National Environmental Research Institute
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Timing of herbicide application

NERI Technical Report, No. 410

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Beate Strandberg
Marianne Bruus Pedersen

Data sheet

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Abstract:	In order to study the effects of herbicide tolerant fodder beets on arable biodiversity four different herbicide regimes were set up: Application of conventional beet herbicides, application of Roundup Ready following label recommendation, early application of Roundup Ready, and Roundup Ready applied as late as possible without any expected reduction in root yield. The 2001 results revealed that the Roundup Ready treatments may result in significant improvements of weed flora and arthropod fauna during June and early July compared to conventionally treated plots. A prerequisite for this environmental benefit is that Roundup Ready applications follow the label recommendation or are further delayed. The production of weed seeds was reduced or lacking as a consequence of the effectiveness of Roundup Ready. This may both have an effect on the availability of seeds as food for the birds and may also change the weed flora significantly. To conclude about the long-term consequences of transgenic herbicide tolerant crops on arable land biodiversity it will be necessary to study the effects over several seasons in relevant crop rotations.
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Foreword

During the last three years with the moratorium for marketing of transgenic crops in the EU, Monsanto, DLF-Trifolium and Danisco Seed have run demonstration trials with glyphosate tolerant fodder beets in Denmark. These trials were performed in collaboration with The Danish Agricultural Advisory Centre, and The National Environmental Research Institute, Department of Terrestrial Ecology took the opportunity to follow the trials and collect data on effects of introduction of transgenic herbicide tolerant crops to the arable flora and fauna.

The results from that work are reported in Bruus Pedersen and Strandberg (2000) available at the homepage http://www.sns.dk/natur/bioteknologi/roundup_art.htm, in Strandberg et al. (2002) and in the Technical Report No. 349 by Elmegaard and Bruus Pedersen available on the NERI homepage, http://www.dmu.dk/1_viden/2_Publikationer/3_fagrapporter/rapporter/FR349.pdf.

Based on the results from the years 1999 and 2000 we decided to design our own field experiment in 2001, so that the effects of herbicide application on the flora and fauna were studied more thoroughly and during the entire growing season. The results from the work in 2001 are presented in this report.

The study took place on the island Mors in northern Jutland, and we are most grateful to the farmers for access to their fields and for their very competent handling of the field work that was performed in cooperation with the local agricultural advisory consultants, Frank Løvendal and Børge Andersen.

We appreciate the help with field work we have received from the technical staff at the Department and send a special thank to Inger Møller and Birgit Kristensen who have been responsible for the identification of the arthropods. We also want to thank Dr. F. Tencalla for pleasant collaboration, Niels Elmegaard and Christian Kjær for comments on an earlier draft of the report, and Tinna Christensen for assistance with setting up the report.

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Summary

The present study clearly elucidated some of the agronomic advantages, e.g. the flexibility in herbicide application and a more reliable weed control, and potential environmental benefits of the herbicide tolerant fodder beet, viz. more weeds and arthropods in the field in early summer. However, it also focused on a potential drawback of the strategy that needs further investigation.

Previous studies on flora and fauna in conventional and transgenic Roundup tolerant fodder beets have revealed that the implementation of Roundup tolerant fodder beets may increase biodiversity in beet fields in the early summer period (Bruus Pedersen and Strandberg 2000, Elmegaard and Bruus Pedersen 2000). However, the improvement was indicated to rely on a delayed weed control. Whereas these studies aimed at a comparison of the effects of conventional and genetically modified fodder beet management strategies on biodiversity, the aim of the present experiment was to study the relationship between the timing of the herbicide application (whether conventional beet herbicides or Roundup Ready) and biodiversity further.

Four different herbicide regimes were included in the experiment: Application of conventional beet herbicides (May 10 and 22, June 14), application of Roundup Ready following label recommendation (June 6 and July 3), early application of Roundup Ready (May 25 and June 27), and Roundup Ready applied as late as possible without any expected reduction in root yield (June 27 and July 16). The study was designed as a split block design with four replications of each treatment and took place at the field site on Mors, which has also been included in the former studies. Each plot was 20 by 20 metres of which the central 18 by 18 metres were used for biological samplings. Weeds were sampled five times during the season and at three of these sampling occasions arthropods were collected for later identification in the laboratory.

The 2001 results revealed that the Roundup Ready treatments may result in significant improvements of weed flora and arthropod fauna during June and early July compared to conventionally treated plots. A prerequisite for this environmental benefit is that the first application of Roundup Ready is delayed relative to the application of conventional beet herbicides, i.e. Roundup Ready applications should follow the label recommendation (Monsanto Europe 1999) or be further delayed.

Until mid-summer the weed flora was more abundant and diverse compared to conventional herbicide treatment when Roundup application followed label recommendation. The largest improvements, however, were found when the first application of Roundup Ready was delayed by 48 days relative to the first application of conventional beet herbicides. The obvious consequence of this late treatment is that weeds are available in the field for a much longer period. Furthermore, a significant weed biomass built up, which benefitted the fauna in the field. The present year the weed biomass as well as

the arthropod density, measured on June 21 i.e. a week before the first Roundup Ready application of that treatment, was ten-foldly higher than when following label recommendation. The long period with weeds in the field did not result in any yield reduction the present year, but the farmers' readiness to accept weeds below the economic threshold for a longer period will be decisive for the positive effect on biodiversity during early summer.

Early application of Roundup Ready resulted in an extremely effective weed control with very few species, low densities and a very low weed biomass during the entire season. As shown in previous years, application of conventional beet herbicides resulted in a poor weed flora and few arthropods in the plots until the end of June. However, weeds were recruited in these plots succeeding the spraying period. Although they were not numerous, the weeds grew big and may thus present a problem at harvest. Furthermore, these weeds produced plenty of seeds, whereas no production of weed seeds was recorded in any of the plots receiving Roundup.

Generally, arthropod abundance and diversity also benefitted from label-recommended or late Roundup Ready treatment compared to conventional and early Roundup Ready treatment, but the response was delayed compared to the flora. The positive effect on arthropods disappeared in July in the plots treated according to label recommendations, whereas arthropods remained more numerous in the late-treated Roundup plots. The higher arthropod abundance and diversity especially in plots sprayed late with Roundup Ready compared to conventionally treated plots are likely to benefit arable birds, provided that the positive effect is not counteracted by insecticide treatments.

The reduced or lacking production of weed seeds that is a consequence of the effectiveness of Roundup Ready may both have an effect on the availability of seeds as food for the birds and may also change the weed flora significantly. Species composition may be changed towards species that are less sensitive to glyphosate, and density and biomass may be altered, too. Thereby, the basis for the positive effect on arable land flora and fauna in the early summer may also change. To conclude about the long-term consequences of transgenic herbicide tolerant crops on arable land biodiversity it will be necessary to study the effects over several seasons in relevant crop rotations.

Dansk resumé

Konklusioner

Undersøgelsen viste klart, at der er mulighed for forbedringer for plante- og dyreliv i marken i forsommeren ved dyrkningen af glyphosat-tolerante foderroer sammenlignet med konventionelt dyrkede roer blandt andet som følge af fleksibiliteten i sprøjtetidspunkt. Vi fandt imidlertid også, at den meget effektive ukrudtskontrol ved anvendelsen af glyphosat-midlet Roundup Ready (RR) bevirker, at der produceres meget få eller ingen ukrudtsfrø i marken i år, hvor der dyrkes glyphosat-tolerante roer. Langtidskonsekvenserne af en begrænset eller manglende produktion af ukrudtsfrø kendes ikke og bør undersøges for relevante sædskifter. En begrænset eller manglende frøproduktion må forventes at have betydning ikke kun for de fugle, der spiser ukrudtsfrø efterår og vinter, men også for ukrudtsfloraen i marken de efterfølgende år. Artssammensætningen vil ændres således at arter, der er mindre følsomme overfor glyphosat, bliver hyppigere, og tæthed og biomasse kan også blive påvirket. Dermed ændres grundlaget for de forbedringer af ukrudtsflora og leddyrfauna, vi har fundet i forsommeren.

De fundne forbedringer af forholdene for ukrudt og leddyrfauna i forsommeren i marker, hvor der dyrkes gensplejsede, glyphosat-tolerante foderroer sammenlignet med konventionelle foderroer stemmer overens med tidligere undersøgelser (Bruus Pedersen og Strandberg 2000, Elmegaard og Bruus Pedersen 2000) og indikerer, at agerlandets fugle vil kunne nyde godt af en sådan ændring i dyrkningspraksis, idet fuglene især i yngleperioden er afhængige af insekter som føde. I nærværende undersøgelse fandt vi, at tidspunktet for første Roundup-sprøjtning er afgørende for forskellen mellem de to typer roer. Hvis man følger den behandlingsstrategi for Roundup Ready, som anbefales (Monsanto Europe 1999), er ukrudtsfloraen mere talrig og diversiteten højere sammenlignet med ukrudtsfloraen i roer behandlet med konventionelle roeherbicider. Roer behandlet med Roundup Ready efter anbefalingerne blev i 2001 sprøjtet første gang 35 dage senere end de konventionelt dyrkede roer. Langt de største forbedringer for ukrudt og leddyrfauna blev dog fundet i roer sprøjtet første gang 48 dage senere end de konventionelt dyrkede roer og vel at mærke uden reduktion i roeudbyttet. Ukrudtsbiomassen såvel som tætheden af leddyr var 10 gange højere i felter med sen Roundup-behandling sammenlignet med felter behandlet efter anbefalingen. Den umiddelbare følge er, at der er ukrudt og dermed føde for herbivorerne i længere tid. Desuden er jorden dækket af planter (roer og ukrudt) gennem hele forsommeren, hvilket forbedrer mikroklimaet væsentligt til gavn for leddyrfaunaen. Hvis første Roundup-sprøjtning derimod foretages tidligere end anbefalet, opnås ingen forbedringer for flora og fauna. Ukrudtsbekæmpelsen i disse roer var så effektiv, at kun meget få og små ukrudtsplanter blev fundet gennem hele sæsonen. Driftslederens motivation for gennem juni måned at acceptere ukrudt i afgrøden under den økonomiske tærskelværdi er derfor afgørende for den positive effekt, dyrkningen af glyphosat-tolerante foderroer kan have på biodiversiteten i marken.

Vi så, at også problematiske ukrudtsarter som Liden nælde og flere pileurt-arter kunne kontrolleres selv med den sene Roundup-sprøjtning. De store planter døde ikke umiddelbart ved første sprøjtning, men deres vækst blev sat i stå, og sammen med den stigende konkurrence fra afgrøden og en efterfølgende anden behandling bevirkede det, at planterne forsvandt hen over sommeren.

Uanset tidspunktet for Roundup-behandlingen skete der ingen eller meget begrænset rekruttering af nye ukrudtsplanter. Alle Roundup-behandlede parceller havde en meget begrænset ukrudtsflora fra midten af juli og ind til høst, og der blev ikke produceret ukrudtsfrø i disse parceller. I konventionelt behandlede parceller skete der i modsætning hertil en løbende etablering af ukrudtsplanter, og fra slutningen af juni og indtil høst var ukrudtsfloraen væsentlig rigere i disse parceller. Flere ukrudtsarter, f.eks. Lugtløs kamille, Hvidmelet gåsefod og Snerle-pileurt, var i stand til at klare sig i konkurrencen med roerne. De var meget store og producerede mange frø.

Formål

Det er tidligere vist, at der med dyrkningen af glyphosat tolerante foderroer er en mulighed for en væsentligt rigere ukrudtsflora og leddyrfauna i marken, og at sprøjtepraksis sandsynligvis var afgørende for de fundne forbedringer. Formålet med forsøget har derfor været at undersøge sammenhængen mellem sprøjtepraksis og biodiversitetsforandringer i foderroemarker gennem hele vækstsæsonen.

Metoder

Forsøget blev gennemført på en mark på Mors. Denne lokalitet blev valgt, da både driftsledere og landbrugskonsulenter havde erfaringer med forsøgsarbejdet med glyphosat-tolerante foderroer fra tidligere års demonstrationsforsøg. Undersøgelsen var en kontrolleret undersøgelse af betydningen af sprøjtemiddel og sprøjtetidspunkt med 4 gentagelser af hver af følgende fire ukrudtsbehandlinger: 1) behandling med konventionelle roe-herbicider, 2) behandling med Roundup Ready (RR) som anbefalet i brugsanvisningen, 3) behandling med RR tidligere på sæsonen end anbefalet, og 4) behandling med RR så sent, som driftslederen fandt forsvarligt for opretholdelsen af udbyttet. Alle behandlinger blev gennemført i parceller med den gensplejsede foderroevarietet Simplex.

Ukrudtssammensætning, tæthed og biomasse blev undersøgt fem gange fordelt over sæsonen, og leddyrfaunaen, der er betydelig mere ressourcekrævende både ved indsamling og identifikation, blev indsamlet tre gange. I oktober blev roerne høstet, og rodudbyttet bestemt.

Resultater

Kun i parceller, der var behandlet med Roundup Ready efter anbefalingerne (Monsanto Europe 1999) eller senere, var der gennemsnitlig flere ukrudtsarter, højere plante tæthed og større ukrudtsbiomasse frem til midten af juli end i parceller behandlet med konventionelle herbicider (Figur 3.1) og kun i disse felter blev der fundet væsentligt bedre forhold for leddyrfaunaen, dvs. flere arter og større individtæthed (Figur 3.3 og 3.4).

Fra begyndelsen af juli og resten af perioden var plantetætheden lav for alle behandlinger, men i de konventionelt behandlede parceller blev ukrudtet stort og bidrog til en betydelig biomasse, navnlig ved målingen i august og kun i konventionelt behandlede parceller blev der produceret ukrudtsfrø. Antallet af leddyr steg i juli sammenlignet med juni (Figur 3.3), formentlig fordi leddyrenes udvikling er forsinket i forhold til floraen, selvom der for nogle arter, både herbivore og ikke-herbivore, var en sammenhæng mellem ukrudtsbiomassen og antallet af dyr. Der vedblev at være flere leddyr i de sent behandlede RR-parceller end i de konventionelt dyrkede parceller, hvorimod antallet af dyr i de øvrige RR-parceller var lavere eller på størrelse med antallet i de konventionelt dyrkede parceller i juli. Leddyrenes artsantal fulgte samme udvikling (Figur 3.4). Det gennemsnitlige rodudbytte adskilte sig ikke statistisk mellem behandlinger (Figur 3.2). Selv når første Roundup-behandling lå i slutningen af juni, var rodudbyttet ikke reduceret.

1 Background and aims

Generally, an effective weed control is presumed a prerequisite for a high yield in beet fields. Weeds in conventional beet fields are controlled by use of a mixture of herbicides that usually includes the active ingredients phenmedipham, ethofumesate and metamitron (Jensen et al. 2001). Because of the sensitivity of beets to the conventional herbicides, they can only be used until the crop starts to develop true leaves. Moreover, conventional beet herbicides are only effective when the weeds are at the cotyledon stage. Roundup, on the contrary, is highly effective even when applied to later growth stages (Kudsk and Mathiassen 1998, Bückmann *et al.* 2000). The development of a glyphosate tolerant beet gives the farmer the opportunity of a more optimal herbicide strategy.

Comparative studies of conventional and genetically modified glyphosate tolerant fodder beet have revealed that implementation of glyphosate tolerant fodder beets may increase biodiversity in beet fields in early summer (Bruus Pedersen and Strandberg 2000, Elmegaard and Bruus Pedersen 2001). Both studies found higher weed density, diversity and biomass in plots with glyphosate tolerant fodder beet than in conventional fodder beet plots. Data on arthropods showed the same patterns, i.e. an increase in species diversity and higher density in plots with glyphosate tolerant fodder beet, although the response was less clear. Farm management was crucial to the potential environmental benefit of growing glyphosate tolerant fodder beets. Delayed weed control in particular resulted in higher weed and arthropod diversity, whereas use of insecticides counteracted the benefits (Bruus Pedersen and Strandberg 2000).

Whereas the previous studies aimed at a comparison of the effects of conventional and genetically modified fodder beet management strategies on biodiversity, the focus of the present study was on the relationship between the timing of the herbicide (conventional beet herbicides and Roundup Ready) application and biodiversity. To study the potential for biodiversity improvements, four different weed control strategies were tested: early application (conventional beet herbicides or Roundup Ready), Roundup Ready application according to label recommendation, and late Roundup Ready application. However, it was important for the relevance of the study that the delay in herbicide application did not result in a yield reduction of the fodder beet.

2 Methods

The experiment took place at a field on the island Mors in Limfjorden. This site has been included in former years of biodiversity monitoring in fields with fodder beets (Bruus Pedersen and Strandberg 2000, Elmegaard and Bruus Pedersen 2001). The area selected for the experiment has been managed as one field unit during the last 10 years and the previous crop was spring-barley.

2.1 Experimental setup

The experiment was set up as a split block design with 4 replicates of each of the following herbicide treatments (conventional beet herbicides or the glyphosate product Roundup Ready):

Treatment A: application of conventional beet herbicides, 3 applications.

Treatment B: 2 applications of Roundup Ready (RR) following label recommendation (Monsanto Europe 1999).

Treatment C: 2 applications of RR, first application early compared to treatment B.

Treatment D: 2 applications of RR, first application late compared to treatment B.

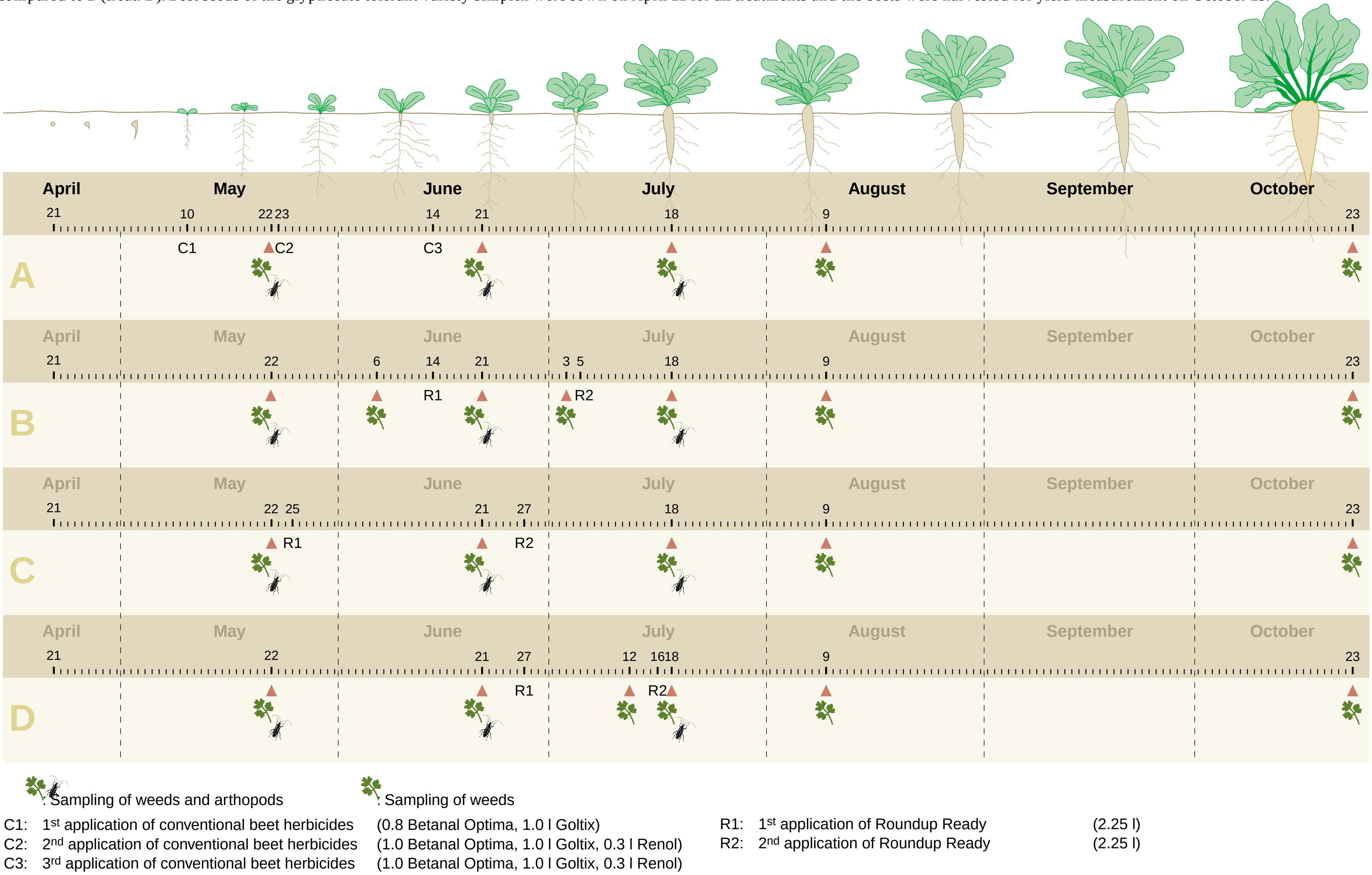
The glyphosate tolerant fodder beet of the cultivar Simplex was used for the entire experiment, also where conventional beet herbicides were used for weed control. The seeds were sown on the 21 of April 2001. Information on herbicides, dosages, and application time is given in Figure 2.1.

2.2 Biological samplings

The idea behind the biological sampling scheme was to measure the effects of the different herbicide treatments on weeds and arthropods over the crop season and, therefore, the sampling dates were determined by the spraying schemes for each of the four different treatments. Ideally, the samples should be taken before and after every herbicide application and a couple of times later in the season. Moreover, common sampling dates for all treatments would optimise the data analysis and interpretation. However, the available resources made no more than three arthropod samplings for each treatment possible. Therefore, sampling during the spraying period followed the application schemes for each of the four treatments. However, three common sampling dates for all treatments became possible (May 22, June 21 and July 18). The sampling on July 18 was undertaken succeeding the spraying period (Figure 2.1).

Each experimental plot was 20 m x 20 m of which the central 18 m x 18 m was used for the biological sampling. The plots were surrounded by a 5 m wide buffer zone treated with conventional beet herbicides.

Figure 2.1 Calendar showing the project activities (herbicide applications, biological samplings) on the four herbicide treatments viz. application of conventional beet herbicides (treat. A), application of Roundup Ready following standard label recommendation (treat. B), application of Roundup Ready, first application early compared to B (treat. C), and application of Roundup Ready, first application late compared to B (treat. D). Beet seeds of the glyphosate tolerant variety Simplex were sown on April 21 for all treatments and the beets were harvested for yield measurement on October 23.



Weeds were sampled within 10 randomly chosen but non-overlapping squares (0.75 m x 0.75 m). At each sampling, the weeds rooted within the squares were identified to species or genus and the number of plants was counted for each species or genus separately. Subsequently, the above ground biomass was harvested. Total biomass per square was recorded for the sampling on May 22, and June 6, whereas biomass was sampled for each species separately on June 21, July 3, July 18, and August 9. Weed density at beet harvest was estimated visually. The production of weed seeds was checked at each sampling and harvested separately for each of the seed producing weeds when the seeds were ripe. All samples were dried at 80°C for 24 hours, and dry weight determined. Nomenclature follows Hansen (1991).

Within each plot 10 arthropod samplings consisting of 10 suctions lasting 10 seconds each were collected by a Dietrick Vacuum Sampler (D-vac) (Dietrick 1961). The suctions were alternately carried out in the beet rows and between rows. The samples were stored at -18°C until identification. In order to optimise the resources the level for identification (species, genus, family or order) depended on both importance of the species as food for arable birds and taxonomic difficulties.

2.3 Data analysis

The collected data were tested for normality, and since data on weeds and arthropods were found not to fulfil the criterion, effects of herbicide treatment and block position on number of weed species, weed density and biomass as well as arthropod abundance were analysed by the non-parametric Kruskal-Wallis test. Effects of herbicide treatment on beet yield were tested by analysis of variance, and means were compared using a t-test. All tests were evaluated at the 5 % level.

2.4 Crop yield

On 23rd October beet roots for yield estimation were harvested by a two-row beet digger for agricultural trials that collected and weighed the roots. Within each plot beets from 48 m^2 were harvested.

2.5 Risk management

The field trials were approved according to § 9 Article 1 Law No 356 of June 6 1991 Lov om miljø og genteknologi (Ministry of Environment and Energy, 1999). Weekly the field was checked for bolters that had to be removed. Supervision of the field trials has been performed by the County of Viborg according to § 20, Law No. 356 of June 6 1991. The supervision performed by the County did not raise any comment (Viborg Amt 2002). The RR beets have not yet been allowed for marketing in Denmark and EU and therefore the fodder beets could not be used for feeding cattle and were cut and mulched on the soil to be destroyed by frost during winter time, except for the samples taken for yield measurements that were destroyed afterwards.

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3 Results

3.1 Weeds

Generally, the weed flora developed differently when subjected to conventional herbicide application (Treatment A) and the three Roundup Ready (RR) treatments (Treatment B, C and D) (Figure 3.1a, b, c). The conventional treated plots had a poor weed flora early in the season as a consequence of the early herbicide applications, but new plants were recruited succeeding the applications and they grew big later in the season. Some species produced plenty of seeds. In contrast, the weed flora in the RR treated plots peaked before the applications and decreased in all measured parameters (diversity, density and biomass) after the RR applications, i.e. there was little or no recruitment after the applications.

Only very few and inconsistent effects of block number on plant parameters were identified, and these will not be described any further.

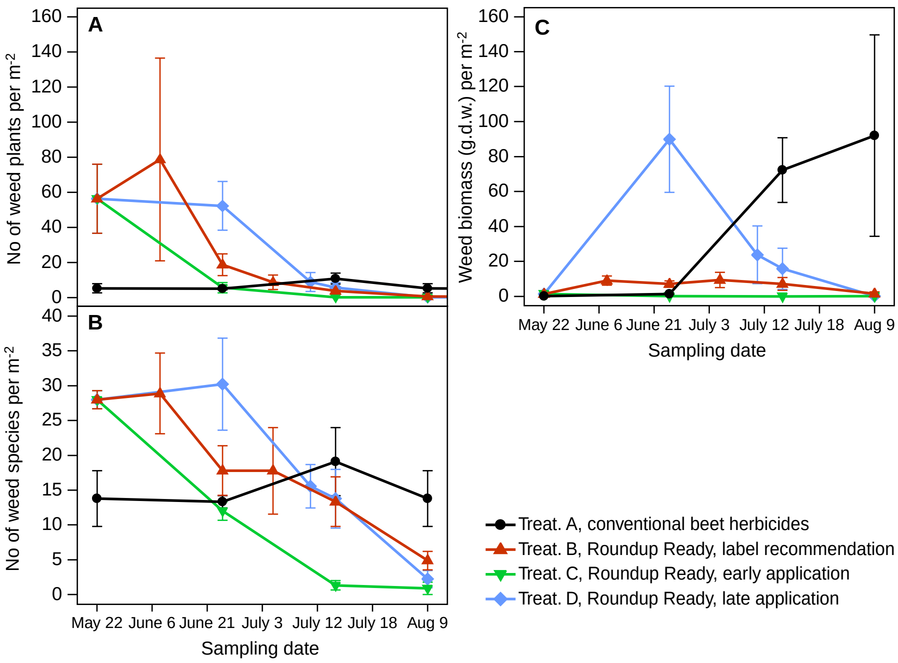

Figure 3.1. Weed flora measured as (A) weed density (No of weed plants per m^2), (B) weed diversity (No of weed species per m^2), and (C) weed biomass (g d. w. per m^2) for the four different herbicide regimes, viz. application of conventional beet herbicides (Treat. A), application of Roundup Ready following label recommendation (Treat. B), application of Roundup Ready, first application early compared to B (Treat. C), and application of Roundup Ready, first application late compared to B (Treat. D). Information on sampling dates and herbicides applications is given in Table 2.1. All data shown are means ± S.D. Note that data on weed density at harvest is not shown in figure A. Data is given in chapter 3.1.1.

3.1.1 Weed density

For all Roundup Ready-treated plots (Treatment B, C and D) the average weed density was highest before the first RR application and decreased rapidly after the application (Figure 3.1a). This decrease continued over the summer as a consequence of the second RR application, the increasing competition from the beet plants, and the lack of recruitment of new plants. Independent of application time few plants were left in the field, and the differences between RR treatments on August 9 were small (Table 3.1). This pattern persisted for the rest of the season (data not shown on Figure 3.1a) and at harvest the weed densities were 0.2 ± 0.002 plants / m^2, 0.05 ± 0.001 plants / m^2, and 0.1 ± 0.002 plants / m^2 for the treatments B, C, and D, respectively. The plots that received a first application early in the season (Treatment C) had lower densities than the other RR treated plots throughout the sampling period (Figure 3.1, Table 3.1).

Contrary to this, plots treated with conventional beet herbicides had few weed plants throughout the season. No pre-application measurements were taken within this treatment (see, Figure 2.1); however, the weed density before the first application (May 10) presumably was higher than on May 22, although not as high as measured for the B, C and D treatments. Some recruitment following the applications was observed, and the density was 5.3 ± 1.5 and 4.4 ± 1.5 plants / m^2 at the samplings August 9 and October 23, respectively, i.e. a factor 10 higher than in any RR treated plots ($p<0.0001$). Although relatively few, these plants grew big and contributed to a significant weed biomass at the end of the season (Figure 3.1 and Chapter 3.1.3.).

Table 3.1 Outcome of the nonparametric Kruskal-Wallis test of effects on the weed flora measurerd as density (no. of plants per m2), diversity (no. of species per m2), and aboveground biomass (g dw per m2)of the 4 different herbicide treatments, viz. 3 applications of conventional beet herbicides (Treat. A), 2 applications of Roundup Ready following standard label recommendation (Treat. B), 2 applications of Roundup Ready, first application early compared to B (Treat. C), and 2 applications of Roundup Ready, first application late compared to B (Treat. D). Significance is given as p-value, and the means are shown for each of the four treatments.

Sampling date	p-value	Mean density (no of plants per m^2)			
		Treatment A	Treatment B	Treatment C	Treatment D
May 22	<0.0001	5.3		56.4	
June 21	<0.0001	0.5	18.8	5.7	52.3
July 18	<0.0001	10.8	3.8	0.2	5.6
August 9	<0.0001	5.3	0.6	0.09	0.2
		Mean diversity (no. of species per m^2)			
May 22	<0.0001	13.8		28	
June 21	<0.0001	13.3	17.8	12	30
July 18	<0.0001	19.1	13.3	1.3	13.8
August 9	<0.0001	13.8	4.9	0.9	2.2
		Mean aboveground biomass (g dw per m^2)			
May 22	<0.0001	0.1		1.3	
June 21	<0.0001	1.3	7.1	0.2	89.9
July 18	<0.0001	72.2	7.1	0.009	15.8
August 9	<0.0001	92.0	1.6	0.05	0.2

3.1.2 Diversity and dominance

The development in weed diversity given as number of weed species per sample followed the same pattern as described for weed density (Figure 3.1b, Table 3.2). The average number of species per sample was highest before the first herbicide application for all RR treated plots (Treatment B, C and D) and decreased continuously for the rest of the sampling period. The picture for the plots treated with conventional beet herbicides was somewhat different. No pre-application data are available, but presumably the number of species was higher before the first application (May 10) than found at the first sampling (May 22). After the applications, a recruitment of new species was seen in June and the largest number of species was recorded in mid-July. At that time the weed diversity was higher in the conventionally treated plots than in RR treated plots and this difference persisted for the rest of the sampling period (Table 3.1).

Poa annua, Chenopodium album, Veronica arvensis, Lamium spp., *Tripleurospermum inodorum*, and *Polygonum spp.* were the most common weeds. The three most abundant species accounted for more than 50% of the total weed number (Table 3.3). Grasses were most abundant early in the season. In plots with application of Roundup Ready *Poa annua* was the most abundant plant whereas *Lolium multiflorum* dominated in plots with application of conventional herbicides. Following the RR applications some weeds e.g. *Lamium purpureum, Trifolium repens* and *Polygonum* spp. became more abundant and accounted for a larger part of the total weed number. Following application of RR *Poa annua* was only found in significant numbers in the plots with late application of RR (Treatment D), and most individuals were found underneath the beet plants where they to some extent were protected from herbicide application. Only few species tolerated the early application of RR (Treatment C) and little or no recruitment took place over the season in these plots. Therefore, the weed cover was extremely sparse (0.2 plant per m^2) from early July onwards. *Trifolium repens, Veronica arvensis, Lamium amplexicaule* and *Euphorbia peplus* were the only species found and the individuals were small and depauparate.

Table 3.2. List of all the weed species occurring within the plots before and during the period of herbicide applications and succeeding the applications for the four different herbicide treatments viz. 3 applications of conventional beet herbicides (Treat. A), 2 applications of Roundup Ready following standard label recommendation (Treat. B), 2 applications of Roundup Ready, first application early compared to B (Treat. C), and 2 applications of Roundup Ready, first application late compared to B (Treat. D). For treatment A the first weed sampling was performed immediately after the first herbicide application.

	Treatment A		Treatment B		Treatment C		Treatment D	
	during appl.	after	before and during appl.	after	before and during appl.	after	before and during appl.	after
Anagalis arvensis			+		+		+	
Brassica napus		+					+	
Capsella bursa-pastoris		+	+	+	+		+	+
Cerastium sp.			+		+		+	
Chamomilla suaveolens	+	+	+				+	
Chenopodium album	+	+	+	+	+		+	+
Cirsium arvense	+		+					
Crataegus sp.							+	
Elytrigia repens	+	+	+	+	+		+	+
Euphorbia peplus	+	+	+		+	+	+	
Fumaria officinalis			+	+	+		+	
Galeopsis tetrahit							+	
Galinsoga quadriradiata				+			+	
Galium aparine		+	+	+			+	
Gnaphalium uliginosum			+					
Hordeum vulgare	+	+						
Juncus tenuis			+		+		+	
Lamium sp	+							
Lamium amplexicaule	+	+	+	+		+	+	+
Lamium purpureum		+	+	+			+	+
Lolium multiflorum		+						
Lolium perenne	+	+	+					
Myosotis arvensis			+					
Plantago major		+	+				+	
Poa annua	+	+	+	+	+		+	+
Poa pratensis	+		+					
Polygonum aviculare	+	+	+	+	+		+	+
Polygonum convolvulus	+	+	+	+	+		+	+
Polygonum persicaria		+	+				+	+
Ranunculus sp.			+					
Rumex acetosa			+					
Sinapis arvensis			+		+		+	
Solanum nigrum			+		+		+	
Sonchus sp.			+		+		+	
Spergula arvensis			+		+		+	
Stellaria media	+	+	+		+		+	+
Thlaspi arvense			+				+	
Trifolium repens			+	+	+	+	+	+
Tripleurospermum inodorum	+	+	+	+	+		+	+
Urtica urens	+	+	+	+	+		+	
Veronica arvensis	+	+	+	+	+	+	+	+
Viola arvensis	+	+	+	+	+		+	+
Total number of species	17	22	35	16	21	4	32	14

Table 3.3. The three weed species per herbicide treatments occurring at highest densities and biomass, respectively. The four herbicide regimes are: Treatment A) application of conventional beet herbicides, Treatment B) application of Roundup Ready following standard label recommendation, Treatment C) application of Roundup Ready, first application early compared to B, and Treatment D) application of Roundup Ready, first application late compared to B. The contribution (%) of the species to the total no. of plants and total biomass, respectively, is indicated.

		Density		Biomass	
Date	Treatment	Species	Dominance	Species	Dominance
June 21	A	*Lolium perenne*	42	*Lolium perenne*	93
		Chenopodium album	12	*Veronica arvensis*	3
		Poa annua	11	*Chenopodium album*	1
	B	*Poa annua*	44	*Capsella bursa-pastoris*	45
		Capsella bursa-pastoris	13	*Poa annua*	20
		Chenopodium album	6	*Veronica arvensis*	15
	C	*Poa annua*	70	*Poa annua*	35
		Chenopodium album	7	*Chenopodium album*	21
		Urtica urens	5	*Euphorbia peplus*	21
	D	*Poa annua*	56	*Chenopodium album*	30
		Chenopodium album	10	*Capsella bursa-pastoris*	14
		Veronica arvensis	5	*Tripleurospermum inodorum*	13
July 18	A	*Poa annua*	22	*Tripleurospermum inodorum*	48
		Lolium multiflorum	15	*Lolium multiflorum*	16
		Triplerospermum inodorum	15	*Chenopodium album*	13
	B	*Capsella bursa-pastoris*	27	*Capsella bursa-pastoris*	25
		Lamium purpureum	14	*Lamium purpureum*	25
		Veronica arvensis	11	*Urtica urens*	25
	C	*Trifoilum repens*	86	*Trifoilum repens*	98
		Veronica arvensis	14	*Veronica arvensis*	2
	D	*Poa annua*	53	*Urtica urens*	30
		Veronica arvensis	7	*Triplerospermum inodorum*	22
		Polygonum convolvulus	6	*Lamium purpureum*	13
		Trifolium repens	6		
August 9	A	*Tripleurospermum inodorum*	20	*Tripleurospermum inodorum*	42
		Polygonum aviculare	18	*Chenopodium album*	23
		Chenopodium album	17	*Lolium multiflorum*	20
	B	*Capsella bursa-pastoris*	38	*Urtica urens*	54
		Urtica urens	15	*Veronica arvensis*	24
		Viola arvensis	15	*Capsella bursa-pastoris*	16
	C	*Euphobia peplus*	50	*Lamium amplexicaule*	98
		Lamium amplexicaule	50	*Euphobia peplus*	2
	D	5 species, all found one time		*Polygonum convolvulus*	28
				Lamium amplexicaule	25
				Anagalis arvensis	20

3.1.3 Weed biomass

Although data on weed biomass showed the same pattern as described for density and diversity, some obvious differences were also found (Tables 3.3. and 3.4). Throughout the sampling period, the weed biomass in the plots with early Roundup application (Treatment C) was very low and did not differ from plots with application of conventional beet herbicides (Treatment A) until end of June. Plots treated with RR following label recommendation (Treatment B) or later (Treatment D) had a higher weed biomass during the early summer period than plots treated with either conventional beet herbicides or with early RR application (Fig 3.1c, Table 3.1). In the present experiment the first application of the late Roundup treatment (Treatment D) was delayed by 48 days compared to the conventional application. Weed biomass decreased following the first applications of RR to Treatment B and most rapidly for Treatment D and from mid-July and the rest of the sampling period, weed biomass did not differ between the different RR treatments. *Capsella bursa-pastoris, Urtica urens,* and *Lamium spp.* accounted for most of the biomass in RR treated plots (Table 3.3).

The weed biomass in plots treated with conventional beet herbicides increases rapidly from the end of June. Weeds were not numerous, although more plants were found than in Roundup treated plots (Table 3.1), but they grew big. Among the about 10 species recorded in conventionally treated plots, some species accounted for a significant amount of the biomass. *Tripleurospermum inodorum* accounted for more than 40% of the total biomas and the species *Chenopodium album, Lolium multiflorum, Veronica arvensis* and *Chamomilla suaveolens* also had a significant biomass (Table 3.3).

3.1.3.1 Production of weed seeds

The production of weed seeds started early in the season. Species like *Poa annua* and *Viola arvensis* set seeds from mid-June and on. In RR-plots most of these seeds, however, never ripened as the plants died after the herbicide applications and consequently the seeds were not harvested for biomass determination. In the present experiments ripened seeds were only produced within plots treated with conventional beet herbicides. The few weed plants found underneath the beet plants within the RR treated plots were small and did not produce seeds. *Poa annua* and *Viola arvensis* had ripe seeds at the sampling on July 18. In plots treated by conventional herbicides these species, however, were not very abundant and the seeds accounted for less than 1% of the biomass. Later in the season the tall-growing weeds like *Tripleurospermum inodorum* and *Chenopodium album* and climbing species like *Polygonum convolvulus* escaped the closing beet cover and succeeded to produce plenty of seeds. These species contributed to the largest part among the eight seed producing species (*Chamomilla suaveolens, Chenopodium album, Hordeum vulgare, Lolium multiflorum, Polygonum convolvulus, Tripleurospermum inodorum, Veronica arvensis* and *Viola arvensis*) in conventional plots. At the sampling on August 9, weed seeds accounted for 10-20% of the total biomass.

Table 3.4. Average aboveground weed biomass (g dw per sample) on July 18 in the four blocks of each of the four herbicide treatments.

	A, conventional herbicides				B, Roundup, label recommend.				C, Roundup, early application				D, Roundup, late application			
Block	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Brassica napus			0.90													
Capsella bursa-pastoris	0.97		1.28	0.31	0.27	0.68	1.37	1.72					0.35			
Chamomilla suaveolens	1.40	0.52	4.71													
Chenopodium album	14.47	5.69	0.64			0.32									0.19	
Elytrigia repens					0.48								0.02	0.25	0.04	
Euphorbia peplus						0.02										
Galinsoga quadriradiata						0.001										
Galium aparine		0.64				0.38		0.25								
Hordeum vulgare	0.33			2.19												
Lamium amplexicaule		0.05			0.04	0.10							0.24	0.01		
Lamium purpureum	0.67	0.19		0.01	0.02	1.67	1.28	0.908					1.04	1.64	1.74	0.37
Lolium multiflorum	4.70	5.27	2.61	13.90												
Lolium perenne	1.37															
Plantago major			0.11													
Poa annua	0.70	0.20	0.28		0.02								0.08	2.52	1.64	0.06
Poa pratensis																
Polygonum aviculare	1.23	0.13	0.87		0.01	0.25								0.10	0.26	
Polygonum convolvulus	0.69	2.28		0.46		0.06		0.07					0.51	1.11	1.12	
Polygonum persicaria	0.06														0.91	
Stellaria media	0.71														0.13	
Thlaspi arvense																
Trifolium repens							0.03	0.03			0.001	0.02			0.18	0.29
Tripleurospermum inodorum	7.24	16.73	15.59	38.37				0.41					1.01		6.81	
Urtica urens	2.85					4.03								10.38		
Veronica arvensis	9.66	1.61	0.21		0.02	0.60	0.89		0.003				0.56	1.56	0.15	
Viola arvensis	0.10				0.17										0.20	
Average biomass per sample	46.8	33.31	27.19	55.23	1.03	8.1	3.56	3.39	0.003		0.001	0.02	3.81	17.57	13.36	0.72
Total no of weed species	16	11	9	7	8	11	4	7	1	0	1	1	8	8	12	3

3.2 Fodder beet yield

No significant differences in root yield were found among treatments (Figure 3.2) although mean root biomass tended to be higher in plots that have received early application of Roundup Ready (Treatment C). The plots that had the first Roundup application delayed by 48 days relative to the conventional treatment (Treatment D) had a root yield comparable to the ones that received conventional beet herbicides (Treatment A).

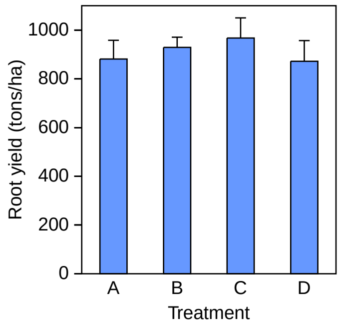

Figure 3.2. Root yield (means ± S.D) of fodder beet of the transgenic, glyphosate tolerant variety Simplex measured on October 23, 2001 for the four different herbicide treatments: Treatment A) application of conventional beet herbicides, Treatment B) application of Roundup Ready following label recommendation, Treatment C) application of Roundup Ready, first application early compared to B, and Treatment D) application of Roundup Ready, first application late compared to B. Information on herbicides used and applications is given in Table 2.1.

3.3 Arthropods

3.3.1 Abundance

For all groups and species of which more than just a few specimens were found, frequencies in the plots representing the different treatments were compared for the dates May 22, June 21 and July 18.

On May 22, before any Roundup treatment took place, only few arthropods were found in the samples (149 in 30 samples, aphids and thrips not included). Diptera (Brachycera / Cyclorrhapha and Nematocera) and Staphylinidae predominated, and there were no significant effects of treatment (plots treated with conventional herbicides versus the rest, i.e. the plots that were to be treated with Roundup Ready, Figure 3.3).

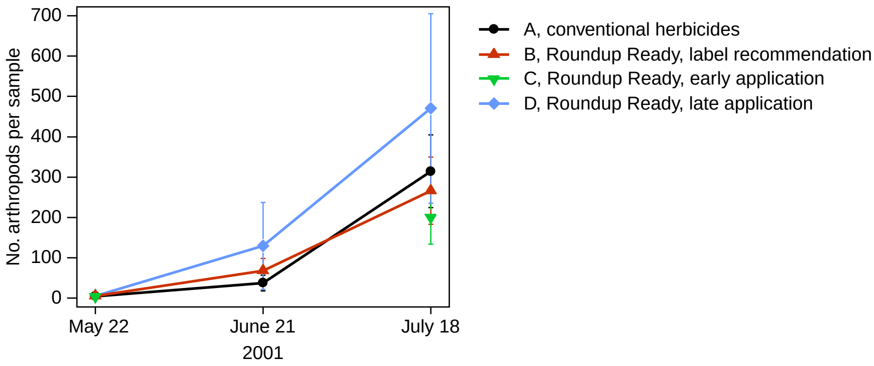

Figure 3.3. Total numbers of arthropods (means and SD) per sample collected on three dates in fodder beets treated with different herbicide regimes. Treatment C was not sampled on June 21.

The samples collected on June 21 contained more arthropods, 8715 animals in 110 samples. Most of these were Diptera, but also Apocrita, Staphylinidae and Aleocharinae were found in considerable numbers. For most groups of arthropods there were more animals in the plots treated with RR late in the season (Treatment D) and fewer in the plots treated with conventional herbicides (Table 3.5, Figure 3.3), but for some groups (Carabidae, Aleocharinae, Staphylinidae as a whole and Curculionidae) abundance was higher in plots treated with RR according to the label recommendation (Treatment B). For several groups there was a block effect, meaning that arthropod abundance differed between the different plots of identical treatment. However, there was no clear trend in this interactive effect.

In the samples collected on July 18, more than 50,000 arthropods were counted in 160 samples. The dominant groups were Diptera, Linyphiidae, Apocrita, Aleocharinae, Staphylinidae and Clavicornia. Wherever significant effects of treatment were identified, more animals were found in the plots treated late with Roundup Ready than in the other plots (Figure 3.3, Table 3.6), except for the pollen beetle *Meligethes aeneus*. There were no consistent differences in arthropod fauna between plots treated with Roundup Ready earlier than the "Treatment D" plots and plots treated with conventional herbicides, even though for most groups means were generally lower in the plots treated with conventional herbicides. For the samples collected on July 18 there were also several cases of block effects, but without clear trends.

Average total arthropod abundance was related to average weed biomass with an R^S of 0.68 (Table 3.7) for the linear relationship for all sampling dates together. Arthropod abundance was only slightly (negatively) related to average weed density, with an R^S of 0.15. Relations between arthropods and weed biomass was fairly well described by linear equations for adult Delphacidae and Crysomelidae, but the relationship was generally better described by a quadratic equation, $ax^2 + bx + c$, with a negative value for "a" (Table 3.7), indicating an optimum arthropod abundance at intermediate weed biomass values.

3.3.2 Diversity

Diversity of arthropods measured as number of species or groups per sample followed the pattern described for abundance (Figure 3.4), with a higher diversity in the plots treated late with Roundup Ready. There were only small differences between the other RR-treated plots and the conventionally treated plots, with the trend that diversity was lower in the conventionally treated plots.

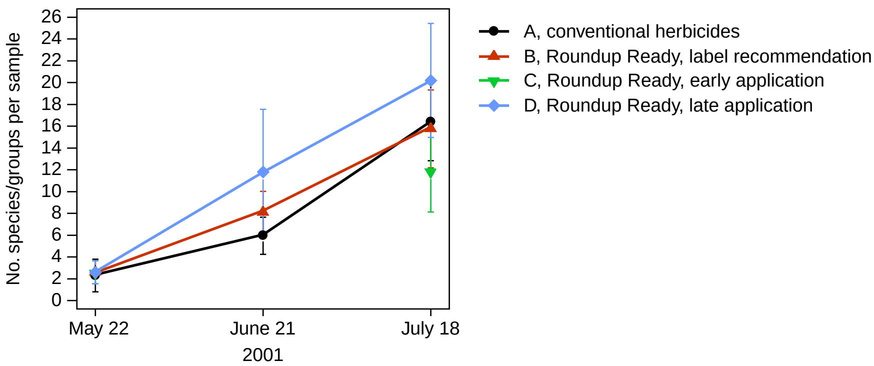

Figure 3.4. Number of arthropod species or groups (means and SD) per sample collected on three dates in fodder beets treated with different herbicide regimes. Treatment C was not sampled on June 21.

Table 3.5. Mean abundance of selected arthropod species and groups (no. per sample of 0.9 m^2) collected in the fodder beet field on June 21. In the "treatment" column p-values from the Kruskal-Wallis test are presented. A = conventional herbicide treatment, B = Roundup treatment according to label recommendations, D = late Roundup treatment. Treatment C, early Roundup treatment, was not sampled on this date.

Order	Group / species	Treatment	A	B	D
Araneae	Linyphiidae	0.01	0.6	1.5	1.6
Diptera	Brachycera / Cyclorrhapha	<0.0001	2.7 c	14.3	43.5
	Nematocera	0.0009	26.6	38.2	62.7
Lepidoptera	Lepidoptera larvae	too few to analyse			
Hymenoptera	Apocrita	<0.0001	2.0 c	4.6	12.9
	Symphyta larvae	0.09	0	0.03	0.2
Hemiptera	Delphacidae	<0.0001	0	0.1	0.7
	D. nymphs	too few to analyse			
	Cicadellidae	0.04	0	0.03	0.1
	C. nymphs	too few to analyse			
	Heteroptera	0.2	0	0	0.05
Coleoptera	*Agonom dorsale*	0.4	0	0	0.03
	Carabidae larvae	too few to analyse			
	Carabidae, total	0.07	1.2	1.3	0.7
	Aleocharinae	<0.0001	2.8	5.1	2.1
	Staphylinidae, total	<0.0001	3.3	6.2	3.4
	Crysomelidae	0.02	0	0.03	0.2
	Curculionidae	0.002	0.1	0.6	0.5
	Coccinelidae larvae	too few to analyse			
	Meligethes aeneus	<0.0001	0	0.03	1.7
	Lathridius sp.	0.03	0	0	0.1
	Corticaria sp.	too few to analyse			
	Atomaria sp.	0.04	0	0.2	0.2
	Clavicornia*, total	0.005	0.2	0.2	2.0

*Here used as a collective term for Coccinellidae, Nitidulidae, Lathridiidae, Phalacridae, Cryptophagidae and Byburidae.

Table 3.6. Mean abundance of selected arthropod species and groups (no. per sample of 0.9 m^2) collected in the fodder beet field on July 18. In the "treatment" column p-values from the Kruskal-Wallis test are presented. A = conventional herbicide treatment, B = Roundup treatment according to label recommendations, C = early Roundup treatment, D = late Roundup treatment.

Order	Group / species	Treatment	A	B	C	D
Araneae	Linyphiidae	0.005	27.2	27.6	22.8	32.9
Diptera	Brachycera / Cyclorrhapha	<0.0001	189	162c	131 c	280
	Nematocera	<0.0001	16.0c	19.3	7.2 c	35.2
Lepidoptera	Lepidoptera larvae	0.004	6.4	4.5c	3.4 c	8.3
Hymenoptera	Symphyta larvae	<0.003	0.2	0.2	0.05	0.6
	Apocrita	<0.0001	21.1	14.0	10.0	26.4
Hemiptera	Delphacidae	0.3	0	0.03	0.03	0.08
	D. nymphs	0.6	0	0.1	0	0.03
	Cicadellidae	0.4	1.2	0.7	1.0	1.3
	C. nymphs	0.1	0	0	0	0.1
	Heteroptera	0.001	0.8	1.3	0.2	0.8
Coleoptera	*Agonom dorsale*	0.003	0.1	0.2	0.1	0.5
	Carabidae larvae	0.3	0.05	0.3	0.03	0.5
	Carabidae, adult total	0.03	3.3	3.1	3.1	5.6
	Aleocharinae	0.03	16.0	11.8	15.8	19.2
	Staphylinidae, total	<0.0001	20.6	18.9	18.3	41.6
	Crysomelidae	0.04	0.4	0.03	0.03	0.2
	Curculionidae	0.0003	1.2	1.9	0	1.1
	Coccinelidae larvae	<0.0001	1.2	0.3	0.05	1.8
	Meligethes aeneus	<0.0001	15.1	0.7	0.3	1.5
	Lathridius sp.	<0.0001	0.4	1.0	0.2	5.2
	Corticaria sp.	<0.0001	0.03	0.6	0	3.1
	Atomaria sp.	<0.0001	6.4	5.5	2.4	17.6
	Clavicornia*, total	<0.0001	23.3	8.3	3.0	29.7

*Here used as a collective term for Coccinellidae, Nitidulidae, Lathridiidae, Phalacridae, Cryptophagidae and Bybturidae.

Table 3.7. Relations between abundance of selected arthropods and weed biomass. R^2 of linear fits and fits of quadratic equations, $ax^2 + bx + c$, are given. "a" of the quadratic equation is negative, except for the groups marked with an asterix.

Group / species	R^2 weed biomass (linear)	R^2 weed biomass (quadratic)
Apocrita	0.26	0.72
Linyphiidae	0.0076	0.55
Brachycera / Cyclorrhapha	0.035	0.62
Nematocera	0.35	0.37*
Delphacidae, adults	0.52	0.76*
Cicadellidae, adults	0.033	0.52
Heteroptera	0.020	0.54
Carabidae, total	0.0011	0.58
Aleocharinae	0.0068	0.53
Staphylinidae, total	0.0019	0.60
Crysomelidae, total	0.52	0.93
Curculionidae, total	0.057	0.53
Clavicornia, total	0.091	0.85
Arthropods, total	0.68	0.71*

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4 Discussion

The results on weed flora and arthropod fauna in transgenic herbicide tolerant fodder beets are largely in accordance with the monitoring data from previous years, where flora and fauna in conventional fodder beet fields were compared with that of transgenic glyphosate tolerant fields (Bruus Pedersen and Strandberg 2000, Elmegaard and Bruus Pedersen 2001). Thus, the hypothesis stating that farm management and timing of the herbicide applications in particular is most important for biodiversity improvements is sustained. However, the results emphasise that a positive effect on flora and fauna can only be obtained when Roundup Ready applications are delayed relative to application of conventional beet herbicides, i.e. Roundup Ready applications should follow the label recommendation (Monsanto Europe 1999) or be further delayed (Figure 3.1, 3.3 and 3.4). Early application of Roundup Ready resulted in an extremely effective weed control with low weed diversity, density and biomass during the entire season. The present study also clarifies that the effects on flora and fauna within the fields are due to the herbicide application and not related to the crop itself as the transgenic variety was used for all herbicide treatments including application of conventional beet herbicides.

In 2001 the "window" between the first application of conventional beet herbicides and the first application of Roundup Ready when applied following label recommendation was 35 days. The previous year more narrow "windows", viz. 15-20 days, between the applications resulted in significant improvements in the weed biomass in Roundup treated plots (Elmegaard and Bruus Pedersen 2001). Between-year and between-field variation in weed flora will determine the weed infestation and thereby the weed control needed. Weed control in conventional fodder beets is only possible until the crop starts to develop true leaves because of the sensitivity of beets to the conventional herbicides. Moreover, conventional beet herbicides are only effective when the weeds are at the cotyledon stage (Jensen 1998). Therefore, the weed control in conventional beets is unreliable. Weed problems may arise in early summer when the competitive ability of the beet is low and may result in yield reductions. In 2001, no yield reduction was found in plots treated by conventional beet herbicides. However, the recruitment of weeds in these plots succeeding the spraying period resulted in large weed plants at harvest, which from a farmers point of view is undesirable.

Previous data from the Danish demo-trials (available on the web site: http://www.lr.dk/planteavl/informationsserier/gmoroer/index.htm) showed that the difference in number of weeds between conventional plots and GMO plots decreased during June and might even be slightly reversed about two weeks after the second Roundup application. The present study confirms that the weed flora in fields that have received conventional herbicides is richer from early July and the rest of the season with regard to density, diversity and in particular weed biomass than plots receiving Roundup Ready at recommended level (Figure 3.1). This may be a direct result of the superior efficacy of glyphosate compared to conventional beet herbicides as shown by e.g. Bückmann et al. (2000) and Jensen (1998). We found that irrespective of the time of the first application the weeds were effectively controlled by Roundup Ready. Weeds, such as *Urtica urens* and *Polygonum convolvulus*, known to be difficult to control by glyphosate application when treated at larger growth stages (Kudsk and Mathiassen 1998) contributed significantly to the biomass in plots where the first application was delayed by 48 days (Table 3.3). However, we observed that although these "problematic weeds" were not killed immediately by the RR applications their growth stunted, and this in combination with the increasing competition from the beets resulted in the plants dying between the samplings in mid-July and August (Tables 3.2 and 3.4). The fact that only very few weeds, if any, were recruited following the Roundup applications further demonstrated the efficacy of the Roundup treatments, and the very limited weed flora throughout the season in plots receiving Roundup early in the season is a direct consequence (Figure 3.1). In previous years, some recruitment of weeds were seen between the first and second Roundup application, especially in plots receiving reduced dosages, but these plants had reduced growth (Bruus Pedersen and Strandberg 2000) and they contributed little to the total weed biomass (Elmegaard and Bruus Pedersen 2001).

Following the herbicide applications some changes in species composition were found, reflecting the sensitivity of the different species to Roundup Ready and conventional beet herbicides, respectively. Species that are acknowledged to require higher dosages of glyphosate for control especially at later growth stages, e.g. *Urtica urens, Polygonum* spp., *Tripleruspermum inodorum*, and *Lamium* spp. (Bückmann et al 2000, Kudsk and Mathiassen 1998), became more abundant and accounted for a larger part of the biomass after the applications (Table 3.3). *Poa annua* that was the most abundant species before the herbicide applications seemed to be very sensitive to Roundup Ready and nearly disappeared following the applications, whereas it became the most abundant weed in the July sampling in plots that received conventional beet herbicides.

Based on model simulations for skylark populations in farmland Watkinson et al. (2000) predicted that genetically modified herbicide tolerant crops will be a serious problem for farmland birds. Their presumptions, however, made it an extremely conservative case. They modelled a five-year crop rotation with one year of sugar beet followed by four years of cereals and assumed that weed seeds were only produced in sugar beets. Furthermore, they based their predictions on the assumptions that seeds of *Chenopodium album* control the skylark (*Alauda arvensis*) populations being the principal food for wintering birds, and that a relation exists between present weed infestation (weed density) and the likelihood that the farmer will implement the transgenic herbicide tolerant crop. The present study confirms that weed seeds are not produced or only produced in very limited amounts when the field is treated by Roundup Ready despite of time of application. Effect of glyphosate on weed seed set has also been demonstrated by Clay and Griffin (2000) and for conventionally used herbicides as well (e.g. Andersson 1995, Fawsett and Slife 1978). In contrast, we found that plenty of weed seeds were produced in plots treated with conventional beet herbicides. We did not count the actual number of seeds, but they contributed to 10-20% of the total weed biomass. *Chenopodium album* seeds are small (< 1 mg per seed, Green 1980a) and therefore might have been numerous. However, Green (1980a, b) showed that small seeds like those of *Chenopodium album* constituted less than 10% of the weed seeds eaten by skylarks, and a Danish study of the importance of weed seeds for foraging birds showed that less than 5% of the arable birds' food consisted of weed seeds if waste grain was available (Berthelsen et al. 1997). This conclusion has recently been supported in a four year survey by Esbjerg and Petersen (2002) who considered waste grain biomass to be tenfold or more the weed seed biomass. The conclusions by Watkinson and coworkers on the consequences of the herbicide tolerant beets for skylark populations, therefore, are based on doubtful assumptions: The consumption of *Chenopodium* seeds. Presumably, these seeds are of minor importance as food for the skylarks especially in crop rotations where waste grain from neighbouring cereal fields will be available and represent a more attractive food source. On the other hand we cannot rule out that the total lack of seed production may have a negative effect on farmland birds. Moreover, also unripe inflorescences of e.g. *Poa annua* may serve as high-quality food sources of arable birds such as skylark and partridge in early summer (Elmegaard pers. com.). *Poa annua* was found at high densities particularly in Roundup Ready-treated plots in June and had plenty of unripe inflorescences.

On June 21 and July 18, when arthropod densities were high, most species and groups displayed similar patterns with higher frequencies in the late-treated RR plots compared to all other plots. Since no insecticides were used, the differences in arthropod densities between treatments are caused by indirect effects. Total arthropod abundance, Nematocera, Delphacidae and Crysomelidae were reasonably well linearly correlated with weed biomass (Table 3.7), indicating that the occurrence of arthropods in general and the mentioned groups in particular depends on the available weed biomass. For most groups, however, there was no linear relation between weed biomass and arthropod distribution (Table 3.7), but the distribution pattern for arthropods seemed to be delayed compared to the weeds (Figures 3.1 and 3.3). This seems reasonable, as the animals need time to respond to the vegetation in terms of e.g. reproductive success. The poor correlation with weed biomass for some arthropod groups corresponds with the findings of Esbjerg and Petersen (2002), who in beet fields only found a significant correlation for Chrysomelidae in their study of the effect of reduced pesticide dosages on arable wildlife. These authors only tested the correlation between herbivorous insects and weed biomass. However, it is striking that in the present project herbivorous and non-herbivorous groups display the same response pattern. This suggests that something beside the vegetation as food source determines arthropod distribution. One possibility is the indirect effect of differences in plant cover, i.e. differences in microclimatic conditions. The microclimate in dense vegetation tends to be more humid and stable than on bare soil, thus offering better conditions to the arthropods. Obviously, the growing crop biomass interferes with the effects of differences in weed cover on microclimate and thereby with the general picture for the arthropods. The preference of Meligethes for conventionally treated plots on July 18 can be explained by the presence of rape, *Brassica napus*, in these plots. The arthropod results of the present study show that a larger and more diverse arthropod fauna may result from especially delayed Roundup treatment compared to conventional herbicide treatment of fodder beet. Such an effect is likely to benefit arable birds while they rear their chicks. However, it is of course prerequisite that insecticides are not used.

Large between-year variation in weed flora in conventionally managed row-crops such as beet is common and the reliance on weather conditions (time of sowing, spaying weather, general growth conditions) is obvious. Transgenic, herbicide-tolerant crops may decrease the sensitivity of the management strategy, as the period for effective weed control is not restricted to a short period around sowing. The present study shows that yield might be maintained even when herbicide application is substantially delayed and more weeds are present in the field during the early summer period. The farmers' readiness to accept weeds below the economic threshold for a longer period will be decisive for the positive effect on biodiversity during early summer. However, the effect of reduced or lacking production of weed seeds may not only have an effect on the availability of seeds as food for the birds, but may also change the weed flora significantly. Species composition may be changed towards less sensitive species, density and biomass may be altered, and thereby the basis for the positive effect on arable land biodiversity may disappear. If the amount and composition of the weed flora changes, this is likely to be reflected in the abundance and diversity of the arthropod fauna. This aspect of growing herbicide-tolerant crops needs further investigation in relevant crop rotations.

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Denmark
Tel: +45 46 30 12 00
Fax: +45 46 30 11 14

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PO Box 314
DK-8600 Silkeborg
Denmark
Tel: +45 89 20 14 00
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Department of Freshwater Ecology
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