maturity and rainfall affected tuber yield and inulin content (Puttha et al., 2012; Kocsis et al., 2008). Ruttanaprasert et al. (2013) also reported that photoperiod and growing degree days affected dry matter and partitioning of Jerusalem artichoke. However, there is limited information of genotypic response to different seasons for inlulin content and inulin yield in Jerusalem artichoke. Variation in genotype, season and genotype × season interaction can affect tuber yield, biomass, inulin content and inulin yield of Jerusalem artichoke.

Information of genotypic response for inulin content and inulin yield to different environments is important for breeding programs. High variation in genotype and genotype × year interactions (G × Y) were observed for inulin content and tuber yield and extensive evaluation in many years is necessary (Pimsaen et al., 2010; Puttha et al., 2012). However, the information on genotypic response to different seasons for inulin traits is rather limited and the extent to which the genotypic interact with the seasons has not been adequately researched. This information can then be used to develop appropriate management strategies for suitable production of inulin yield between growing seasons. The objective of this study was investigating the variability in genotypic response to seasons for inulin content and inulin yield of Jerusalem artichoke.

Materials and Methods

Plant materials and experimental design

Four Jerusalem artichoke genotypes representing three maturity classes (early, intermediate and late) were studied in both seasons for two years. CN 52867 is early maturity, JA 89 and KT 50-4 are intermediate maturity and HEL 65 is late maturity. CN 52867 and JA 89 were kindly donated from the Plant Gene Resources of Canada (PGRC), HEL 65 was kindly donated from the Leibniz Institute of Plant Genetics and Crop Plant Research (IPK) of Germany and KT 50-4 is a hybrid clone from Khon Kaen University, Thailand.

Field experiments were conducted during the early-rainy season from June to September 2011 and the post-rainy seasons from September to December in 2011 and the two seasons were repeated in 2012 at the Field Crop Research Station of Khon Kaen University. A randomized complete block design (RCBD) with five replications was used. Plot size was 6×6 m with a spacing of 50 cm between rows and 50 cm between hills in a row.

Crop management

Pre-sprouted seed tubers were used as planting materials. To prepare the sprouted seed tubers, the tubers were cut into small pieces each of which had two or three buds. The tuber pieces were incubated in plastic bags containing moist coconut peat at the bottom and the top of the bags for 7 days. The plastic bags were kept opened for good aeration. The tuber pieces with active buds and roots were further transferred to plug plastic trays containing a mixture 1:1 soil: burnt rice husk medium for about 7 days for germination. The fourth leaf-sprouted (V4) seedlings were then suitable for transplanting in the plot. One seedling was transplanted per hill. Fertilizer grade 15-15-15 was applied at 30 days after transplanting (DAT) at a rate of 156 kg per ha^{-1}. A Terraclor (quintozene 24% W/V EC) was applied monthly for 3 months after transplanting at the rate 25 mL to 20 L of water for control of stem rot (*Sclerotium rolfsii*). Supplementary irrigation was applied to the crop with an overhead sprinkler system at two day intervals.

Tuber yield, biomass and harvest index

At harvest, five plants in each plot were sampled randomly and used for determination of tuber yield, biomass, harvest index (HI) and inulin content. The stems were cut at the crowns, tubers were dug and roots were discarded. Tubers were washed in tap water to remove the potting medium. Fresh shoot weight and tuber fresh weight were determined in the field and then the weights were converted to fresh weights per area. A one kg random sample of shoot and tuber fresh weight was taken and oven-dried at 80 °C for 72 hours or until constant weight and weighed. Biomass was calculated from shoot dry weight and tuber dry weight. HI was calculated as tuber dry weight divided by the total biomass.

Total soluble solids

Three tubers of each plot were randomly selected and used for determination of total soluble solids. The tubers were cut longitudinally into small pieces. Juice containing total soluble solids was extracted from small fresh samples of tubers using a hydraulic press. A juice sample of a few milliliters was used to determine total soluble solids using a digital refractometer “model FG 103” (ATAGO, Tokyo, Japan).

Inulin content and inulin yield

Inulin content was analyzed using the methods described by Saengkanuk et al. (2011). Briefly, the tubers were longitudinally sliced into thin pieces at the middle part of the tubers. Fifty grams of sliced tuber was soaked in absolute ethanol at 4 °C for 24 h and the samples were stored at -20 °C until analyzed. The samples were oven dried at 60 °C for 10 hours. To extract inulin, 2 g of dried sample was mixed with distilled water at 80 °C for 20 minutes. The solution was cooled to room temperature and filtered through a 0.45 µm membrane filter.

The extracts (500 µl) were pipette into 25 ml volumetric flasks containing 3% HCl and diluted to 25 ml with water. The mixtures were then heated at 80 °C in a water-bath for 45 minutes. After cooling, the solutions were stored in plastic bottles before being analyzed by spectrophotometer. The detailed method for measurement for inulin content has been reported by Saengkanuk et al. (2011). Inulin analysis was shown as a percentage of inulin content on a dry weight basis and inulin yield was computed by the following formula (Puangbut et al., 2011).

Inulin yield (kgha^{-1})= inulin content (%) × tuber yield (kgha^{-1})

Statistical analysis

Analysis of variance was performed for individual seasons and error variances were tested for homogeneity by Bartlett’s test (Hoshmand, 2006). Because of, genotype × seasons interactions were significant for all characters (Table 1), data were reported for individual seasons. Duncan’s multiple range tests (DMRT) was used to compare means within genotypes and Least significant difference (LSD) was used to compare means between seasons for each genotype. Calculations procedures were done using MSTAT-C package (Bricker, 1989).

Table 1. Mean square from combine analysis of variance for brix value, inulin content, inulin yield, tuber yield (TY), biomass (BIO) and harvest index (HI) of four Jerusalem artichoke genotypes in two years and two seasons.

SOV	df	Brix value	Inulin content	Inulin yield	TY	BIO	HI
Year (Y)	1	14.0^{ns}	39.8^{ns}	19251^{ns}	143059^{ns}	4347315^{ns}	0.003^{ns}
Season (S)	1	6.4^{ns}	2010.0**	839210*	1.2E+07**	2.1E+08**	1.601**
Y × S	1	3.8^{ns}	25.9^{ns}	2.2E+07**	4.0E+07**	1.7E+08**	0.001^{ns}
Reps. within Y and S	16	4.5	80.6	326231	1025222	9971915	0.002
Genotypes (G)	3	72.5**	111.7**	1112169*	1774666*	2.2E+07*	0.036**
G × Y	3	2.3^{ns}	15.8^{ns}	242053^{ns}	512100ns	5427433^{ns}	0.005^{ns}
G × S	3	25.4**	96.2**	3029435**	3999924**	2.6E+07*	0.001^{ns}
Y × S × G	3	3.3^{ns}	16.4^{ns}	412364^{ns}	387817^{ns}	9253937^{ns}	0.004^{ns}
Pooled error	48	284.4	8.2	556286	880234	9990803	0.006

ns, * and ** non-significant, significant at $P<0.05$ and significant at $P<0.01$.

Results and Discussion

Weather conditions

Daily temperatures and photoperiod during growth in the early-rainy season and the post-rainy season in each year are shown in Figure 2. The seasonal means of maximum and minimum air temperatures ranged between 22.9 and 31.3 ºC in the early-rainy season and 20.2 and 30.6 ºC in the post-rainy season in 2011 (Figure 2a). Maximum and minimum air temperatures ranged between 23.0 and 32.4 ºC in the early-rainy season and 21.7 and 32.0 ºC in the post-rainy season in 2012 (Figure 2b). The averaged photoperiod in the early-rainy season was 13.0 h and 11.6 h in the post-rainy season in both years (Figure 2).

The mean temperature during the growing season in the early-rainy season was higher than in the post-rainy season in both years. Photoperiod in the post-rainy season was shorter than in the early-rainy season in both years. Differences in temperature and photoperiod between the seasons might have affected inulin content, inulin yield and tuber yield. Our results demonstrated that vegetative growth and tuber growth are significantly different between seasons (Figure 1). Ruttanaprasert et al. (2013) also reported that Jerusalem artichoke grown during high temperature (32.3 ºC) and long photoperiod (13.0 h) took longer to mature than when grown during low temperature (17.5 ºC) and short photoperiod (11.2 h). In addition, short photoperiod conditions gave higher rate of partitioning of dry matter to tuber (HI) and enhance tuber dry weight (Ruttanaprasert et al., 2013). The present study revealed that low temperature and short photoperiod were favorable for tuber growth and resulted in a rapid increase in dry matter in tuber.

Figure 1. Vegetative growth and tuber growth of HEL 65 Jerusalem artichoke grown in the early-rainy season (a, b) and in the post- rainy season (c,d).

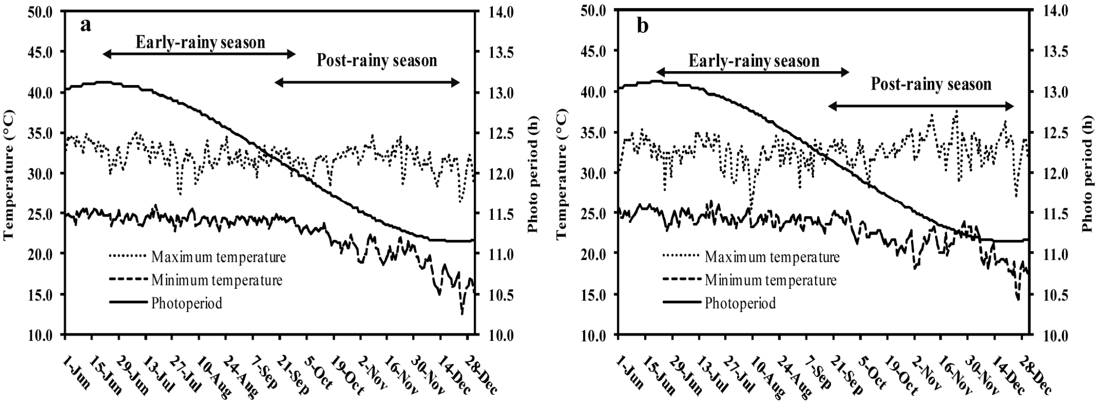

Figure 2. Daily maximum temperature, minimum temperature and photoperiod during the early-rainy and post-rainy seasons in 2011 (a) and in 2012 (b).

Combined analysis of variance

Variations between years (Y) were not significant for brix value, inulin content, inulin yield, tuber yield, biomass and harvest index (HI) (Table 1). Seasons were significant difference for all traits except brix value. This result indicated the effect of season was larger than year. The results also revealed that brix value was more stable than other traits across seasons. Jerusalem artichoke genotypes were significantly different for all traits and there were significant genotype × season interactions (G × S) for these traits. The contribution of season was higher than that genotype for inulin content and HI while genotype contributed a larger portion to the variations in Brix value, inulin yield, tuber yield and biomass. The second level interactions (Y × S × G) were not significant for all traits.

Seasonal variations in brix value and inulin content

The crop grown in the early-rainy season had higher inulin content than did the crop grown in the post-rainy season (Table 2), while brix value showed no significant differences between seasons. The results revealed that brix value was more stable than inulin content across seasons. This was in agreement with Puangbut et al. (2011) who reported that brix values were stable across years with different environments and this trait should be applicable in Jerusalem artichoke breeding programs. In contrast, Puttha et al. (2012) reported that inulin content was stable across seasons. The different observations on the stability of inulin content may be due to the use of different genetic material. Significant differences among Jerusalem artichoke genotypes were found for brix value and inulin content with CN 52867 having the highest brix value and inulin content in both seasons (Table 2).

The results indicated that inulin contents were increased in the early-rainy seasons. This could be due to high photosynthesis that resulted in high assimilate (sucrose) (Schubert and Feuerle, 1997; Legnani and Miller, 2001). Sucrose, fructose and glucose have been reported as the main parameters influencing the regulation of the activities of inulin-metabolizing enzymes (Vandoorne et al., 2012; De Roover et al., 2000). Inulin synthesis is initiated by sucrose: sucrose1-fructosyltransferase, which catalyses the transfer of a fructose moiety between two sucrose molecules to produce glucose and the trisaccharide 1-kestose (Vandoorne et al., 2012). Plant appearance in the early-rainy seasons in comparison to the post-rainy season (Figure 1) illustrated enhanced leaf area which would be expected to lead to high photosynthesis that could have contributed to the increased inulin content. Enhanced photosynthetic capacity would also enhance accumulation of fructose resulting in relatively high inulin content (Monti et al., 2005; Vandoorne et al., 2012). Jerusalem artichoke tubers produced during the early-rainy season in Thailand should be expected to have higher inulin content than those produced during the post-rainy season.

Table 2. Brix value and inulin content of four Jerusalem artichoke genotypes grown during the early-rainy seasons (ERS) and the post-rainy seasons (PRS) for two years.

Genotype	Brix value		LSD	Inulin content (%)		LSD
	ERS	PRS		ERS	PRS	
JA 89	21.1^{b}	21.9^{ab}	ns	83.0^{b}	74.9^{ab}	**
HEL 65	19.8^{b}	21.6^{ab}	ns	81.4^{b}	75.2^{ab}	**
CN 52867	25.7^{a}	23.5^{a}	ns	90.0^{a}	78.1^{a}	**
KT 50-4	21.8^{b}	19.1^{b}	ns	88.0^{a}	70.3^{b}	**
Mean	22.1	20.9		85.6	74.6	

Means in the same column with the same letters are not significantly different by DMRT ($P \leq 0.05$).
ns and ** non-significant and significant at $P \leq 0.01$, respectively by LSD.

Seasonal variations on tuber yield, inulin yield, biomass and harvest index

The effect of season was significant for tuber yield, inulin yield, biomass and harvest index (HI) (Table 1). On average, tuber yield and inulin yield in the post-rainy season was higher than the early-rainy season (Table 3). There were significant differences among Jerusalem artichoke genotypes for tuber yield and inulin yield in both seasons. The highest for tuber yield was KT-50 in both the early-rainy and the post-rainy seasons. KT 50-4 was also highest inulin yield in the early-rainy seasons while CN 52867 had the highest inulin yield in the post-rainy seasons.

Over all genotypes, biomass in the early-rainy season was higher than the post-rainy seasons while HI in the post-rainy season was higher than the early-rainy season (Table 4). Significant differences among Jerusalem artichoke genotypes were found for biomass and HI in both seasons. The highest biomass was observed in HEL 65 and CN 52867 in the early-rainy and the post-rainy seasons, respectively, whereas KT 50-4 had the highest HI in both seasons.

Table 3. Tuber yield and inulin yield of four Jerusalem artichoke genotypes grown during the early-rainy seasons (ERS) and the post-rainy seasons (PRS) for two years.

Genotype	Tuber yield (kg ha^{-1})		LSD	Inulin yield (kg ha^{-1})		LSD
	ERS	PRS		ERS	PRS	
JA 89	3,478^{b}	4,510^{ab}	**	2,834^{b}	3,378^{b}	*
HEL 65	3,655^{b}	4,140^{b}	ns	3,076^{b}	3,113^{b}	ns
CN 52867	3,324^{b}	4,775^{a}	**	2,990^{b}	3,729^{a}	*
KT 50-4	4,572^{a}	4,780^{a}	ns	4,028^{a}	3,360^{b}	ns
Mean	3,757	4,630		3,232	3,454	

Means in the same column with the same letters are not significantly different by DMRT ($P \leq 0.05$).
ns, * and ** non-significant, significant at $P \leq 0.05$ and significant at $P \leq 0.01$, respectively by LSD.

Table 4. Biomass of four Jerusalem artichoke genotypes grown during the early-rainy seasons (ERS) and the post-rainy seasons (PRS) for two years.

Genotypes	Biomass (kg ha^{-1})		LSD	Harvest index		LSD
	ERS	PRS		ERS	PRS	
JA 89	9,271^{ab}	7,128^{ab}	**	0.39^{ab}	0.65^{b}	**
HEL 65	12,656^{a}	6,532^{ab}	**	0.35^{b}	0.65^{b}	**
CN 52867	7,816^{b}	7,433^{a}	ns	0.43^{ab}	0.65^{b}	**
KT 50-4	10,151^{ab}	6,397^{b}	**	0.45^{a}	0.74^{a}	**
Mean	9,973	6,872		0.40	0.67	

Means in the same column with the same letters are not significantly different by DMRT ($P \leq 0.05$).
ns and ** non-significant and significant at $P \leq 0.01$, respectively by LSD.
Harvest index (HI) was calculated as tuber dry weight divided by the total biomass of the plant.

The present study supported previous findings that Jerusalem artichoke grown in the early-rainy season with high temperatures and long photoperiod produced higher biomass but lower tuber yields than when grown in the post-rainy season (Ruttanaprasert et al., 2013; Puangbut et al., 2012). This is similar to potato where Kozia et al. (1995) reported that total dry matter increased with long photoperiod. Jerusalem artichoke grown in the post-rainy season with low temperature and short photoperiod had higher tuber yield which contributed to high inulin yield. Our results demonstrated that the post-rainy season was favorable for inulin yield while the early-rainy season promoted accumulation of inulin content in tubers.

The present study revealed that seasonal variation was more effect on inulin content, tuber yield and inulin yield but not for brix value. The results indicated that the post-rainy season were favorable conditions for tuber yield and inulin yield while the early-rainy season was induced inulin content. This information will enable growers to make decisions and choose suitable growing seasons for Jerusalem artichoke production for high inulin content or high inulin yield in Thailand.

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