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Characterization of Four Molokhia (*Corchorus olitorius*) Landraces by Morphology and Chemistry

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ABSTRACT

Molokhia (*Corchorus olitorius* L., family Tillaceae) is a leafy, green, summer vegetable and medicinal plant that is a traditional dish in the Middle East and parts of Asia and Africa. The plant, which contains phenolics, polysaccharides, carotenoids, minerals, sugars, proteins, and the vitamins B1, B2, C, and E, is also consumed as a folk remedy for treatment of aches, pains, and swellings. Plant leaves are used for treatment of ascites, cystitis, dysentery, dysuria, fever, gonorrhea, pectoral pains, piles, tumors, thick mucus, and female infertility. A cold infusion of molokhia is used to restore appetites and strength. The current study monitored the growth and development of four molokhia genotypes, using fresh and dry weights, plant height, leaf area, and the number of branches, leaves, internodes, and herb yield. Chlorophyll, phenolics, sugars, and tannins were extracted and measured spectrophotometrically. Fresh weight, phenolics, and rosmarinic acid were highest in the wild genotype.

INTRODUCTION

The genus *Corchorus* (Tillaceae family) contains an estimated 40 to 100 species of flowering plants native to tropical and subtropical regions throughout the world (Ahmed, et al., 1998; Mbaye et al., 2001; Palve and Sinha, 2005). Several different common names for the plant are used in various contexts. For example, the name jute applies to the fiber produced from the plant and the name mallow leaf molokhia (with many similar name transliterations) is associated with the plant leaves used as a vegetable.

The leaves of the *Corchorus* plant have been a food staple in Egypt since the time of the pharaohs, when the plant gained recognition and popularity. Young molokhia leaves were and are still used as a green leaf vegetable with *C. olitorius* used primarily in southern Asia, Egypt, and Cyprus. When cooked, the plants have a mucilaginous texture similar to okra. Seeds of the plant are used as a flavoring agent and dried leaves are used in herbal tea. As a food, molokhia leaves can alleviate micronutrient deficiencies, especially low levels of zinc and iron that are associated with decreased human growth, pregnancy failures, and impaired immune functions. The plant is also rich in β-carotene, iron, calcium, and vitamin C. The plant also has antioxidant activity with a significant level of α-tocopherol that is readily absorbed by humans.

In addition to high levels of minerals, leaves of molokhia contain carbohydrates (70 mg/100 g dry matter), ascorbic acid (57 mg/100 g fresh weight) and a total acidity at 6% fresh weight (Abd-Allah and Nasr, 2006). The most common anti-nutrients in the plant leaves are phytate, hydrocyanic acid, oxalic acid, and tannins observed at relatively high levels in seeds of wild-type molokhia as compared with cultivated plants (Abd-Allah and Nasr, 2006; Ndelovu and Afolayan, 2008; Helaly et al., 2008; Yamazaki et al., 2009).

Even though several landraces of *Corchours* are morphologically close, each is reproductively isolated through pre and post zygotic barriers (Swaminathan et al., 1961; Morakinyo, 1997). The purpose of the current study was to discern the most productive selections among four collected landraces and to ascertain the variability among these molokhia for vegetative growth and nutrition under local environmental conditions in the Governorates of Egypt.

MATERIALS AND METHODS

Plant material. Molokhia (*Corchorus olitorius* L., family Tillaceae) landraces were used in this study. The plant material was grown from seeds of four molokhia landraces sourced from selected locations in Northern, Southern, and Western Egypt (Table 1). The collected seeds were sown in a randomized complete block design on May 1, 2006, in 2 m^2 plots, using 5 g of seeds per plot in an experimental field of the Horticulture Department at Al-Azhar University, Cairo, Egypt. Each plot was first harvested 50 days after seeding followed by second and third sequential harvests at 25 days apart for the Behira, Sohag, and Falahy genotypes. The Saidy genotype did not regrow after the first harvest.

Table 1. Molokhia landraces.

Landrace[1]	Source	Seed color	Cutting	Plant height
Wild type	Behira	Black	Several times	1.75 m
Balady	Sohag	Green	Several times	1.50 m
Saidy	Giza	Green	Once	2.00 m
Falahy	Giza	Green	Several times	1.00 m

[1]Landraces were sourced by Dr. Alaa El-Din and Dr. Adel Abd El-Aziz.

Physical characteristics of the molokhia plants were monitored as the seeds germinated and the plants developed. Hypocotyl length was measured from the junction point between root and hypocotyl to the cotyledonary leaves. At the time of harvest, plant height was measured and the number of leaves per plant was counted. Leaf area was measured on the fifth leaf below the plant tip using a portable digital leaf area meter (LI-300 meter, LI-COR, Inc., Lincoln, NE, USA). Dry matter production was determined by drying 100 g fresh weight of leaves at AOAC (1980).

Chemical characteristics of the molokhia landraces were studied using leaves six through ten below the terminal bud. Chlorophyll a (Chl. A), chlorophyll b (Chl. B), and total chlorophyll levels were determined according to the spectrophotometric method described by Hipkins and Baker (1986). Total phenolics were measured using the method outlined by Chandler and Dodds (1983). Tannin levels were determined on leaf tissue samples following the modified vanillin method of Burns (1971).

Rosmarinic acid was quantified using a UV assay as modified by Lopez-Arnaldos et al. (1995). Approximately 50 mg (fresh weight) samples of leaf tissue were extracted in 3 mL of a 50:50 methanol-water mixture and incubated at 55 ^oC for 2 h. A one mL aliquot of the incubated extract was subsequently diluted with an additional 5 mL of methanol the absorbance was measured at 333 nm. Concentration of the rosmarinic acid in the diluted extract was calculated according to the equation $A=\in bc$ with an extinction coefficient of $\in$=19,000 L/mol$*$cm and the width of quartz cuvettes b= 1 cm.

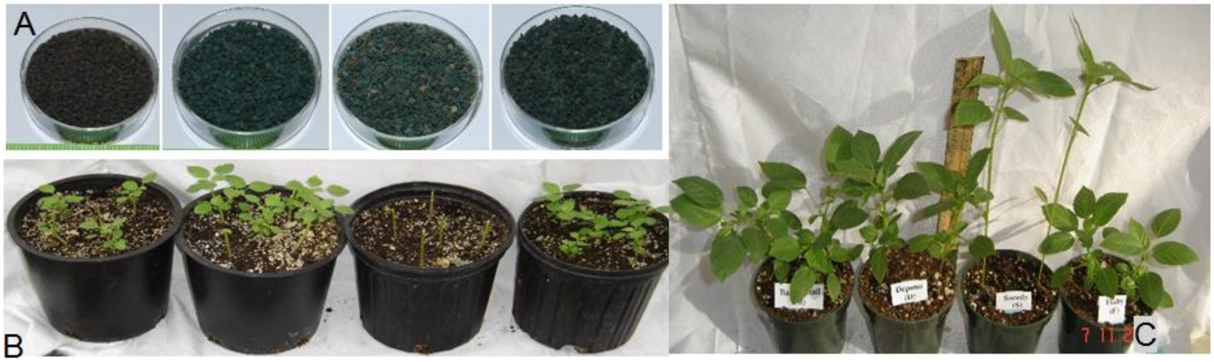
Total sugars were determined colorimetrically as g/100 g of dry plant tissue, according to the method of Dubois et al. (1956).

Statistical analysis. Significant differences among experimental plants were observed. Harvesting dates were compared using the least significant difference (LSD) at 5% probability, according to Snedecor and Cochran (1980).

RESULTS

Physical analysis. The wild type molokhia seeds had a brownish color that differed from the primarily black seeds of the other molokhia seeds (Figure 1). No differences, however, in seed germination among the landraces were noted. Measures of vegetative growth, including plant height, and number of branches and leaves, among the studied molokhia, demonstrated morphological differences among the landraces (Table 2).

The Wild type landrace produced more branches, more leaves, and a greater leaf area per plant as compared with the other plant landraces. The landrace Saidy was the tallest plant with the longest hypocotyl and longest internodes. In contrast, the landrace Falahy was the shortest plant with the shortest hypocotyl and shortest internodes.



Wild type Balady Saidy Falahy Wild type Balady Saidy Falahy

Figure 1. Molokhia landraces. A, B, &C: Seeds, potted plants, and growing plants.

Chemical analysis. The concentrations of chlorophyll (chlorophyll a, chlorophyll b, and total chlorophyll) among the molokhia landraces differed significantly (Table 3). The highest measured levels of chlorophyll were in the Wild type and Falahy landraces. Both of these plants had visibly dark-green leaves as compared with the Balady and Saidy selections. Balady had about 20% less chlorophyll content than Falahy.

The concentration of phenolics, tannins, and rosmarinic acid in the plant tissues were highest in the Wild type landrace during both harvest years (Table 4). Although the differences in the landrace constituents were quite small, results were consistent across the two harvest seasons. Soluble sugar levels were highest in the Balady and Saidy landraces and lowest in the Falahy landrace.

Crop yields. The molokhia landrace Saidy produced the highest yield for the first harvest, but did not regrow and thus did produce additional harvests within the same growing season (Table 4). The other landraces produced both a second and third harvest, producing total harvests greater than the landrace Saidy. The Wild type and the Balady produced the most tissue weight during the growing season. Failure of Saidy to regrow may have been due to cutting the plant hypocotyl during harvest.

Table 2. Vegetative growth of molokhia landraces during 2006 and 2007.

Landraces	Dry weight (%)		Plant height (cm)		Hypocotyl length (cm)		Internode length (cm)		Branches (number/plant)		Leaves (number/plant)		Leaf area (cm^2)	
	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007
Wild type	33.29	34.16	40.9	42.1	3.2	3.4	6.0	5.7	6.8	7.3	16.4	17.2	32.2	33.9
Balady	32.21	33.04	36.3	37.6	2.7	2.7	7.6	5.7	4.8	6.6	14.9	15.4	29.3	31.9
Saidy	34.90	35.73	50.4	54.0	7.2	8.0	15.8	13.9	3.2	3.9	9.8	10.3	34.4	36.8
Falahy	28.02	31.61	34.4	36.2	2.3	2.3	5.2	5.4	6.6	6.6	14.6	14.0	27.7	28.2
L.S.D. at 5%	N.S.	N.S.	3.3	5.1	1.0	1.5	3.4	2.6	1.1	1.8	3.1	2.8	4.4	5.4

Table 3. Concentrations of chlorophyll in molokhia landraces.

Landraces	Chlorophyll a µg/g fresh weight		Chlorophyll b µg/g fresh weight		Total Chlorophyll µg/g fresh weight	
	2006	2007	2006	2007	2006	2007
Wild type	1815	1884	663.8	555.1	2500.8	2460.9
Balady	1634	1729	501.2	559.4	2153.8	2308.7
Saidy	1696	1676	504.4	497.6	2220.1	2193.0
Falahy	1897	1916	877.0	855.5	2799.5	2772.0
L.S.D. at 5%	147	184	160.6	200.2	315.1	155.8

Table 4. Constituent concentration in molokhia landraces.

Landraces	Total phenolics (mg/g FW)		Tannins (mg/g FW)		Rosmarinic acid (mg/g FW)		Total sugars (g/100g DW	
	2006	2007	2006	2007	2006	2007	2006	2007
Wild type	6.05	6.32	4.30	4.28	1.31	1.38	40.47	42.09
Balady	4.89	4.89	4.27	4.16	1.21	1.26	41.18	48.83
Saidy	5.24	5.54	3.75	3.58	1.17	1.19	45.53	47.07
Falahy	4.99	5.22	3.71	3.66	1.07	1.14	35.88	36.07
L.S.D. at 5%	0.85	0.72	N.S.	N.S.	0.15	0.16	6.18	5.81

Table 5. Crop yield of molokhia landraces.

Landraces	2006				2007			
	1st cut kg/m^2	2nd cut kg/m^2	3rd cut kg/m^2	Yield kg/m^2	1st cut kg/m^2	2nd cut kg/m^2	3rd cut kg/m^2	Yield kg/m^2
Wild type	0.69	0.95	0.81	2.45	0.65	0.84	0.72	2.21
Balady	0.65	0.86	0.77	2.28	0.60	0.73	0.68	2.01
Saidy*	1.13	-	-	1.13	0.97	-	-	0.97
Falahy	0.61	0.74	0.66	2.01	0.56	0.67	0.60	1.83
L.S.D. at 5%	0.22	N.S.	N.S.	N.S.	0.15	0.06	N.S.	N.S.

DISCUSSION

Wild plants in the Mediterranean region have a long history of being utilized for food, medicine, fuel, shelter, and other applications (Helaly, et al., 2014). Familiarity with the benefits of local plant materials undoubtedly enabled the Egyptians, Greeks, and Romans to establish civilizations while learning to cultivate and domesticate plants for support of expanding populations. *Corhorus* spp. would have been among these plant materials due their usefulness as food and medicine.

While *Corhorus* plants continue to be grown and used, domestication and improvement of the plant as a food and medicinal plant have been limited. Only a few studies (Olaniyi and Ajibola, 2008 and Ogunkanmi et al., 2010) have examined whether differences in vegetative growth and nutritional value of *Corchorus olitorius* (molokhia) landraces might be due to the status of soil fertility, soil moisture, geographical location, and/or variety inheritance.

Helaly et al. (2008) observed that seeds of molokhia wild type landrace, contained more phenolics and tannins than tested landraces, suggesting that plant selection could improve the nutritional value of the plant. A study on the heritability of Jews mallow, *Corchorus capsularis*) by Ahmed et al. (1993) and Abd-Allah et al. (2006) enabled plant improvement. Enhanced stem length and leaf area are desirable components in molokhia breeding pro-grams for increased yields (Ogunkanmi et al., 2010).

While the Saidy landrace produced the tallest plant and the largest leaf area, only one crop was produced each season. Apparently, in harvesting the first crop the stem may have been cut below the hypocotyl leaves, eliminating any buds that could regrow the plant. Considerable variability was noted in the other landraces, differing in plant height, internode length, branches, leaf numbers, leaf area and chlorophyll levels that would affect plant yield (Buttery and Buzzell, 1977). In addition, Huang, et al. (2014), using *Arabidopsis*, have demonstrated that ecotype production is sensitive to differences in temperature.

The noted interrelationships among *Corchorus olitorius* plants by Abd-Allah (2006) support the concept of plant breeding could be used to improve molokhia production and nutrition. The highest levels of phenolics, tannins, and rosmarinic acid of the current study were in the Wild type landrace, suggesting that this landrace could be used to improve other landraces through a breeding program.

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