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COMPARISON OF YIELD AND MORPHOLOGICAL CHARACTERS OF RHUBARB (*Rheum rhaponticum* L.) 'KARPOW LIPSKIEGO' PLANTS PROPAGATED *in vitro* AND BY CONVENTIONAL METHODS

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Abstract. The present study investigated different methods of propagation of the rhubarb cultivar 'Karpow Lipskiego': vegetatively from tissue cultures and by division of the mother plant as well as generatively from seed. The study also evaluated the usefulness of propagation material for establishing a rhubarb plantation. To produce *in vitro* plantlets, shoots were placed on modified Murashige and Skoog medium containing different cytokinins: benzyladenine (BA – 4.4, 11.1, 22.2 μmol·dm^{-3}), kinetin (4.7, 11.6, 23.3 μmol·dm^{-3}), isopentenyl adenine (2iP – 4.9, 12.3, 24.4 μmol·dm^{-3}) as well as on control medium without growth regulators. Cuttings were obtained by division of crowns, while seedlings from seeds. In the period 2004–2006, vegetatively and generatively propagated plants were grown in a nursery. The obtained propagation material was used to establish a rhubarb plantation. Yield of field-grown plants was evaluated in the years 2007–2009. In the first year of cultivation in the nursery, plants propagated *in vitro* in the medium with the addition of kinetin at a concentration of 11.6 μmol dm^{-3} or 2iP at 12.3 μmol dm^{-3} developed crowns with the highest average weight of 1.48 and 1.05 kg, respectively. In the second year of cultivation in the nursery in the treatment with grown regulators applied, the average rhubarb crown weight ranged from 2.51 to 3.33 kg, while for the control treatment it was 1.78 kg. To characterize the population of *in vitro* plants, they were compared with plants obtained by division of the mother plant and from seeds. Plants propagated vegetatively from *in vitro* plantlets produced crowns with the highest average weight (0.83 kg), followed by those obtained from division of mother plants (0.79 kg), while plants produced from seeds had crowns with a much lower average weight (0.54 kg). In the second year of cultivation in the nursery, vegetatively and generatively propagated plants were characterized by a similar size and greater uniformity than in the first year. In the first year of planting (2007), petiole yield obtained from micropropagated plants was higher by 0.7 kg·plant^{-1} compared to generatively propagated plants, whereas in the second year of cultivation this difference was smaller and amounted to 0.6 kg· plant^{-1}. In the third year of the plantation, plant productivity was more equal. During the study years, by far fewer leaves were harvested from generatively propagated plants compared to plants propagated

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vegetatively. In the second and third year of the plantation, intensive plant growth was observed; in effect, there were no significant relationships between petiole length and width and propagation method. The cultivation of vegetatively propagated plants gave the best effects in productivity; the average petiole yield was higher by 13.3 $t \cdot ha^{-1}$ in the case of micropropagation and by 16.0 $t \cdot ha^{-1}$ in the case of division of the mother plant compared to the yield obtained from rhubarb cultivation from seeds. Early yield at a level of 11.2 $t \cdot ha^{-1}$ was obtained from tissue cultured plants and its proportion accounted for 25% of total petiole yield.

Key words: rhubarb, *in vitro*, propagation method, nursery cultivation, yielding

INTRODUCTION

Garden rhubarb (*Rheum rhaponticum* L.) is a winter-hardy perennial perfectly adapted for growing in the cool and temperate climate [Rumpunen and Heneriksen 1999, Sałata 2005]. The selection of the method of obtaining propagation material to establish a plantation is a very importance in rhubarb cultivation, since the quality of propagation material determines yield and yield quality.

Rhubarb can be propagated generatively, from seedlings prepared in a nursery or in a greenhouse, but also vegetatively by division of the mother plant or from plantlets obtained from *in vitro* cultures. Generatively propagated rhubarb plants are usually not very uniform and do no repeat parental traits. Plants of one cultivar differ from one another in height, habit, shape and intensity of petiole colour, as well as in the number of petioles per plant. In horticultural practice, seed propagation is not recommended. On the other hand, production of rhubarb division of crowns is limited by the low yield of mother plants and the risk of transfer of pathogens, in particular viral diseases [Norman 1978, Maynard 1990, Ma and Gang 2006].

The growing interest in rhubarb cultivation from micropropagated plants is often stressed in the literature. This method gives the possibility of obtaining genetically stable plants of high vigour [Roggemans and Claes 1979, Barna et al. 2007, Shahzad et al. 2007, Kozak and Sałata 2011] and, additionally, free from viral diseases [Roggemans and Boxus 1988, Lassus and Voipio 1994]. A significant problem in tissue culture propagation of plants is their subsequent field acclimatization in nurseries and adaptation to the environment as well as morphological, anatomical, and biochemical changes that take place [Pospisilova et al. 1999, Kadlecek et al. 2001, Hazarika 2003, Mishra et al. 2010]. In *in vitro* propagated rhubarb, after plants were transferred to the field, high somaclonal variation was observed with respect to their morphological characters such as plant habit, petiole shape and colour as well as low winter hardiness [Zhao et al. 2003]. According to Lassus and Voipio [1994], the number of leaves per plant and the leaf blade size have a decisive influence on the winter survival rate of tissue cultured plants. Zhao et al. [2009] observed a high frequency of changes in micropropagated plants grown under field conditions.

In research relating to evaluation of cultured progeny, it has been established that morphological changes can occur in plants in successive years of open field cultivation [Zhao et al. 2005, Misalova et al. 2009].

Tissue culture conditions, in particular the concentration and type of growth regulators used, were a direct cause of stress and resulted in physiological changes in plants, unlike conventional propagation methods [Mokotedi et al. 2009, 2010].

The aim of the present study was to evaluate the possibility of using in horticultural production different methods of rhubarb propagation: vegetative propagation from tissue cultures and by division of the mother plant as well as generative propagation from seeds. The usefulness of propagation material for establishing a rhubarb plantation was evaluated.

MATERIALS AND METHODS

The investigations of rhubarb micropropagation were carried out at the Tissue Cultures Laboratory of the Department of Ornamental Plants, whereas the experiments on seedling production, vegetative propagation by division of crowns and the experiments on open field rhubarb cultivation were carried out at the Felin Research Station belonging to the Department of Vegetable Crops and Medicinal Plants of the University of Life Sciences in Lublin.

Plants of the cultivar 'Karpow Lipskiego' were the subject of the study. This cultivar is very popular among producers who run commercial plantations of this vegetable for the needs of the processing industry speak in favour of this choice.

To produce plantlets, 10 mm long shoots were taken from *in vitro* propagated shoot clusters. The shoots were placed on modified Murashige and Skoog [1962] medium 40.3 mg·dcm^{-3} NaFeEDTA instead $FeSO_4$ + Na_2EDTA containing different cytokinins: benzyladenine (BA – 4.4, 11.1, 22.2 µmol·dm^{-3}), kinetin (4.7, 11.6, 23.3 µmol·dm^{-3}), isopentenyl adenine (2iP – 4.9, 12.3, 24.4 µmol·dm^{-3}) as well as on control medium without growth regulators. 30 shoots were placed in each medium. The obtained plants were transferred to plastic boxes (16 × 10 × 9.5 cm) containing a mixture of peat and perlite (1:1); they were acclimated for 4 weeks and then planted in the field nursery.

To produce stem cuttings, the underground part of the plant, five-year-old rhubarb crowns, were dug out of the ground in 2004, in the first decade of September. The crowns, which have very short internodes, were cut longitudinally with a sharp knife into pieces in such a way that each piece had at least one viable bud. Before planting in the nursery, the surface of the cut was dried to prevent rotting. On average, cuttings had a diameter of 5.0 cm and a weight of 100 g.

Rhubarb seed material was obtained in 2003 from seed plants that had been reproduced vegetatively by division of a 5-year-old crowns only from one plant. The seed plants were in an isolated field. In March 2004 the collected seeds were sown in a greenhouse in boxes filled with peat substrate in rows 5 cm apart. At the seedling stage, the first identification evaluation of the cultivar was carried out based on the morphology and colour of the hypocotyl. After initial selection, the plants were transplanted into plug trays. At the 4–6 true leaf stage, the next morphological identification of young plants was made on the basis of the parameters such as: petiole length and colour, leaf blade length, width and shape as well as leaf margin teeth.

Plants derived from *in vitro* cultures, from crown division and from seeds were planted in September 2004 in the nursery at a spacing of 40 × 40 cm and the crop was grown during the period 2004–2005 and 2005–2006. This experiment was conducted as a single-factor experiment in a randomized block design in three replications. In the group of *in vitro* propagated plants, there were 10 plants in each replication, whereas in the other two groups (plants derived from crown division and from seeds) one replication comprised 100 plants.

In the first and second year of rhubarb cultivation in the nursery, biometric measurements were made of some morphological characters of the underground part of plants after the end of their growth. All plants were sampled for analysis from the plots with *in vitro* plants (30 plants from each type of medium), whereas 30 plants were randomly selected from each replication from the plots in which plants derived from crown division and from generative propagation were grown. The plants were dug out with their roots and subsequently the following traits were determined: crown fresh weight and diameter, number of rosette buds on the stem surface (photo 1).

Photo 1. Underground part – crown of *Rheum rhaponticum* 'Karpow Lipskiego' after the end of growth in the nursery in 2006; on the left after generatively propagated, on the right after *in vitro* propagated

Fot. 1. Części podziemna – karpa *Rheum rhaponticum* 'Karpow Lipskiego' po zakończeniu wegetacji w szkółce w roku 2006; po lewej po rozmnażaniu generatywnym, po prawej po rozmnażaniu *in vitro*

During the further part of the experiment (in the years 2007, 2008, and 2009), the obtained propagation material was used to establish a rhubarb plantation. Plants derived from tissue cultures, from stem cuttings obtained from plant division and from seedlings were planted in the field, at a spacing of 0.67 × 0.40 m, which had been previously prepared properly and fertilized with manure (0.6 t·100 m^2). This experiment was established as a single-factor experiment in a randomized block design in three replications.

The experiment included the same levels of the factor as in the nursery. There were 50 plants in each replication. In each successive year of the study, 4 harvests of rhubarb leaves were carried out on the same 10 randomly selected plants from each experimental treatment. The results on yield and early yield of rhubarb propagated from tissue cultures on media with different supplements were similar during the study years (2007–2009), therefore they are presented in this paper as mean values. The following plant traits were determined for all leaves collected from the plants: number of leaves, petiole length and width (measured in half of the leaf stalk). After the leaf blade was cut off, petiole yield and early petiole yield per plant were determined. Petiole yield from the first two leaf harvests was considered to be early yield. On the basis of yield per plant, petiole yield and early yield per 1 ha as well as the proportion of early yield in petiole yield were calculated.

Analysis of significance of the treatments was determined by one-way ANOVA at a significance level of 0.05.

RESULTS AND DISCUSSION

One of the more difficult stages of micropropagation is the transfer of plants from *in vitro* cultures to the field. The present experiment evaluated the effect of the action of cytokinins added to the medium (tab. 1). In the first year of cultivation after overwintering in the nursery, plants from micropropagated plantlets grown in the medium with the addition of kinetin at a concentration of 11.6 μmol·dm^{-3} or 2iP at 12.3 μmol dm^{-3} developed crowns with the highest average weight of 1.48 and 1.05 kg, respectively. These

Table 1. Growth of *in vitro* propagated *Rheum rhaponticum* 'Karpow Lipskiego' plants 12 months after planting in the field nursery. Plants were planted in September 2004 and evaluated in 2005

Tabela 1. Wzrost *Rheum rhaponticum* 'Karpow Lipskiego' rozmnażanego *in vitro* 12 miesięcy po posadzeniu w szkółce na polu. Rośliny posadzono we wrześniu 2004 r. i oceniano w 2005 r.

Growth regulators Regulatory wzrostu	Concentration Stężenie (μmol·dm^{-3})	Crown weight Masa karpy (kg)	Crown diameter Średnica karpy (cm)	Number of rosette buds Liczba pąków rozetowych (pcs·stem^{-1})
Control Kontrola	0	0.62b	9.6c	5.3e
BA	4.4	0.69b	9.5c	8.3d
	11.1	0.71b	15.0a	11.6a
	22.2	0.60b	9.0c	7.0de
Kinetin	4.7	0.80b	8.2c	9.2cd
	11.6	1.48a	13.7ab	11.0bc
	23.3	0.95b	8.5c	7.0d
2iP	4.9	0.80b	11.3b	9.5c
	12.3	1.05ab	16.2a	11.2b
	24.4	0.66b	11.1bc	9.5c

* Means followed by the same letter are not significantly different – Wartości oznaczone tą samą literą nie różnią się istotnie

plants had a uniform and compact habit, which is evidenced by the large number of rosette buds (on average 11.0 and 11.2 pcs.) from which leaf rosettes grew out. These results are evidence that in the first year after overwintering plants derived from *in vitro* plantlets did not fully regain the hormonal balance between the aerial and underground part. Zhao et al. [2009] found high variation in the structure and size of micropropagated rhubarb plants after twelve months of their acclimatization. Zhao et al. [2004, 2006] express an opinion that growth regulators applied and their different concentrations may have an effect on the change of morphological characters of rhubarb plants as well as on the process of their growth. Similar relationships were found in the case of application of different growth regulators in micropropagation of cabbage [Cristea et al. 2009] and strawberry [Debnath 2009]. A consequential effect of growth regulators applied in tissue cultures on plant flowering after 112 days of cultivation was observed in the case of chicory crops [Demeulemeester and Proft 1999]. The degree of differentiation of *in vitro* cultured plants is associated with their high sensitivity to stress conditions during the initial period of their acclimatization [Zhao et al. 2009]. This phenomenon is not fully understood and is explained by competition for food between the aerial part of the plant and the young growing roots [Costa et al. 2009].

Table 2. Growth of *in vitro* propagated *Rheum rhaponticum* 'Karpow Lipskiego' plants 24 months after planting in the field nursery. Plants were planted in September 2004 and evaluated in 2006.

Tabela 2. Wzrost *Rheum rhaponticum* 'Karpow Lipskiego' rozmnażanego *in vitro* 24 miesiące po posadzeniu w szkółce na polu. Rośliny posadzono we wrześniu 2004 r. i oceniano w 2006 r.

Growth regulators Regulatory wzrostu	Concentration Stężenie ($\mu mol \cdot dm^{-3}$)	Crown weight Masa karpy (kg)	Crown diameter Średnica karpy (cm)	Number of rosette buds Liczba pąków rozetowych ($pcs \cdot stem^{-1}$)
Control Kontrola	0	1.78c	16.5c	7.0d
BA	4.4	2.51b	28.5a	14.0a
	11.1	2.62b	27.6a	9.6c
	22.2	2.90ab	27.0a	8.2c
Kinetin	4.7	2.77b	26.2ab	13.5a
	11.6	2.57b	28.5a	13.5a
	23.3	3.27a	25.5ab	12.5ab
2iP	4.9	2.96a	34.0a	10.0bc
	12.3	2.72b	36.0a	12.5ab
	24.4	3.33a	34.0a	10.8b

* Means followed by the same letter are not significantly different – Wartości oznaczone tą samą literą nie różnią się istotnie

In the second year of cultivation in the nursery, plants from *in vitro* cultures with growth regulators applied showed higher growth potential than those without any supplements (control). In the treatment with growth regulators applied at different concentrations, the average rhubarb crown weight ranged from 2.51 to 3.33 kg, whereas in the

control treatment (without the application of plant hormones) the average crown weight was lower and it was 1.78 kg (tab. 2). The number of rosette buds per plant greatly changed; when the addition of cytokinin to the medium was used in *in vitro* cultures, this number was from 8.2 to 14.0 pcs., while in the case of the control treatment it was 7.0 pcs. In plants derived from plantlets grown on media without growth regulators, a limited number of buds always developed, adjusted to the plantlet size and food resources in the rhubarb crown.

Table 3. Effect of the method of propagation of *Rheum rhaponticum* 'Karpow Lipskiego' on morphological characters of the crown 12 months after planting in the field nursery. Plant were planted in September 2004 and evaluated in 2005.

Tabela 3. Wpływ metody rozmnażania *Rheum rhaponticum* 'Karpow Lipskiego' na cechy morfologiczne karpy 12 miesięcy po posadzeniu w szkółce na polu. Rośliny posadzono we wrześniu 2004 r. i oceniano w 2005 r.

Propagation method Metoda rozmnażania	Crown weight Masa karpy (kg)	Crown diameter Średnica karpy (cm)	Number of rosette buds Liczba pąków rozetowych (pcs·stem^{-1})
Micropropagation Mikrorozmnażanie	0.83a*	11.2a	8.9a
Crown division Podział karpy	0.79ab	9.6b	8.5a
Generatively Generatywnie	0.54b	8.9b	6.0b

* Means followed by the same letter are not significantly different – Wartości oznaczone tą samą literą nie różnią się istotnie

Table 4. Effect of the method of propagation of *Rheum rhaponticum* 'Karpow Lipskiego' on morphological characters of the crown 24 months after planting in the field nursery. Plants were planted in September 2004 and evaluated in 2006.

Tabela 4. Wpływ metody rozmnażania *Rheum rhaponticum* 'Karpow Lipskiego' na cechy morfologiczne karpy 24 miesiące po posadzeniu w szkółce na polu. Rośliny posadzono we wrześniu 2004 r. i oceniano w 2006 r.

Propagation method Metoda rozmnażania	Crown weight Masa karpy (kg)	Crown diameter Średnica karpy (cm)	Number of rosette buds Liczba pąków rozetowych (pcs·stem^{-1})
Micropropagation Mikrorozmnażanie	2.7a*	28.3a	11.1a
Crown division Podział karpy	2.9a	27.9ab	10.9a
Generatively Generatywnie	2.2b	23.9b	10.7a

* Means followed by the same letter are not significantly different – Wartości oznaczone tą samą literą nie różnią się istotnie

To characterize the population of tissue cultured plants, some of their traits were compared with those of plants from division of mother plants and from seeds (tab. 3). Plants propagated vegetatively from *in vitro* plantlets produced crowns with the highest average weight (0.83 kg), followed by those derived from stem cuttings (0.79 kg), while those produced from seeds had crowns with a much lower average weight (0.54 kg). The underground part of the obtained population of vegetatively propagated plants was characterized by a larger diameter and higher number of rosette buds compared to generatively propagated plants. In the second year of cultivation, seed-derived plants and vegetatively propagated plants (by division of the mother plant and by micropropagation) were characterized by a similar size and greater uniformity than in the first year (tab. 4). These differences were less noticeable in average weight and diameter of the underground part of plants but, at the same time, no significant differences were observed in the number of rosette buds in the shortened crowns. Zhao et al. [2009] explained that in rhubarb, after a certain number of embryonic roots have developed and rosette buds have been stimulated to grow, a correlation relationship is established between them that prevents the formation of an excessive number of roots. If there is a deficiency of reserve substances in the rhubarb crown, the aerial and underground parts of the plant start to compete with each other. In a study on blueberry yield, still after 16 years of cultivation significant differences were found in the development of the underground parts of plants and in fruit yield between *in vitro* propagated plants, plants from green cuttings, and seed-derived plants [Jamieson and Nickerson 2003].

In open field cultivation, yields were evaluated under optimal growing conditions in the years 2007–2009 (tab. 5). In the first year of rhubarb growing, petiole yield obtained from micropropagated plants was higher by 0.7 kg·plant^{-1} compared to generatively propagated plants, whereas in the second year of cultivation this difference was smaller and amounted to 0.6 kg· plant^{-1}. In the first year of cultivation, the difference in yield between plants from crown division and seed-derived plants was 0.9 kg·plant^{-1}, while in the second year it was 1.1 kg· plant^{-1}. In the third year of the plantation, plant yields were more equal. Petiole yield from tissue cultured plants was on average higher by 0.3 kg·plant^{-1} than that from plants from division of mother plants and higher by 0.7 kg·plant^{-1} compared to the yield from generatively propagated plants; this difference was statistically significant (LSD = 0.43). In an experiment on different methods of rhubarb propagation conducted by Lepse et al. [2009], no differences were observed in yield and yield quality in the second year of the plantation.

During study, by far fewer leaves were harvested from generatively propagated plants compared to those propagated vegetatively. In the first year of cultivation, the number of leaves obtained from seed-derived plants was lower on average by 5.0 pcs·plant^{-1}, in the second year of cultivation by 11 pcs·plant^{-1}, while in the third year by 10 pcs·plant^{-1} compared to plants obtained by division of the mother plant. A statistically significant difference between the number of leaves harvested from *in vitro* propagated plants and the number of leaves from generatively propagated plants was found only in the second year of planting. In the first year of cultivation, generatively propagated plants developed leaves with short and thin stalks compared to the leaves of vegetatively propagated plants. Generatively propagated rhubarb plants were phenotypically different; they exhibited large morphological differences and were not very uniform.

About 25% of seed-derived plants in the plantation had untypical colour of the petiole epidermis compared to the typical colour for the cultivar 'Karpow Lipskiego'.

In the second and third year of the plantation, intensive plant growth was observed; in effect, there were no significant relationships between petiole length and width and propagation method. In the northern regions of Europe, plants propagated by division of the mother plant were found to produce more small leaves with thin stalks, whereas tissue cultured plants produced fewer leaves but with a larger size [Lepse et al. 2009]. The below-mentioned authors report that higher yield can be obtained in the cultivation of the following *in vitro* propagated plants compared to their cultivation from seedlings: *Capsicum annum* [Song et al. 2009], eucalyptus [Mokotedi et al. 2009], banana [Chavan-Patil et al. 2010], anthurium [Gantait and Sinniah 2011].

Table 5. Effect of the method of propagation of field-grown *Rheum rhaponticum* 'Karpow Lipskiego' on yield and some morphological characters of petioles, in 2007–2009

Tabela 5. Wpływ metody rozmnażania *Rheum rhaponticum* 'Karpow Lipskiego' na plonowanie oraz wybrane cechy morfologiczne ogonków liściowych w uprawie na polu w latach 2007–2009

Growing season Sezon wegetacji	Propagation method Metoda rozmnażania	Petiole yield Plon ogonków liściowych ($kg \cdot plant^{-1}$)	Number of leaves per plant Liczba liści z rośliny	Petiole length Długość ogonka liściowego (cm)	Petiole width Szerokość ogonka liściowego (cm)
2006/2007	micropropagation mikrorozmnażanie	1.7	15.6	30.0	2.1
	crown division podział karpy	1.9	18.6	35.3	2.1
	generatively generatywnie	1.0	12.6	23.0	1.8
		$\alpha_{0.05} = 0.31$	$\alpha_{0.05} = 3.12$	$\alpha_{0.05} = 4.51$	$\alpha_{0.05} = 0.22$
2007/2008	micropropagation mikrorozmnażanie	2.4	21.5	33.5	2.2
	crown division podział karpy	2.9	25.5	39.5	2.1
	generatively generatywnie	1.8	14.5	35.8	2.1
		$\alpha_{0.05} = 0.46$	$\alpha_{0.05} = 4.11$	n.s. – n.i.	n.s. – n.i.
2008/2009	micropropagation mikrorozmnażanie	3.0	24.6	38.0	2.2
	crown division podział karpy	2.7	31.3	40.5	2.2
	generatively generatywnie	2.3	21.3	37.8	2.1
		$\alpha_{0.05} = 0.43$	$\alpha_{0.05} = 5.11$	n.s. – n.i.	n.s. – n.i.

n.s. – no significantly – n.i. – nieistotne

A synthesis of the results on rhubarb productivity in the period 2007–2009 shows high variations in yield depending on the propagation method (tab. 6). The cultivation of vegetatively propagated plants gave the best effects in productivity; the average petiole yield was higher by 13.3 $t \cdot ha^{-1}$ in the case of micropropagation and by 16.0 $t \cdot ha^{-1}$ in the case of division of the mother plant compared to the yield obtained from rhubarb cultivation from seeds. A measure for evaluation of vigour of rhubarb plants is the number of leaves in yield. Vegetatively propagated plants produced on average more leaves than seed-derived plants. The literature data confirm that higher yield can be obtained from *in vitro* propagated plants compared to the conventional methods: sugarcane [Sood et al. 2006], tomato [Bhatia and Ashwath 2004], pepper [Song et al. 2009], blueberry [de Souza et al. 2011].

Table 6. Effect of the method of propagation of field-grown *Rheum rhaponticum* 'Karpow Lipskiego' on petiole yield, early yield, and the proportion of early yield in petiole yield, mean for 2007–2009

Tabela 6. Wpływ metody rozmnażania *Rheum rhaponticum* 'Karpow Lipskiego' na plon ogonków liściowych i plon wczesny oraz udział plonu wczesnego w plonie ogonków w uprawie na polu średnio w latach 2007–2009

Propagation method Metoda rozmnażania	Petiole yield Plon ogonków liściowych ($t \cdot ha^{-1}$)	Number of leaves per plant Liczba liści z rośliny	Early petiole yield Plon wczesny ogonków ($t \cdot ha^{-1}$)	Proportion of early yield in petiole yield Udział plonu wczesnego w plonie ogonków (%)
Micropropagation Mikrorozmnażanie	47.3	20.6	11.2	24.7
Crown division Podział karpy	50.0	25.1	6.5	12.9
Generatively Generatywnie	34.0	16.1	2.5	7.7
	$\alpha_{0.05} = 8.3$	$\alpha_{0.05} = 4.3$	$\alpha_{0.05} = 4.1$	

An important property that determines the usefulness of rhubarb for horticultural cultivation is the fast growth rate of biomass and early yield [de Souza et al. 2011].

The proportion of early yield in petiole yield was shown to be dependent on the method of plant propagation. Tissue cultured plants produced yield at a level of 11.2 $t \cdot ha^{-1}$ and its proportion accounted for 25% of total petiole yield. In plants propagated vegetatively by division of the mother plant, the proportion of early yield in petiole yield was 50% lower than in the cultivation of micropropagated plants. The lowest percentage of early yield in petiole yield (7.7%) was found in seed-derived plants. Issues concerning the effect of rhubarb propagation methods on early yield are not very well known yet. In the cultivation of other micropropagated species, higher early yield is generally obtained [Sandhu et al. 2009].

In vitro rhubarb propagation proved to be effective. Therefore, the presented results give grounds for a conclusion that under the Polish conditions rhubarb micropropagation is an effective and proper method to produce planting material for establishing plantations.

CONCLUSIONS

In the first year of cultivation in the nursery, plants propagated *in vitro* in the medium with the addition of kinetin at a concentration of 11.6 μmol·dm^{-3} and 2iP at 12.3 μmol·dm^{-3} developed crowns with the highest average weight. In the second year of cultivation in the nursery in the treatment with growth regulators applied, the average rhubarb crown weight ranged from 2.51 to 3.33 kg, while for the control treatment it was 1.78 kg. To characterize the population of *in vitro* plants, they were compared with plants obtained by division of the mother plant and from seeds. Plants propagated vegetatively from *in vitro* plantlets produced stems with the highest average weight (0.83 kg), followed by those obtained from division of mother plants (0.79 kg), while plants produced from seeds had crowns with a much lower average weight (0.54 kg). In the second year of cultivation in the nursery, vegetatively and generatively propagated plants were characterized by a similar size and greater uniformity than in the first year. In the first year of field cultivation (2007), higher petiole yield was obtained from micropropagated plants than from generatively propagated plants, whereas in the second year of cultivation this difference was smaller. In the third year of the plantation, plant productivity was more equal. During the study years, by far fewer leaves were harvested from generatively propagated plants compared to plants propagated vegetatively. In the second and third year of the plantation, intensive plant growth was observed; in effect, there were no significant relationships between petiole length and width and propagation method. The cultivation of vegetatively propagated plants gave the best effects in productivity compared to the yield obtained from rhubarb cultivation from seeds. Early yield at a level of 11.2 t·ha^{-1} was obtained from *in vitro* propagated plants and its proportion accounted for 25% of total petiole yield. In plants propagated vegetatively by division of the mother plant, the proportion of early yield in petiole yield was 50% lower than in the cultivation of micropropagated plants. The lowest percentage of early yield in petiole yield (7.7%) was found in seed-derived plants.

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PORÓWNANIE PLONOWANIA ORAZ CECH MORFOLOGICZNYCH ROŚLIN RABARBARU (*Rheum rhaponticum* L.) 'KARPOW LIPSKIEGO' ROZMNAŻANEGO *in vitro* ORAZ METODAMI TRADYCYJNYMI

Streszczenie. Badania dotyczyły różnych metod rozmnażania rabarbaru odmiany 'Karpow Lipskiego': wegetatywnie z kultur tkankowych oraz przez podział roślin matecznych i generatywnie z siewu nasion. Oceniano również przydatność materiału rozmnożeniowego do założenia plantacji rabarbaru. W celu wyprodukowania mikrosadzonek *in vitro* pędy wykładano na zmodyfikowaną pożywkę Murashige i Skooga zawierającą różne cytokininy: benzyloadeninę (BA – 4,4, 11,1, 22,2 μmol·dm^{-3}), kinetynę (4,7, 11,6, 23,3 μmol·dm^{-3}), izopentyloadeninę (2iP – 4,9, 12,3, 24,4 μmol·dm^{-3}) oraz pożywkę kontrolną niezawierającą regulatorów wzrostu. Sadzonki otrzymano przez podział karp, a rozsadę z siewu nasion. W latach 2004–2006 rośliny otrzymane w wyniku rozmnażania wegetatywnego i generatywnego uprawiano w szkółce. Uzyskany materiał rozmnożeniowy wykorzystano do założenia plantacji rabarbaru. W latach 2007–2009 w uprawie polowej przeprowadzono ocenę plonowania. W pierwszym roku uprawy w szkółce karpy o największej średniej masie 1,48 i 1,05 kg wykształciły rośliny otrzymane *in vitro* z pożywki z dodatkiem kine-

tyny w stężeniu 11,6 µmol dm^{-3} oraz 2iP w stężeniu 12,3 µmol dm^{-3}. W drugim roku uprawy w szkółce w przypadku zastosowanych fitohormonów średnia masa karpy rabarbaru zawierała się w przedziale od 2,51 do 3,33 kg, gdy z obiektu kontrolnego wynosiła 1,78 kg. Dla scharakteryzowania populacji roślin *in vitro* porównano je z roślinami otrzymanymi przez podział roślin matecznych i z nasion. Karpy o największej średniej masie wykształciły rośliny rozmnażane wegetatywnie: z mikrosadzonek (0,83 kg) i sadzonek uzyskanych przez podział roślin matecznych (0,79 kg), o zdecydowanie mniejszej średniej masie sadzonki otrzymane z nasion (0,54 kg). W drugim roku uprawy w szkółce rośliny otrzymane w wyniku rozmnażania wegetatywnego i generatywnego charakteryzowały się zbliżoną wielkością i większym wyrównaniem niż w pierwszym roku. W pierwszym roku prowadzenia uprawy (2007) większy plon ogonków liściowych o 0,7 kg·rośl.$^{-1}$ zebrano z roślin otrzymanych z mikrosadzonek aniżeli z roślin rozmnażanych generatywnie, natomiast w drugim roku uprawy różnica ta była mniejsza i wynosiła 0,6 kg·rośl.$^{-1}$. W trzecim roku prowadzenia plantacji plonowanie roślin było bardziej wyrównane. W latach prowadzenia badań z roślin rozmnażanych generatywnie zebrano zdecydowanie mniej liści w porównaniu z roślinami rozmnażanymi wegetatywnie. W drugim i trzecim roku prowadzenia plantacji obserwowano intensywny wzrost roślin, czego efektem był brak istotnych zależności pomiędzy długością i szerokością ogonków liściowych a sposobem rozmnażania roślin. Najlepsze efekty plonowania dała uprawa roślin rozmnażanych wegetatywnie, z której plon średni ogonków liściowych był większy o 13,3 t·ha^{-1} w porównaniu z mikrorozmnażaniem, oraz o 16,0 t·ha^{-1} przy podziale roślin matecznych w porównaniu z plonem uzyskanym w uprawie rabarbaru z nasion. Z uprawy roślin rozmnażanych za pomocą kultur tkankowych uzyskano plon wczesny na poziomie 11,2 t·ha^{-1}, a jego udział stanowił 25% plonu ogółem ogonków liściowych.

Słowa kluczowe: rabarbar, *in vitro*, metoda rozmnażania, uprawa w szkółce, plonowanie

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