Management of Herbicide-Resistant Weed Beet: a Simulation Study

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Abstract

Accidental bolting of a few genetically-modified herbicide-tolerant (GMHT) sugar beet plants would result in seed production and risk pollen-mediated transgene flow towards weed beets that have escaped the herbicide in the same field and in the neighbourhood. The coexistence concern, here, applies to the spread of herbicide-resistant weed beets, which could make it necessary to modify the cropping system in the neighbourhood. We use a spatio-temporal simulation model (GENESYS-Beet) for comparing three production systems with different crop rotation and cultural practices to look at the risk of obtaining herbicide-tolerant weed beet populations, at the scale of a small agricultural region where GMHT and conventional sugar beet coexist.

Introduction

Sugar beet is grown for its root and harvested before flowering, so that cross-pollination occurring among fields in a beet-producing region is not a direct concern for the coexistence of genetically-modified (GM) and non-GM crops. However, adopting GM sugar beet varieties can have serious consequences for the cropping systems of a farming region. In regions with sugar beet cultivation, weed beet infestations are responsible for economic losses. Weed beet belongs to the same species as the crop plant, thus rendering herbicide control impossible and requiring costly practices such as manual weeding. GM herbicide-tolerant (GMHT) sugar beet varieties, which were initially designed to allow more efficient, less labour-consuming and less expensive practices than current herbicide programs (Richard-Molard et al., 1996), could be a unique opportunity to create selectivity between the crop and its weedy relative. Thus, it would be possible to kill weed beets while preserving the sugar beet crop. However, this benefit would be lost if the weed beets acquired the resistance through sexual cross-hybridisation with accidentally bolting crop plants and/or through seeds produced by these accidental bolters. In addition, the newly resistant weed beet could spread throughout the region where weed control strategies currently use the same herbicide, thus jeopardising operational cropping systems even in the fields that had never been cultivated with GM crops.

Sugar beet is a biennial plant requiring vernalization for bolting, so that it is not expected to flower during the growth season, but a few cases of abnormal bolting always occur (Perarnaud et al., 2002). This phenomenon results in pollen flow in the field and the region. The recipient plants could be the few weed beets that escaped the herbicide treatment in the same field, but also bolting sugar beets and weed beets growing in other fields in the region. Evidence of this kind of cross-pollination was found in long-term experiments at the farm scale with weed beets growing in the same field and in adjacent and nearby set-aside (Darmency et al., 2007a,b). Table 1 shows the average percentage of transgenic hybrids produced by susceptible sugar beet bolters and susceptible weed beets growing within the field and in neighbouring fallow. This phenomenon is probably not easy to contain, as long-distance airborne pollen flow was recently demonstrated to contribute greatly to the fertilising pollen deposition at any given point in the fields (Darmency et al., 2009), and genetic studies showed that weed beet populations several km apart can exchange genes (Fénart et al., 2007).

Table 1: Average percentage (%) of resistant hybrid seed produced by susceptible sugar beet bolters and weed beets growing within the field, and weed beets growing in adjacent to 100 m distant fallow, in two locations (North East France): over 6 years in Châlons, and 4 years in Dijon (data from Darmency et al., 2007b). The lower value recorded at Dijon for in-field weed beet was certainly due to the lack of spontaneous indigenous weed beet that had been replaced by transplanted individuals.

Location	Susceptible bolters	In-field weed beet	Weed beet in fallow
Châlons	3.12	3.02	0.57
Dijon	2.67	0.57	0.41

Predicting gene flow at a regional scale is certainly a difficult task because it depends on numerous variables: pollen dispersal, plant biology, spatial arrangement of the fields, landscape components, weather, and cropping systems. No experimental design is suitable to account for all the combinations of all these variables. Therefore, we developed a spatio-temporal simulation model based on the previous GENESYS model released for modeling the fate of volunteer oilseed rape in the GM context (Colbach et al., 2001a,b). The main interest of the GENESYS model family is to rank the effects of the different components of the cropping system on the population dynamics of the weed populations, the gene flow and the genotype proportions for genes of interest (Sester et al., 2007, 2008).

In the present paper, the first objective was to test a large range of possible combinations of crop successions and cultivation techniques, in order to rank the various cropping system components and to quantify their effects, at the field scale, on weed beet dynamics, particularly on herbicide-tolerant populations. To reach this goal, a global sensitivity analysis of the GENESYS-Beet model to cropping system variables was run, based on Monte Carlo simulations where all input variables were made to vary randomly and simultaneously (Tricault et al., 2009). This procedure has already been used in a previous sensitivity analysis with a similar model and is useful for ranking variables over a large range of situations while taking into account the interactions between the variables (Colbach et al., 2004a). The simulation model is also used in a case study of a French sugar beet production region where three types of intensive production systems had been identified: (1) "potato grower"; (2) "beet grower"; and (3) "cereal grower". This provides an opportunity for testing contrasting production systems with different crop successions and cultural practices to evaluate the risk of herbicide-tolerant weed beet populations arising in a small agricultural region where GMHT and conventional sugar beet coexist. The three production systems were successively simulated on a real 149-field map extracted from the studied region.

Materials and Methods

GENESYS-Beet model

Model inputs are (1) the regional field pattern, (2) the crop sequence in each field, (3) the cultivation techniques used to manage each crop, (4) the genotype of the sugar beet varieties, and (5) daily climate and soil conditions (see Sester et al., 2007, 2008 for detailed description). The model simulates the life cycle of weed beet in every field as a function of abiotic constraints and cropping system variables (i.e. crop, tillage operations, herbicide treatment etc.: Sester et al., 2004, 2006). The model operates on a daily time step with life-stage densities and genotype proportions as state variables. Genotype proportions change when the herbicide associated with the transgene is applied (i.e. glyphosate in case of GM glyphosate-tolerant sugar beet) and during pollination. Gene flow is calculated from the pollen production of flowering plants in each field and the individual pollen dispersal function (Darmency et al., 2009) integrated over the whole region for every recipient field.

Sensitivity analysis at field scale

Simulations started with an empty weed seed bank and a GM sugar beet crop, lasted for 20 years, and always finished with a non-GM sugar beet (as the most favourable crop for weed beet). All other input variables (notably crops and techniques) were randomised (see Tricault et al., 2009, for full description). The simulation plan consisted of 100 000 random cropping systems, and only the weed output data (total density and GM density) of the last year (with non-GM sugar beet) were kept for analysis. There were 55 crop management variables per year including options for tillage (17), herbicide applications (11), mechanical weeding (3), cutting (3), hand-pulling (5), crop characteristics (5), together with the set of weather scenarios plus soil texture and the width of the various items of farm machinery (11 variables). Because of the huge amount of data generated, a classification method (segmentation tree) was used first to identify the major input variables. These variables were then introduced as input variables into a linear regression model (with output data as dependent variable) for years 0 (the analysed non-GM sugar beet), -1 (the previous crop) and -2 (the pre-previous crop), and more if the input variables were significant for additional past years. This estimation method allowed determining the "persistency" of each variable, i.e. for how many past years they must be taken into account to understand current weed beet infestations and genotypes.

Farm type simulations at regional scale

The GeneSys-Beet model was used for farm type simulations based on a survey of a French sugar beet production region (Picardie) where sugar beet covers 18 % of the arable land. In this region, three farm types were identified:
(1) "potato grower", with a seven-year rotation: sugar beet / potato / winter wheat / legume / winter wheat / potato / winter wheat.
(2) "beet grower", with a five-year rotation: sugar beet / winter wheat / set aside / pea / winter wheat.
(3) "cereal grower" with a four-year rotation: sugar beet / winter wheat / pea / winter wheat.

The management of the various crops in the three farm types was kept the same except for the strategy of weed beet control and the tillage option. The strategy of weed beet control determined the choice of sugar beet varieties (GM vs. non-GM sugar beet), with the use of the GMHT variety restricted to fields already infested by weed beets (note that all this is virtual as GM sugar beet is prohibited in France). Beet bolters were left uncontrolled, sprayed with glyphosate (in case of GM sugar beet), or eliminated by manual weeding. Tillage either included mouldboard ploughing or was restricted to superficial tillage only. Weed beet control and tillage options resulted in four strategies (Table 2). Each strategy was assigned to a given area of the cropping region. For instance, strategy A concerned 11 % of the simulated area, used GM sugar beet varieties because its fields were infested by weed beet (300 seeds m^{-2} in the soil seed bank at the onset of the simulation), and fields were tilled with a mouldboard plough.

Table 2. Different weed beet control strategies

Input variables	Strategies			
	A	B	C	D
% of region	11%	45%	22%	22%
Initial weed beet infestation	300 seeds/m²	0	0	0
Sugar beet variety	GMHT	non-GM	non-GM	non-GM
Mouldboard ploughing	yes	yes	yes	superficial
Manual bolter weeding	none	none	twice	none

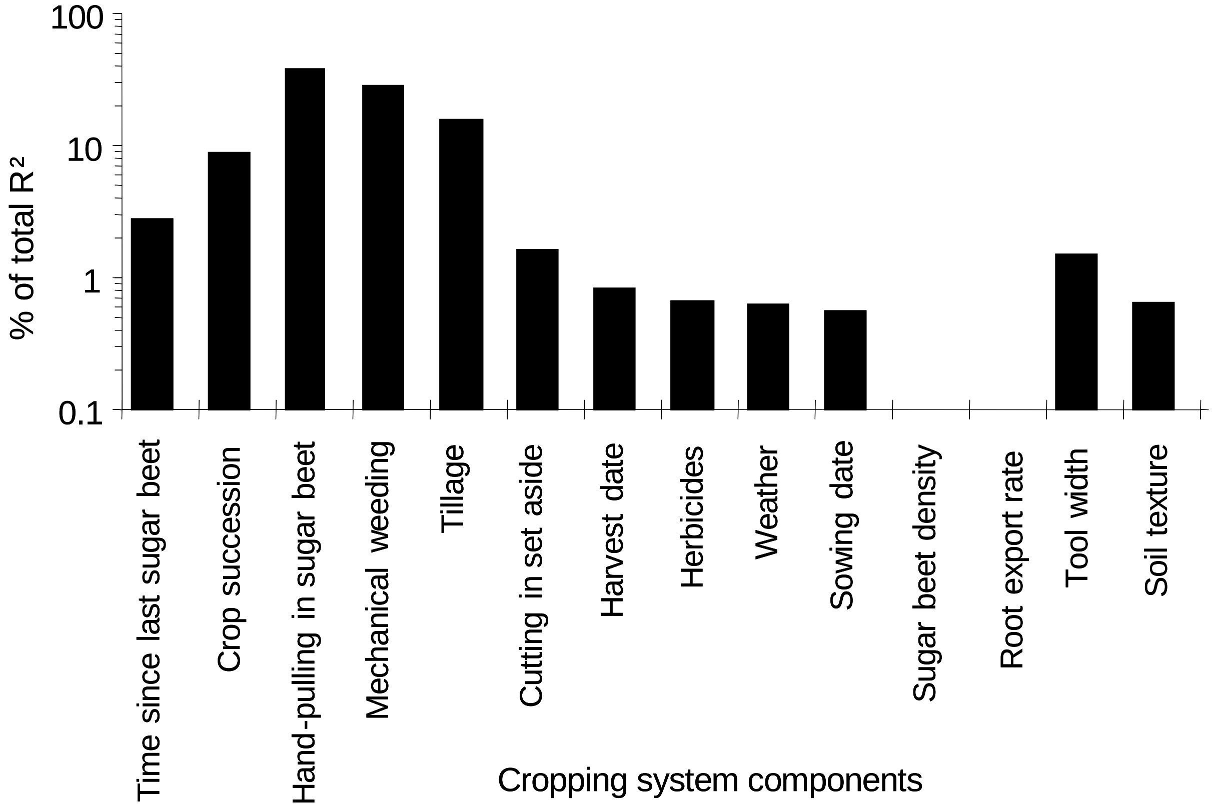
The three farm types were separately simulated on a real 149-field map extracted from the studied region (see Figure 4). At the onset of each farm type simulation, the various strategies were randomly allocated to these fields according to their proportion, and the starting crops of the rotation were distributed randomly in each field. During the subsequent years, the strategy remained fixed in each field; in particular, a field once grown with a GM variety was never cultivated with a non-GM variety and vice versa. Simulations lasted 15 years, with the GMHT weed seed bank in each field as the output variable.

Results

Sensitivity analysis at field scale

The linear regression analysing bolter densities in the last non-GM sugar beet as a function of cropping system components and environmental conditions indicated that sugar beet frequency (over the 20 years) was not the only factor explaining bolter variability (Figure 11). Hand-pulling of bolters and mechanical weeding (i.e. the only two bolter control options in non-GM sugar beet) were the two most important variables in terms of explained variability (i.e. partial R^2, Figure 1). Tillage and crop succession (i.e. the time since the last GM and non-GM sugar beets and the nature of the other crops) were the next two major variables. The remaining variables, including soil texture and tool width, only explained a little variability while sugar beet sowing density and the root export rate (i.e. the harvest efficiency) in sugar beet were not significant.

Figure 1. Contribution of explicative variables to explaining the variability in bolter densities at field scale simulated by GeneSys after 20 years. Percentage of total R^2 of linear regression model, all rotations confounded (adapted from Tricault et al., 2009).



Crop succession presented the longest persistency, i.e. the times since the last GM and non-GM sugar beet were significant during the 19 years preceding the analysed non-GM sugar beet and the nature of the preceding crops during the preceding four years. Harvest date was the only other variable with a four-year persistency. All other cultivation techniques were only significant up to two years prior to analysis except sowing date, which was only significant for one year. The total R^2 (0.43) explained by the selected variables within a linear additive regression model was rather low, pointing to important interactions between the selected variables and to minor long-term effects of the unselected variables.

The ranking of cropping system variables was similar for GM bolters (not shown), except that the time since the previous sugar beet (with three quarters of that effect due solely to the time since GM sugar beet), tillage, cutting, herbicides and sowing dates saw their impact increased. In contrast, the importance of hand-pulling decreased. The persistency of most variables remained unchanged except for tillage, herbicides and sowing date, which were significant for an additional year. The total R^2 was slightly lower (0.40) than for total bolter density.

Farm type simulations at regional scale

Whatever the production system, all the fields were infested with GM seeds after 15 years. GM seed density was largest in GMHT sugar beet fields. Among the fields where no GMHT sugar beet had ever been grown, GM seed density was largest in ploughed fields and without any manual weeding (strategy B, Figure 2). In unploughed and twice-weeded fields (strategies C and D), GM seed density decreased considerably. The no ploughing strategy was even more efficient in the "cereal grower" system because the seeds were left to germinate on the soil surface after sugar beet harvest, thus reducing seed survival in soil. This option is, however, only feasible when the following crop is a crop such as winter wheat where weed beet hardly reproduces and sets very few seeds.

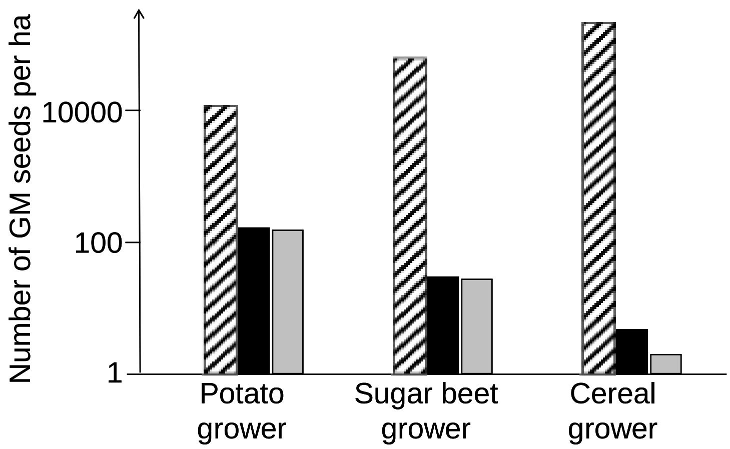


Figure 2. Mean simulated infestations with GMHT weed beet seeds in fields that had never been sown with GMHT sugar beet, as a function of crop management strategy (B, C or D), after 15 years and for each farm type: B with ploughing (hatched bar); C with hand pulling (filled bar); D without ploughing (grey bar).

Farm type also has a marked effect on seed density. The infestation level is inversely proportional to the frequency of sugar beet in the crop rotation of the fields managed with strategy B, while it is simply proportional with strategies C and D (Figure 2). In the first case, more frequent sugar beet in the cereal grower farm type than in the sugar beet and potato grower types (1/4, 1/5 and 1/7, respectively) creates more favourable conditions for the presence of target weed beets that can catch the GM pollen coming from the GMHT fields, and then produce GM seeds. In the two other cases, hand pulling and seed bank depletion due to not ploughing reduce the number of weed beet flowering in the sugar beet, resulting in the opposite trend.

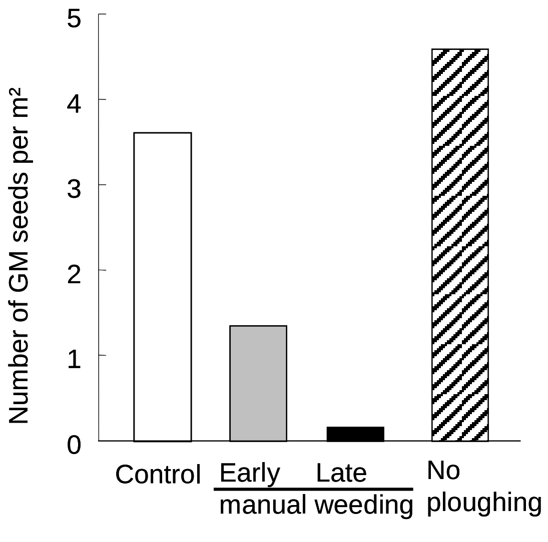


Figure 3. Effect of the management of GM sugar beet on the GM seed bank averaged over the fields that had never been sown with GMHT sugar beet in the region after 15 years of GM cultivation in a traditional sugar-beet region adopting the "potato grower" farm type.

Changing the management of the GM sugar beet in the GM fields may also modify the density of GM seeds in non-GM fields (case of “potato grower” type: Figure 3). Hence, this only arises as a consequence of gene flow. For instance, manual weeding of GM bolters, particularly when carried out at a time when most potential bolters had indeed bolted, was very efficient in reducing the GM seed bank. In contrast, abandoning mouldboard ploughing before GM sugar beet sowing increased the GM seed bank because the seeds of weed beets easily emerge in a very favourable crop.

When considering the 149-field landscape of the small studied region, the most impressive result was the presence, in some fields that have never been sown with GMHT sugar beet, of herbicide-resistant weed beet at densities as high as in the fields with the cropping strategy A including GMHT sugar beet (Figure 4). The location of the most infested fields is not simply proportional to the distance to the closest fields where GMHT sugar beet was cultivated but also depends on the coincidence of the years devoted to sugar beet cropping in GM and non GM fields.

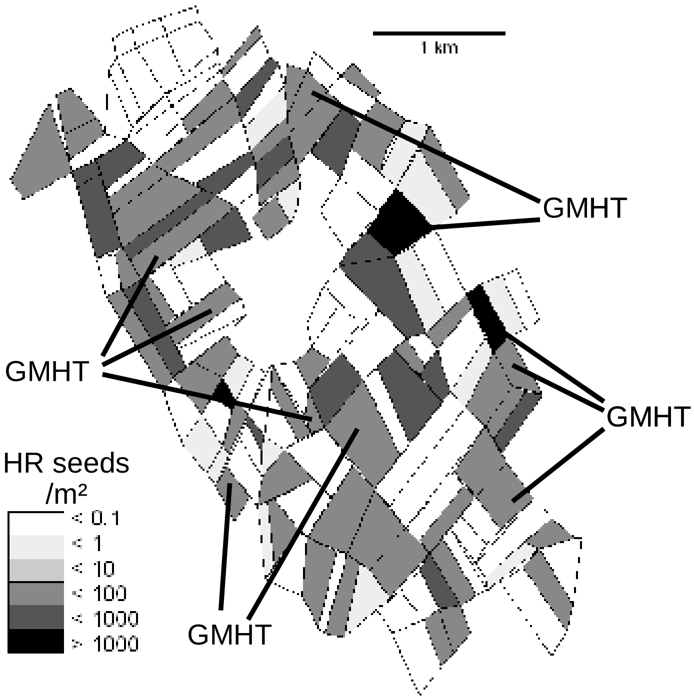


Figure 4. Map of the arable fields of a small district of the sugar beet production region. The intensity of the grey painting corresponds to the density of herbicide resistant weed beet seeds in the soil 15 years after the first use of GMHT sugar beet. The arrows indicate the fields where GMHT sugar beet is grown (cropping system A).

Discussion

The present simulations suggested that the occurrence of GM weeds is inevitable when some proportion of bolting sugar beet, even as minute as possible, is present in the sown GMHT seed lot, because no management practice is able to destroy all of the bolting plants. The ranking of the different crop management variables in the sensitivity analysis made evident the major effect of hand pulling and mechanical weeding on the weed beet demography. Because of high seed longevity (Sester et al., 2006), the time since the last sugar beet grown in the same field had a significant effect on the level of infestation up to 19 years later. This suggests that, even when GM sugar beet has not been grown for several years, GM individuals persist in the weed beet seed population and could thus infest the field when conditions become favourable again. Indeed, as in the case of total bolter densities, the present study showed that the successful management of GM populations is greatly dependent on the right choice and combinations of crops as well as cultivation techniques. The main advice proposed for bolter densities also applies to GM beet, with a few somewhat predictable differences: the careful choice of herbicide options is much more important for GM weeds while hand-pulling loses its key role, mostly because it is no longer the sole option for managing bolters in sugar beet. In addition, it will take longer to control GM weed population than non-GM infestations because the former have significant selective advantage in herbicide-based cropping systems.

As an example, the model was used to evaluate cropping strategies for managing weed beet and for limiting the advent of herbicide-tolerant populations in a small sugar beet root production region. The main result was that gene flow towards non-GM fields is not strictly proportional to distances between donor (GM) and recipient fields (non GM), thus underlying the importance of both the cropping system variables at field scale and the temporal synchronisation of crops between fields (i.e. GM and non GM sugar beets grown during the same years). Increased delay between two successive sugar beet crops and systematic hand-pulling of bolters are efficient and often essential measures. Introduction of perennial crops where weed beet hardly survive and reproduce could be another efficient way for keeping these weeds at low density (Tricault et al., 2007). Obviously, the primary measures must be applied in the fields where GMHT sugar beets are grown (e.g. the use of certified seeds of a bolting resistant variety, two manual weeding operations, and improved control in spring crops). However, appropriate cropping systems and management practices must be also adopted in the whole region if we aim at limiting the spread of the herbicide resistance. Thus, the coexistence concern for sugar beet is definitely not a matter of root admixture in the harvest, but it must include the agronomic and economic consequences of growing GMHT sugar beet for the cropping systems of the neighbouring farms.

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