An Innovative Vegetative Propagation System for Large-scale Production of Globe Artichoke Transplants.

Part II. Propagation System Validation

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ADDITIONAL INDEX WORDS. offshoot, validation, propagation, globe artichoke, cost

SUMMARY. Research was conducted at the University of Tuscia (central Italy) to validate the propagation system for globe artichoke (*Cynara cardunculus* var. *scolymus*) described in a previous paper for a 1-year production cycle. The resulting globe artichoke plants were used in a 2-year field trial to investigate the field response of plantlets obtained with our propagation technique in comparison with plantlets produced by in vitro propagation and by offshoots harvested in commercial fields. The total number of artichoke plantlets obtained with our propagation system was 62.7 plantlets/m² per year. In the first year, the globe artichoke production (bud number and fresh bud weight) was higher in plants obtained with our propagation system and by micropropagation than in those obtained from offshoots harvested in commercial fields. The production cost of plantlets obtained with our propagation technique was 52% lower than those of the micropropagated plantlets. This could lead to a significant reduction of production costs for artichoke growers, while preserving the advantages of in vitro propagation (disease-free plants and plant uniformity).

Globe artichoke usually is vegetatively propagated by offshoots harvested in commercial fields at the end of the production cycle. This method often causes physiological, pathological, and economic disadvantages (Basnisky and Zohary, 1987; Calabrese et al., 2000). Micropropagation has solved many of these problems. However, in vitro propagation is expensive, and efficient micropropagation protocols are only available for a few cultivars. In a previous paper (Cardarelli et al., 2005), we set up a new propagation system for the large-scale production of globe artichoke transplants. The purpose of the current study was to validate our propagation technique carried out on a 1-year production cycle. The resulting plants were used in a 2-year field trial to investigate the field response of plantlets obtained with our propagation system in comparison with plantlets produced by in vitro propagation and by offshoots harvested in commercial

fields. In addition, the production cost of plantlets obtained with our propagation system was compared with those of micropropagated plantlets.

Materials and methods

EXPT. 1. Pathogen-free micropropagated plantlets of 'C3' artichoke (Vitroplant, Cesena, Italy) were transplanted into 20-cm-diameter pots filled with pumice (particle diameter from 1 to 3 mm) on 17 Aug. 2001. Stock plants were placed at a plant density of 5.1 plants/m² in a polyethylene-covered greenhouse equipped with insect-proof nets on the vent openings to prevent plant virus vectors [e.g., aphids (*Brachycaudus cardui*, *Myzus persicae*, *Protrama radicum*)] from

entering the greenhouse. The cultural practices were the same as those reported in a previous paper (Cardarelli et al., 2005). The stock plants were grown until June 2002 and during the growing period three production cycles of offshoots were forced (Table 1). In each production cycle, 6-benzylamino purine (BA) was applied to stock plants at a concentration of 200 mg·L⁻¹. Offshoots were continuously harvested when they reached four to five leaves (>30 g), leaving the last well-formed offshoot, which was used to regrow the stock plant. Harvested offshoots were counted, trimmed (removing the upper half of the leaves), and immersed for 3 min in a watery solution containing 1 g·L⁻¹ carbendazim (Bavistin; Basf Agro, Milano, Italy). Before rooting, cuttings from the I and II production cycles were cold-stored in plastic bags at 2 ± 0.5 °C, and 90 ± 5% relative humidity for approximately 180 to 75 d, respectively. In June, cuttings from the I, II, and III production cycles were rooted in separate individual benches (one bench for the cuttings of the same production cycle) filled with commercial potting soil (Metro-mix 360; W.R. Grace and Co., Cambridge, Mass.) and placed in high humidity tents. After 30 d, the number of rooted cuttings was counted.

EXPT. 2. A 2-year field trial was conducted on sandy-loam soil at the Viterbo experimental farm, central Italy (lat. 42°25'N, long. 12°08'E) to evaluate the effects of the plantlet production system on the yield and quality of the artichokes. The treatments were: 1) plantlets obtained from cold-stored cuttings (Expt. 1); 2) plantlets obtained from cuttings that were not cold-stored (Expt. 1); 3) plantlets obtained from apparently healthy offshoots (without visual symptoms of disease) harvested from commercial fields; and 4) micropropagated plantlets. The treatments

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Units			
To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
0.4047	acre(s)	ha	2.4711
0.3048	ft	m	3.2808
0.0929	ft ²	m ²	10.7639
0.0283	ft ³	m ³	35.3147
2.5400	inch(es)	cm	0.3937
25.4000	inch(es)	mm	0.0394
1.1209	lb/acre	kg·ha ⁻¹	0.8922
28.3495	oz	g	0.0353
70.0532	oz/acre	g·ha ⁻¹	0.0143
7.4892	oz/gal	g·L ⁻¹	0.1335
1	ppm	mg·L ⁻¹	1
(°F - 32) ÷ 1.8	°F	°C	(1.8 × °C) + 32

Table 1. Basic information on the globe artichoke propagation system in 2001–02.

Date	Operation
I production cycle	
17 Aug. 2001	Micropropagated plantlets transplanted
10 Oct. 2001	Stock plants foliar-sprayed with 6-benzylamino purine (BA)
17 Oct. 2001	Stock plants cut back
15 Nov.–17 Dec. 2001	Offshoots harvested and cold-stored
II production cycle	
17 Dec. 2001	Stock plants re-growth from the last well-formed offshoot
22 Jan. 2002	Stock plants foliar-sprayed with BA
29 Jan. 2002	Stock plants cut back
1 Mar.–1 Apr. 2002	Offshoots harvested and cold-stored
III production cycle	
1 Apr. 2002	Stock plants re-growth from the last well-formed offshoot
30 Apr. 2002	Stock plants foliar-sprayed with BA
7 May 2002	Stock plants cut back
31 May–24 June 2002	Offshoots harvested
31 May–24 June 2002	Rooting of cuttings which have been cold-stored (I and II cycles) and harvested from the third cycle

were arranged in a randomized complete-block design with three replicates. Each plot consisted of 30 plants. Plantlets were transplanted on 22 Aug. 2002 in single rows of plants 1.1 m apart (8264 plants/ha). Fertilizers (diammonium phosphate and sulfate of potash) were preplant spread and disked at the following rates: 54 kg·ha⁻¹ of nitrogen, 61 kg·ha⁻¹ of phosphorus, and 130 kg·ha⁻¹ of potassium. Plants were side-dressed with 100 kg·ha⁻¹ of nitrogen (ammonium nitrate) on 15 Dec. 2002 and on 17 Dec. 2003. Sidedress was applied in bands 30 cm to either side of the plants and 10 cm deep. Plots were sprinkler-irrigated “as needed” by visually monitoring the water status of the crop and soil. Weeds were controlled with 4 L·ha⁻¹ of pendimethalin (Stomp 330E; Basf Agro) at planting and mechanical and hand hoeing during both growing seasons. Slugs were controlled by methiocarb (Mesurol N-Esca; Bayer CropScience, Milano, Italy) at the rate of 160 g·ha⁻¹, while powdery mildew was controlled by three sulfur applications (Zolfo Bagnabile; Bayer CropScience) at the rate of 1.6 g·L⁻¹. The artichoke buds were hand-harvested from the center 12 plants in each replication when the buds achieved maximum size, but before the bracts began to spread open. In 2003, the harvest period started on 14 Feb. and ended on 18 Apr., while in 2004, the growing season harvest started on 20 Feb. and ended on 29 Apr. The fresh weight and number of buds were determined.

STATISTICAL ANALYSIS. All data were statistically analyzed by analysis of variance using SPSS (version 10 for Windows; SPSS, Inc., Chicago). Duncan’s multiple range test was performed at $P \leq 0.05$ on each of the significant variables measured.

Results and discussion

The total number of cuttings per stock plant obtained in Expt. 1 decreased over the three production cycles with a higher number observed in the first cycle (7.2 cuttings/stock plant) followed by those recorded in the second and third cycles (6.1 and 4.3 cuttings/stock plant, respectively).

The percentage of rotten cuttings was lower in cuttings from the II production cycle (5%) in comparison with those obtained in the I production cycle (14%). This is similar to results we observed in a previous experiment in which the number of rotten cuttings

increased with increased cold storage time (Cardarelli et al., 2005). Moreover, the rooting percentage was lower in cuttings of the I cycle (61%) than in those of the II and III cycles (82% and 88%, respectively). The total number of artichoke plantlets obtained with our propagation system was 62.7 plantlets/m² per year. In Expt. 2, globe artichoke plants obtained with our propagation technique and with micropropagation produced significantly more buds (secondary, tertiary, and total) than those obtained with offshoots harvested in commercial fields (Table 2). The average fresh weight of buds was higher in plants obtained with our propagation technique and micropropagation than in those obtained with offshoots harvested in commercial fields (Table 2). No significant differences were observed in the treatments with regards to the number of offshoots produced per plant (average = 12 offshoots/plant), indicating that there were no residual effects of the previous applications of BA to the stock plants. This is very important because of the high operational cost of removing offshoots in the field.

In 2004, no significant differences were observed between treatments with regards to the fresh weight and number of buds. The average bud number of the first, second, third order, and the total were 1.0, 3.2, 4.4, and 8.7 buds/plant, respectively. The average fresh bud weight of the first, second, and third order were 280.3, 193.2, and 120.7 g, respectively.

To summarize, we can conclude that growing artichokes from plantlets obtained with our propagation technique is a valid alternative to micropropagation for the Romanesco type of globe artichoke because of the similarities in plant performance. In addition,

Table 2. Effect of propagation technique on primary, secondary, tertiary, and total bud number and fresh weight of globe artichokes during 2002–03 growing season (Expt. 2). Values are the mean of three replicate samples.

Treatment ^y	Buds				Mean wt (g/plant) ^z		
	No. (no./plant)						
	Primary	Secondary	Tertiary	Total	Primary	Secondary	Tertiary
C1	1.0 a ^x	3.0 a	4.0 a	8.1 a	245.0 a	180.7 a	121.7 a
C2	1.0 a	2.8 a	4.2 a	8.0 a	240.3 a	175.9 a	112.6 a
T	1.0 a	2.2 b	3.3 b	6.4 b	230.1 b	151.3 b	114.9 a
M	1.0 a	3.1 a	4.3 a	8.3 a	250.2 a	183.5 a	120.1 a

^z1 g = 0.0353 oz.

^yC1 = plantlets obtained by cold stored cuttings of the I and II production cycle; C2 = plantlets obtained by cuttings of III production cycle; T = plantlets obtained by offshoots harvested from commercial field; M = micropropagated plantlets.

^xIn each column, values followed by the same letter do not differ significantly at $P \leq 0.05$.

Table 3. Production cost of 1 ha (2.47 acres) using the globe artichoke transplant production system.^z

Item	Description	Total cost (\$/transplant)
Fixed expenses ^y	Depreciation, repairs and insurance of fixed investments (e.g., greenhouse, fertigation system, cold storage, building)	0.10
Nonlabor variable expenses ^y	Transplants, substrate, fertilizers, pesticides, hormones, plastic bags, styrofoam trays, electricity, oil	0.20
Labor expenses ^x	Maintenance, transplanting, spraying, cutting, and harvesting	0.10
Interest expenses ^w	Fixed and variable expenses	0.04
Total fixed and variable expenses including interest		0.44

^zCalculations based on 1 year with 0.6 ha (1.48 acre) of greenhouse for growing stock plants, 0.3 ha (0.74 acre) of greenhouse for offshoot rooting, 100 m³ (3531.5 ft³) of cold storage, and 100 m² (1076.4 ft²) of building.

^yInput prices were obtained from local greenhouse suppliers and building material suppliers.

^xNational nursery industries were consulted to obtain information regarding the operating costs associated with propagation.

^wArtichoke transplant charged a 10% rate of interest on capital.

the production costs of plantlets obtained with our propagation technique are significantly lower (Table 3) than those of micropropagated plantlets [~\$0.9 per transplant; this cost was provided by national commercial companies that produce around 1 million artichoke transplants per year], which could lead to a significant reduction of the production cost for artichoke growers. Our propagation technique may also be applied to early cultivars, while in vitro propagation mainly leads to late-ripening plants with more deeply divided leaves. The frequency of this occurrence increased linearly with the number of subcultures, sometimes reaching 100% after 20 subcultures (Pécaut and Martin, 1993). In order to produce healthy plants from early cultivars, the late-ripening plants must be discarded and an accurate selection of micropropagated stock plants should

be used. Finally, our propagation technique can be used in breeding programs to obtain, quickly and easily, a large number of plants per inbred line, which is a basic requirement for producing a large quantity of hybrid seed.

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