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Modeling the development of a target site resistant *Apera spica-venti* (L.) P. Beauv. population – A comparison of model output and field data

Modellierung der Entwicklung einer Target-site resistenten Apera spica-venti Population – Ein Vergleich von Modelergebnissen und Felddaten

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Abstract

A population dynamic model was combined with a genetic model and embedded into a cellular automaton. The model was evaluated with data from two three year field trials which were conducted on commercial fields in Lower Saxony and Saxony where target-site resistance to acetolactate synthase (ALS) inhibitors was detected in *Apera spica-venti*. The cropping system consisted of continuous winter wheat in the trial period. On four plots different herbicide strategies were tested. These were continuous application of a soil herbicide, alternation between ALS inhibitor and soil herbicide, continuous use of an ALS inhibitor and two applications per growing season with different mode of actions (MoA). In the beginning of the trial soil samples were taken to estimate the number of viable seeds in the seed bank. This data was used to produce seed distribution maps by interpolating the estimated seed data over the field. These seed maps were then used as the initial seed bank in the model and simulations over three years were executed with the assumption of herbicide use as conducted in the field trial. A comparison of the model output with the field data showed very good analogies in the weed density. Also the development of resistance was reproduced well. The model can now be used to assess herbicide management strategies concerning the development of herbicide resistance for *A. spica-venti*.

Keywords: ALS resistance, genetical model, population dynamic model, loose silky bent

Zusammenfassung

Ein populationsdynamisches Modell wurde mit einem genetischen Modell verbunden und in einen zellularen Automaten eingebunden. Das Modell wurde mit Daten von zwei dreijährigen Feldversuchen evaluiert. Diese wurden auf Resistenzstandorten (Target-Site Resistenz gegen Acetolactat Synthase (ALS) Hemmer) in Niedersachsen und Sachsen durchgeführt. Angebaut wurde Winterweizen während des Versuchszeitraums. In vier verschiedenen Versuchsgliedern wurden unterschiedliche Herbizidstrategien untersucht. Diese waren kontinuierlicher Einsatz eines Bodenherbizids, Wechsel zwischen einem ALS-Hemmer und einem Bodenherbizid, kontinuierlicher Einsatz eines ALS-Hemmers und zwei Herbizidapplikationen mit Produkten verschiedener Wirkmechanismen innerhalb einer Vegetationsperiode. Zu Versuchsbeginn wurden Bodenproben entnommen und das Samenpotential im Boden bestimmt. Diese Daten wurden genutzt um durch Interpolation Samenverteilungskarten zu erstellen. Diese Daten wurden dann als Anfangssamenverteilung für das Modell genutzt und Simulationen über drei Jahre durchgeführt mit denselben Herbizidstrategien aus den Feldversuchen. Der Vergleich der Felddaten mit den Modellausgaben zeigte eine gute Übereinstimmung in der Unkrautdichte. Auch die Resistenzentwicklung konnte wiedergegeben werden. Das Modell kann jetzt dazu genutzt werden Herbizidmanagementstrategien in Bezug auf ihr Resistenzentwicklungspotential zu bewerten.

Stichwörter: ALS Resistenz, genetisches Modell, populationsdynamisches Modell, Windhalm

Introduction

The world population is steadily increasing and will reach nearly 10 billion in the year 2050. Therefore the cultivated area per person is decreasing and the efficiency of agricultural production needs to be increased to ensure enough food, feed and fibers. To maintain high yield herbicides are an important tool in modern agriculture, to minimize the weed competition. But herbicide use

can lead to resistant weed population, especially when using repeatedly the same herbicide or mode of action (MoA). Only a limited number of MoA's are available. For the treatment of *A. spica-venti* in cereals 8 MoA's are available in Germany (BUNDESAMT FÜR VERBRAUCHERSCHUTZ UND LEBENSMITTELSICHERHEIT, 2013). To three of them (HRAC groups A, B and C2) resistance has already been developed (HEAP, 2013). A good resistance management is needed in intensive agriculture where weed control is sometimes exclusively depending on herbicides. Models can be useful to study the long-term dynamics of herbicide resistance development in a weed population in a short time span. In the following a model will be presented which simulates the development of target-site resistance in an *A. spica-venti* population, and data of two field trials are reported. For each trial, the resistance development was monitored over three years on four plots with different herbicide treatments. On both fields target-site resistance to an ALS inhibitor was detected before the beginning of the trial. The model output was compared to the field data from the two long-term field trials.

Material and methods

Field trial

From fall 2008 to summer 2011 two field trials were conducted in Lower Saxony and Saxony, from now on called site A and B respectively. Both *A. spica-venti* populations showed target-site resistance to sulfonylureas a subgroup of ALS inhibitors. On the two commercially used fields an area of approximately 1 ha was divided into four plots and valuation and sampling occurred on a 10 m grid in the first year and on a 6 m grid in the second and third year respectively. Winter wheat was cultivated over the three years on both fields. On the four plots different herbicide management systems were conducted (Tab. 1).

Tab. 1 Application rates and active ingredients for the four plots.

Tab. 1 Aufwandmengen und Wirkstoffgehalte für die vier Versuchsglieder.

growing season	plot 1 (soil herbicide)	plot 2 (ALS inhibitor)	plot 3 (alternation inhibitor and soil herbicide)	plot 4 (different MoA's)
2008 / 2009	Bacara Forte 1.0 l/ha	Husar OD 0.1 l/ha + Mero 0.6 l/ha	Husar OD 0.1 l/ha + Mero 0.6 l/ha	IPU 3.0 l/ha Axial 50 1.2 l/ha
	Flufenacet, 120 g/l Flurtamone, 120 g/l Diflufenican, 120 g/l	Iodosulfuron, 100 g/l	Iodosulfuron, 100 g/l	Isoproturon, 500 g/l Pinoxaden, 50 g/l
2009 / 2010	Bacara Forte 1.0 l/ha	Husar OD 0.1 l/ha + Mero 1.0 l/ha	Bacara Forte 1.0 l/ha	Bacara Forte 1.0 l/ha Ralon super 1.2 l/ha
	Flufenacet, 120g/l Flurtamone, 120g/l Diflufenican, 120g/l	Iodosulfuron, 100g/l	Flufenacet, 120g/l Flurtamone, 120g/l Diflufenican, 120g/l	Flufenacet, 120g/l Flurtamone, 120g/l Diflufenican, 120g/l Fenoxaprop-P-ethyl, 69g/l
2010 / 2011	Bacara Forte 1.0 l/ha	Husar OD 0.1 l/ha + Mero 1.0 l/ha	Husar OD 0.1 l/ha + Mero 1.0 l/ha	IPU 3.0 l/ha Axial 50 1.2 l/ha
	Flufenacet, 120g/l Flurtamone, 120g/l Diflufenican, 120g/l	Iodosulfuron, 100g/l	Iodosulfuron, 100g/l	Isoproturon, 500 g/l Pinoxaden, 50g/l

In Table 1 the application rates and the active ingredients are listed. The aim of this design was to investigate the development of resistance under different herbicide strategies. In plot one and

four no ALS inhibitors were used over the three years. Here the development of the resistance was surveyed under the influence of a soil herbicide treatment and the application of two herbicides with different MoA's within the growing season. In plot two and three ALS inhibitors were in use. Plot two was treated with an ALS inhibitor in all three growing seasons, whereas in plot three the treatment altered with a soil herbicide. The valuation of *A. spica-venti* plant density as well as the sampling of plants was done before and after the application in every growing season. The sampled plants were analyzed for a mutation on the ALS gene at position Pro₁₉₇.

Generating seed distribution maps

In the beginning of the trial period soil samples were taken at site A at 16 grid points in plots two and four. The soil samples were taken by sampling four soil cores up to 30 cm depth around the grid points with a geological drill (Pürckhauer). At location B at 22 grid points, in plots two and four, five soil cores were taken up to 20 cm depth. The soil cores for each grid point were mixed. The different sample numbers and depths resulted from the different depths of the cultivated soil layers through the different tillage systems on both sites.

At location A the field was ploughed each year and at site B conservation tillage was conducted. To obtain enough soil at location B, one additional soil core was taken. The sampling was done to estimate the number of *A. spica-venti* seeds in the seed bank. Therefore the soil samples were laid out in the greenhouse and the germinated plants were counted and sampled to analyze them for ALS resistance. Sampling occurred only on two plots, because the first plot was already treated at the time of the sampling and it was assumed that the number of seeds in the seed bank were homogenous over the field. The number of germinated *A. spica-venti* plants was converted to viable seeds per m² at the analyzed grid points. To generate seed distribution maps for the sampled plots the data were interpolated using the kriging method (Fig. 1). The interpolation was done with the software SURFER Version 11 (Golden Softare, Inc., Golden, Colorado, USA, 2013). To get kriging weights semivariograms were created and models were fitted to them (OLIVER, 2010). Variogram calculation and the model fitting were carried out by the software GS+ Geostatistics for Environmental Sciences Version 9 (Gamma Design Software, LLC, Plainwell, Michigan, USA 1989-2013).

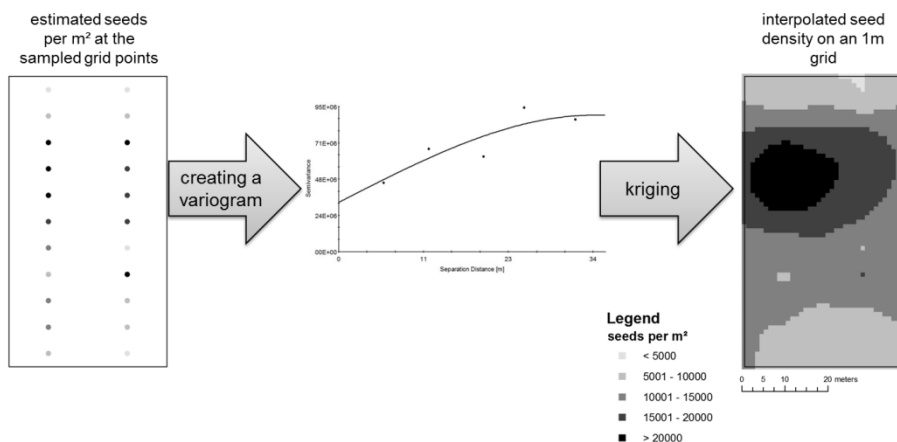


Fig. 1 Example for the creation of the seed distribution maps for one plot.

Abb. 1 Beispiel für die Erstellung der Samenausbreitungskarten für ein Versuchsglied.

Model structure

The model design was described in (RUMMLAND *et al.*, 2012). A population dynamic model was combined with a genetic model and embedded into a cellular automaton (Fig. 2). The population dynamic model is divided into 5 development stages, which are connected via transition probabilities. The three genotypes (susceptible, homozygous resistant and heterozygous resistant) are connected through the inheritance which is modeled using the Hardy-Weinberg-Law. A part of the produced seeds in the last stage are distributed into the neighboring cells. The model starts with an initial seed bank. For the initial number of seeds in the soil the previous created seed distribution maps were used. The simulated area had the same size like the plots in the field trial. The survival probability depending on the used herbicide was set for every herbicide to 5%, except for the ALS resistant individuals whose survival probability was set to 95% for the treatment with an ALS inhibitor. All parameters were set to the same values for both locations.

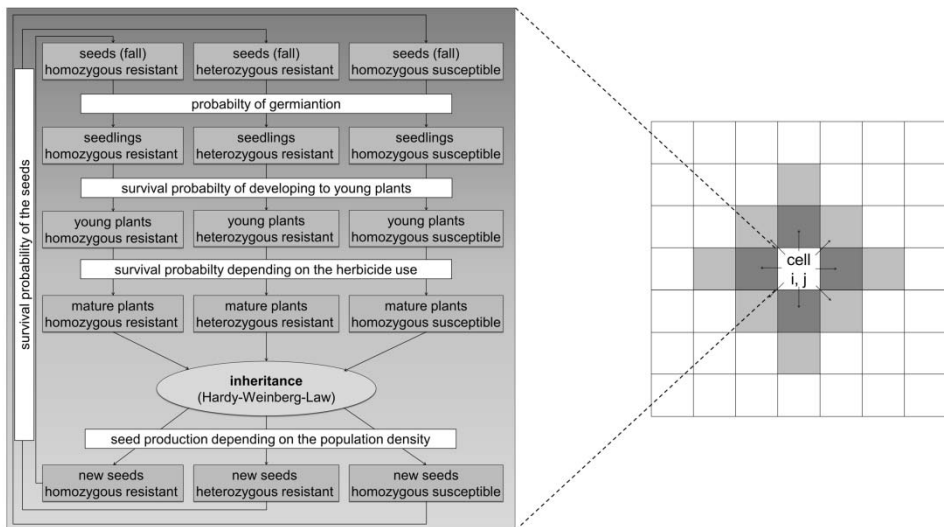


Fig. 2 Model design: a population dynamic model embedded into a cellular automaton.

Abb. 2 Modelldesign: ein populationsdynamisches Modell eingebunden in einen Zellularen Automaten.

Results

In Figures 3 and 4 the mean density of *A. spica-venti* in the field was compared to the modeled densities. For the field data the standard deviation is also shown. At both locations the field data showed an increase in *A. spica-venti* density in plots with ALS inhibitor treatment, as well as a decrease in plots without ALS inhibitor use. Furthermore the field data showed years with very high densities in all four plots compared to the other years. This is especially seen in plots with higher plant densities in the previous years. The germination seemed to be favored in this year. For location A this was the case in the third year of the trial period and location B showed high *A. spica-venti* densities in the second year. This could only be reproduced by the model through raising the value for the germination probability. In the years with elevated germination the standard deviations of the plant density data from the field are the greatest. Here the differences between model results and field data are also the greatest.

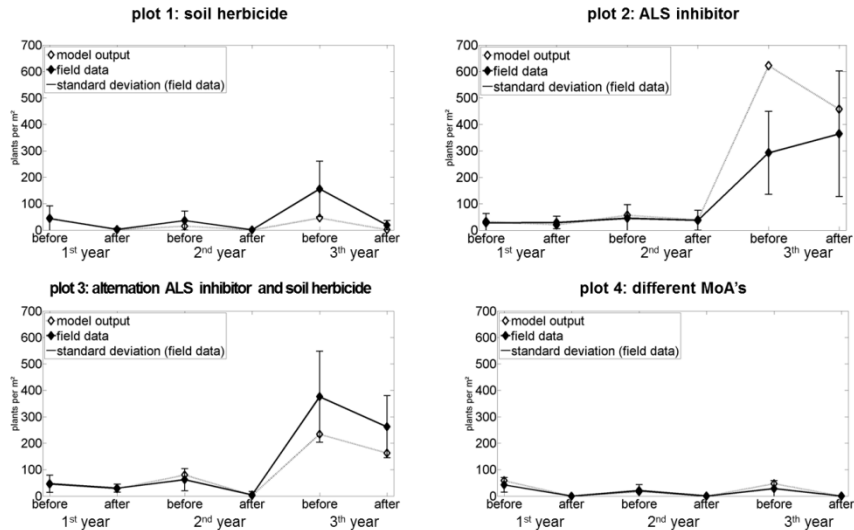


Fig.3 Comparison of the mean *A. spica-venti* density with the model output for location A.

Abb. 3 Vergleich der mittleren *A. spica-venti*-Dichte mit der Modellausgabe am Standort A.

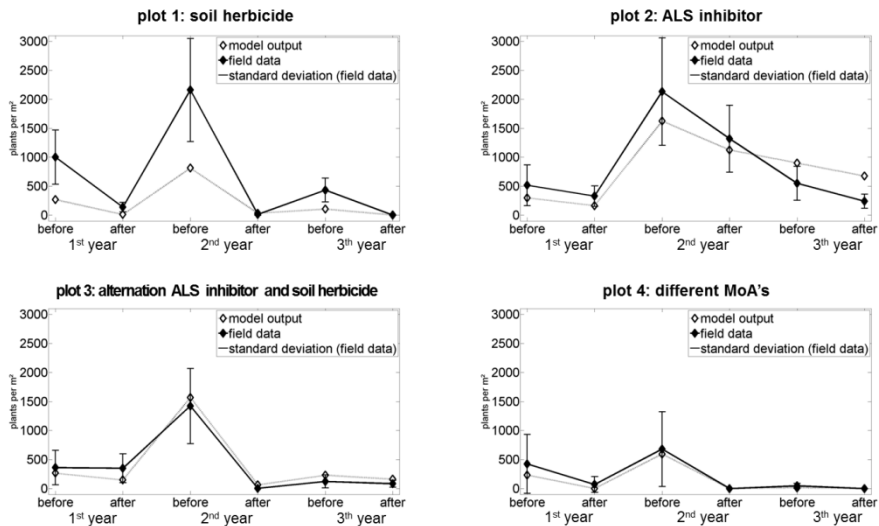


Fig. 4 Comparison of the mean *A. spica-venti* density with the model output for location B.

Abb. 4 Vergleich der mittleren *A. spica-venti*-Dichte mit der Modellausgabe am Standort B.

The highest discrepancy between field data and model output is seen in the third year before the application at location A in the plot with continuous use of an ALS inhibitor. Here the model overestimates the plant density by twice as much. But the field data showed also that the density increased slightly after the application. Probably more plants germinated after the valuation was done. A trend is not seen concerning the over or under estimation of *A. spica-venti* density. In general it can be stated that the model is capable of reproducing the *A. spica-venti* density, but only by altering the germination probability.

The development of target-site resistance in the field and in the model is shown in Figures 5 and 6. The graphs illustrate the proportion of ALS resistant and susceptible plants. The resistance development in plots with the use of ALS inhibitors was reproduced well by the model. In the model as well as in the field a reduction in susceptible plants is seen. In plots without ALS inhibitor use the model showed no changes in the proportion of susceptible plants in the population, this could not be seen in the population from the field trial. In the field the proportions of susceptible and resistant plants in plots without ALS inhibitor use are fluctuating. This could not be reproduced by the model.

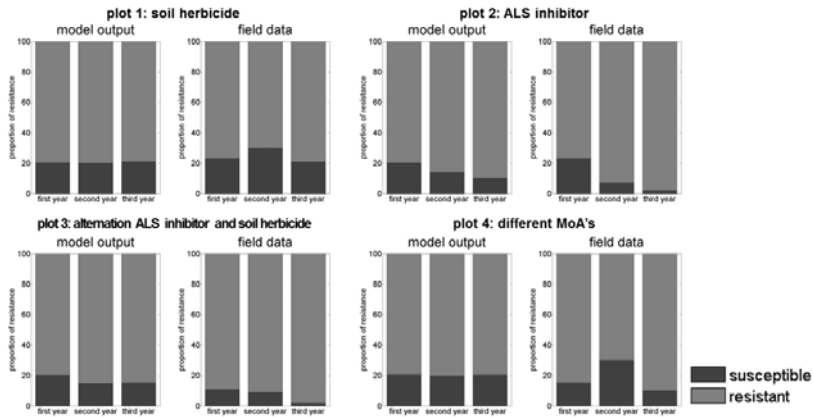


Fig. 5 Comparison of the portion of target-site resistant and susceptible *A. spica-venti* plants in the field and in the model, for location A.

Abb. 5 Vergleich des Anteils Target-Site resistenter und sensibler *A. spica-venti* Pflanzen im Feld und im Modell, für den Standort A.

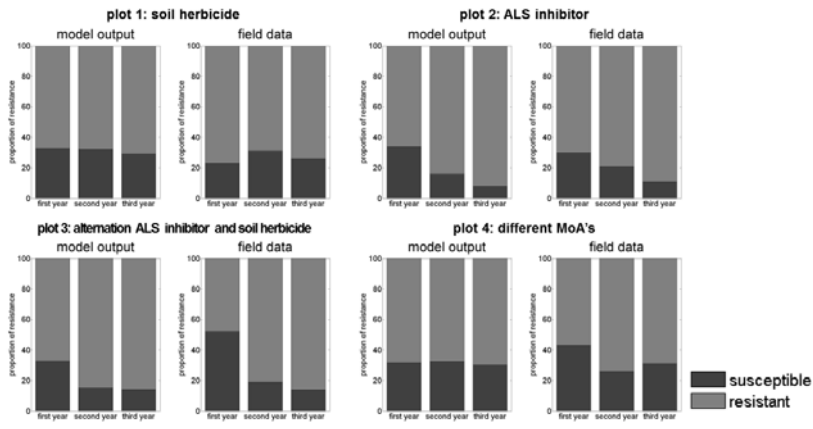


Fig. 6 Comparison of the portion of target-site resistant and susceptible *A. spica-venti* plants in the field and in the model, for location B.

Abb. 6 Vergleich des Anteils Target-Site resistenter und sensibler *A. spica-venti* Pflanzen im Feld und im Modell, für den Standort B.

Discussion

In general the model is capable of simulating *A. spica-venti* density and the development of target-site resistance. Germination varies between years. If the seed density is low in the soil this has only minor impacts on the plant density, like it is seen in Figure 3 in the graph in the bottom right-hand site (the absolute variant). In the third year the *A. spica-venti* density only exceeds slightly the densities of the previous years. In the three other plots the increase in weed density was much higher. It seems that the number of seeds have had been reduced through the effective weed control in the two years before and the thereby associated inhibition of new seed input. But if the number of seeds in the seed bank is high weed density is increasing dramatically in such years. Weather has a great influence on the germination characteristics of *A. spica-venti* (KAMPE, 1976). Wet and warm weather situations favor the germination (GERHARDS and MASSA, 2011; KRYSIAK *et al.*, 2011). Unfortunately it is not possible to predict weather conditions years in advance. So it complicates the simulations of *A. spica-venti* development into the future. Simulations of scenarios should therefore consider at least two runs with different germination probabilities, one low value and a higher probability, to get a possible range of results.

The model describes the development of ALS target-site resistance well, if the selection pressure, due to the use of an ALS inhibitor still exists. The fluctuations in the field data could not be reproduced in plots without the use of an ALS inhibitor. The survival of susceptible and resistant individuals in the field is random, if they are not resistant to the used herbicide. Perhaps a more stochastic model approach could help to reproduce the noted fluctuation.

Summarizing it can be said that the model is reproducing the field data and that it can be used to test herbicide management strategies for their resistance development potential. It is not capable of predicting exact plant densities, but trends can be predicted. It is not only applicable for *A. spica-venti*. It could also be applied for other annual weed species as long as there is enough data about the species to feed the model. Needed input data are for example seed production, germination rates, survival rates for seedlings and seed viability in the seed bank.

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