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Yield performance of Russian dandelion transplants (*Taraxacum koksaghyz* L. Rodin) in flat bed and ridge cultivation with different planting densities

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ABSTRACT

As shown for other valuable crops, transplanting of wild rubber-producing *Taraxacum koksaghyz* (Tks) could be an option to counteract poor field emergence and stand establishment after direct seeding. Field trials (spring planting, autumn harvest) were conducted in 2012 and 2013 on a loamy soil (Quedlinburg, Saxony-Anhalt, Central Germany) to investigate the influence of different planting beds (flat, ridge) and planting densities (222,222 plants/ha, 88,889/133,333 plants/ha) on the yield performance of Tks. Between planting date and harvest significant plant losses of 43–48% occurred across all treatments in both trial years. Major plant losses occurred within the first month after transplanting. The achieved planting density (APD) at harvest was significantly influenced by year, targeted planting density (TPD) and type of planting bed. Nearly all yield parameters were significantly influenced by the year of cultivation. There was a significant advantage of ridge over flat bed cultivation regarding achieved planting density, fresh root yield per hectare, and fresh/dry root yield per plant in the season 2013 and within the reduced planting density. In ridge cultivation root yield parameters were increased by 5–13%. Furthermore planting density had a significant effect on biomass yield. There were weak to strong positive linear correlations ($r = -0.35$ to 0.75) between achieved planting density and biomass per hectare (fresh/dry root/leaf) and moderate negative correlations ($r = -0.49$ to 0.57) with individual plant yield (fresh/dry root/leaf, rubber/inulin). The presented study demonstrates that transplanting of Tks on ridges can be an option to increase root yield of Tks and that the focus of future research activities should be laid on an optimization of Tks transplant production and management in the field.

1. Introduction

Russian dandelion (*Taraxacum koksaghyz* L. Rodin, Tks), also named Kazakh dandelion or Buckeye Gold, belongs to the worldwide spread genus of *Taraxacum* and is considered as a promising candidate for the domestic rubber and inulin production in many countries worldwide e.g. Russia, Canada, United States and Germany (Krotkov, 1945; Whaley and Bowen, 1947; Ulmann, 1951; Van Beilen and Poirier, 2007a).

Natural rubber (NR) is a biopolymer that consists of hundreds to ten thousands of isoprene units (C_5H_8) linked in 1,4 *cis*-configuration, occurring in many different plant species (Schulze Gronover et al., 2011). The average molecular weight (mw) of the poly-*cis*-isoprene of NR from the rubber tree, *Hevea brasiliensis*, is about 1300 kD (Van Beilen and Poirier, 2007b). The mw of poly-*cis*-isoprene isolated from Tks roots is in a similar range or even higher (Van Beilen and Poirier, 2007b; Kreuzberger et al., 2016). The rubber amount of Tks ranges between 3 and 28% of the dry root mass depending on plant material and growing conditions (Lipshitz, 1934). The Tks material available to the authors (progeny of the USDA Tks germplasm collection) showed a rubber concentration of 3–9% in the dry root depending on the developmental stage of plants (Kreuzberger et al., 2016). NR is a strategic material. 154,000 tons of NR originating from the *Hevea* tree were used for tire production and In 2015, 64,000 tons of NR were processed as latex (medical devices, hygiene products, baby articles) in Germany (Wdk, 2015). Despite the high quality attributes of NR gained from other rubber-producing plants such as Tks or Guayule (*Parthenium argentatum)*, to date *Hevea* NR remains the sole economically important source (Van Beilen and Poirier, 2007b).

Aside from polyisoprenes, 25–40% of the dry root is composed of inulin (Ulmann, 1951; Whaley and Bowen, 1947) which is a polydisperse fructan linked in 2,1 *beta*-configuration used in food industry for several applications such as sweetening or improving texture (Mensink et al., 2015). Commercially available inulin is mostly obtained from *Cichorium intybus* and exhibits an average polymerization degree of 10–20 (Flamm et al., 2001). The polymerization degree of Tks

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inulin ranges from 8 up to 30 depending on growth stage/harvest time of the plants and the inulin quality is therefore comparable to that from commonly available plant sources (Kreuzberger et al., 2016). However, recent studies on the agronomic performance of Tks have shown that biomass, rubber and inulin yields are low and currently not competitive with established commercial sources like *Hevea* (rubber) and *Cichorium* (inulin) (Arias et al., 2016a; Kreuzberger et al., 2016; Arias et al., 2016b).

Publicly funded research activities in academic institutions and industry in Europe (EU-PEARLS – Grant Agreement (GA) No. 212872, DRIVE4EU- GA No. 613697) and Germany (TARULIN GA No. 0315971, TAKOWIND GA No. 22002312; EVITA GA No. 031A285A) aim at turning Tks from a wild plant into a new commercial crop with the general objective to support the local production of renewable raw materials (e.g. rubber, inulin, latex). The field study described here was part of a research collaboration (TARULIN) covering diverse aspects of Tks breeding, agronomy, processing and product development. The agronomic research activities of the project aimed at the cultivation of Tks under various field conditions and the documentation of its yield performance under the influence of different agronomic measures. The temporal yield performance of a Tks stand established by sowing was described by Kreuzberger et al. (2016). The establishment of Tks stands via direct seeding has been the most common approach in the former Soviet Union and the United States (Whaley and Bowen, 1947). However, also transplanting (planting seedlings) is conceivable for large scale Tks cultivation even though the former is obviously the less labor (Suomela, 1950) and cost-intensive approach. However, to date plant establishment and subsequently yield performance of Tks via direct seeding faces several obstacles (e.g. low field emergence, slow juvenile plant development, overlapping growth stages) that need to be overcome to gain a dense crop and stable yield. The study of Kreuzberger et al. (2016) emphasized the need for improving the seeding technique and the need for the development of seeds with high seed vigor. These drawbacks might partially be overcome by establishing Tks in the field with transplants. The production of Tks transplants includes the production of Tks seedlings under controlled environmental conditions (e.g. in the greenhouse) and their transfer to the field. E.g. in sugar beet, transplanted beets showed higher root yield than seeded beets, due to an increased length of the growing season which enhanced an earlier and faster development of the leave canopy and roots of transplants (Theurer and Doney, 1980). This faster growth was associated with an increased photosynthetic activity and hence increased assimilate transport to the roots. In vegetables, reasons for transplanting are earlier harvest, better control of abiotic and biotic stresses in the greenhouse, as well as an optimal stand with clearly defined plant spacing and uniform physiological plant age in the field compared to direct seeding (Schrader, 2000).

This study aimed at 1) establishing Tks in the field by transplanting and 2) exploring the impact of this cultivation regime on the yield performance of the transplants with regard to biomass, rubber, and inulin after one season (spring planting, autumn harvest) with the overarching goal of improving/maximizing Tks yields. The latest documented experiments with Tks stands established with planted seedlings were made in Finland in 1944–46 on small areas with sand soil (Suomela, 1950).

Based on the soil texture at the trial site (heavy loam) and the available harvesting technique (potato harvester) for Tks roots, it was decided to grow Tks in two types of planting beds, flat beds and ridges. Due to reduced soil compaction and subsequently increased harvest depth, a higher amount of root mass was expected to be harvested from the ridges when compared to the flat bed situation. Additionally, seedlings were planted in two planting densities (222,000 plants/ha versus 89,000 plants/ha (2012), 133,000 plants/ha (2013)) in order to investigate a potential impact caused by the competition between plants on the yield performance. Different ridge cultivation systems were already investigated worldwide for different crops (e.g. maize, cotton millet, cowpea, and soybean) with the aim to conserve water (Hulugalle, 1990) or to prevent soil erosion (Liu et al., 2008; Pikul et al., 2001). In Germany the cultivation of potatoes, asparagus and carrots on ridges is common practice. Ridge cultivation of sugar beets resulted in increased beet yield by 5–10% in Northern Germany compared to conventional flat cultivation (Schlinker et al., 2007). In further field trials with sugar beet, white sugar yield was increased by 8.4% compared to flat cultivation (Krause et al., 2009). To the authors' knowledge, there are no suggestions neither regarding planting density for Tks transplants nor their economic production and establishment in the field until today. Hence, this is the first study presenting data on the establishment of Tks via transplanting under field conditions. Targeted planting densities were chosen on the basis of available mechanical equipment at the experimental station.

2. Material and methods

2.1. Study site and crop management

The trials were conducted in two consecutive years (2012, 2013), each trial during one season (spring planting, autumn harvest). The study site was located at the experimental field station of the Federal Research Centre for Cultivated Plants (JKI) in Quedlinburg, Germany. The site (51.4N, 11.8E, 140 m of elevation) is characterized by a temperate climate, influenced by a nearby mountain range (Harz). The long-term mean for air temperature is 8.9 °C and for precipitation 497 mm. The monthly values for temperature and preciptiation during the trial periods are given in Table 1. The soil at the study site was a Chernozem with a loamy texture, a humus content of 2.1% and a pH value of 7.1. Since the soil was well supplied with nutrients (8.2 (P_2O_5), 10 (K_2O), 11 (Mg) mg 100 g soil^{-1}), no fertilizers were applied. The soil mineral nitrogen content (N_{min}) at the time of transplanting was 142 (2012) and 49 (2013) kg N ha^{-1} in the depth of 0–90 cm. Tks was grown at this site for the first time. The previous crop was grass-clover. Weeds were controlled manually.

2.2. Plant material and trial design

The parental population of the Tks seeds used for the field trial in 2012 was gained from 14 Tks accessions (Plant ID W6-35 -156, -159, -160, -164, -166, -168, -169, -170, -172, -173, -176, -178, -181, -182) which were collected by Barbara Hellier in the high valleys of the Tian Shan mountains in Kazakhstan in 2008 (Hellier, 2011) and which were received by the authors from the USDA-ARS National Germplasm System. For seed propagation, seeds of these accessions were germinated under greenhouse conditions and random manual crossings between the accessions were performed. The progeny of these crossings represented a random part of the collected wild individuals which were flowering under these conditions since they were not selected for any other trait. Single Tks seeds had a weight of 0.5 ± 0.06 mg and a germination rate of 77 ± 6%. In 2013, transplants were grown from the seed progeny harvested during the flowering period in 2012. There transplants flowered openly and pollination occurred naturally in the

Table 1
Monthly and average values of air temperature and precipitation for trial period in 2012 and 2013.

	Apr	May	Jun	Jul	Aug	Sep	Oct	Av
Air temperature				°C				
2012	8.8	14.6	15.6	18.1	19.0	14.9	9.6	14.4
2013	8.6	12.6	16.6	20.2	18.8	13.4	11.6	14.5
Precipitation				mm month^{-1}				
2012	26	65	115	90	35	40	2	373
2013	23	53	4	32	30	57	86	285

field. Seed harvest in the field was performed with a vacuum cleaner.

For seedling production in the greenhouse, seeds were germinated on peat/perlite substrate two months before transplanting. Seedlings/transplants were cultivated in 200 well trays in turf sand substrate (Co. Fruhnstorfer, Anzuchterde) at 14/10 h day-night-cycle (high pressure sodium light of Philips Powertone SON-T AGRO 400 W) at 18/16 °C and with a relative air humidity of 70%. Seedlings were watered as needed from below to enhance root growth. The young plants were hardened off outside two weeks before planting into the field. Planting was conducted on 18/4/12 and 22/4/13. Plants were irrigated immediately after planting, and then every two days for two weeks to allow rooting.

The trial was conducted in two subsequent years (2012, 2013) as a two-factorial randomized complete block design with six replications. Soil was prepared by mouldboard ploughing in previous autumn and field cultivator in spring before planting bed preparation with seed bed combination (flat beds) and ridge former (Typ 2 1650, Groenewegen's Landbouwwerk Rozenburg Tuigen B. V., Holland). Each experimental plot (total size 35.1 m^2) contained six rows of 7.8 m length with a row width of 75 cm (Fig. 4). Ridges had a height of 20 cm, a crown of 18 cm and a foot of 50 cm. Targeted planting density (TPD) was either narrow (TPD 1) with 130 plants/row (one plant every 6 cm or 222,222 plants/ha) or wide (TPD 2) with 52 plants/row (one plant every 15 cm or 88,889 plants/ha) in 2012. Due to a toothed stand in 2012, the plant number per row in TPD 2 was increased to 78 (one plant every 10 cm or 133,333 plants/ha) in 2013 in order to achieve a denser stand.

2.3. Harvest and plant analyses

Plants were harvested with rosettes still attached to the roots 181 (16/10/12) and 183 (22/10/13) days after transplanting with a single-row sifting belt harvester (Wühlmaus, Kartoffel-Vorratsroder KVR 750T, Maschinenfabrik Niewöhner GmbH & CoKG, Germany), cutting the roots in a depth of 0.15–0.25 m in both types of planting beds. Out of six rows only the four central rows were harvested for yield estimation (core-harvesting). The number of plants was counted for each plot and used to calculate the achieved planting density at harvest (APD). All plants were washed and rosettes were removed with secateurs. Fresh root and leaf weight per plot was determined to estimate fresh root (FRY) and fresh leaf yield (FLY) in tons per hectare. In order to determine dry root yield (DRY) and dry leaf yield (DLY) in tons per hectare for each plot, a subsample of 1 kg of fresh roots or rosettes (including about 1 cm of the root crown) was dried until constant weight in a drying oven at 120 °C to determine dry root/leaf mass. Fresh (FRYP) and dry (DRYP) root yield per plant as well as fresh (FLYP) and dry (DLYP) leaf yield per plant were calculated by the division of FRY, DRY, FLY and DLY with APD.

Rubber concentration of dry root mass (RC) was determined by a solvent-assisted extraction and gravimetric measurement as described in Kreuzberger et al. (2016). Rubber yield per hectare (RY) was calculated from RC and DRY. Rubber yield per plant (RYP) was calculated by the division of RY with APD.

Inulin concentration (IC) of dry Tks roots was analyzed by enzymatic hydrolysis following hot water extraction and HPLC analysis of resulting monosaccharides (fructose, glucose) as described in Kreuzberger et al. (2016). Inulin yield per hectare (IY) was calculated from IC and DRY. Inulin yield per plant (IYP) was calculated by dividing IY with APD.

2.4. Statistics

For statistical analysis of the data, the SigmaPlot for Windows Version 13.0 was used. Data of all 14 parameters were checked for normality (Shapiro-Wilk) and equal variance (Brown-Forsythe). All data showed normal distribution and equal variance except DLY and IY. DLY and IY were $\log_{10}$-transformed for further analysis. For all parameters, a three-way general linear model was calculated with the raw or transformed data with model factors year, planting bed and planting density. Differences between means were compared by using Tukey's honestly significant difference (Tukey's HSD) with an alpha error probability (p) level of $p < 0.05$ or Dunn's test (if group sizes were unequal).

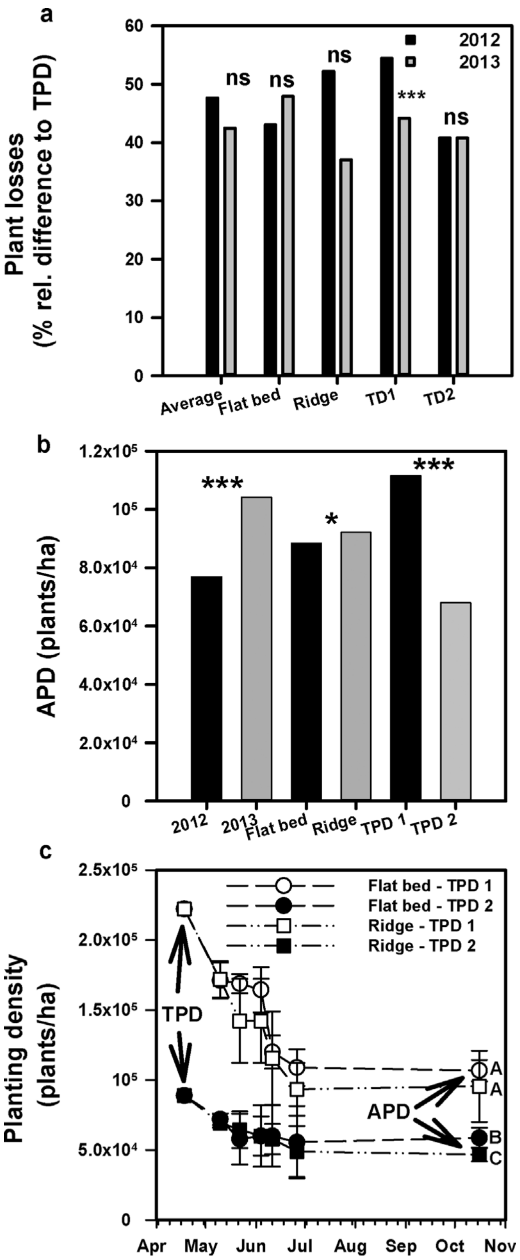

Fig. 1. Plant losses in two years in different planting beds (flat bed, ridge) and planting densities (TD1, TD2) (a). Two-tailed *t*-test showed no significant (ns) difference between years within groups at a p level < 0.05. Results of three-way analysis of variance for the fixed effect of the main factors year, planting bed and targeted planting density (TPD) on achieved planting density (APD) (b). Two-tailed *t*-test showed a significant difference *, *** between years, type of planting beds and planting density at a p level < 0.05, < 0.001. Development of planting density over the season 2012 from the date of planting (18/04/12) over the summer until harvest date (16/10/12) in two types of planting beds (flat bed, ridge) and two targeted planting densities (TPD1–222,222 plants/ha, TPD 2–88,889 plants/ha) (c). Symbols show means of six experimental plots and standard deviation. Differences of means between treatments were calculated for achieved planting density (APD) at harvest with Tukey test. Altered capital letters indicate a significant difference between treatments at a p level of < 0.05.

Table 2

Relationship between planting density and yield parameters across two years and all treatments (n = 48) analyzed with Pearson‘s coefficient of correlation (r). $r > 0.45$ have a significant level of $p < 0.001$.

	APD	FRY	DRY	FRYP	DRYP	FLY	DLY	FLYP	DLYP	RC	RY	RYP	IC	IY	IYP
TPD	0.78	0.64	0.34	−0.48	−0.54	0.41	0.32	−0.45	−0.44	0.11	0.07	−0.54	−0.24	−0.19	−0.51
APD		0.75	0.48	−0.49	−0.54	0.35	0.23	−0.57	−0.57	0.16	0.22	−0.49	−0.41	−0.25	−0.57
FRY			0.65	0.05	−0.35	0.63	0.52	−0.23	−0.24	0.04	0.23	−0.36	−0.25	−0.07	−0.40
DRY				0.07	0.25	0.30	0.21	−0.19	−0.21	0.03	0.68	0.19	−0.29	0.43	−0.05
FRYP					0.55	0.27	0.32	0.76	0.74	−0.34	−0.11	0.39	0.41	0.40	0.55
DRYP						−0.12	−0.06	0.54	0.53	−0.21	0.31	0.88	0.30	0.76	0.94
FLY							0.98	0.46	0.44	−0.24	−0.10	−0.26	0.27	0.23	−0.05
DLY								0.55	0.56	−0.37	−0.22	−0.26	0.42	0.31	0.05
FLYP									0.99	−0.44	−0.29	0.32	0.63	0.49	0.63
DLYP										−0.51	−0.36	0.27	0.69	0.52	0.64
RC											0.63	0.17	−0.53	−0.37	−0.35
RY												0.57	−0.48	0.22	0.08
RYP													0.03	0.57	0.76
IC														0.61	0.57
IY															0.84

TPD/APD – targeted/achieved planting density, FRY/DRY – fresh/dry root yield per area, FRYP/DRYP – fresh/dry root yield per plant, FLY/DLY – fresh/dry leaf yield per area, FLYP/DLYP – fresh/dry leaf yield per plant, RC/IC – rubber/inulin concentration, RY/IY – rubber/inulin yield per area, RYP/IYP – rubber/inulin yield per plant

3. Results

3.1. Plant establishment – relationship between targeted and achieved planting density

There was a large difference between TPD and APD in both years. Plant losses were 48% (2012) and 43% (2013) across all treatments (Fig. 1a). In total, 26% more plants could be harvested in 2013 than in 2012 (Fig. 1b). APD was 76,887 (2012) and 104,189 (2013) plants/ha across all treatments. It appeared that APD was 4% higher in ridge cultivation than in flat bed cultivation across all years and treatments (Fig. 1b). Yet, there was an interaction between year and type of planting bed for APD (Fig. 3a). In 2013, significantly more plants (18%) could be harvested on ridges than in flat beds, whereas this number was slightly but not significantly decreased in 2012. TPD was highly correlated to APD (Table 2). Higher TPD resulted in significantly higher APD. The relative difference between TPD 1 (111,561 plants/ha) and TPD 2 (68,008 plants/ha) was 39%. TPD also had a significant effect on the development and the level of plant losses over the season (Fig. 1c). In 2012, it was apparent that higher TPD caused higher absolute plant losses. This observation was made independently of the type of planting bed (Fig. 1a and c). For TPD 2, major plant losses occurred within the first 34 days after planting (end of May) but remained static during the rest of the season. For TPD 1, APD continuously decreased until the end of June (Fig. 1c) and remained stable until harvest. In 2013, plant losses were not measured periodically.

A significant difference in the number of plants between TPD1 and TPD 2, as originally intended, could be observed in both years and in both types of planting beds. The intended differences between TPD 1 and TPD 2 were 60% and 40% in 2012 and 2013, respectively. The achieved relative differences at harvest between the treatments were reduced to 48 and 27% over the season in 2012 and 2013, respectively. Hence, yield parameters of individual plants were calculated on the basis of APD (Fig. 1b).

3.2. Yield parameters — the influence of year, planting bed and planting density

All yield parameters, except DRY, DRYP and RYP, were significantly influenced by year of cultivation (Fig. 2a). FRY of 2.3 (2012) and 2.6 t/ha (2013) was equivalent to a DRY of 0.5 t/ha in both years. RC was 26% higher in 2013 (5.4% dm) when compared to 2012 (4.0% dm). In contrast, IC was significantly decreased by 41% in 2013 (15.9% dm) compared to 2012 (26.8% dm). The type of planting bed had a significant effect on FLY, FRYP and DRYP (Fig. 2b). The absolute differences between flat bed and ridge cultivation in FLY were 0.2 t/ha and 3.5 g/root for FRYP. DRYP of 6.3 g was about 17% higher in ridges than in flat beds across all treatments. TPD had a significant effect on FRY, FRYP, DRYP, FLY, DLY and FLYP (Fig. 2c). FRY, FLY and DLY were significantly higher in TPD 1 than in TPD 2. For FRYP, DRYP, RYP and IYP this effect was reversed. The absolute differences between TPD 2 and TPD 1 were 5, 1.4, 0.2 and 1 g for FRYP, DRYP, RYP and IYP respectively. For FRY, FLY and FRYP, interactions between year and between agronomic measures became evident. For FRY, there was an interaction between year and type of planting bed (Fig. 3b). While in 2012 there was no significant difference in FRY between flat bed and ridge cultivation, in 2013, FRY was about 16% higher in ridge cultivation than in flat beds. There was a further interaction between type of planting bed and TPD for FRYP (Fig. 3c) and for FLY (Fig. 3d). FRYP was significantly higher (13%) in TPD 2 than in TPD 1, however this difference was only apparent in ridge cultivation. FLY was significantly higher (36%) in TPD 1 than in TPD 2, however only in flat bed cultivation.

3.3. Relationship between achieved planting density and yield parameters and among yield parameters

There were numerous significant linear correlations between APD and yield parameters as well as among yield parameters (Table 2). There was a strong positive correlation between APD and FRY ($r = 0.75$) and a moderate correlation of APD with DRY ($r = 0.48$). Moderate negative relationships ($r = -0.41$ to -0.57) existed between APD and FRYP, DRYP, FLYP, DLYP, RYP, IC and IYP, respectively. FRY was positively correlated to DRY, FLY and DLY. There was also a strong correlation between DRY and RY ($r = 0.68$). DRYP showed a moderate positive correlation with FLYP ($r = 0.54$) and DLYP ($r = 0.53$) and strong relationship to RYP ($r = 0.88$), IY ($r = 0.76$) and IYP ($r = 0.94$). Interestingly, DLYP and RC were negatively correlated ($r = -0.51$) as well as RC to IC ($r = -0.51$).

4. Discussion

To develop Tks into a profitable and competitive crop, its agronomic performance needs to be improved. Rubber and inulin yield of Tks is determined by the level of dry root yield and the concentration of rubber/inulin in the dry root mass. Both parameters are influenced by the genetic background of the Tks species, environmental conditions and agronomical practices. In this study, the effect of the year of cultivation and two agronomic measures, planting bed and planting density, on APD and on the performance of 14 yield parameters of Tks was

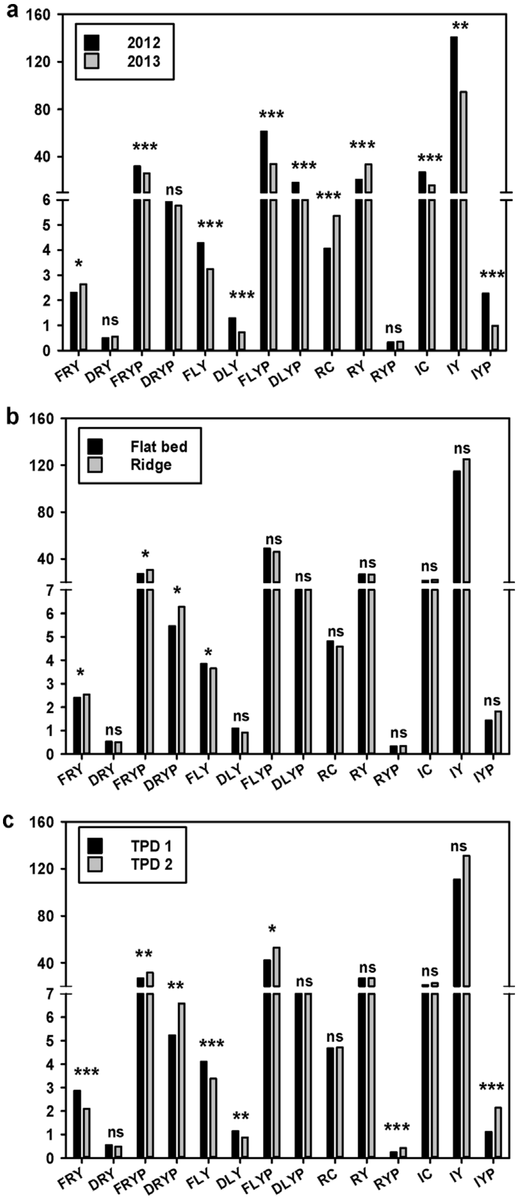

Fig. 2. Results of three-way analysis of variance for the fixed effect of the main factors year (a), planting bed (b) and targeted planting density (TPD) (c) on yield parameters of Tks (FRY – fresh root yield per hectare (t/ha), DRY – dry root yield per hectare (t/ha), FRYP – fresh root yield per plant (g/plant), DRYP – dry root yield per plant (g/plant), FLY – fresh leaf yield per hectare (t/ha), DLY – dry leaf yield per hectare (t/ha), FLYP – fresh leaf yield per plant (g/plant), DLYP – dry leaf yield per plant (g/plant), RC – rubber concentration (% of dry matter), RY – rubber yield per hectare (kg/ha), RYP – rubber yield per plant (g/dry root), IC – inulin concentration (% of dry matter), IY – inulin yield per hectare (kg/ha), IYP – inulin yield per plant (g/dry root)). ***, **, *indicate a significant effect of the main factor on the parameter at a p value level of < 0.001, < 0.01, < 0.05. ns indicates no significant effect ($p > 0.05$).

investigated.

4.1. The success of stand establishment by Tks transplants

As for direct seeding (Kreuzberger et al., 2016), improvement of Tks production by transplants can only be achieved if the stand establishment is accomplished successfully. Since each individual Tks plant contributes to the total crop yield, plant losses over the season will lead to a reduced yield. Despite careful preparation of the planting bed and sufficient watering of young plants within the first two weeks after planting, there was a significant average plant loss of 48 and 43% in 2012 and 2013 across all treatments (Fig. 1a). These results were adverse to the experiments of Suomela (1950) who observed slow but 90–100% rooting of the seedlings even though plantings were not irrigated. In 2013, the overall APD was significantly higher than in 2012 (Fig. 1b). This was mainly due to higher TPD 2 in that year compared to the previous year. Additionally, less plant losses occurred over the season 2013 by trend (Fig. 1a). As no protocol for a successful transplant production in Tks for field cultivation is available, transplant production was done according to the commonly practiced procedures for transplant production of the institute‘s experimental greenhouse and similar to the seedling production described by Suomela (1950). Hence, the reason for the high plant losses in the Tks plants is not known and can only be speculated. Maybe slightly cooler temperatures during rooting in April and May (Table 1) enhanced plant establishment (less transpiration via the canopy) and minimized transplant shock in 2013. Yet, warmer air temperatures and subsequently warmer soil temperatures enhance rooting of seedlings (Kaspar and Bland, 1992). However, from the vegetable productions it is known that transplant production and transferring seedlings into the field is challenging. In Tks the major losses occurred within the first 30 days after planting (Fig. 1c) which might be a result of a severe transplant shock. Transplant shock is generally described as the stagnation of seedling growth and development due to root and leave injuries during the transplanting process (Li et al., 2016). This phenomenon generally occurs in all plants grown from transplants including rice (Li et al., 2016), trees (Struve, 2009), ornamental plants (Franco et al., 2016) and all kinds of vegetables (Schrader, 2000). However, plants suitable for transplanting do recover from this period which implies for Tks that it is either not suitable for transplanting on large scale or the practiced transplanting procedures need optimization. The high plant losses observed in the presented study indicate that the factors facilitating a successful Tks stand establishment via transplants need further investigation. It would be necessary to identify promoting/inhibiting factors such as container type, substrate, production system, irrigation, fertilization, transplant age, hardening conditions and watering that might influence the success of the plant establishment in the field (Boyan and Granberry, 2010), which have not yet been investigated in detail for Tks. Furthermore, abiotic (Franco et al., 2011) and biotic stress may inhibit plant growth after recovery of transplant shock. In the course of this study, various pathogenic fungi were isolated and identified by ITS region sequencing from the roots of younger and older Tks plants e.g. *Phoma exigua*, *Alternaria* sp., *Fusarium* sp., *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, *Erwinia* sp. (unpublished results), where each species can cause commonly known root diseases in many major crops. These may have added to the plant loss over season. Suomela (1950) also observed that 10–15% of Tks plant losses mainly occurred due to *Sclerotinia* sp., however during overwintering.

4.2. Year and agronomic measures influenced the achieved plant density at harvest

Overall, transplanting in ridges resulted in higher APD than in flat beds (Fig. 1b), however, this effect was solely highly significant (Fig. 3a) in 2013. This might be caused by a reduced soil compaction enhancing the establishment of the root system of seedlings as e.g. described by Krause et al. (2009) and Blazewicz-Wozniak and Konopinski (2012). Furthermore and as shown by Hulugalle (1990), the advantage of ridge cultivation over flat beds becomes more obvious under arid conditions due to the capacity of the furrows to conserve water. This would explain the more pronounced advantage of the ridges over the flat bed as observed during the drier season in 2013 (Table 1).

Even though higher TPD appeared to be associated with higher plant losses over the season (Fig. 1a), higher TPD was significantly associated to higher APD (Fig. 1b). E.g. in 2012, 55% and 44% plant losses occurred in TD1 and TD2, respectively, indicating that this was a result of lessened competition in the wider stand. This argument is

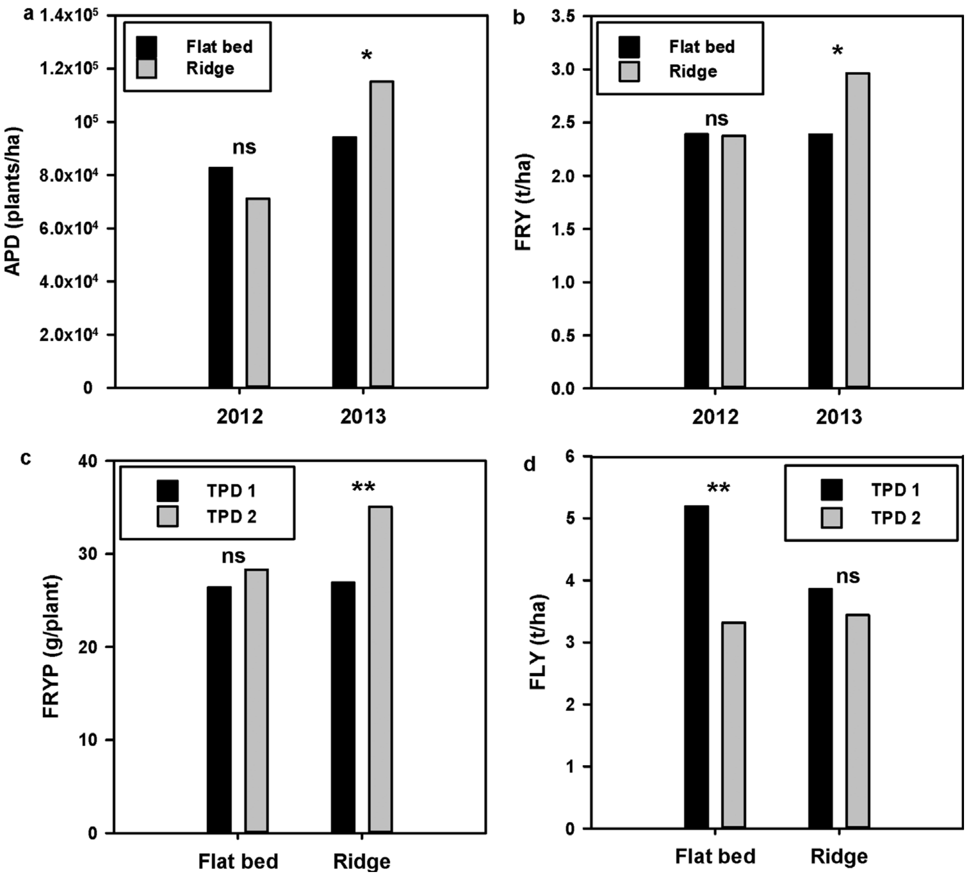

Fig. 3. Interaction between year of cultivation (2012, 2013) and type of planting bed (flat bed, ridge) on (a) achieved planting density (APD) at harvest and b) fresh root yield (FRY). Interaction between type of planting bed (flat bed, ridge) and targeted planting density (TPD 1, TPD 2) on (c) fresh root weight per plant (FRYP) and (d) fresh leaf yield (FLY). Differences between group means were analyzed with *t*-test or Mann-Whitney rank sum test (if normality or equal variance of data was not given). ns, *, ** indicate no/a significant difference between means at a p level > 0.05, < 0.05, < 0.01.

strengthened by the findings of Harper (1977) that very high sowing densities lead to self-thinning in cereals.

4.3. *Most yield parameters were significantly influenced by year*

The trial year had a significant effect on 11 of the 14 measured yield parameters (exceptions were DRY, DRYP and RYP) (Fig. 2a). This

Fig. 4. Tks transplants on ridges (a, c) and flat beds (b, d) with a planting density of 130 plants per row at 30/5/2013 (a, b – beginning of flowering) and at 09/07/2013 (c, d – peak of flowering). In the early seedling stage, a fence was erected as protection against rabbits. Pictures: JKI, Eggert.

indicated that most of the parameters were strongly influenced by environmental conditions. The average DRYP was about 6 g and did not vary significantly between years across all treatments. Subsequently, DRY and RYP appeared not to be influenced by year. RYP ranged only between 0.32–0.35 g in both years even though RC was significantly higher in 2013 (5.4% of dm) than in 2012 (4.1% of dm). Since DRY was not affected by year, significant differences in RY and IY between the two years resulted from significant absolute differences of RC and IC between years. Interestingly, there seemed to be a moderate negative correlation between IC and RC ($r = -0.53$) and RY ($r = -0.48$) (Table 2). Reduced RY was associated with higher IY in 2012 compared to 2013 and vice versa (Fig. 2a). This relationship indicates that inulin and rubber biosynthesis in Tks may be associated and even be linked antagonistically. This was already indicated by an observed seasonal drop of inulin during overwintering and a synchronic increase of rubber in field Tks by Kreuzberger et al. (2016) as well as by Cornish et al. (2013) who showed that cold induction decreased inulin and increased rubber concentrations in large stored Tks root systems. However, the underlying mechanisms need to be unraveled (Iaffaldano et al., 2016).

4.4. Root yield was increased in ridges

The type of planting bed significantly affected FRY, FRYP, DRYP and FLY (Fig. 2b). Considering both years and planting densities, FRY (5%), FRYP (11%) and DRYP (13%) were significantly higher in ridge cultivation than in flat bed whereas FLY (5%) was increased in the flat beds compared to ridges (Fig. 2c). However, increase of FRY and individual root yield did not result in a simultaneous increase of DRY, RY or IY in ridges. Nevertheless, there was both an interaction between year and type of planting bed for APD and FRY (Fig. 3a and b) indicating that higher FRY (16%) in ridges compared to flat beds was a result of higher APD (16%) in ridge cultivation compared to flat bed in 2013. In 2012, there was no significant difference for APD or FRY between the two types of planting beds. Since increased FRY was not associated positively with any other yield parameter, except with DRY (Table 2), a possible advantage of ridge cultivation with regard to yield was clearly related to APD and subsequently the factors influencing APD. Significantly higher FRYP in ridges than in flat beds was solely apparent in 2013 (Fig. 3b). Additionally, FRYP was only significantly increased in TPD 2 (Fig. 3c). Obviously, individual plants developed higher root biomass in ridges under certain environmental (year) and agronomic conditions (wider stand), probably due to looser soil in ridges compared to flat bed and reduced plant competition. The benefits of ridge cultivation for root crops have been demonstrated for e.g. carrots (Sady and Cebulak, 2000), sugar beet (Krause et al., 2009), and parsnip (Konopinski et al., 2011). According to these studies, ridges have higher soil temperatures, a deeper root penetration and hence an advanced root development over flat beds. As described for other root crops (e.g. Krause et al., 2009; Blazewicz-Wozniak and Konopinski, 2012), it is likely that the establishment of an efficient ridge cultivation system for Tks was hampered in the present study by the high plant losses as observed in both planting seasons. To fully explore the potential of such a planting system further research would be necessary. The advantages and disadvantages of the two types of planting beds in Tks cultivation need clarification through further trials with different soil types, e.g. with a higher sand content. The soil at the study site had a very high clay content which is associated, e.g. with a greater water holding capacity and higher soil crusting potential than more sandy soils. At the same time, a loamier soil has slower water infiltration rates and warms up slower in spring than a sandy soil (Orzolek, 1999). All these aspects will most likely affect the root biomass formation of Tks in prospective cultivation regions.

4.5. Achieved planting density affected yield per area and of individual plants

TPD was highly positively correlated to APD ($r = 0.78$) (Table 2, Fig. 1b) and APD was negatively correlated with yield of individual plants (Table 2, Fig. 2c). Because of the significant seasonal plant losses, it was decided to calculate individual plant yield based on APD. An adverse relationship between plant density (TPD and APD) and individual plant yield was underlined by the negative correlation coefficients for FRYP, DRYP, FLY, DLYP, RYP and IYP ($r = -0.49$ to 0.57) (Table 2). This negative relationship is an indication for the competition between plants, originally induced by different TPD. Individual Tks plants had significantly higher FRYP, DRYP, FLYP, RYP and IYP in TPD 2 compared to TPD 1 (Fig. 2c). This strongly implies that reduced TPD led to higher yield formation of individual plants compared to plants in a denser stand. E.g. in TPD 2, DRYP was about 21% higher than in TPD 1. Subsequently, higher DRYP also led to higher RYP and IYP in TPD 2 compared to TPD 1 since RC and IC were equal in both treatments. This increased root development going along with decreased plant spacing implies that there was competition between the plants. Since higher TPD resulted in higher APD, higher TPD was also associated to significantly higher FRY, FLY and DLY in TPD 1 compared to TPD 2 (Fig. 2c). This is in agreement with the statement by Weiner and Freckleton (2010) who concluded that biomass per area increases linearly with planting density up to a critical crop density. It is clear that this critical point was not yet reached for Tks under the study conditions. In any case, there were significant positive correlations between APD and biomass yield per area. For FRY, DRY, FLY and DLY the coefficients of correlation r were 0.75, 0.48, 0.35 and 0.23, respectively (Table 2), indicating that each plant contributed significantly to these yield parameters. As reported by Hecht et al. (2016), until today there is no comprehensive understanding of how sowing/planting density affects root systems because most studies in this field focus on the aerial part of the plant. It is known that individual shoot biomass decreases with increasing density (Harper, 1977), which is reflected by reduced FLYP in the denser TPD 1 stand compared to the wider TPD 2 stand of this study (Fig. 2c). Abdollahi and Mahna (2012) showed that increasing planting density (125,000–500,000 plants/ha) in *Cichorium intybus* led to a significant reduction of leaf area per plant, fresh root weight per plant, root length and root diameter. Yonts and Smith (1997) already showed that increasing planting populations (TPD 25,000–150,000 plants/ha) led to an increasing number of small roots in sugar beet. This study demonstrated that Tks is capable to compensate root yield per area in stands with reduced planting densities by an increase of root yield of the individual plants. This may have led to the effect that DRY, RY and IY did not differ significantly between TPD 1 and TPD 2 despite significantly higher APD (Figs. 1b and 2c). An optimal planting density for Tks transplants still needs to be defined.

5. Conclusions

In order to establish the resource intensive transplanting of Tks as an efficient cultivation and management system, further investigations considering all essential aspects for a successful transplant production would need to be performed on a spatial and temporal level. The advantages of transplanting compared to field seeding could be reduced weed protection during the sensitive seedling stage and a longer growth period. The later benefit may be specific for loam soils as on this study's trial site because of slow warming up in spring and slow germination of Tks seeds under cool conditions. Hence, further investigations should focus on the optimal production of Tks transplants and their transfer to the field and subsequent management. Of course many aspects of vegetable transplant production summarized in Orzolek (1999) could be adapted to this uprising industrial crop. A direct comparison of the two systems, transplanting versus direct seeding, might be a feasible approach if an optimized crop management system is established for each

Fig. 5. Differences in root system of Tks after transplanting (left) and after direct seeding (right). Both root types (fresh root weight about 10 g) stand exemplarily for the roots harvested in two different trials using transplants (this study) or direct sowing (described in Kreuzberger et al., 2016). Both roots developed from spring (transplants: greenhouse, February 2012, field, April 2012; direct seeding: field, March 2012) to middle of October 2012. Pictures: JKI, Eggert.

of them. Suomela (1950) assumed that field-sown Tks stands with a planting density of 40–95 plants per row meter give higher yields of rubber than transplants were the growth period was equal, due to the higher planting density in sown stands. At status quo, poor/unstable stand established in both systems, absent field techniques and heterogeneous plant material are the major obstacles for the performance of meaningful agronomic field studies in Tks. However, careful (not completely meaningful due to different planting densities) comparison of the yields from transplants with the results of sown stands in the same environment (Kreuzberger et al., 2016) support the presumption of Suomela (1950). In the presented study, achieved planting density at harvest was directly linked to fresh/dry root yield per hectare and also influenced the yield of individual plants. Aside from the (uncontrollable) environmental conditions, yield parameters of Tks transplants were influenced by both agronomic measures, choice of planting bed and planting density. There was a significant advantage of the ridge system over flat bed cultivation regarding APD, FRY, FRYP and DRYP under specific environmental conditions (drier season) and planting density (reduced). The most relevant yield parameters (DRY, RY, IY) were not influenced by these measures. RY and IY are both positively related to high DRY, and high DRY is achieved by high APD (Table 2). Therefore, RY and IY will be increased if the harvested plant number is increased. A predictable APD will also help to optimize the planting density within the range of the yield potential of an individual plant, i.e. the time before root competition reduces single root mass significantly. This would also facilitate a reliable prediction of root yield at harvest. Future planting density should consider the yield potential of the individual plants that varied independently of year and agronomic measures, from 2.8–28.3 g DRYP, around 0.1–1.1 g RYP and from 0.5–9 g IYP in the Tks material available. Due to the high and not yet predictable plant losses over the season, it is suggested to transplant as many Tks plants per area as technically feasible because this ensures higher APD.

As a further aspect, the architecture of the Tks roots has to be considered as well when transplanting. In general, Tks develops a tap root with few lateral roots (Lipshitz, 1934; Kreuzberger et al., 2016). It was observed that the development of the tap root was apparently impaired by transplanting, leading to the formation of numerous lateral roots close to the root crown compared to plants that had developed from direct seeding (Fig. 5). This effect of branched tap roots is also known from transplanted sugar beets (Theurer and Doney, 1980). These lateral roots broke off during harvest and lead to reduced yield. Root processing also became more difficult due to increased soil and gravel attachments between root branches. Hence, this lateral root formation also appears undesirable in Tks and should be minimized.

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