Population density and soil seed bank of weed beet as influenced by crop sequence and soil tillage

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ABSTRACT

Dynamics of population density and soil seed bank of weed beet was studied in a 5 year crop rotation consisting of spring barley, and sugar beet. Beside the crop rotation experiment, the seeds of weed beet were studied for their dormancy and viability in soil seed bank over the period of four years. The obtained data indicates that weed beet was able to produce seeds only in sugar beet, but not in barley. In sugar beet, its reproductive potential allows weed beet to restore and increase the soil seed bank of glomerules rapidly. Common infestation of sugar beet is able to persist over more than the 2-year period between repeated introductions of sugar beet in crop rotation. The experiment has also proven the negative effect of weed beet presence on sugar beet yield. The sugar beet root yield decreased of 0.4 t/ha with every 1000 weed beet plants per hectare. The yearly loss of viable seeds was about 75%. The number of surviving seeds decreased exponentially in time. Less than 2% of seeds remained viable after three years in the soil. Seasonal fluctuations of seed dormancy were observed. Seeds were dormant in autumn, lost dormancy in winter and recovered it in late summer.

Keywords: *Beta* spp.; crop rotation; population dynamics; soil seed bank; sugar beet; weed beet

Wild and cultural forms of genus *Beta* (beet) are highly sexually compatible and create so called *Beta*-complex (De Bock 1986, Bartch et al. 1999). Annual weedy hybrids of wild and cultural forms of beet referred to as 'weed beet' were first described in the 1920s in the USA (McFarlane 1975). In Europe, the problem of weed beet raised in the 1960s when sugar-beet seed multiplication moved to the Southern European regions (Northern Italy and South-western France) for the reason of more favourable natural conditions for seed production. However, these areas are also habitat of wild seed beet (*Beta vulgaris* ssp. *maritima* Arcang.) and feral beet populations which often flourish in the neighbourhoods of sugar beet seed multiplication fields (Bartsch and Schmidt 1997, Boudry et al. 2002, Van Dijk 2004). The wild forms are ancestors of several cultural forms of beet, especially sugar beet (*Beta vulgaris* ssp. *vulgaris* var. *altisimma* Döll, syn. var. *saccharifera*), most important beet crop widely grown all over the world (Statistical Yearbook 2009). The offspring is predominantly annual carrying the dominant 'bolting gene' inherited from their wild ancestor (Boudry et al. 2002). The seeds are distributed as sugar beet seed impurity and establish first primary populations when this contaminated sugar beet seed is sown in production areas. Because of annuality, the weed beet plants produce seeds in the first year; after shedding they enrich soil seed bank being a base for secondary populations infesting the fields for many years (Müncher et al. 2000).

Weed beet populations are difficult to control in sugar-beet fields for their close botanical relation to the crop which does not allow the use of herbicides. The only highly effective regulation methods demand high amount of manual work (repeated hand pulling of the flowering plants,

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when the bolted plants are removed from the field) (Bond and Turner 2004, Van Dijk 2004). That is the reason why the hybrid plants often escape regulation and in many European countries the weed beet is becoming a major problem for sugar beet production (Longden 1993, Soukup et al. 2002). Presence of weed beet in the sugar beet fields causes not only technological problems, but also high yield losses both in quantity and quality (Fayed et al. 1997). Weed beet occurs also in other crops; however, it can be easily regulated by selective herbicides (Sester et al. 2004).

The weed beet population dynamics are affected by both environment and agronomy. The bolting rate is getting lower and the flowering time later with growing weed beet densities. The first can be explained by the importance of light quality and quantity for bolting induction, the others are caused by the competition of plants for nutrients (Sester et al. 2004).

The seed production of weed beet is highly dependent on the interspecific and intraspecific competition pressure, which the plants are exposed to, and also on the time of flowering. The plants flowering sooner produce more seeds than the plants which flower later (Soukup et al. 2002). When the plants are not destroyed before they complete their life cycle, weed beet plants produce on average 1000 to 2000 glomerules per plant, each carrying on average 2 or 3 seeds (Soukup et al. 2002).

The weed beet seed bank dynamics are determined by the primary dormancy of the weed beet seeds. The cultural plants have been bred for rapid and simultaneous germination. On the other hand weed beet have retained the dispersal mechanisms of their wild relatives, so their seeds germinate within several years. Hence the once established weed beet population can persist in the seed bank between the sugar beet crop rotation cycles (Sester et al. 2006).

The survival rate is even higher and its duration even longer when the seeds are buried in deep layers of soil, as the seeds become dormant in such conditions. Longden (1974) reported almost 100% weed beet seed mortality after 8 years when buried in 5 cm of soil, and after 15 years when buried in 50 cm of soil. If we expect a linear dependence between the depth in which the seed is buried and length of survival, under the usual 20 cm tillage practice the weed beet seeds keep their germinability for about 10 years.

The difference in seed survival in various burial depths is the ground of the tillage depth effect on population dynamics of weed beet. The deeper the tillage the higher percentage of weed beet individuals will survive until the following sugar beet crop and reproduce (Bond and Turner 2004). Analogously, the survival rate will be lower in longer sugar beet crop rotations (Bond and Turner 2004). Maughan (1984) reported infestation lower by 30% in fields with 4-year sugar beet rotations and by 80% in fields with 8-year sugar beet rotations compared to fields with 3-year rotations.

The weed beet occurrence can be also lowered by other agronomical practices. In general, all practices stimulating the weed beet germination have negative effects on the weed beet population. Despite the seed-time of sugar beet does not have a direct effect on the weed beet occurrence (Maughan 1984), a sowing delay of 7 or 10 days after the presowing tillage can cause destruction to up to 30% of all germinated weed beet seedlings in the year (Longden 1980).

In the sugar beet crop the weed beet control consists mainly of repeated manual or mechanical removing of the inter-row beet plants and all bolters. According to Colbach et al. (2009) a successful weed beet control depends more on optimal timing rather than the high efficiency of these operations.

An early harvest can prevent the later flowering bolters shedding seeds. Also, the longer the seeds will be left on the surface of the soil after the harvest the more of them will germinate and later can be destroyed by tillage or predation (Bond and Turner 2004). The early harvest time can affect the number of viable weed beet seeds as it prevents the later flowering plants to produce matured seeds.

Despite the fact that weed beet has been occurring in sugar beet fields and in other beet crops for decades, it was for a long time overlooked by the growers and its relevancy underestimated. Therefore, its prevalence has risen on many fields to relevant levels and in some cases it has even become a menace to profitable sugar beet production on the particular fields or farms (Bartsch et al. 1996, Soukup et al. 2002, Darmency et al. 2007). Therefore there is a need for detailed research on weed beet biology and population dynamics to identify its actual weediness, dispersal ability, and the best practice to reduce and prevent the infestation in agro-ecosystems.

The problem of weed beet has become even more prevalent in light of the possible introduction of GM sugar beet, as the possible hybrid forms between GM sugar beet and wild beet can establish new transgenic weed beet populations (Bartsch et al. 1996, Bartsch et al. 2003).

MATERIAL AND METHODS

Field experiment

The small plot experiment (randomised blocks with four replications) was established in the field without natural occurrence of weed beet in soil seed bank, in natural conditions typical for growing sugar beet. Soil conditions: loamy soil, chernozem, pH 6.99, humus content 3%, capillarity 51.66. Climatic characteristics: normal annual precipitation 470 mm, yearly average temperature 8.4°C (Table 1).

A locally common crop sequence was used as follows: spring barley – spring barley – sugar beet, these three crop sequences were established in 2004, 2005, and 2006. Potatoes were used instead of sugar beet as forecrop at the beginning of the crop rotation to avoid accidental contamination of seed bank by seeds of weed beet. The weed beet seeds were harvested from a long-term established population in a Central Bohemian field at Libochovicky (50°10'22.831''N, 14°14'40.384''E). Each crop rotation trial was divided into 3 sections where weed beet seeds were distributed at three densities: 100, 1000 and 10 000 glomerules (gl.) per plot, respectively. Each section had 4 replications/plots each having 30 m^2 in size. The introduction of weed beet seeds was conducted before the beginning of the crop sequence in the autumn after ploughing. A shallow incorporation of 10 cm was used to simulate the natural seed bank establishment by soil tillage after harvest of sugar beet infested by weed beets (Table 2).

Common soil tillage practices were used – ploughing in autumn and shallow seedbed preparation (5 cm) in spring. Barley was sown at the density of 350 seeds per m^2.

Assessments

1. Occurrence of weed beet seedlings in crop stand. Abundance of weed beets was assessed in each plot. In cereals the occurrence was noticed only twice because of strong crop competition: first in the spring before canopy closure, the second in autumn, counting the seedlings emerged in the period between stubble tillage and autumn ploughing. Weed beet occurrence in sugar beet crop was assessed once before the canopy closure.

2. Evaluation of soil seed-bank. Evaluation was conducted only at the end of the whole crop sequence in sugar beet in the autumn soon after harvest because the probability of finding the seeds in soil samples would be very low especially at the lowest initial densities. Soil samples were taken up randomly in each subplot by the Eijkelkamp piston sampler from the whole soil profile 30 cm (ploughing depth) and collected in one mixed sample (10 l) from each replication. Each sample was eluted through metal sieves of 1.5 mm size. The soil skeleton was dried and then manually examined for presence of weed beet seeds. Seed viability (germination) was tested on Petri dishes in a climatic chamber of regulated conditions (20°C, 16 h light/8 h darkness).

3. Impact of weed beet occurrence on sugar-beet yield. Sugar beet was harvested and weighed up; samples of 20 beet roots from each subplot were examined for qualitative parameters – digestion and ash contents in accredited laboratory. Correlations between weed beet occurrence and sugar beet yield were calculated using simple linear regression.

Longevity of soil seed bank

The seed bank longevity and dormancy of weed beet glomerules was studied in two experiments; the 1st experiment describing the long-term decrease of weed beet viability during four consecutive years (2004–2007) and the 2nd experiment focused on dormancy cycle/s within one year (2005).

The weed beet seeds were collected after ripening at the beginning (exp. 1) and the end of October (exp. 2) in Pohorelice (48°58'0.233''N, 16°29'35.941''E, designated as location 1), Litobratrice (48°52'44.883''N, 16°23'42.239''E, designated as location 2), Branisovice (48°57'55.303''N,

Table 1. Climatic characteristics of the years of experiments for Prague and Central Bohemia

Year	2004	2005	2006	2007	2008
Annual mean air temperature (°C)	8.5	8.4	8.9	9.8	9.4
Deviation from long-term normal 1961–1990 (°C)	0.3	0.2	0.7	1.6	1.2
Annual precipitation totals (mm)	566	595	592	604	527
Annual precipitation totals as percentage of the long-term normal 1961–1990 (%)	96	101	100	102	89

Table 2. Time scheme of the field experiment

Year	1st rotation	2nd rotation	3rd rotation
2004	spring barley		
2005	spring barley	spring barley	
2006	sugar-beet	spring barley	spring barley
2007		sugar-beet	spring barley
2008			sugar-beet

16°25'4.576''E, designated as location 3) and Unetice (50°9'27.403''N, 14°21'6 .801''E, designated as location 4) in sugar beet crops. Seeds originated from Libochovicky (50°10'22.831''N, 14°14'40.384''E) were used in the 2nd experiment. The seeds were dried in room conditions for 5 days. A hundred glomerules (each containing 3 seeds) were wrapped with 100 g chernozem soil into pieces of nylon fabric with 0.24 mm mesh size to stop seed predation. The design of the experiment was set in four replications per location and per sampling period. The samples were buried at a depth of 15 cm. During April/November (exp. 1) and monthly intervals (exp. 2), four bags per location were exhumed and the percentage of germinating seeds was established at the most favourable temperature: 20°C and 14 h photophase. All 4 × 100 glomerules per replication were placed on moist filter paper in a Petri dish of 12 cm in diameter. The percentage of germinating seeds was recorded 14 days afterwards. Soil temperatures were recorded at hourly intervals by Tinitag dataloggers (Gemini Data Loggers, Chichester, UK) located adjacent to the experimental site at 15 cm in depth. The data were fitted to either exponential (continuous steps) or non-linear model (discontinuous course).

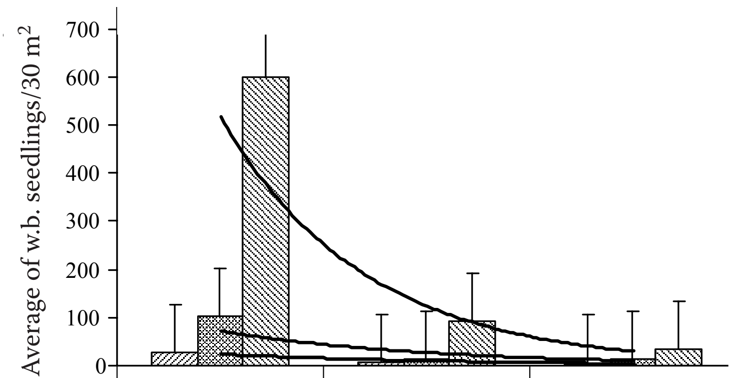
Data analysis. Raw data were analyzed by ANOVA, and linear regression and non-linear models using the Statistica software program, version 9.0 (StatSoft 2009).

RESULTS

Field experiment

1. Occurrence of weed beet plants during the crop rotation. The highest number of weed beet seedlings was usually found in barley crops that were established the following year after distribution of weed beet seeds in experimental fields. In the second year of the crop rotation (subsequent barley) the number of weed beet seedlings decreased on average by 82% compared to the occurrence in the first year of the rotation. Analogously, in the last, third year of the rotation (sugar beet), the numbers of weed beet seedlings decreased by another 5.3% compared to the first year (Figure 1 and Table 3).

Moreover, the sum of emerged weed beet plants over the crop rotation was much smaller than the initial input considering that each glomerule added to the soil seed bank was carrying about 3 seeds of weed beet. Relatively higher percentages of plants germinated from the lower initial seed bank than



	s. barley 1	s. barley 2	sugarbeet
100 gl./30 m²	28	7	5
1 000 gl./30 m²	103	13	15
10 000 gl./30 m²	599	92	34

Figure 1. Numbers of weed beet plants that emerged in the three rotations of the field experiment in the three different initial seed bank trials. The vertical bars indicate the standard errors. The numbers of emerged weed beet seedlings decreased exponentially over the years of crop rotation (initial seed bank 10 000 gl./30 m^2: $y = 2152.7e^{-1.4283x}$; $R^2 = 0.9684$; initial seed bank 1000 gl./30 m^2: $y = 191.82e^{-0.9782x}$; $R^2 = 0.7133$; initial seed bank 100 gl./30 m^2: $y = 56.954e^{-0.8673x}$; $R^2 = 0.8976$)

Table 3. The field experiment results – average emergence of weed beet seedlings per plot (plot = 30 m^2)

Initial seed bank (gl./plot)	1st rotation			2nd rotation			3rd rotation		
	100	1 000	10 000	100	1 000	10 000	100	1 000	10 000
2004	14.75	42.50	382.50	–	–	–	–	–	–
2005	3.00	7.00	47.50	60.75	234.00	1 065.00	–	–	–
2006	1.25	2.50	25.75	14.75	26.25	203.50	9.50	33.00	349.25
2007	–	–	–	2.00	8.75	24.25	3.75	6.50	24.50
2008	–	–	–	–	–	–	11.75	32.50	53.25

from the higher initial seed bank (for the average of 3 seeds/glomerule: 13.5% of the 100 gl./30 m^2 seed bank, 4.5% of the 1000 gl./30 m^2 seed bank and 2.5% of the 10 000 gl./30 m^2 seed bank).

The differences in weed beet emergence observed among the crop rotations were most likely caused by differences in the crop stand quality. Where the crop stand was weaker, the numbers of emerged weed beet seedlings were higher.

In the barley crop the highest intensity of weed beet emergence was noticed in spring. First weed beet seedlings emerged within days after sowing and the emergence continued until the canopy closure. However, the weed beet seedlings were soon overgrown by the crop plants and died out in the growth phase of leaf rosette, or in the phase of extension growth while trying to reach for light. The second wave of weed beet emergence appeared after the crop harvest, when the soil surface was again exposed to light. Nevertheless, these newly emerged weed beet plants were destroyed by soil tillage in their early growth stages and therefore could not reproduce and enrich the weed beet seed bank.

The last, third year of crop rotation with sugar beet was the most important from the viewpoint of weed beet occurrence. The weed beet plants emerged in spring with the sugar-beet plants bolted, and produced high numbers of seeds. First flowers opened at the beginning of July and at the end of July seeds started to develop. At the beginning of September almost all seeds were fully matured and started to shelter from the stems. By the time of the sugar-beet harvest most of the seeds were already sheltered down on the soil surface.

However, there was no significant difference in weed beet occurrence in sugar beet after three years crop rotation ($P = 0.11$) between the initial infestation levels by weed beet seeds. Perhaps, due to the high mortality of glomerules and their distribution in soil profile by tillage over the three years of the crop rotation.

2. Evaluation of soil seed bank. The number of glomerules found in the soil samples were calculated for the size of a plot = 30 m^2 and 20 cm of plough layer so the infestation could be compared to the original level. The resulting seed bank varied between the rotations. In the 1st rotation, the number of weed beet seeds in the soil seed bank often decreased from its original level. On average the weed beet seed bank in the 1st rotation rose 4.26 times and in the 2nd rotation 82.49 times (Table 4).

There were wide variations among the replications of the same trials in numbers of weed beet glomerules found in the seed bank at the end of each rotation shown. There were also differences between the data from the parallel rotations of the experiment. At the end of the 2nd rotation, 9 times more weed beet glomerules was found in the soil than at the end of the 1st rotation, even though the initial soil seed bank was the same, the numbers of weed beet plants in the sugar-beet crop were similar (Figure 2) and the numbers of weed beet plants emerged over the three years of the rotations were also similar (see above).

The germination rate of the beet glomerules found in the soil samples at the end of the 1st rotation was just an average of 25.2%, which is much

Table 4. The field experiment results – average resulting seed bank of weed beet glomerules calculated per plot (plot = 30 m^2)

Initial seed bank (gl./plot)	1st rotation			2nd rotation		
	100	1 000	10 000	100	1 000	10 000
2006	600	5 550	12 300	–	–	–
2007	–	–	–	19 950	37 200	107 700

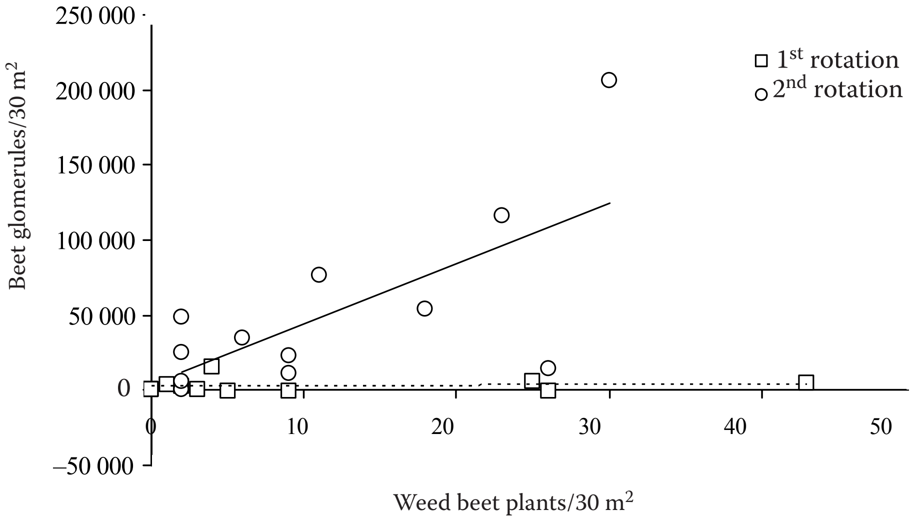


Figure 2. Correlation between the number of weed beet plants per plot (30 m^2) and the balance (decrease/increase) of glomerules in soil seed bank at the end of the modelling crop rotation (found beet glomerules calculated for the size of a plot = 30 m^2 and 20 cm of plough layer, decreased for the initial seed bank on the plot). Regression functions: 1st rotation: $y = 42.057x + 2036.4$, $R^2 = 0.0157$; 2nd rotation: $y = 4019.9x + 4350.8$; $R^2 = 0.4743$)

lower than the germination of the beet glomerules from the 2nd rotation (an average of 40.03%).

3. Impact of weed beet occurrence on sugar-beet yield. The correlation between the number of weed beet plants and the sugar beet yield (Figure 3) was on the significance level $\alpha = 0.05$. On the contrary, the qualitative parameters of sugar beet yield showed no statistically relevant correlations with the number of weed beet plants per plot.

Longevity of soil seed bank

The weed beet seed lost viability during a three year period; less than 2% of viable non-dormant seeds were observed after that time. The proportion of surviving seeds decreased exponentially ($y = 76.332\exp(-0.604x)$). The rapid loss of viability within the first three years was recorded (≥ 90%), followed by a slower rate of decline with a low proportion of seeds remaining viable. The annual decline rate of the weed beet seed bank was 75% each year (Figure 4). Even if this number seems to be very small, in fact, the weed beet seed bank is large, thus, emergence of a small fraction of the seed bank would result in a high seedling density.

Seeds were in primary dormancy at the moment of their burial. The high dormancy level occurred immediately after dispersal, at about 5% of buried seeds that were able to germinate. In the course of one year they passed through a pattern of dormancy change. Seasonal fluctuations were

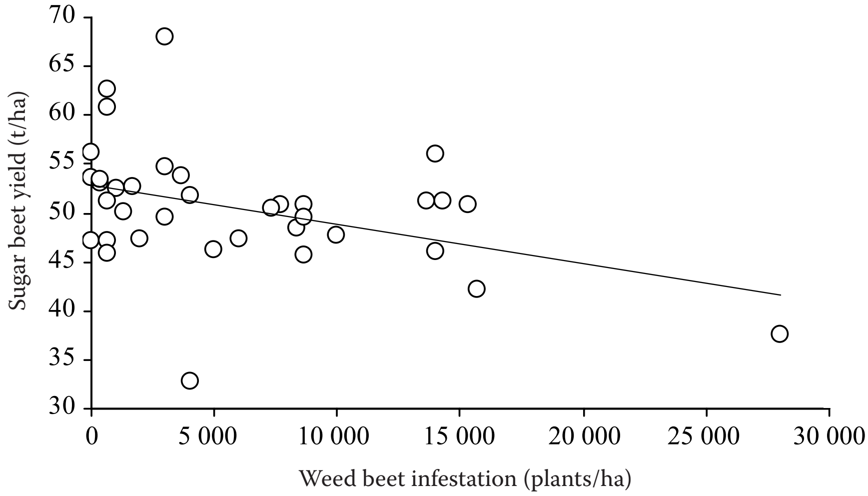


Figure 3. Correlation between the number of weed beet plants calculated per hectare and the sugar beet yield calculated per hectare. (regression function: $y = -0.0004x + 52.811$, $R^2 = 0.1608$). Set of data from all tree rotations

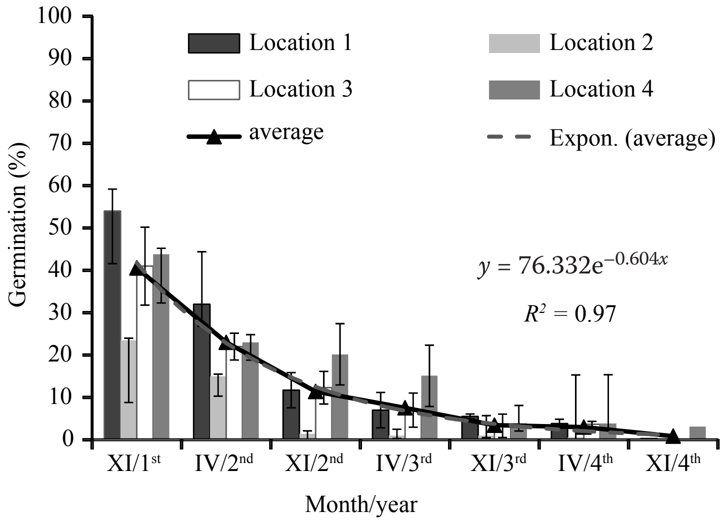


Figure 4. Weed beet seeds survival in the soil as decreased with the time

observed, when seeds were dormant in autumn, lost dormancy in winter (germination up to 70%) and recovered it in late summer. The factor that most likely affected this process was the soil temperature. The reintroduction of dormancy was triggered by high temperatures during the spring and summer (Figure 5). Because the germination in the darkness was considerably lower than under light conditions, the data here are presented only for the photophase, nevertheless the course of the dormancy was very similar.

DISCUSSION

As expected, the experiment has reaffirmed that the weed beet plants occurring in a closed stand of cereals were not able to compete with crop and produce seeds even if not regulated by selective herbicides. The weed beet seedlings emerged also after the crop harvest but those plants were always destroyed by tillage before they reproduced. Therefore, none of these weed beet plants contributed to the soil seed bank. Moreover, the existing seed bank decreased from the emerged seeds. In spite of that, the weed beet seedlings are very sensitive to crop competition especially in the early stages of their development (Sester et al. 2004). The low competitive strength of weed beet in narrow-row crops is compensated by its ability to survive in the soil seed bank (Sester et al. 2006). The surviving rate is even higher and its duration longer when the seeds are buried in the deep layers of soil as in such conditions the seeds become dormant. Dormancy is an important feature of weed seed bank dynamics. Sester et al. (2007) modelled dormancy, germination and emergence processes in detail. They found that the mortality during autumn was high; the rate of dormant seeds was highest in autumn and lowest in late winter-early spring. It increased with seed age and seed depth and was lower for

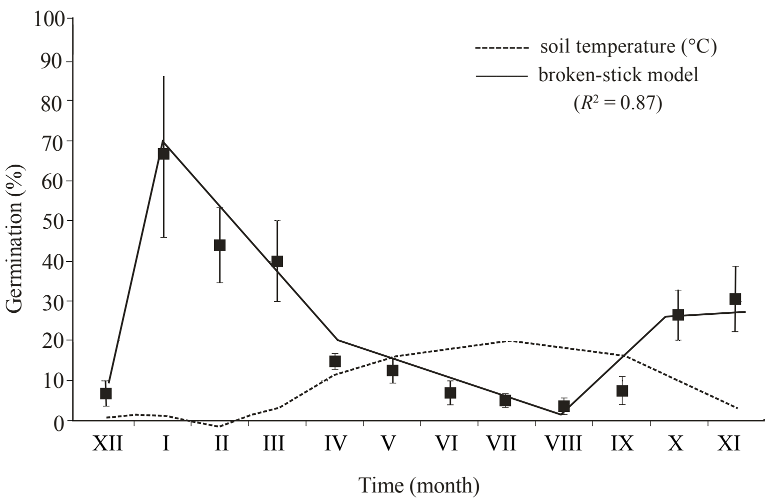


Figure 5. Proportion of non-dormant seeds of weed beet fitted to non-linear model. Symbols indicate mean values of four replicates and vertical bars of standard errors

light-stimulated seeds. In their studies, the total of seed bank due to natural mortality was approximately 20% each year. Our results showed a faster decrease of seed viability in soil. There are many difficulties in comparing the results of this study to other long-term seed longevity trials, because of differences in burial methods and procedures of viability testing. Additionally, artificial seed-bank studies may not accurately estimate seed longevity – decrease seed predation, modify soil temperature, moisture fluctuation, increase of pathogens and ignore tillage effects and many others. Therefore, such studies provide an indication of seed survival capability. Even populations within the same species can either present a cyclic dormancy pattern or not, depending on the origin of the population, which indicated that a cyclic dormancy pattern could be inherent to ecotypes from environments with marked seasons (Benech-Arnold et al. 2000). Temperature cannot be regarded as the only factor that can induce secondary dormancy. The term of seeds burial is very important and there are many environmental factors influencing seed dormancy status insufficiently explained. On the other hand, the data from such experiment could help to determine stages in the life cycles of weed that are crucial in the regulation of weed persistence.

Knowledge of the seed longevity may allow us to forecast if weed would be problematic in subsequent rotation, and the potential density that could be expected. Longden (1974) reported almost 100% weed beet seeds mortality after 8 years when buried in soil in 5 cm, and after 15 years when buried in 50 cm. If we expect a linear dependence between the depth in which the seed is buried and length of survival, under the usual 20 cm tillage practice the weed beet seeds keep their germinability for about 10 years.

In other words, weed beet commonly persists in the soil between cycles of sugar beet crop rotation, and also weed beet populations are growing faster in fields where sugar beet crop is grown in a closer rotation (Maughan 1984, Bond and Turner 2004). The data we obtained from our field experiment has also shown the decreasing rate of weed beet seedlings emergence most likely as a result of seed mortality over the years. Despite of that, relatively many weed beet plants emerged in the sugar beet crop at the end of the model crop rotation and successfully produced high numbers of seeds.

Weed beet has a huge reproductive potential – one plant can produce up to 20 000 glomerules (Bartsch et al. 1996) depending on the conditions. However, the number of seeds one plant produces is limited by competition of other plants (Laxander 1981, Sester et al. 2004) and usually oscillates from 1000 to 2000 seeds. Of course the seed production is also limited by other abiotic and biotic factors.

According to Van Dijk (2004) a single weed beet plant left in the sugar-beet crop to mature and produce seeds can in the next cycle of the sugar-beet rotation (3 years) cause a rise of the weed beet number to 300 plants per hectare. In our experiment the seed bank rose on average by 326% in the 1st rotation and by 7870% in the 2nd rotation. Also the germination rate of the weed beet glomerules was higher at the 2nd rotation.

The differences in the results were probably caused by the weather as the year 2007 when sugar beet was grown in the 2nd rotation was much warmer than the previous year 2006 (Table 1). Therefore the weed beet at the end of the 2nd rotation produced bigger and more branched plants and higher numbers of seeds than the weed beet at the end of the 1st rotation in the year 2006. However, our results do prove the above mentioned conditional. They also show that the rise can be even higher if the weed beet plants have favourable conditions for development.

Longden (1979) reported that the infestation of one weed beet plant per m^2 causes a decrease of white sugar yield between 9 to 15%, although our data did not prove this result. On the other hand, the field experiment data proved a negative correlation between the number of weed beet plants and sugar-beet root yield (Bitner 2001).

To conclude, the field experiment has demonstrated the high weediness of weed beet in sugar beet crop and its high population dynamics under sugar beet crop rotation and normal farming practices. Therefore an attentive monitoring of weed beet occurrence in sugar beet fields and its immediate elimination is essential for sustainable sugar beet production.

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