Influence of Harvest Intervals on Growth Responses and Fatty Acid Content of Purslane (*Portulaca oleracea*)

Desmond G. Mortley²

G.W. Carver Agricultural Experiment Station, Tuskegee University, 214 Milbank Hall, Tuskegee, AL 36088

Jun-Hyun Oh and Damicca S. Johnson¹

Department of Plant and Food Sciences, Sangmyung University 300 Anseo-Dong, Dongnam-Gu, Cheonan, Chungnam, 330-720, South Korea

Conrad K. Bonsi and Walter A. Hill

G.W. Carver Agricultural Experiment Station, Tuskegee University, 214 Milbank Hall, Tuskegee, AL 36088

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Abstract. A greenhouse study was conducted to evaluate the influence of harvest intervals on biomass yield and omega fatty acids of ‘Golden purslane’ (*Portulaca oleracea*). Nutrients were supplied as a modified full-strength Hoagland solution two to three times weekly. Plants were harvested sequentially at 20, 40, and 60 days after transplanting (DAT) corresponding to 42, 63, and 84 days after sowing. Fatty acids were determined using a gas chromatography–mass spectrometry. Harvest intervals significantly influenced foliage fresh and dry weight, leaf number and plant height, and root length and fresh weight and were greatest at 60 DAT. Fatty acid analysis verified the presence of myristate, palmitate, linoleate, and linolenate at 20 DAT and in all three harvests, whereas stearate and oleate were detected only in the last two harvests (40 and 60 DAT). Linoleate, palmitate, and linolenate were the most abundant fatty acids in purslane with levels in excess of 300 mg·kg⁻¹. Those for myristate, stearate, and oleate were in excess of 200 mg·kg⁻¹. The ratio of omega-6/omega-3 ranged from 0.44 for Harvest 1 to 1.1 for Harvest 3, whereas ratios for harvest intervals two and three were equal to or greater than the recommended daily human requirement. Results showed qualitative and quantitative differences of harvest intervals of purslane, suggesting that an optimal ratio of omega-6 to omega-3 fatty acids can be achieved ≈20 DAT.

Purslane (*Portulaca oleracea*) is a member of the portulacaceae family and is an annual that produces hundreds of seeds that remain dormant until conditions are favorable for germination. Over the last 10 years, purslane has gained added attention as a result of the discovery that it possessed the highest content of omega-3 fatty acid among several green leafy and succulent vegetables such as spinach, red leaf lettuce, buttercrunch lettuce, or mustard greens (Alamazan and Adeyeye, 1998; Liu et al., 2000; Palaniswamy et al., 1997, 1998, 2001; Simopoulos, 2002). Guil-Guerrero and Rodriguez-Garcia (1999) showed that leaves of purslane yielded a greater amount

of lipids/100 g dry weight compared with other edible species.

There is a fair amount of variation in the lipid content of purslane based on time of harvest. For example, Omara-Alwala et al. (1991) reported that lipid content of purslane leaves and stems varied depending on harvest time [30, 49, or 59 d after planting (DAP)]. Palaniswamy et al. (2001) reported that the concentration of polyunsaturated essential fatty acids in the leaves of purslane harvested at six-, 10- and 14-true-leaf developmental stages (corresponding to 35, 49, and 60 DAP) varied significantly. They found a lower concentration of lipids at the 10-true-leaves stage vs. at six- or 14-true-leaf stages. They also reported differences with respect to the ratio of omega-3 to omega-6 regardless of stages of development.

Liu et al. (2000) also showed that leaves, stems, and seeds of Australian purslane were an excellent source of alpha linolenic acid compared with American varieties harvested at different stages of growth (45, 60, or 70 DAP) regardless of whether plants were grown in the field or a greenhouse. Total fatty acids

and alpha linolenic acid were greater in the seeds, whereas longer-chain fatty acids were not detected in any of the samples. These reports suggest that the amount of omega-3 fatty acids in purslane depends largely on the developmental stage at which the plants are harvested as well as the plant parts analyzed.

There are several sources of omega-3 fatty acids that are available commercially, including from fish and flax seed oils. However, these are relatively expensive to obtain and adequately consumed in proper dosages. Growing and consuming purslane would be a relatively inexpensive source of this essential fatty acid. There is limited information available concerning the content and form of omega-3 and omega-6 fatty acids present in purslane at intermediate harvest intervals.

Our objective was to evaluate the influence of sequential intermediate harvest intervals on growth responses and omega-3 fatty acid content of purslane.

Materials and Methods

Experiments were conducted in a greenhouse in a completely randomized design with three replications. Each replication contained three plots with 18 plants each. Seeds of ‘Golden purslane’ (Nicholas Garden Nursery, Albany OR) were sown in moist commercial Jiffy Mix (Batavia, IL) medium in TLC Pro-Trays transplant flats (TLC Polyform, Inc., Plymouth, MN) and covered with ≈0.6 cm of the medium. Flats were placed in a greenhouse, watered as needed, and seeds germinated after 3 d. Seedlings were grown for ≈3 weeks after which they were transplanted into greenhouse beds containing a 1:1:1 (v/v) mixture of Jiffy mix, sand, and soil. Seedlings were placed 20 cm within and 25 cm between rows totaling ≈18 plants per plot. Plants were fertilized with a modified full-strength Hoagland nutrient solution (Hoagland and Arnon, 1950) two to three times weekly supplying 84, 31, and 234 μg·g⁻¹ nitrogen, phosphorus, and potassium, respectively.

Greenhouse conditions included a 12-h light/12-h dark period by supplementing natural sunlight with cool white fluorescent lamps at sundown and a matching thermoperiod of 28 °C light/22 °C dark cycle. Relative humidity was 50 ± 5% and photosynthetic photon flux at canopy level averaged 300 ± 25 μmol·m⁻²·s⁻¹.

Fourteen plants each were sequentially harvested at 20, 40, and 60 DAT and separated into tops and roots. Both plant parts were placed under running tap water followed by two successive deionized water rinses to remove adhering soil. Shoots and roots were blot-dried on paper towels after which fresh weights were taken. Plant tissues were dried at 70 °C for 72 h and dry weights of component plant parts determined. Samples of shoots for gas chromatographic analysis were frozen and stored at –80 °C before freeze drying for at least 24 h before analysis. After freeze drying, samples were ground to a fine powder in a Waring Commercial Laboratory Blender (Torrington, CT).

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¹Former Graduate Student.

²To whom reprint requests should be addressed; e-mail mortleyd@tuskegee.edu.

A gas chromatograph linked to a mass spectrometer (Shimadzu, Osaka, Japan) was used to analyze for omega-3 fatty acids according to the method of Nielson (2003). A 0.5-g sample was added to 5 mL of hexane and incubated at room temperature for 30 min on a shaker at 50 rpm followed by centrifugation at 5000 rpm for 10 min and filtered through a funnel containing 1 g of anhydrous sodium sulfate to remove excess water. Sodium methoxide reagent (1.5 mL) was added to the hexane solution and the mixture shaken for 1 min to methylate the fatty acids, to which 5 mL of NaCl solution was added. Before analyses, all samples were filtered to pass a 0.2- μ m filter to remove impurities. To identify peaks on the chromatogram, retention times, peak heights, and areas were compared with standard fatty acids as well as with those in the mass spectrometry library. Fatty acids were expressed as a percentage and compared with established standards (Nu-Chek Prep, Elysian, MN). Data were combined by treatments and tested by analysis of variance using the General Linear Model procedure (SAS Institute, 2009) with significance determined at the 0.05 level of probability.

Results and Discussion

Harvest interval significantly influenced both above- and below ground biomass (Table 1). Foliage fresh and dry weight, number of leaves per plant, plant height, root length, and root fresh weight all increased substantially as plants matured and were highest at 60 DAT. These increases in biomass with harvest intervals are consistent with those reported by Palaniswamy et al. (2001); they characterized the developmental stages in terms of the appearance of true leaves rather than DAT.

At 20 DAT, four fatty acids were identified: myristate (C14:0), palmitate (C16:0), linoleate (C18:2), and linolenate (C18:3; Fig. 1), whereas six were identified at 40 and 60 DAT (stearate C18:0 and oleate C18:1) inclusive of the four identified at 20 DAT. Five of the fatty acids increased substantially with harvest intervals. However, myristate (C14:0) remained virtually unchanged between 40 and 60 DAT. All the fatty acids except myristate (C14:0) increased an average of 1.5- to 2.5-fold from 20 to 60 DAT. That myristate (C14:0), palmitate (C16:0), linoleate (C18:2), and linolenate (C18:3) were identified at 20 DAT suggests that they are apparently synthesized during early developmental stages, whereas stearate and oleate are synthesized during later stages of growth. Linoleate, palmitate, and linolenate were the most abundant fatty acids in purslane with levels in excess of 300 mg·kg⁻¹. Those for myristate, stearate, and oleate were in excess of 200 mg·kg⁻¹ (Fig. 1).

Similar fatty acid profiles of purslane have been observed by others (Guil et al., 1996; Guil-Guerrero and Rodriguez-Garcia, 1999; Liu et al., 2000; Omara-Awala et al., 1991; Palaniswamy et al., 2001). Indeed, Palaniswamy et al. (2001) identified palmitate, stearate, oleate, linoleate, and linolenate

in purslane leaves at different developmental stages (six-, 10-, and 14-true-leaf stages) but reported that the increase in fatty acids was not steady. In contrast, our results showed a steady increase in fatty acids from 20 to 40 DAT and substantial increases up to 60 DAT. Palaniswamy et al. (2001) used leaves, however, whereas we used the entire above-ground plant parts. Omara-Alwala et al. (1991) analyzed individual parts as well as whole plants and obtained fatty acid profiles, which decreased after the first harvest and subsequently increased at final harvest in every plant part, generally, except the stems. However, whereas Omara-Alwala et al. (1991) did not provide an explanation for their findings, Palaniswamy et al. (2001), who obtained similar results at the 10-true-leaf stage (49 DAT) compared with the six-true-leaf stage (35 DAT), suggested that this response could be attributable in part to a "dilution effect" or use of the fatty acids synthesized to repair cell damage as a result of oxidative stress. It is probable that because the 40 DAT harvest time

used by both researchers was outside the range (49 DAT; 10-true-leaf stage) combined with the varying environmental conditions may have influenced responses of the plants.

The contents of major fatty acids, linoleate (C18:2; omega-6) and linolenate (C18:3; omega-3), increased significantly with harvest intervals (Fig. 2). At 20 DAT, the content of omega-3 (linolenate) was more than twice that of the omega-6 (linoleate) fatty acid. The reason for this is unclear; however, Ezekwe et al. (1999) found a similar trend during their first harvest of purslane. The difference between omega-6 and omega-3 fatty acids was substantially reduced at 40 and 60 DAT but remained significant.

Although individual availability of both omega-6 and omega-3 in food can benefit human health significantly, extensive research results (Brown, 2004; Ezekwe et al., 1999; Klurfeld, 1998) have emphasized that the ratio at which they are present is equally important. In our study, the ratio of omega-6 to omega-3 fatty acids increased significantly between 20

Table 1. Foliage and root yield of purslane in response to harvest intervals.

Harvest interval (d)	Foliage fresh wt	Foliage dry wt	Leaf no.	Plant ht	Root length	Root wt (g)
	----- (g) -----	----- (g) -----		----- (cm) -----		
20	1.5	0.45	11	4.8	9.4	0.10
40	7.2	2.63	27	6.1	12.0	0.38
60	27.2	15.28	69	9.3	18.6	0.81
Significance	**	**	**	**	**	**

**Significant at the 0.01 level of probability.

Fig. 1. Fatty acid contents of purslane in response to harvest intervals.

Fig. 2. Omega-3 and omega-6 fatty acid content of purslane in response to harvest intervals.

and 40 DAT but was similar between 40 and 60 DAT, being 1.08 and 1.1, respectively (data not shown). Our results fall within the 1:1 to 1:7 ratio suggested by Klurfeld (1998).

The lower rates of coronary heart disease and cancer in populations consuming a higher ratio of omega-6 to omega-3 fatty acids have been well documented (Thorsdottir et al., 2004; Youdin et al., 2000). It is generally agreed that the optimal ratio of omega-6 to omega-3 fatty acid for human diets ranges between 1:1 and 7:1 (Klurfeld, 1998) with healthy ratios ranging from 1:1 to 1:4 (Lands, 2005; Tribble, 2007). In fact, because most agricultural crops are richer in omega-6 than omega-3 fatty acids, increased intake of omega-3 from major crops may not be achieved. In the typical American diet, the ratio of omega-6 to omega-3 fatty acids is believed to range between 10:1 and 40:1 (Hibbeln et al., 2006) instead of the 1:1 to 7:1 ranges (Klurfeld, 1998; Lands, 2005; Tribble, 2007). Our data demonstrate that an optimal ratio of omega-6 to omega-3 fatty acids can be achieved as early as 20 DAT.

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