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Monitoring, warning, and decision support in winter wheat

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Authors:

Jørgen Aagard Axelsen¹,
Lise Nistrup Jørgensen²,
Christian F. Damgaard¹,
Annemarie Fejer Justesen²,
Marianne Bruus¹

¹) Institute of Bioscience, Aarhus University

²) Institute of Agroecology, Aarhus University

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Summary

Background

Winter wheat occupies a large part of the conventionally cultivated area in Denmark, and farmers commonly treat against fungal diseases 2 - 3 times per season. At least 1-2 of these treatments are specifically aimed at controlling Septoria. When treating with fungicides in June, farmers often add an insecticide to the application in order to be sure to avoid problems with aphids, even though the number of aphids has not exceeded the given threshold. Therefore, treatments with fungicides in winter wheat may be the reason for unnecessary treatments with insecticides, and it makes sense to develop a decision support system targeting Septoria and aphids at the same time.

In an earlier project, a detailed, mechanistic ecosystem model (SeptoriaSim) was developed to simulate the growth of both winter wheat and Septoria and the damage caused by Septoria to winter wheat, depending on temperature, solar radiation, humidity, and rainfall. The output from this model was both the yield of winter wheat with and without fungicide treatment and the revenue of the treatments, taking sales price of wheat and treatment costs into account. The revenue of a treatment is an essential parameter when deciding on pesticide treatments, and the model was found to perform well in trials in 2015.

Objective

The overall objective of this project was to develop a combined decision support system for Septoria and aphids in winter wheat by improving SeptoriaSim and extending it with an aphid module. The work to fulfill this objective was split into four immediate objectives:

1. Develop new monitoring methods for the two pests
2. Provide a stronger scientific foundation for warning models against Septoria and aphids
3. Describe the spatial variation of aphids
4. Evaluate the reliability of the warning method/decision support tools in field trials.

Investigations

Four types of investigations were carried out with the aim of both providing data for calibration of the model and enforcing the scientific background:

1. Investigations of the background level of Septoria spores causing the initial infestation of winter wheat
2. Investigations of the growth of Septoria in the winter wheat leaves
3. Investigations of the population development of all three cereal aphids in winter wheat from late May to mid-July
4. Detailed growth analysis of winter wheat under field conditions.

In 2016 and 2017, controlled trials were carried out, where treatments against Septoria were timed by: 1) SeptoriaSim using the original calibration, 2) the well-known Danish decision support system Crop Protection Online (CPO), 3) a humidity model, and 4) a number of fixed predefined treatments. The controlled trials were supplemented with a few trials in farmers' fields, where the farmers sprayed a small part of their fields according to SeptoriaSim.

Results

All controlled experiments showed positive gross effects of fungicide treatments. Concerning the net yield, which is most relevant for the farmers, all treatments showed very limited responses in 2016, ranging from averages of 3.97 hkg/ha in the triple standard scheme to 1.67 hkg/ha following the recommendations of CPO. Of the three models, CPO performed best in

2016 with the humidity model being second, but generally treating against Septoria in 2016 appeared not to increase net yields much.

In 2017, the average net yields of the Septoria treatments ranged between averages of 1.25 hkg/ha using the humidity model to 4.25 hkg/ha following SeptoriaSim. Following CPO gave the second best net yield, 3.2 hkg/ha. Following both the standard treatments and the three models gave higher average net yields in 2017 than the best performing one in 2016.

Four out of the seven experiments showed generally negative or neutral net yield responses to fungicide treatments, while the only ones showing clear positive treatments were the experiments with susceptible varieties from Flakkebjerg (Nakskov in 2016, and Hereford in 2017) and the medium susceptible variety Torp in Holeby in 2017. Both experiments with Sheriff in 2017 showed generally negative net yields. However, using the models produced slightly increased net yields in Sheriff in all cases, except in Flakkebjerg in 2017.

Looking at the averages of the 2016 treatments (across varieties), CPO performed best, the humidity model was number two and SeptoriaSim number three. In 2017, SeptoriaSim performed best, with CPO being second and the humidity model third. Generally, the net yields produced using the decision support systems were even to (susceptible varieties) or slightly higher (resistant variety) than the ones produced when treating according the standard schemes with three treatments. This especially applies to SeptoriaSim and CPO.

Concerning the number of treatments, SeptoriaSim produced fewest treatments against Septoria in the trials in 2017 (average 1.80) and most treatments (average 1.33) in 2016. CPO behaved oppositely by triggering most fungicide applications in 2017 (average 2.25) and fewest in 2016 (0.67). Over the two years, SeptoriaSim and CPO triggered a very similar number of treatments (average 1.46 and 1.55, respectively), while the humidity model triggered 1.6. The results from the trials in farmers' fields show low and non-significant increases in yield and 1000-grain weight in the trial plots, and the Septoria treatment frequency was slightly lower in the trial plots than outside the trial plots. The aphid treatment frequency was the same in the trial plots as outside the trial plots.

The field data from the project was used to re-calibrate SeptoriaSim, and the recalibrated version was tentatively tested by repeating the projections carried out in May - June 2016 and 2017 to decide on timing of treatments. The number of treatments using weather files from 2016 reduced the average treatment frequency by 0.33 compared to the original version, while the new version reduced the average treatment frequency by 0.5 treatments in 2017. This means that the recalibrated version of SeptoriaSim would have released clearly fewer treatments than the originally used version, making it environmentally speaking clearly the most attractive model.

The aphid module of SeptoriaSim was only tentatively tested in field trials at five farmers' field tests, where the average yield in the trial plots was even to the yield outside the trial plots. The average number of treatments against aphids was 0.2, which was reduced slightly by using the recalibrated version of SeptoriaSim. SeptoriaSim did not justify tank mixes at any time. The results on the spatial distribution of aphids suggest that the initial density does not vary strongly between fields within a distance of 10 km.

Discussion and conclusion

The number of treatments is discussed in relation to national averages in number of treatments and/or Treatment Frequency Indices, and it is concluded that using all three decision support systems can reduce the number of Septoria treatments and that SeptoriaSim also can reduce the number of aphid treatments.

The results from the investigation of the spatial distribution of aphids suggest that it is possible to use regional estimates of initial aphid densities as input for the aphid module of Septoria-Sim.

Perspectives

This project has shown that a decision support system based on biological knowledge and projections of economic revenue of treatments may produce net yields equivalent to or better than the empirically based CPO. One important difference between the two decision support systems is that SeptoriaSim does not tell the farmer to spray. Instead, it gives predictions of the economic revenue of treating, and then it is up to the farmer to decide whether to treat or not. This decision may depend on the farmer's economic situation and his environmental attitude. This type of decision support system is an innovation that has the capacity to avoid treatments that may be economically beneficial, but may only produce very low revenues that farmers do not regard worth the effort.

The model system of SeptoriaSim is a general model type that can be used for other crops and other pests. Thus, it will be relatively simple to add other winter wheat pests to the system, and a similar system can be established for oilseed rape and its insect pests without much effort.

Administrative perspectives

One of the immediate objectives of the project was to develop new monitoring systems for the two pests, Septoria and aphids, and the results might be transformed into actions, making it attractive for farmers to use a decision support system on these two pests.

With one or more well-functioning decision support systems and the result that necessary input parameters can be applied to larger geographical areas, it is possible to qualify the decisions concerning pesticide treatments in winter wheat. The idea is to establish a system consisting of:

1. regional reporters who monitor the basic input of initial aphid densities and Septoria spore influx for SeptoriaSim
2. the reporters enter their results to a central database
3. a central service runs SeptoriaSim weekly, based on the input from the database, and identifies regional demands for control operations
4. the central service emits regional alerts to the farmers.

Such a system might help farmers make qualified decisions concerning Septoria and aphid control to the benefit of both farmers' economy and the environment

Sammenfatning

Baggrund

Vinterhvede optager en stor del af det konventionelt dyrkede areal i Danmark og behandles normalt 2 - 3 gange pr. sæson, hvoraf mindst 1-2 af disse behandlinger normalt er specielt rettet mod Septoria. Ved behandling med fungicider i juni tilsætter landmænd ofte et insekticid til tanken for at sikre sig mod problemer med bladlus, selvom antallet af bladlus ikke har overskredet skadetærsklen. Derfor kan behandlinger med fungicider i vinterhvede forårsage unødvendige insekticidbehandlinger, og det giver derfor mening at udvikle et beslutningsstøttesystem, der retter sig mod Septoria og bladlus på samme tid.

En detaljeret, mekanistisk økosystemmodel (SeptoriaSim) blev udviklet i et tidligere projekt til at simulere væksten af både vinterhvede og Septoria, samt den skade Septoria forårsager på vinterhvede afhængig af temperatur, solstråling, fugtighed og nedbør. Outputet fra denne model var udbyttet af vinterhvede med og uden Septoriabehandlinger, samt nettoindkomsten af behandlingerne. Fortjenesten af behandling er en vigtig parameter for beslutninger om pesticidbehandlinger, og modellen viste sig at fungere godt i forsøg i 2015.

Formål

Det overordnede formål med dette projekt var at udvikle et kombineret beslutningsstøttesystem til Septoria og bladlus i vinterhvede ved at forbedre den oprindelige udgave af SeptoriaSim og udvide det med et bladlusemodul. Projektet opererede med fire delmål:

1. Udvikle nye overvågningsmetoder for de to skadevoldere
2. Styrke det videnskabelige grundlag for varsling mod Septoria og bladlus
3. Beskrive den rumlige variation af bladlus
4. Afprøve pålideligheden af beslutningsstøttesystemet i feltforsøg.

Undersøgelser

Der blev udført fire forskellige undersøgelser, som havde til formål både at levere data til kalibrering af modellen og udbygge den videnskabelige baggrund for modellen:

1. Undersøgelser af baggrundsniveauet af Septoria-sporer, der forårsager infektionen af vinterhveden
2. Undersøgelser af væksten af Septoria i vinterhvedeblade
3. Undersøgelser af populationsudvikling hos alle tre kornbladlusearter i vinterhvede fra slutningen af maj til midten af juli
4. Detaljeret vækstanalyse af vinterhvede under markforhold.

I 2016 og 2017 blev der foretaget kontrollerede forsøg, hvor timingen af behandlinger mod Septoria blev styret af: 1) SeptoriaSim ved hjælp af den oprindelige kalibrering, 2) det velkendte danske beslutningsstøttesystem Crop Protection Online (CPO), og 3) en fugtighedsmodel, i sammenligning med række faste foruddefinerede behandlinger. De kontrollerede forsøg blev suppleret med markforsøg hos landmænd, hvor de behandlede en lille del af en vinterhvedemark for Septoria og bladlus efter varsler fra SeptoriaSim.

Resultater

Alle behandlinger mod Septoria i de kontrollerede forsøg gav et merudbytte, men nettomerudbyttet, som er det mest relevante for landmændenes økonomi, viste begrænsede effekter i 2016. Effekterne lå fra gennemsnit på - 3,97 med tre standardbækæmpelser til 1,67 efter anbefalinger fra CPO. Af de tre modeller fungerede CPO bedst i 2016, hvor fugtighedsmodellen var

anden, men generelt havde Septoriabehandlingerne ikke stor effekt på nettomerudbyttet i 2016.

I 2017 varierede nettoudbyttet af Septoria-behandlingerne mellem gennemsnit på 1,25 hkg / ha ved anvendelse af fugtighedsmodellen til 4,25 hkg / ha ved anvendelse af SeptoriaSim. På andenpladsen gav CPO det næstbedste nettoudbytte på 3,2 hkg / ha. Både standardbehandlingerne og behandlinger efter de tre modeller gav højere gennemsnitlige nettoudbytter i 2017 end den bedste i 2016.

Fire ud af de syv eksperimenter viste generelt negative eller neutrale nettoudbytter på fungicidbehandling, mens de eneste, der viste klare positive effekter, var eksperimenterne med modtagelige sorter fra Flakkebjerg (Nakskov i 2016 og Hereford i 2017) og den medium modtagelige sort, Torp, i Holeby 2017. Begge eksperimenter med Sheriff i 2017 viste generelt negative nettoudbytter. Ved brug af beslutningsmodellerne forøgedes nettoudbyttet i Sheriff lidt i alle tilfælde, undtagen i Flakkebjerg 2017.

Ser man på gennemsnittet af 2016-behandlingerne (på tværs af sorter), klarede CPO sig bedst, fugtighedsmodellen var nummer to, og SeptoriaSim nummer tre. I 2017 klarede SeptoriaSim sig bedst med CPO som anden, og fugtighedsmodellen tredje. Generelt var nettoudbytterne produceret ved hjælp af beslutningsstøttesystemerne nogenlunde ens med (modtagelige sorter) eller lidt højere (resistent sort) end dem, der blev opnået ved standardbehandlingerne med tre behandlinger. Dette gælder især for SeptoriaSim og CPO.

Med hensyn til antallet af behandlinger udløste SeptoriaSim færrest behandlinger mod Septoria i forsøgene i 2017 (gennemsnitlig 1,80) og fleste behandlinger (gennemsnitlig 1,33) i 2016. Omvendt udløste CPO flest fungicidbehandling i 2017 (gennemsnitlig 2,25) og færrest i 2016 (0,67). I løbet af de to år udløste SeptoriaSim og CPO stort set samme antal behandlinger (gennemsnitlig henholdsvis 1,46 og 1,55), mens fugtighedsmodellen udløste 1,6.

Resultaterne fra forsøgene i landmændenes marker viste lave og ikke-signifikante stigninger i udbytte og 1000-kornvægt i forsøgsfelterne, og Septoria-behandlingsfrekvensen var lidt lavere i forsøgsfelterne end i resten af marken. Bladluse-behandlingsfrekvensen var den samme i forsøgsplottene som uden for forsøgsplottene.

Feltdata fra projektet blev brugt til at re-kalibrere SeptoriaSim, og den re-kalibrerede version blev tentativt testet ved at gentage de simuleringer, der var blevet udført i maj - juni 2016 og 2017. Antallet af behandlinger ved hjælp af simuleringer med vejrfiler fra 2016 reducerede den gennemsnitlige behandlingsfrekvens med 0,33 i forhold til den oprindelige version, og antallet af behandlinger blev reduceret med gennemsnitlig 0,5 i 2017. Det betyder, at den re-kalibrerede version af SeptoriaSim ville have udløst klart færre behandlinger end den oprindelige brugt version, hvilket gør den til den miljømæssigt klart mest attraktive model.

Bladluse-modulet i SeptoriaSim blev tentativt testet i feltforsøg hos fem landmænd, hvor gennemsnitsudbyttet i forsøgsplottene var på samme niveau som udbyttet uden for forsøgsplottene. Det gennemsnitlige antal behandlinger mod bladlus ved brug af SeptoriaSim var 0,2, hvilket blev reduceret en smule ved anvendelse af den re-kalibrerede version. SeptoriaSim retfærdiggjorde ikke tankblandinger på noget tidspunkt

Resultaterne angående den rumlige fordeling af bladlus antyder, at de oprindelige tætheder ikke varierer voldsomt inden for en afstand af 10 km.

Diskussion og konklusion

Antallet af behandlinger diskuteres i forhold til det nationale gennemsnit og / eller behandlingshyppighed, og det konkluderes, at antallet af Septoria-behandlinger kan reduceres ved anvendelse af hvert af de tre beslutningsstøttesystemer, og at SeptoriaSim kan også reducere antallet af behandlinger imod bladlus.

Resultaterne fra undersøgelsen af den rumlige fordeling af bladlus foreslår, at det er muligt at anvende regionale estimater af initiale bladlusetætheder som input for bladlusmodul i SeptoriaSim.

Perspektiver

Dette projekt har vist, at et beslutningsstøttesystem baseret på biologisk viden og fremskrivninger af økonomiske gevinster af behandlinger kan bevirke nettoudbytter af omkring samme størrelse eller højere end den empirisk baserede PVO. En vigtig forskel mellem de to beslutningsstøttesystemer er, at SeptoriaSim ikke anbefaler landmanden at sprøjte. I stedet giver det forudsigelser om de økonomiske gevinster ved at sprøjte, og så er det op til landmanden at afgøre, om han vil behandle eller ej. Denne beslutning kan afhænge af landmandens økonomiske situation og hans miljømæssige holdning. Denne type beslutningsstøttesystem er innovativ, og den giver mulighed for at undgå behandlinger, der nok kan være økonomisk fordelagtige, men som giver så lave nettoudbytter, at landmændene ikke anser dem for at være indsatsen værd.

Modelsystemet SeptoriaSim er af en generel modeltype, der også kan bruges til andre afgrøder og andre skadedyr. Det vil således være relativt enkelt at tilføje andre af vinterhvedens skadedyr til systemet, og et lignende system kan relativt let etableres for raps.

Administrative perspektiver

Et af projektets mål var at udvikle nye overvågningssystemer for de to skadevoldere, Septoria og bladlus, og resultatet kan bruges til at skabe et beslutningssystem, der er attraktivt for landmændene at anvende.

Med et eller flere velfungerende beslutningsstøttesystemer og det resultat, at nødvendige startparametre (initiale tætheder) kan anvendes for større geografiske områder, er det muligt at kvalificere beslutningerne vedrørende pesticidbehandlinger i vinterhvede. Tanken er at etablere et system bestående af:

1. regionale rapportører, der bestemmer regionale initiale værdier for bladlusetætheder og flux af Septoria sporer.
2. Rapportørerne indtaster deres resultater i en central database
3. En central service kører SeptoriaSim ugentligt, baseret på input fra databasen, og udarbejder regional varsler om bekæmpelse af Septoria og bladlus, evt. i tankblanding.
4. Den centrale tjeneste udsender de regionale varsler til landmændene.

Et sådant system kan hjælpe landmændene med at træffe kvalificerede beslutninger vedrørende bekæmpelse af Septoria og bladlus til gavn for både landmændenes økonomi og miljøet.

1. Introduction

Winter cereals occupy 34% of the conventionally cultivated area in Denmark (Danish Environmental Protection Agency, 2012) and winter wheat constitutes a very large part of the winter cereals and was grown on 617,480 ha in 2015 (SEGES, 2016), corresponding to 23% of total Danish agricultural area. About 50% of the Danish fungicidal load and almost 30% of the insecticidal load in Danish agriculture are due to the cultivation of winter cereals, where Septoria (*Zymoseptoria tritici*) is the dominant fungal disease and aphids are the dominant pests (Videnscenter for Landbrug, 2011, 2012, 2013, 2014). The need for control of Septoria and aphids varies considerably from year to year, and a better and more robust foundation for decisions concerning spraying may reduce the number of unnecessary treatments. Leaf disease control is considered the most profitable treatment in wheat, and it provides a positive net return in most seasons. Wheat is commonly treated 2 - 3 times per season, of which at least 1-2 of these treatments are specifically aimed at controlling Septoria. When treating with fungicides in June, it is common to add an insecticide to the tank in order to be sure to avoid problems with aphids (Axelsen et al., 2012), even when the number of aphids has not exceeded the given threshold. Therefore, treatments with fungicides in winter wheat may be the reason for unnecessary treatments with insecticides, and it makes sense to look at these two problems together.

As applying routine tank mixes with both fungicides and insecticides may lead to economically unjustified insecticide treatments, it is relevant to develop a decision support system in winter wheat that takes both Septoria and aphids into consideration at the same time. In addition, the widespread use of tank mixes with insecticides together with fungicides could also mean that a reduction in fungicide use can be expected to lead to a reduction in insecticide use.

Based on the above assumptions, the first two working hypotheses of the project described in this report are:

1. A decision support system that provides estimates of aphids' significance for the net yield in winter wheat at the farm level can support a reduced use of insecticides.

2. A reduction in the control intensity of Septoria causes a reduction in insecticide use.

Between 2013 and 2015, two new decision support tools were developed to assess the need for controlling Septoria (Bligaard et al., 2017). One model, called the Leaf Moisture Model, seeks to assess the need for control based on the extent of continuous leaf moisture above a given threshold value. The second model, SeptoriaSim, simulates the growth of winter wheat under the influence of light, temperature, and the Septoria fungus, which by its growth reduces the photosynthetic active area of winter wheat, which, in turn, reduces the simulated yield. The performance of these two models was compared with the performance of the existing decision support system Plant Protection Online and three standard treatments in trials in 2015. This trial was not conclusive, but showed significantly better net yields when using the decision support tool instead of the standard treatments. It was therefore considered worthwhile to continue the trials in order to assess the validity of the third working hypothesis:

3. It is possible to reduce the number of Septoria control treatments in winter wheat without economic losses using the SeptoriaSim and / or leaf moisture model for decision support.

Therefore, the overall objective of this project was **to develop a combined decision support system for Septoria and aphids in winter wheat** by improving SeptoriaSim and extending it with an aphid module. The work to fulfill this objective was split into four immediate objectives:

1. Develop new monitoring methods for the two pests
2. Provide a stronger scientific foundation for warning models against the two pests
3. Describe the spatial variation of aphids and develop stochastic spatial models that can be used to include uncertainties in warnings against aphids
4. Try out the reliability of the warning method/decision support tools in field trials.

1.1 Work packages

The work in this project was split into four work packages.

1.1.1 WP 1. Fundamental research on Septoria.

In this WP, the effort was split into two topics: 1) the background load of spores that causes the initial *Septoria* infection was investigated throughout spring and summer, and 2) the development of a *Septoria* infection by measuring Septorial DNA and estimating Septorial cover percentage. The results were used to improve the scientific background of SeptoriaSim and were used for calibration of SeptoriaSim.

1.1.2 WP 2. Aphids

In WP 2, the population development of aphids was followed about weekly from late May to mid-July in 12 fields placed in 3 clusters in two growing seasons. Each cluster contained a central field and fields situated about 3, 6 and 10 km from the centre field.

The data were used for three purposes:

1. Calibration of the aphid module of SeptoriaSim
2. A spatio-temporal stochastic aphid population growth model was developed and fitted to the data using a Bayesian hierarchical fitting procedure. The fitted spatio-temporal population growth model was used to generalize existing deterministic aphid forecasting models with the effect of stochastic spatial variation.
3. The fitted model and complementary spatial statistics were used to investigate the hypothesis that initial aphid population sizes and epidemics may be predicted in fields within a 10 km radius of the nearest aphid-monitoring site.

1.1.3 WP 3. Growth analysis of winter wheat

In this WP, the growth of winter wheat was analysed in detail. The data were used to calibrate the winter wheat module of SeptoriaSim.

1.1.4 WP 4. Field trials

In this WP, the performance of SeptoriaSim was tried out in comparison with the humidity model, Plant Protection On-line, and standard treatments in controlled experiments at three research stations and in trials in farmers' fields.

2. Material and Methods

2.1 Warning models

2.1.1 Crop Protection Online

Decision support systems (DSS) have been developed for various reasons during the past 25 years. Overall, the systems aim at organizing complex information in a user-friendly way. The aim of reducing the input of pesticides in general has also been a leading factor in the Danish system PC-Plant Protection (Secher, 1991), which was introduced to Danish farmers in 1993. The models underlying the system have been adjusted according to the results of validating trials (Henriksen et al., 2000), and in 2002 the system was introduced as a web-based DSS (Rydahl et al., 2003) and is now called Crop Protection Online (CPO).

CPO includes models for most relevant cereal diseases, including Septoria diseases, based on empirical data and include thresholds based on different levels of assessed diseases and/or weather data. The specific thresholds are adjusted according to growth stage and susceptibility to diseases of the grown varieties. For control of Septoria, a threshold of 4 days with precipitation (>1 mm) starting at growth stage 32 is used as the main cut off criteria in susceptible cultivars, and 5 days starting at GS 37 is used for more resistant cultivars (Henriksen et al., 2000).

2.1.2 Humidity model

Septoria attacks are driven by humidity events during the growing season. In a previous project, a new prediction risk model for Septoria leaf blotch was developed based on measurements of relative humidity and leaf wetness parameters. A model based on 20 hours' continuous humidity was developed, and one hour with humidity was counted using either relative humidity above 85% measured at 2 m more than 30 min leaf wetness or rain during an hour. The model was tested based on historical weather data from ten years and 10 Danish sites. The prediction values of the model were linked to disease events and yield responses in specific years in order to see whether the model could adjust to major seasonal variations (Bligaard et al., 2017).

2.1.3 SeptoriaSim

SeptoriaSim was programmed to carry out:

1. biologically detailed simulations of the growth of winter wheat depending on solar radiation, temperature, and available nitrogen.
2. biologically detailed simulations of both the growth of Septoria within the wheat leaves and the vertical propagation of Septoria spores up through the winter wheat canopy
3. biologically based simulations of the damage caused by Septoria on winter wheat
4. calculations of the net yield of one or more fungicide treatments
5. projections of the growth of winter wheat and Septoria fungus based on real weather measurements combined with two-day weather prognoses and historical weather data from 11 years (2003 – 2013)
6. prognoses of the optimal timing of Septoria control applications based on the projected net yields.

SeptoriaSim can be regarded as consisting of four modules: 1) a winter wheat module, 2) a Septoria module, 3) an aphid module, and 4) a decision support module, where the net yield is calculated.

2.1.3.1 Description of simulation model

The simulation model is a physiologically based metabolic pool simulation model (Gutierrez, 1996) created to simulate the growth of annuals, perennials and trees dependent on rainfall, solar radiation and temperature. The model is based on plant growth models published by Gutierrez et al., (1988), Graf et al, (1991, 1992) and Sønderskov et al. (2006). The model type is a general population model, which can be used to simulate the growth of almost any kind of organism, and it is able to handle a large number of interacting species in realistic ecosystems (Axelsen, 2009, Axelsen et al., 2009). Furthermore, it is capable of simulating the growth of plants and the competition between them (Sønderskov et al, 2006). The basic element in the model is the population, and a population of plants consists of interacting populations of roots, stems/trunks, leaves, buds, flowers, fruits/grains, and, in the case of perennial species, storage organs.

2.1.3.1.1 The winter wheat module

When running a metabolic pool type simulation model (Figure 1), the initial step is to read the species specific input parameters of the involved species, the starting conditions, and the relevant weather file. Then, the model will enter a daily loop where the first step is to calculate the amount of carbon compounds (sugar from photosynthesis) required to fulfil the innate temperature dependent demand for growth and reproduction. The second step is to calculate the amount of carbon compounds produced based on the carbon assimilated by photosynthesis dependent on the available solar radiation and the area of leaves and green stems. The third step is to distribute the produced carbon compounds to the different organs of the plant according to where it is required, following an order of priority. The order of priority in woody plants is fruits, flowers and buds (fruits, flowers and buds are not important when simulating young trees only) and then roots, leaves and aerial wooden parts (trunk and branches). In annual species, such as winter wheat, the order of priority is seeds/grains, flowers, buds, stems, and finally roots and leaves proportional to their requirements. The final step in the daily loop is to take care of ageing, growth and mortality, and store the daily numbers and biomass of the involved populations in the memory. Finally, after having run through the daily loop during the simulated period, the model will give graphical output of the results, a graphical output that can be controlled from the user interface.

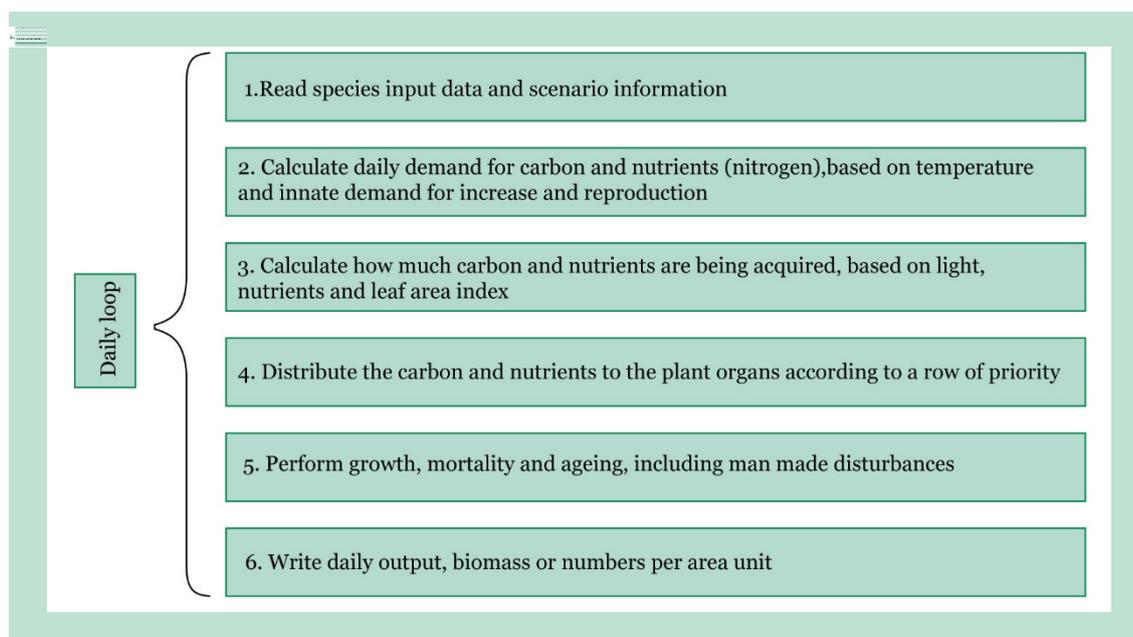


FIGURE 1. A diagram showing the steps in a simulation with SeptoriaSim.

During the calculation of the demand for carbon compounds in step 1 of the daily loop, the demand for respiration, growth, reproduction and storage for each organ type is calculated. Then, the produced carbon compounds (step 2) have to be distributed to the different organs in step 3, and the carbon compounds have to be distributed to cover the different demands, i.e. respiration, growth, reproduction and storage, within each organ. This is done in order of priority 1) respiration, 2) growth for growing organs, 3) creation of new individuals of the organ type, and 4) storage of surplus. This order of priority is important in the model, as it allows the simulation of the impact of shortage of carbon compounds. For instance, in case of competition for sunlight or drought, there will probably not be enough resources for storage. In cases of more serious shortage of carbon compounds, there may not be sufficient resources to create new organs (e.g. leaves) or let the existing ones grow. In case of severe shortage, there may not be sufficient resources for respiration, which means that a part of the population of the organs will die, and only the proportion that can be sustained will survive.

Growth and ageing are taken care of by a series of distributed delay procedures with attrition (Severini et al., 1998). A distributed delay procedure is a bookkeeping device, where a stage, for instance the growing stage of a leaf, is split in a number of sub-stages. As time goes by, the quantity in a sub-stage is transferred to the following sub-stage, and when leaving the last sub-stage of a stage, the quantity is transferred to the following stage, for instance growing leaves will be transferred to mature leaves. This procedure adds variation to the average transient time, a variation that is dependent on the number of sub-stages of a stage; the more sub-stages the smaller variation. The attrition, i.e. the removal or addition of quantities in a sub-stage, is done within the distributed delay procedure. The organs roots, stems, leaves, storage organs and fruits have the stages "growing" and "mature", while buds only have the stage "growing" and flowers only have a "mature" stage.

Time in the model is not in hours, days and months because this is the time scale of humans, not the time scale of plants. Plants are poikilothermic organisms for which all processes are temperature-dependent. It is, in fact, impossible to distinguish between temperature and time for plants. Therefore, the model uses the concept of physiological time and converts daily average temperatures to degree-days, which is a physiological time unit. This means that the time unit within the simulation model is degree-days, and the demand for carbon compounds, for instance, comes in units of grams per degree-day. The degree-day step of a day is calculated as the daily average temperature minus a threshold temperature under which the growth can be ignored. The use of degree-days as the time unit is an automatic way to take into account that the biochemical processes driving plant growth are temperature dependent. The only process in the model that does not follow the degree-day scale is the respiration rate, which is modelled as being dependent on the daily average temperature.

All plant organs in the model have minimum required nitrogen contents, and the photosynthesis efficiency of the leaves is dependent on the nitrogen contentment. Therefore, the uptake of nitrogen is important and is simulated as being dependent on the root biomass and available nitrogen. Consequently, removing roots will reduce the nitrogen uptake efficiency and result in a nitrogen shortage. On the other hand, removing leaves, for instance due to damage by *Sep-toria*, reduces the photosynthetic active leaf area and thereby affects the production of photosynthetic material, resulting in a shortage of carbon compounds available for growth.

The nitrogen dynamics in the model is simulated in nearly the same way as the carbon dynamics. Calculation of the demand for nitrogen is based on the demand for carbon and information on the maximum nitrogen content of the plant organ in question, and the amount of nitrogen acquired by a plant population is distributed to the different plant organs. This distribution is controlled using the same order of priority as the carbon, and there is priority within the different plant organs for nitrogen for respiration, growth, reproduction and storage. If there is not enough nitrogen to fulfil the demands for nitrogen, the growth may cease due to nitrogen

shortage and, in severe cases, death may occur if there is not enough nitrogen to fulfil the demand for respiratory nitrogen (the nitrogen content of the carbon compounds necessary to fulfil the demand for respiration). Altogether, this means that plant organs can die due to both shortage of nitrogen and carbon.

2.1.3.1.2 The Septoria model

The growth and spread of the Septoria fungus is largely simulated the same way as populations of plant organs in the winter wheat model, i.e. by: 1) calculating the need for respiration, growth, reproduction and storage, 2) calculating the resources obtained by growth in the host plant's leaves, 3) distributing resources according to orders of priority, and 4) aging, growth and mortality. This means that it is actually following the daily loop of Figure 1. Based on the described biology of the fungi (Ponomarenko et al 2011), various parameters have been estimated and included as part of the model. The fungus is simulated stage structured with the following stages: 1) spores, 2) penetrating stage (newly germinated hyphae growing on the leaf surface and trying to find a stomata through which they can penetrate the leaves), 3) a biotrophic stage, which is the stage growing inside the leaves, 4) a necrotrophic stage, where the fungus can be seen on the leaves, and finally 5) a mature stage that does not grow, but only takes care of spore production and spreading.

In the model, Septoria acts as an herbivore living within winter wheat leaves and its growth depends on the available amount of leaves. In cases where Septoria has "eaten" all the winter wheat leaves, it will "starve to death". In order to simulate Septoria's vertical spread, the Septoria "population" in the fields is divided into a number of horizontal "population layers" that suit the horizontal layers in the winter wheat module. The current version uses five horizontal layers, which means that there are five Septoria populations in the model.

The spread of spores from one horizontal layer to the next depends on the daily amount of precipitation. Spreading of spores to higher layers is possible only if the daily amount of precipitation exceeds a threshold. Spore spreading also requires the formation of spores in Septoria's mature stage. If the conditions for vertical spread of spores are not met, spores can germinate within the horizontal layer in which they were produced, if the conditions for germination and growth in the penetrating stage are met (i.e. not too dry).

The germination of spores and growth in the penetrating stage is highly dependent on relative humidity, which means that spores do not germinate if the relative humidity is below a threshold, and at the penetrating stage Septoria will die if the relative humidity is below the threshold. Therefore, a good vertical spread of Septoria requires rain that must be followed by a period of approximately two days with relative humidity above the threshold.

In the model, fungicides are programmed to kill the stages that are exposed on the winter wheat surface, i.e. the penetrating stage and the mature stage, and to leave the stages within the leaves untouched. Use of fungicide will protect existing winter wheat leaves against Septoria attacks for ten days, while newly created leaves will be susceptible to infection immediately. The calculation of Septoria's degree of coverage on the winter wheat leaves was based on the balance between the Septoria biomass and the biomass of leaves, modified by a constant. This calculation made it possible to compare simulated Septoria infections with observed infections from the fields.

Model output

When running the model, it carries out at least three simulations: 1) a simulation of winter wheat growth and yield without Septoria attack, 2) a simulation of winter wheat growth and yield under the influence of Septoria, and 3) a simulation of winter wheat growth and yield under the influence of Septoria, where Septoria is being controlled by fungicide applications. For each additional fungicide application, it will run one more simulation. The model stores the

yield of all simulations, and by subtracting the cost of the fungicide/insecticide and the delivery cost, the model calculates the economic revenue of each treatment. The number of treatments and the timing of them are controlled from the user interface.

Prognosis

The model makes a prognosis by calculating the yield based on a number of combined weather files. The combined weather files are composed of: 1) currently measured weather data from sowing date to the actual date, 2) 4-day forecast data from the Danish Meteorological Institute based on zip code; and 3) the rest of the growing season from historical weather files from the nearest weather station. Historical weather files from 2003 to 2013 are used. Forecasts are made by starting out with a simulation using combined weather files using historical weather data from 2003. The next simulation uses a combined weather file with a historical weather file from 2004, the following simulations will use files with historical weather data from 2005, 2006, ... , 2013. After each simulation, the model displays the average output from the simulations that have been carried out. This means that after the simulation using historical data from 2013, a forecast is based on results from ten simulations (with ten different real weather regimes). The results from all ten simulations are also displayed, which gives an impression of the variation between years (depending on the weather of each year). The results come in terms of both yield (t / ha) and farmer's economic revenue.

Assessment of optimal timing of applications

The program can run a series of forecasts of the yield if a treatment is carried out on one of the days in a given period, for example between 5th and 15th of May. The first simulation uses the treatment date given in the user interface, the second simulation will use the treatment date + one, the third simulation will use the treatment day +2, etc. up to treatment day + nine. The timing of the treatment that gives the best economic benefit is displayed as the optimal timing result of the simulation. Such calculations can also be made with several application dates, which in the simulations must be 14 days apart. In cases with optimization of timing of more applications, all applications will be displaced by the same figure. If the date of one or more of the simulations has been exceeded, the exceeded date will not be displaced.

2.1.3.2 SeptoriaSim – mathematical description

2.1.3.2.1 The winter wheat model

The mathematical description follows the daily loop of Figure 1, which means 1) calculation of demands, 2) calculation of supply, 3) distribution of supplies, and 4) book keeping of growth, mortality and ageing in the population.

2.1.3.2.1.1 Calculation of demands

All calculations of demands were made in units of $g \times ^\circ D^{-1}$, where $^\circ D$ signifies degree-days. The daily degree-day step was calculated as

$$\Delta(^{\circ}D) = T_{avg} - T_0 \quad (1)$$

where T_{avg} was the daily average temperature and T_0 was the thermal threshold under which growth was ignored (Begon et al., 1990).

The daily demand for carbon compounds of a plant organ D_i was calculated as the sum of demands for respiration, growth, reproduction, and storage of the particular organ (2)

$$D_i = D_{resp,i} + D_{g,i} + D_{rep,i} + D_{s,i} \quad (2)$$

where $D_{resp,i}$ was the demand for respiration of the organ i , $D_{g,i}$ was the demand for growth of organ i , $D_{rep,i}$ was the demand for reproduction, and $D_{s,i}$ was the demand for storage. The demand for respiration of an organ was generally calculated according to equation (3)

$$D_{\text{resp},i} = M_i \times z_0 \times 2^{(0.1 \times T_{\text{avg}})} \quad (3)$$

where M_i was the mass of the organ i and z_0 was the basic respiration rate.

The demand for growth was calculated as (4)

$$D_{g,i} = M_{g,i} \times r_g^{\Delta(^{\circ}\text{D})} \quad (4)$$

where $M_{g,i}$ was the mass of the growing stage of the organ i and r_g was the innate growth rate in units of $g \times \text{degree-days}^{-1}$. The demand for reproduction, which means the demand for producing new individuals of the plant organ, e.g. new leaves or buds, was made dependent on the mass of the entire plant and not only on the mass of the particular plant organ (5)

$$D_{\text{rep}} = M_{\text{tot}} \times r_{\text{rep}}^{\Delta(^{\circ}\text{D})} \quad (5)$$

where M_{tot} was the mass of the entire plant and r_{rep} was the innate rate of reproduction in units of $g \times ^{\circ}\text{D}^{-1}$. The demand for storage was simply made a constant fraction of the mass of the organ (6)

$$D_s = a \times M_i \quad (6)$$

where a was a constant and M_i was the mass of the organ i . The total demand of the population of plants (D_{tot}) was calculated as (7)

$$D_{\text{tot}} = \sum_{i=0}^{\text{All organs}} D_i \quad (7)$$

2.1.3.2.1.2 Calculation of supply

The supply of carbon compounds (photosynthates) was based on the amount of assimilated solar energy, and the simulation of the assimilation of solar energy was made dependent on the leaf area index in a number of horizons, i.e. the canopy was split into a number of horizontal zones.

The amount of carbon compounds produced by photosynthesis by a plant population (a species) (M^*) was calculated by (8)

$$M^* = D_{\text{tot}} \times \left(1 - \exp\left(\frac{b \times E_p}{D_{\text{tot}}}\right) \right) \quad (8)$$

where E_p was the amount of solar energy available for the population p , and b was a constant converting solar radiation to g organic matter. When the solar radiation comes in W/m^2 and the output in grams, the constant b was 2.23×10^{-4} (Gutierrez et al., 1984). The amount of energy available for a population (a species) was calculated by (9)

$$E_p = \sum_{j=1}^L E_j \times (1 - e^{-\epsilon \times LAI_j}) \quad (9)$$

where j indicated the canopy horizon j , LAI_j was the leaf area index in the horizon j , ϵ was the light extinction coefficient, and L was the number of horizons. The leaf area index of a horizon was calculated at the summation of the leaf area indices of the species in the simulated ecosystem (10)

$$LAI_j = \sum_{sp=1}^N LAI_{sp} \quad (10)$$

where sp indicated the species, and N was the number of competing plant species in the simulated ecosystem. However, the facility to include more species was not used in the Septoria-Sim model, as the only plant species was winter wheat. The amount of solar energy available in the horizons was calculated according to (11)

$$E_i = E_0 - \sum_{j=L}^{i-1} E_j \quad (11)$$

where E_0 was the measured solar radiation above the upper horizon (L) originating from weather files, and E_i was the solar energy captured in the canopy horizon i , which means that the energy available for a horizon was the measured radiation (E_0) minus what was assimilated by the canopy layers above. In order only to operate with photosynthetic active radiation, the measured global solar radiation from weather files was multiplied by 0.55.

The vertical distribution of the plant organs except leaves was simulated by equation (12), which was modified from Graf et al., (1992)

$$p(x) = \left(1 - \frac{x}{h}\right) \times \left(\frac{x^{k_1}}{h^{k_2}}\right) \quad (12)$$

where $p(x)$ was the fraction of the biomass of a plant organ at the height x , h was the actual plant height, and k_1 and k_2 were constants. If $k_1 = 4$ and $k_2 = 5$, the equation distributed the biomass as having the largest part close to the height h , and when using lower values of k_1 and k_2 such as $k_1 = k_2 = 1$, the distribution was almost even up through the vegetation layer.

The vertical distribution of leaves was controlled by a series of leaf populations, a leaf population for each of the horizontal layers mentioned in connection with equation 9 and 10. The layers were populated by new leaves as they emerged in the simulation, and the simulated height of the wheat plant determined into which "layer population" a newly created leaf was placed. If, for instance, the height was within the limits of layer 2, the newly created leaves were placed in the leaf population of layer 2, and they remained in this layer throughout their lifetime.

2.1.3.2.1.3 Allocation of supplies

The resources available for respiration, growth, reproduction and storage (M^*) were distributed to the different demands of the different organs according to the previously mentioned order of priority. The priority between demands within an organ type was always respiration, growth for the growing stage, reproduction for the mature stage and, finally, to put something on stock if anything left. The amount of supplies (carbon compounds) available for the different demands (M_d) was calculated by (13).

$$M_d = d \times \left(\frac{\sigma}{d}\right) \quad (13)$$

where d represented the demand for either respiration, growth, reproduction or storage, σ was the actual supply, and σ/d was the supply – demand ratio controlling the proportion of the demand being satisfied. The actual supply, σ , decreased after allocating resources to a demand and eventually hit zero because equation (7) did not allow acquiring more carbon compounds than demanded.

2.1.3.2.1.4 Book keeping of growth, mortality and ageing

The simulation of how the populations grow in numbers and biomass was done by aid of a distributed delay procedure, where the quantities in transition were described by a series of differential equations (14)

$$\begin{aligned} \frac{dQ_1(t)}{dt} &= x(t) - r_1(t) - \mu_1 Q_1 \\ \frac{dQ_2(t)}{dt} &= r_1(t) - r_2(t) - \mu_2 Q_2 \\ &\dots \dots \dots \\ \frac{dQ_{k-1}(t)}{dt} &= r_{k-1}(t) - y(t) - \mu_k Q_k \end{aligned} \quad (14)$$

where Q_m was the quantity (mass or numbers) in sub-stage $m= 1,2,\dots,k$, t was the time in physiological time units ($^{\circ}D$), $x(t)$ was the input to the stage, $y(t)$ was the output from the stage, which was transferred to the following stage or died if the adult stage was concerned, and r_m was the flow from one sub-stage to the next. The term μQ_i took care of growth and mortality, where μ_i was the stage specific growth rate. The stage specific growth rate was calculated by (15)

$$\mu_i = \lambda_i \frac{Q_i + G_i}{Q_i} \quad (15)$$

where λ_i was the stage specific survival rate and G_i was the growth of the stage i .

2.1.3.2.1.5 The Septoria module

Similarly to the mathematical description of the winter wheat module in appendix 1, the mathematical description of *Septoria* followed the daily loop of Figure 2.1.3.1, which means 1) calculation of demands, 2) calculation of supply, 3) distribution of supplies, and 4) book keeping of growth, mortality and ageing in the population. The description of the *Septoria* module has equations overlapping the similar equations of the winter wheat module, but in order to assure readability, overlapping equations were maintained in this description.

2.1.3.2.1.5.1 Calculation of demands

All calculations of demands were done in units of $g \times ^{\circ}D^{-1}$, where $^{\circ}D$ signified degree-days. The daily degree-day step was calculated as

$$\Delta(^{\circ}D) = T_{avg} - T_0 \quad (1)$$

where T_{avg} was the daily average temperature and T_0 was the thermal threshold under which growth could be ignored (Begon et al., 1990).

The daily demand for carbon compounds of a fungus D was calculated as the sum of demands for respiration, growth, reproduction, and storage (2)

$$D = D_{resp} + D_g + D_{rep} + D_s \quad (2)$$

where D_{resp} was the demand for respiration, D_g was the demand for growth, D_{rep} was the demand for spore production, and D_s was the demand for storage. The demand for respiration was calculated according to equation (3)

$$D_{resp,i} = M_i \times z_0 \times 2^{(0.1 \times T_{avg})} \quad (3)$$

where M_i was the mass of the organ i and z_0 was the basic respiration rate.

The demand for growth was calculated as (4)

$$D_g = M_g \times r_g^{\Delta(^{\circ}D)} \quad (4)$$

where M_g was the mass of the fungus and r_g was the innate growth rate in units of $g \times \text{degree-days}^{-1}$. The demand for spore production was made dependent on the mass of the entire fungus population in the horizontal layer (5)

$$D_{rep} = M_{tot} \times r_{rep}^{\Delta(^{\circ}D)} \quad (5)$$

where M_{tot} was the mass of the entire fungus and r_{rep} was the innate rate of reproduction in units of $g \times ^{\circ}D^{-1}$. The demand for storage was simply made a constant fraction of the mass (6)

$$D_s = a \times M \quad (6)$$

where a was a constant and M was the mass.

Spores and penetrating stages were assumed not to have demands, as the spores are resting stages, and the penetrating stage grows on resources stored in the spores.

2.1.3.2.1.5.2 Calculation of supply

In the model, the acquisition of supplies was the result of a trophic interaction, namely a plant – herbivore interaction, as the fungus was regarded an herbivore feeding on the winter wheat leaves.

The acquired leaf mass by the fungus in the plant-herbivore interaction (M^*) was calculated by (7)

$$M^* = D \times \left(1 - \exp\left(-\frac{s \times M'}{D}\right) \right) \quad (7)$$

where M' was the amount of winter wheat leaf matter available, and s was a constant search rate describing the acquisition efficiency of the fungus consuming leaf matter (Gutierrez et al., 1984).

2.1.3.2.1.5.3 Allocation of supplies

The resources available for respiration, growth, reproduction and storage (M^*) were distributed to the different demands according to an order of priority. The order of priority between demands was respiration, growth for growing stages, reproduction for the mature stage and, finally, to put something on stock if anything was left. The amount of supplies available for the different demands (M_d) was calculated by (8)

$$M_d = d \times \left(\frac{\sigma}{d} \right) \quad (8)$$

where d represented the demand for either respiration, growth, reproduction or storage, σ was the actual supply, and σ/d was the supply – demand ratio controlling the proportion of the demand being satisfied. The actual supply, σ , decreased after allocating resources to a demand and eventually hit zero because equation (7) did not allow acquiring more carbon compounds than demanded.

2.1.3.2.1.5.4 Book keeping of growth, mortality and ageing

The simulation of how the populations grow in numbers and biomass was done by aid of a distributed delay procedure where the quantities in transition were described by a series of differential equations (9)

$$\begin{aligned} \frac{dQ_1(t)}{dt} &= x(t) - r_1(t) - \mu_1 Q_1 \\ \frac{dQ_2(t)}{dt} &= r_1(t) - r_2(t) - \mu_2 Q_2 \\ &\dots \dots \\ \frac{dQ_{k-1}(t)}{dt} &= r_{k-1}(t) - y(t) - \mu_{k-1} Q_{k-1} \end{aligned} \quad (9)$$

where Q_m was the quantity (mass or numbers) in sub-stage $m= 1,2,\dots,k$, t was the time in physiological time units ($^{\circ}D$), $x(t)$ was the input to the stage, $y(t)$ was the output from the stage, which was transferred to the following stage or died if the adult stage was concerned, and r_m was the flow from one sub-stage to the other. The term $\mu_i Q_i$ took care of growth and mortality, where μ_i was the stage specific growth rate. The stage specific growth rate was calculated by (15)

$$\mu_i = \lambda_i \frac{Q_i + G_i}{Q_i} \quad (15)$$

where λ_i was the stage specific survival rate and G_i was the growth of the stage i .

2.1.3.2.1.5.5 Spread of *Septoria* in winter wheat

Spread of *Septoria* upwards in the canopy was simulated only to take place in case of daily rainfall above a threshold or in case of rain intensity above a threshold. Once the weather had induced spreading in the canopy, the spore spreading was calculated according to equation 10

$$S_{i,j} = a_{j-i} \times S_i \quad (10)$$

$S_{i,j}$ was the number or biomass of spores to be relocated from layer i to layer j , S_i was the number or biomass of ripe spores in layer i , and a_{j-i} was the fraction of the ripe spores in layer i being spread to the layer $j-i$; This means that spores were relocated upwards between any layers, e.g. from layer 1 to 2, and 1 to 3, but only if there were leaves present in the receiving layers.

On days that weather conditions did not trigger a spread upwards through the canopy, it was possible to spread within the layer, as it was assumed that spores could spread from infected to healthy parts of leaves.

2.1.3.2.1.5.6 Impact of fungicides

Fungicides were simulated to kill the fungal stages present on the leaf surface (spores, penetrating and mature) on the day of the application and to prevent spore development the following 10 days on the surface of the leaves existing on the day of fungicide treatment. Typically, the fungicide treatments do not completely kill the fungi, but mainly protect the upper leaves. The fungal stages present within the leaves, biotrophic and necrotrophic, were not harmed by the fungicide and were allowed to continue their development and to spread spores normally when reaching the mature stage. Leaves emerging after the application of fungicides (e.g. GS 33) were not protected from infection by newly spread spores coming from windborne ascospores or pycnidia spores placed lower in the crop canopy.

2.2 Fundamental research on *Septoria*

2.2.1 Spore trapping

During the season, spores were trapped in four Burkard spore traps (Figure 3) placed at four different sites, one near Holeby – Lolland, one near Gedsergaard (2016 only) – Lolland, one at the trial site at Horsens, and two at Flakkebjerg – one outside and one within the crop (Table 1). The traps collected airborne particles by impaction onto a sticky tape, which was fixed onto a rotating drum. Every week, the tapes were changed and cut into pieces, each representing one day. DNA from all particles on the tape sections was extracted according to the method described by Duvivier et al. (2013). For each day, a QPCR test specific for *Z. tritici* was run to measure the quantity of *Septoria* spores collected on the tape. This provided a picture of the spore concentration released during the season, which might have an impact on the disease epidemic. Two types of spores are produced by *Septoria* – ascospores and pycnidia spores. Ascospores are wind-spread, while pycnidia spores are mainly splash-borne. The QPCR used in this study was developed and tested by Duvivier et al. (2013). The QPCR method enabled the quantification of *Septoria* DNA in a sample, but it could not distinguish pycnidia spores from ascospores. However, the spore traps were placed so that the orifice of the spore trap was one meter above ground to avoid trapping of pycnidia spores spread in water droplets. The amount of spores per day was calculated by preparing a standard curve based on a dilution series of a pycnidia spore suspension with a known concentration. A pycnidia spore contains four to eight nuclei per spore, whereas an ascospore contains 2 nuclei, thus, the measured numbers of spores per day may be higher if converted to ascospores. The detection threshold was estimated to be approximately 20 pycnidia spores (~70 ascospores) per daily tape section.

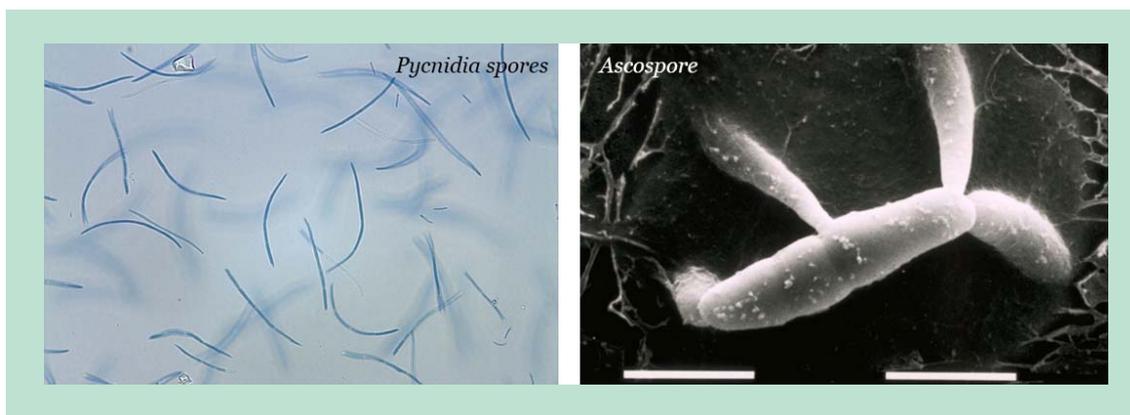


FIGURE 2. Pycnidia-spores and ascospores of *Septoria tritici*.

TABLE 1. Localities and periods at which spores were collected.

Location	2016		2017	
	Start of collection	End of collection	Start of collection	End of collection
Flakkebjerg 1, near wheat crop	21 April	10 November	6 April	28 November
Flakkebjerg 2, placed in a wheat crop	21 April	10 November	6 April	28 November
Horsens, placed close to a wheat field	22 April	12 July	6 April	8 October
Gedsersgaard – Gedser. Used for collection of beet pathogens – but used similarly for <i>Septoria</i>	29 June	30 September	-	-
Holeby - Lolland. Used for collection of beet pathogens – but used similarly for <i>Septoria</i>	8 April	21 October	6 April	3 December

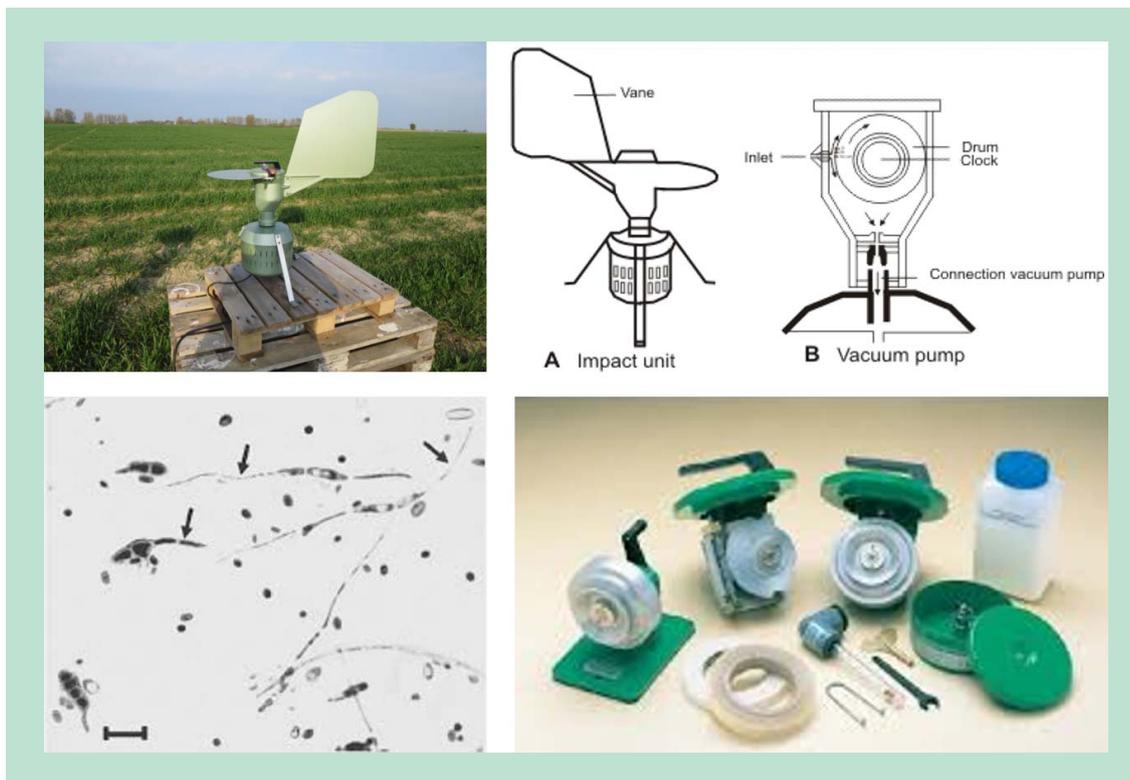


FIGURE 3. Photos of a Burkard 7-day volumetric spore trap located in the field. The trap is linked to a vacuum pump, and airborne particles are impacted onto a sticky tape that rotates at a speed so that it is equivalent to 1 week.

2.2.2 Progression of Septoria attacks – QPCR

During the 2016 and 2017 seasons, leaves were collected at regular intervals and were sorted into leaves with visible attack and leaves without visible attack. At the sampling time, growth stage and the level of *Septoria* were assessed on each of the leaf layers.

The leaves were transported to the laboratory, where they were dried at room temperature and DNA was extracted from the leaf samples. The level of *Z. tritici* DNA was measured by the QPCR method described by (Bearechell et al. 2005).

2.3 Aphids

2.3.1 Field sampling program

The aim of this sampling was to get data for the development of aphid populations for the aphid module of SeptoriaSim and to assess the spatial variation in aphid population growth in order to evaluate the uncertainty in predictions of damage due to aphids when based on data from the official aphid-sampling program (Observation Web, <https://www.landbrug-sinfo.dk/planteavl/plantevaern/varslingsregistrerings-net/-sider/startside.aspx>). In winter wheat fields at different distances to selected Observation Web fields, the percentage of straws infested with aphids was estimated weekly during the aphid seasons 2016 and 2017. Both in 2016 and 2017, 12 fields near Viborg, Borum and Hammel were included in the study (four fields at each place). In case the farmer wanted to spray the study field with insecticide, an unsprayed “window”, 50 m x boom length, was established for our samplings. Aphids were counted at species level for the three main species, the grain aphid (*Sitobion avenae*), the bird cherry - oat aphid (*Rhopalosiphum padi*) and the rose – grain aphid (*Metopolophium dirhodum*).

In 2016, the sampled fields were located app. 0, 3, 6 and 10 km from the Observation Web fields (Figure 4). Aphids were sampled weekly from late May until early July. On each sampling occasion, 100 or 80 wheat straws were carefully examined for aphids at five plots along a gradient in each field. This yielded a total of 400-500 inspected straws per field at each sampling date. The plots were at least 50 m apart.

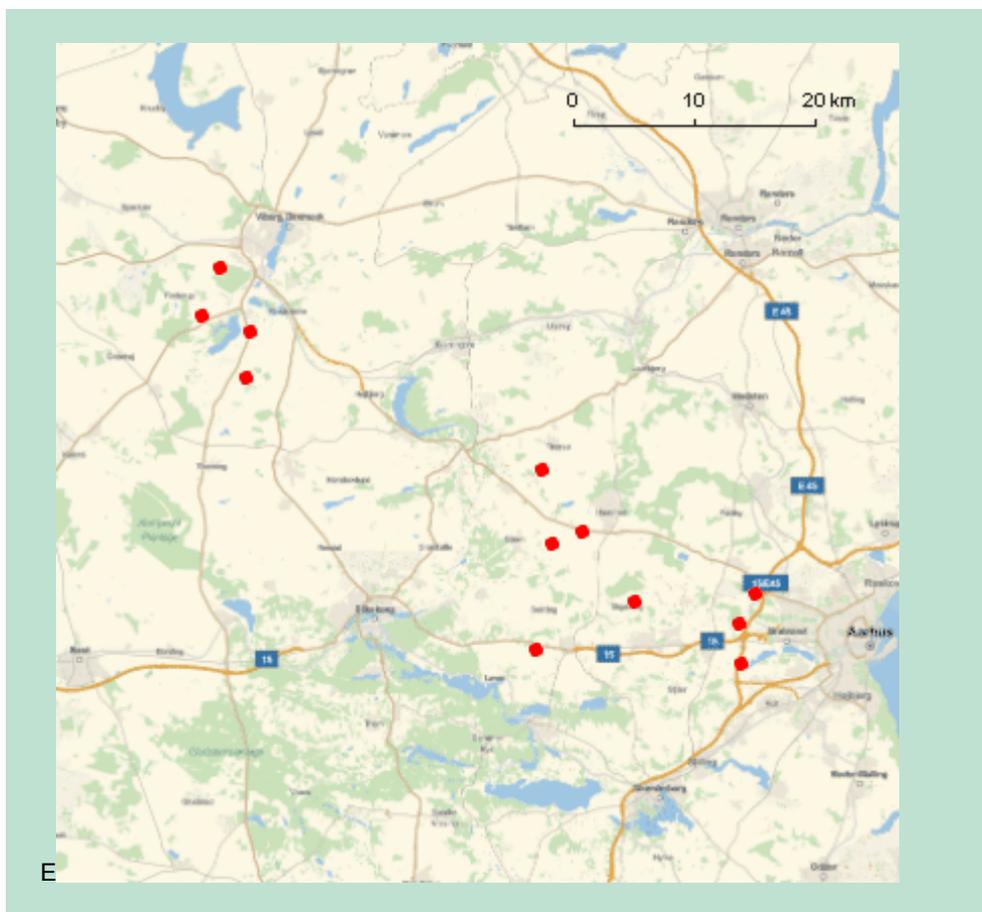


FIGURE 4. The locations of the 12 aphid sampling sites.

In 2017, the sampling strategy was modified to reduce time consumption and, at the same time, ensure a detailed sampling of the initial population growth, which proved the most critical data for reliably modelling aphid population development. The same farmers and the Observation Web fields were involved, but due to the rotation of crops between fields, the distances of other study fields from the Observation Web fields varied in a more irregular manner than in 2016. At the beginning of the aphid season, before any insecticide treatment was performed (end of May and beginning of June), aphid infestation was estimated in 10 plots per field, placed as indicated in Figure 5 and registered by GPS. Three plots were placed in three of the five windows that were to be kept unsprayed (blue). In each plot, aphid infestation was assessed in 10 spots, and in each spot, five wheat straws were inspected. I.e., 50 straws were inspected in each plot, and 500 per field. On the following sampling occasions, the five unsprayed windows were sampled, i.e. 250 straws per field were inspected for aphids.

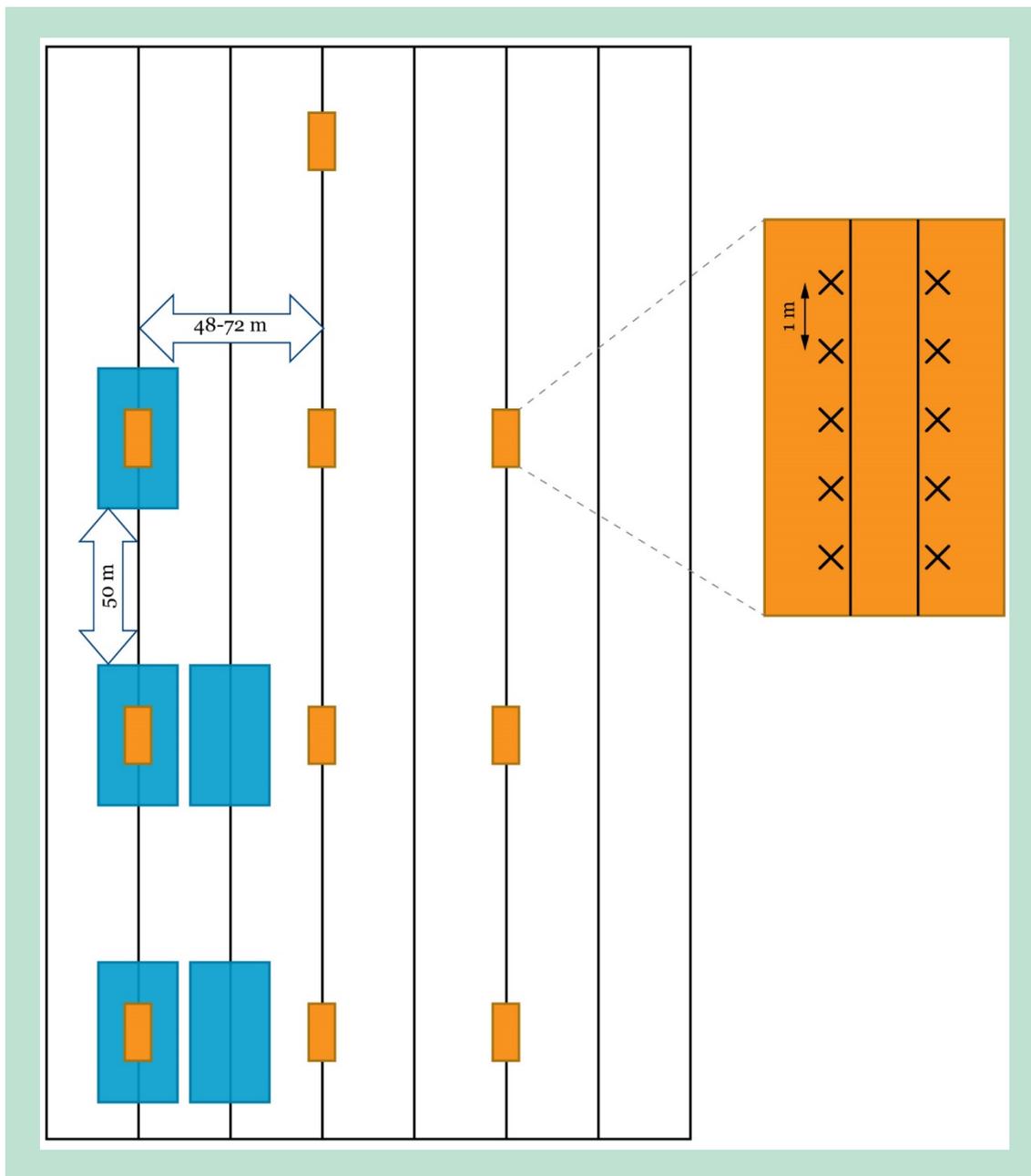


FIGURE 5. Outline of initial sampling in the unsprayed fields (in orange plots) and later sampling (in blue, unsprayed windows, but otherwise the same principle in 2017). In each plot, 50 wheat straws were inspected for aphids.

2.3.2 Spatial variation

In order to complement the result of the spatial variation (aphid sampling) of the initial phases of aphid epidemics obtained in the spatial modelling of the aphid occurrence data sampled in 2016, aphid occurrence was recorded more intensively at the same twelve sites in the beginning of the growth period the following year and analysed in a semivariogram. In 2017, aphid occurrence was recorded in ten sampling plots of 50 wheat plants at each site on 30. Maj and 7. June. The ten sampling plots within a site were laid out along three transects with at least 50 m between all sampling plots, and the exact geographical position of each plot was determined.

2.3.2.1 Statistical modelling of the aphid population

The spatio-temporal aphid occurrence data was modelled using Bayesian hierarchical methods (Clark & Gelfand 2006). The observed number of straws with at least one aphid at site i

and sampling plot k at degree-day t is denoted $y_{i,k,t}$ and is assumed to be binomially distributed with $n_{i,k,t}$, the number of straws sampled, and $p_{i,t}$, the occurrence probability that a straw has at least one aphid at site i at degree-day t ,

$$y_{i,k,t} \sim \text{Bin}(n_{i,k,t}, p_{i,t}) \quad (1)$$

The site-specific occurrence probability is modelled using an exponential function of degree-day t ,

$$p_{i,t} = p_{i,0} \text{Exp}(r_0 + r_1 t + r_2 t^2 + \epsilon_i) \quad (2)$$

where $p_{i,0}$ is the occurrence probability at a fixed initial day, r_0 , r_1 , and r_2 are population growth parameters, and ϵ_i are Gaussian distributed site-specific random effects, $\epsilon_i \sim \text{Gau}(0, \sigma_p^2)$.

The n site-specific initial occurrence probabilities are assumed to arise from a Gaussian process model,

$$p_{i,0} \sim \text{Gau}(\mu_0, \Sigma), \quad \Sigma = \begin{pmatrix} \sigma_0^2 & \dots & \sigma_0^2 \rho(d_{1,n}) \\ \vdots & \ddots & \vdots \\ \sigma_0^2 \rho(d_{n,1}) & \dots & \sigma_0^2 \end{pmatrix} \quad (3)$$

where μ_0 is the mean initial occurrence probability, σ_0^2 is the variance, and $\rho(d_{i,j}) = \rho_0 \text{Exp}(-\frac{d_{i,j}}{\alpha})$ with $d_{i,j}$ being the distance between site i and site j , α is the scale of the spatial effect that is set to 10 km, and ρ_0 is a parameter that measures the spatial covariance (Haran 2011; Ovaskainen et al. 2016). The covariance matrix by definition has to be positive definite, which puts upper and lower bounds on ρ_0 .

2.4 Growth analysis of winter wheat

The aim of this WP was to collect detailed height-zoned growth data on the two varieties of winter wheat, Torp and Mariboss. The data were used for calibration of the winter wheat module of the SeptoriaSim model.

The samples were taken throughout the growing season 2016 – 2017 in two fields situated about 10 km south of Viborg, Denmark (Coordinates: 56.386267, 9.383832). The variety in one of the two fields was Torp, which is susceptible to Septoria, and in the other field, the variety Mariboss, which is moderately susceptible to Septoria.

Samples were taken on the following dates: 10/13, 11/14, 12/20 in 2016, and 2/22, 3/23, 4/25, 5/5, 5/15, 5/31, 6/12, 6/22, 7/3, 7/17, 7/27 in 2017. This means that samples were taken monthly from October to April (except February), and about every 10 days in May, June, and July.

A sample consisted of all winter wheat plants growing in a 25 × 25 cm square, taken in eight replicates. The first replicate was taken at a random place at a tramline at least ten m from the field edge, and the following samples were taken at every 20 m along the tramline. The same tramline was used for all samplings. All samples were taken about ½ m away from the tramline. The sampled squares were delimited by a metal frame, which had one open side. The delimitation on the open side was assessed by eye. All plants were cut at ground level and stored in numbered paper bags for transportation to the laboratory.

The plants were measured in the laboratory right after sampling or at the latest the day after. In the laboratory, the plants were split into height zones and dissected into leaves, straws, and ears one by one. This was done by placing the plant next to a ruler and cutting it into 15 cm zones. A leaf was regarded as belonging to the zone where it was fastened to the straw. Leaves, straws and ears from each zone were counted, dried in an oven for 24 hours at 60 °C, and weighed. This procedure was rather simple during autumn, winter and early spring, when

the wheat plants were lower than 15 cm, but became rather laborious during May and June, when the winter wheat plants reached the height of about 85 cm. At this time, the plants were cut into the zones 0-15 cm, 16-30 cm, 31-45 cm, 46-60 cm, 61-75 cm, and 76-90 cm. The average number of grains per straw was estimated by counting the number of grains in 20 randomly selected ears from each sample.

The data were used for calibration of the winter wheat module of the SeptoriaSim model.

2.5 Decision support system trials

2.5.1 Flakkebjerg, Holeby, Horsens

In the project, an old model and two new models for the control of Septoria were tested. The decision support system Crop Protection Online (CPO) has recommended treatments for control of Septoria based on days with precipitation (Hansen et al 1994, Jørgensen & Nielsen 2003) for many years. Treatments are recommended if four days with rain (> 1 mm) have occurred starting at GS 32. If the crop has been treated, the crop is seen as protected for 10 days before a new risk period is initiated. A new model based on leaf wetness and periods with high relative humidity has been investigated as an alternative to the existing model along with the updated version of SeptoriaSim (Bligaard et al 2017), which was developed further in this project.

In order to test the new models, trials have been carried out at three localities in 2016 and 2017. In each of the two seasons, trials were located at Flakkebjerg, Horsens, and Holeby (Lolland). At Flakkebjerg, two cultivars were tested in 2017.

When treatments were released by the models, the fungicide Bell (Boscalid 233 g/l + Epoxiconazol 67 g/l) was used at all three locations in 2016. In 2017, the trial plan was changed slightly following a general trend towards more fungicide resistance (Heick et al. 2017). The dose of Bell was increased from 0.5 to 0.75 of the recommended dose, and Prosaro (Tebuconazol 125 g/l + Prothioconazol 125 g/l) was applied at GS 32 and GS 55 instead of using a repetition of Bell.

All trials were carried out as field trials in wheat and laid out as randomized block trials with 4 replicates and plot sizes varying between 12 and 22.5 m². Treatments were applied using either backpack sprayers or a self-propelled sprayer using 150-200 l/ha. The trials were assessed for attack of diseases at 14 day intervals, assessing attack on individual leaf layers. At the last assessment, the green leaf area was also assessed. The trials were harvested using a plot harvester, and grain yield per plot and moisture content measured. Following this, yield per ha was calculated adjusting for 15% grain moisture. With the exception of fungicides, all other treatments (PGR, herbicides, nitrogen etc.) were applied as in the rest of the field.

2.5.2 Farmers' field tests

The farmers, who had put wheat fields at our disposal for aphid counts, were asked if they were interested in participating in a trial of the SeptoriaSim decision support system. Five of them answered positively.

These five farmers established a 100 m trial plot along both sides of one of the tramlines. The trial plots were placed along tramlines situated next to the aphid plot tramlines. The trial plots were sprayed with fungicides to control Septoria and/or insecticides to control aphids according to warnings issued after projections of net income made by aid of SeptoriaSim. All five farmers received personal warning messages on 12 May, 22 May, 31 May, and 16 June, telling them whether it was economically beneficial to spray against either Septoria and/or aphids in the coming week and when to do it. The farmers had promised to treat the rest of the field the way they would have done themselves without our warning messages that were directed at the trial plots only.

Shortly before harvest, 10 yield samples were taken in both the trial plots and along the neighbouring tramline representing the rest of the field. The yield samples were taken the same way as the growth analysis samples, i.e. they covered 25 × 25 cm. The samples were stored in paper bags under dry conditions in the laboratory until they were threshed on an automatic small-scale thresher at Nordic Seed. After threshing, the seeds were weighed and the 1000-grain weight was measured. The results on yield and 100-grain weight were analysed by aid of an analysis of variance, where each field was regarded a replicate, i.e. six replicates in total.

3. Results

3.1 Fundamental research on Septoria

3.1.1 Improved monitoring of Septoria – Spore trapping

The analysis and graphs (Figures 6-14) indicate that a minor release of spores takes place during most of the season. In certain intervals, major peaks of release took place. This was particularly seen in late July and August at all locations. At Flakkebjerg, higher numbers of spores were seen in the air samples from the spore trap placed in the field compared to the one placed outside the field. However, both spore traps generally followed the same pattern of spore release. From April to harvest, small peaks of spore release were occasionally seen, which might have been caused by the release of ascospores from pseudothecia developing on the wheat plants. After harvest and throughout the autumn, several, and generally higher, peaks of spore release were detected and were most likely due to the release of ascospores from plant debris.

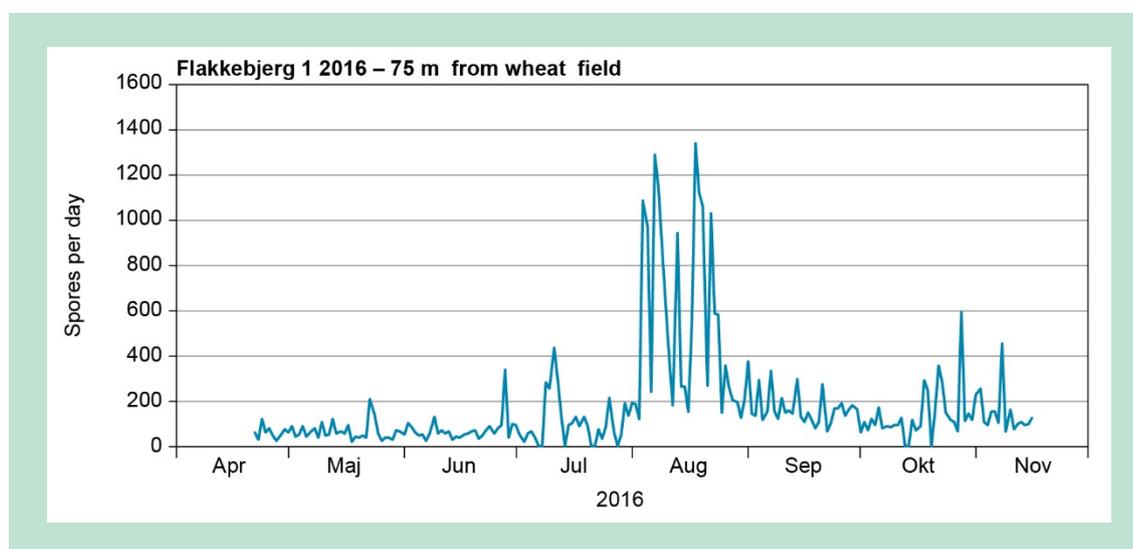


FIGURE 6. Spores collected by spore trap situated outside the winter wheat field at Flakkebjerg during the growing season 2016.

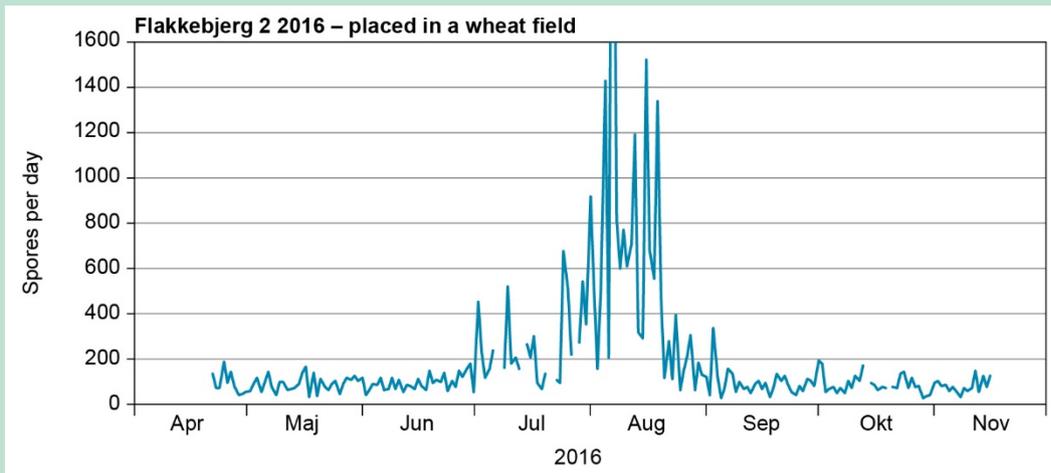


FIGURE 7. Spores collected by spore trap within the winter wheat field at Flakkebjerg during the growing season 2016.

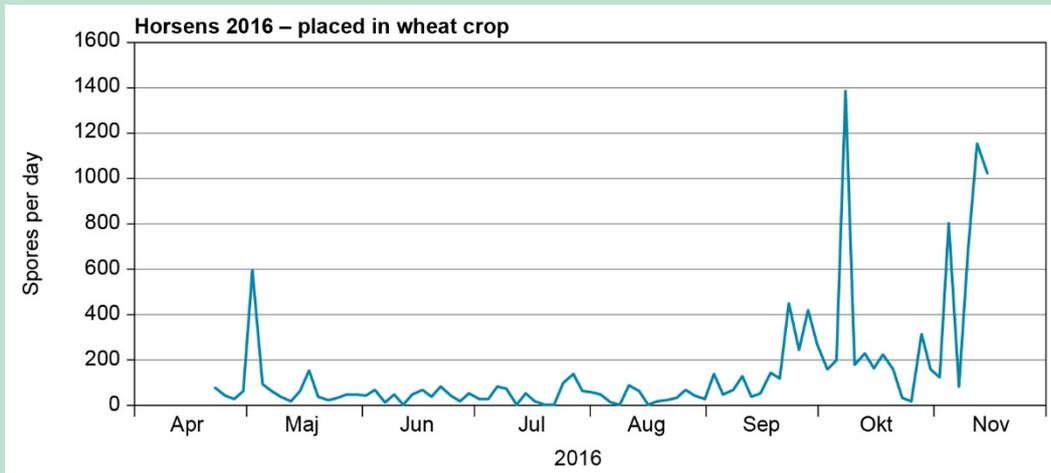


FIGURE 8. Spores collected by spore trap placed near Horsens during the growing season 2016.

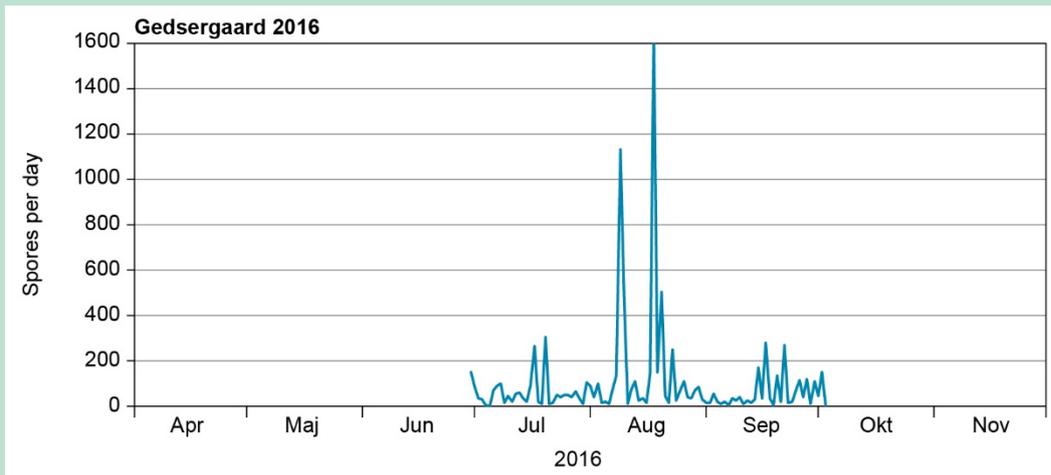


FIGURE 9. Spores collected by spore trap at Gedsergaard in Falster outside the wheat field during the growing season 2016.

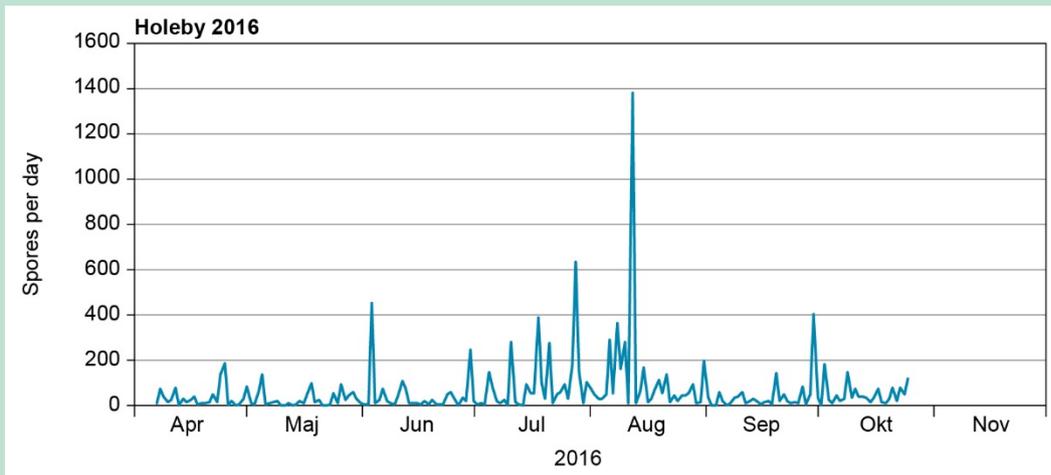


FIGURE 10. Spores collected by spore trap placed near Holeby in Lolland during the growing season 2016. The trap was placed 50-100 meter from a winter wheat field.

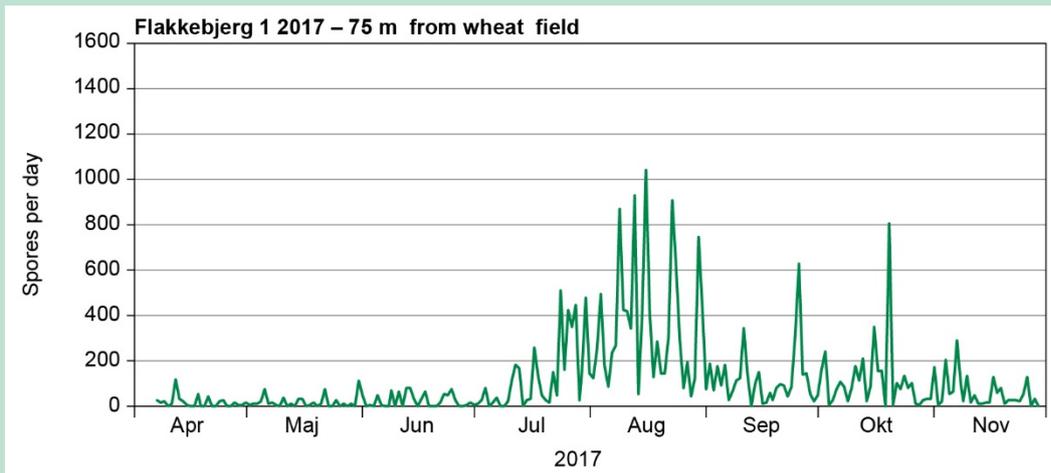


FIGURE 11. Spores collected by spore trap situated outside the winter wheat field at Flakkebjerg during the growing season 2017.

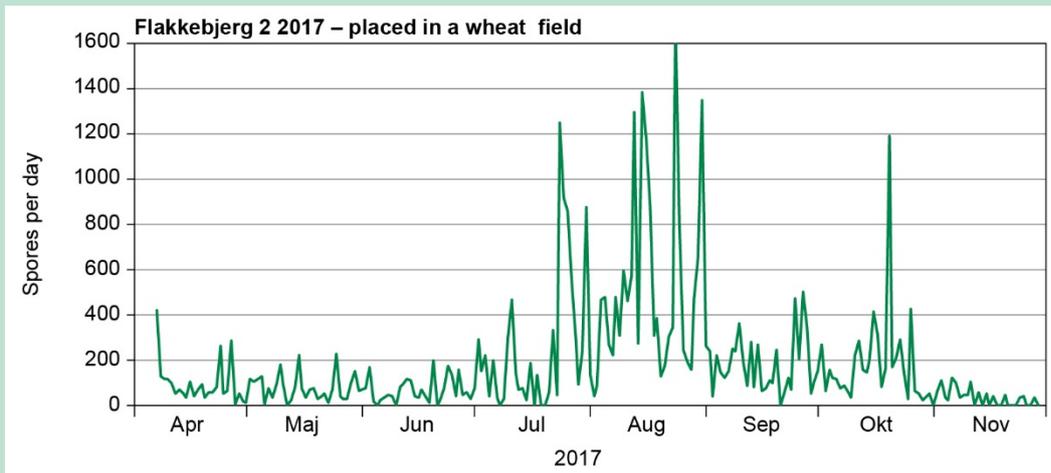


FIGURE 12. Spores collected by spore trap within the winter wheat field at Flakkebjerg during the growing season 2016.

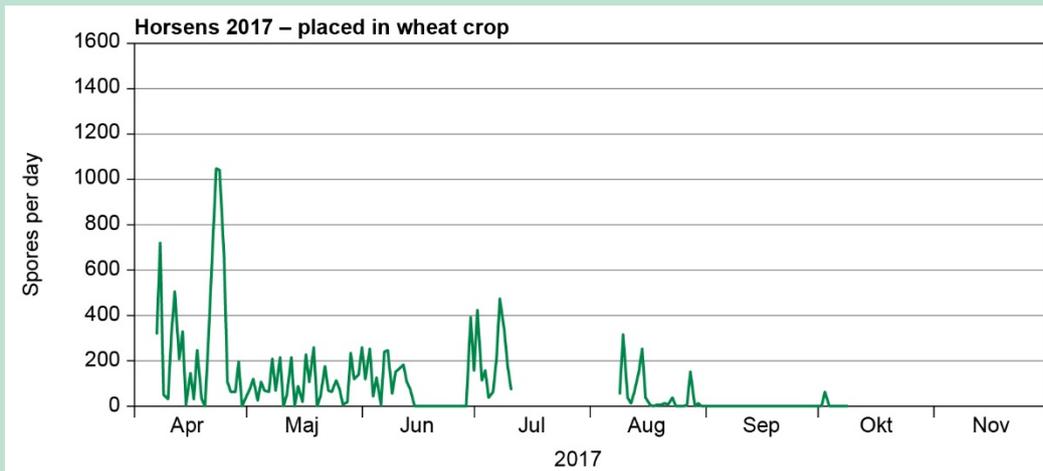


FIGURE 13. Spores collected by a spore trap at Horsens during the growing season 2017. The spore trap was placed in a wheat field. Samples were not collected 14.06.17-28.06.17 and 11.07.17-07.08.17.

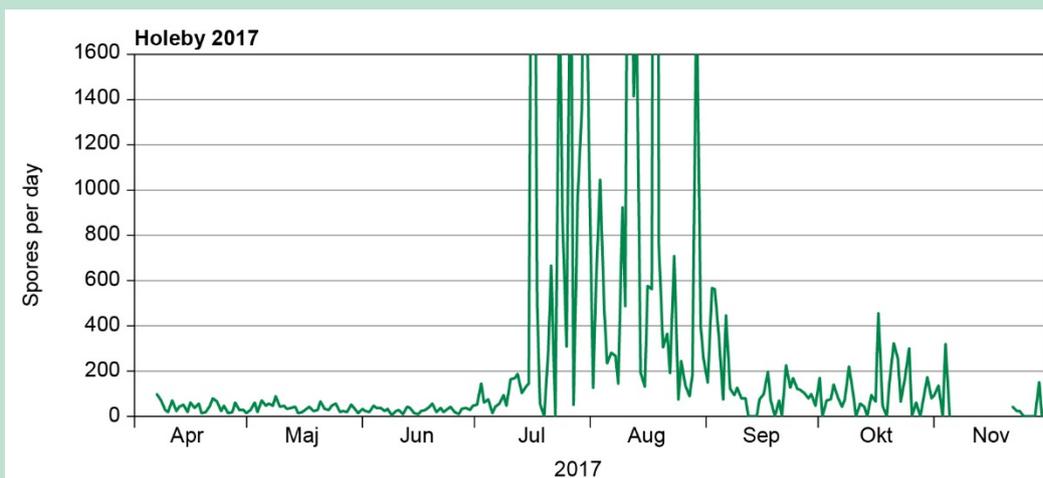


FIGURE 14. Spores collected by a spore trap at Holeby during the growing season 2017. The spore trap was placed 50-100 meter from a wheat field.

The data indicated a variation between localities and years, but a steady flow of spores was trapped every day during the growing season. In all traps, the highest amount of spores was caught in August and during the autumn. The constant flow of ascospores during the growing season indicates that ascospores may play a more important role for the epidemic than previously recognized.

The constant flow of ascospores was introduced in SeptoriaSim, which previously had been programmed to finish the infection by ascospores 1 March.

3.1.2 Progression of Septoria attacks - QPCR

A clear gradient across the canopy (Figures 21-25) indicates a higher level of attack on the lower leaves than on the upper leaves. The DNA analysis gave 18 cases out of 90 of pre-

symptomatic readings in 2016 and 28 cases out of 111 in 2017, where no visible attacks were seen on the leaves, indicating that the DNA method can detect latent attack.

Generally, there was a good link between disease severity and DNA measurement, as shown in Figures 3.2.4 and 3.2.10 from specific cultivars and localities. In a few cases for the late growth stages, only moderate DNA content was seen despite assessments of severe attack (3.2.2, 3.2.5, 3.2.6, 3.2.7, 3.2.8). Part of this poor correlation might be due to the leaves being very dry and senescent for this very late assessment.

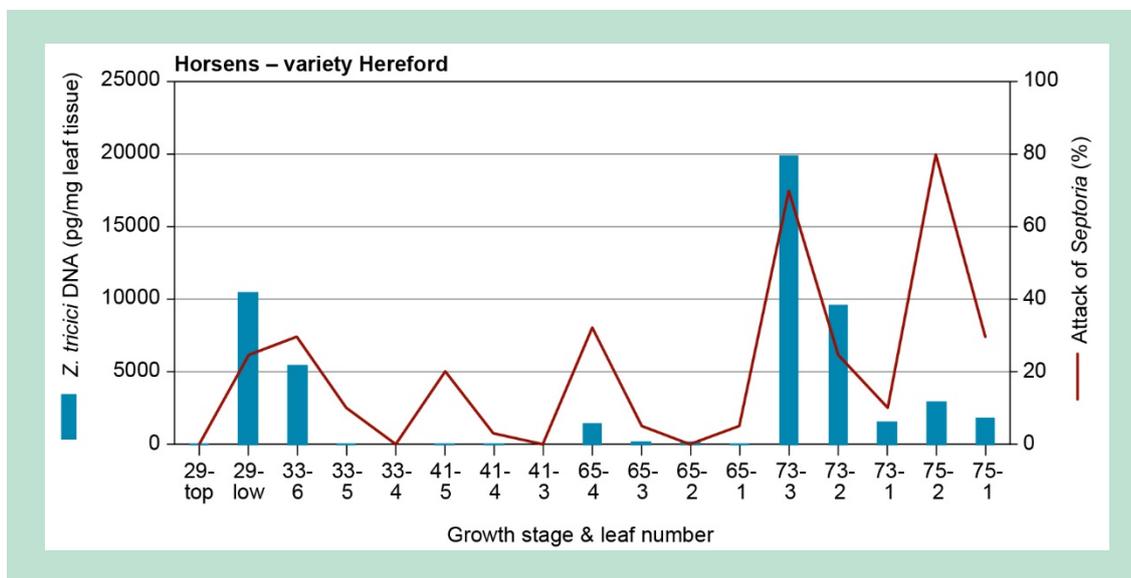


FIGURE 15. Link between DNA and % attack of *Septoria* versus growth stage and leaf number in Hereford. Data from Horsens. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf number, where the lowest number indicates the top leaf number at each growth stage.

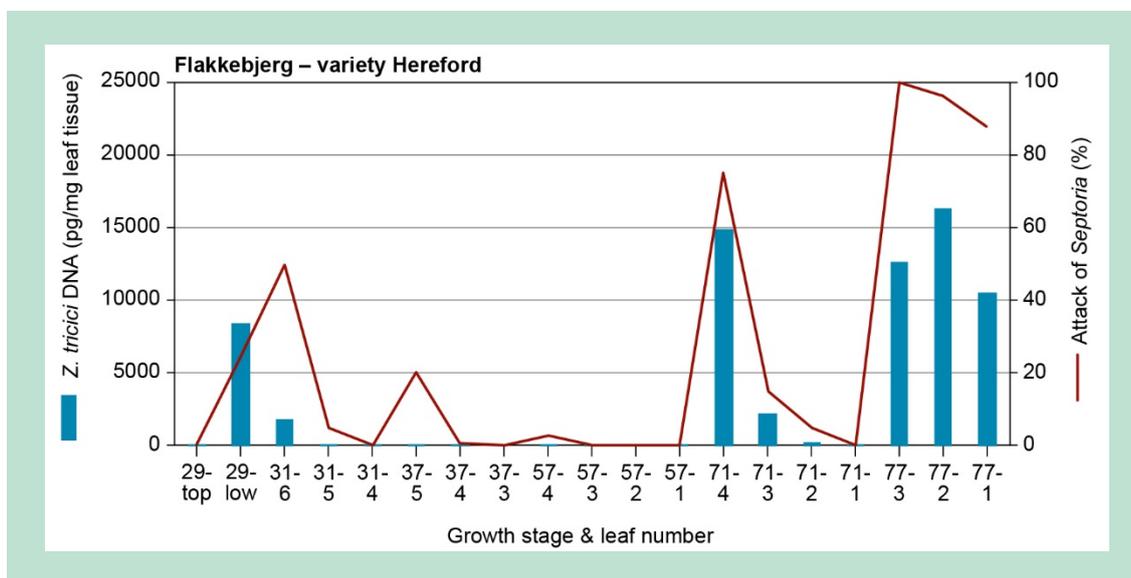


FIGURE 16. Link between DNA and % attack of *Septoria* versus growth stage and leaf number in Hereford at Flakkebjerg. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf number, where the lowest number indicates the top leaf number at each growth stage.

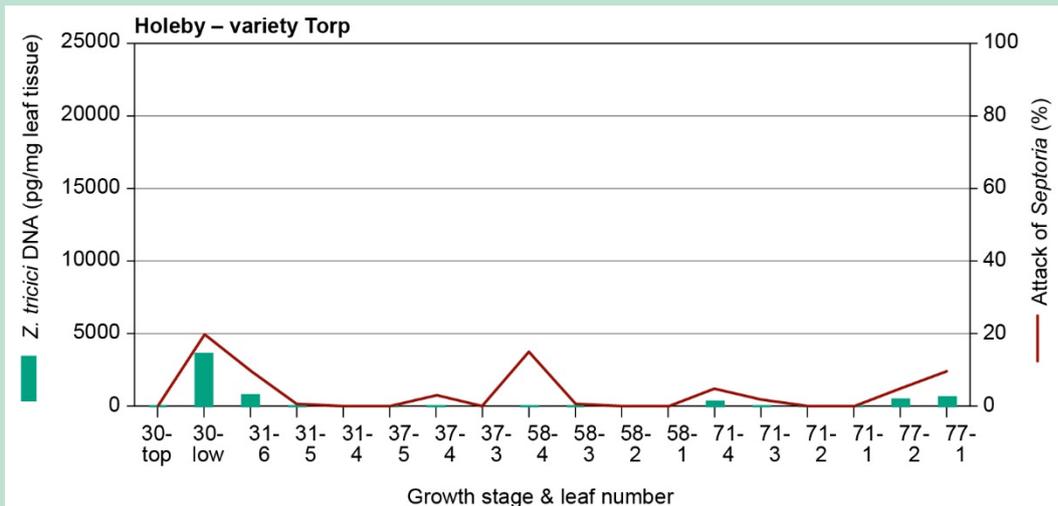


FIGURE 17. Link between DNA and % attack of *Septoria* versus growth stage and leaf number in Torp. Data from Holeby from a locality with low levels of diseases “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

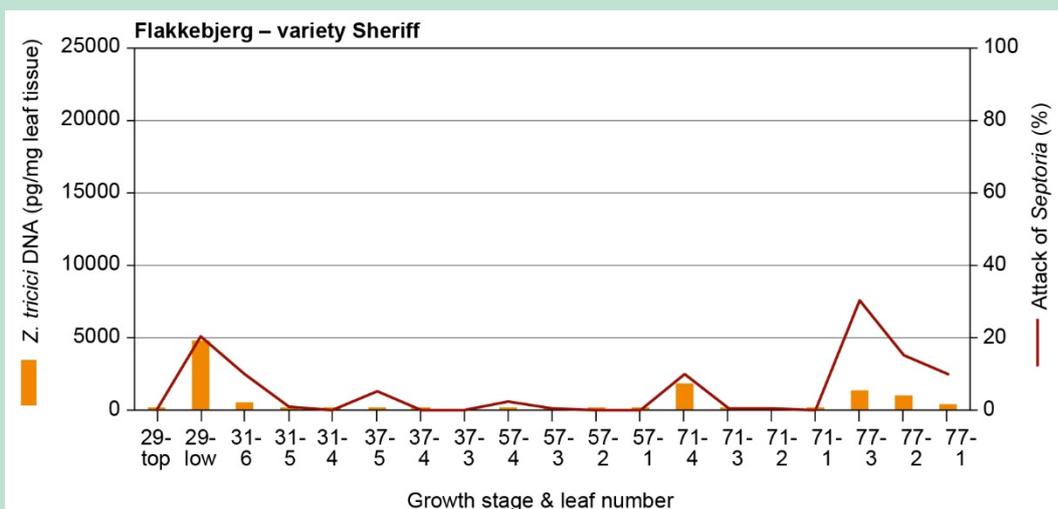


FIGURE 18. Link between DNA and % attack of *Septoria* versus growth stage and leaf number in Sheriff. Data from Flakkebjerg. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

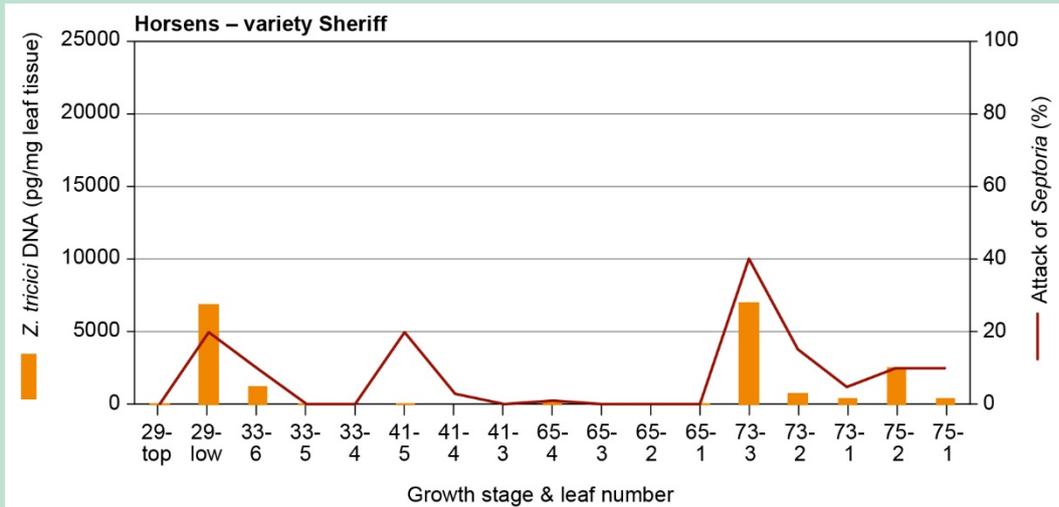


FIGURE 19. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Sheriff. Data from Horsens. Sheriff is much less susceptible than Hereford. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The n numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth.

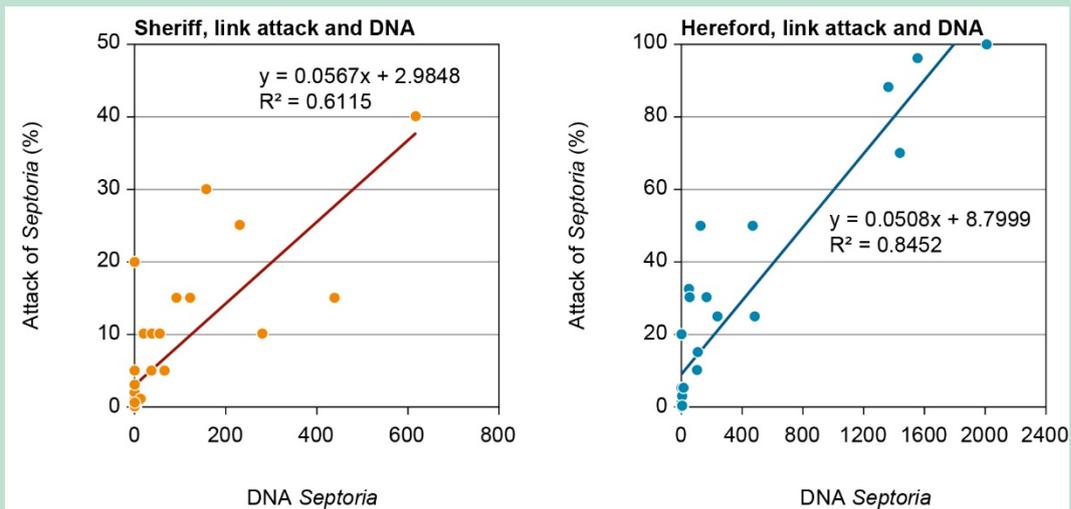


FIGURE 20. Correlation between DNA and visual assessments of *Septoria* in two cultivars (DNA 1). Link between two DNA measurements.

Data from 2017

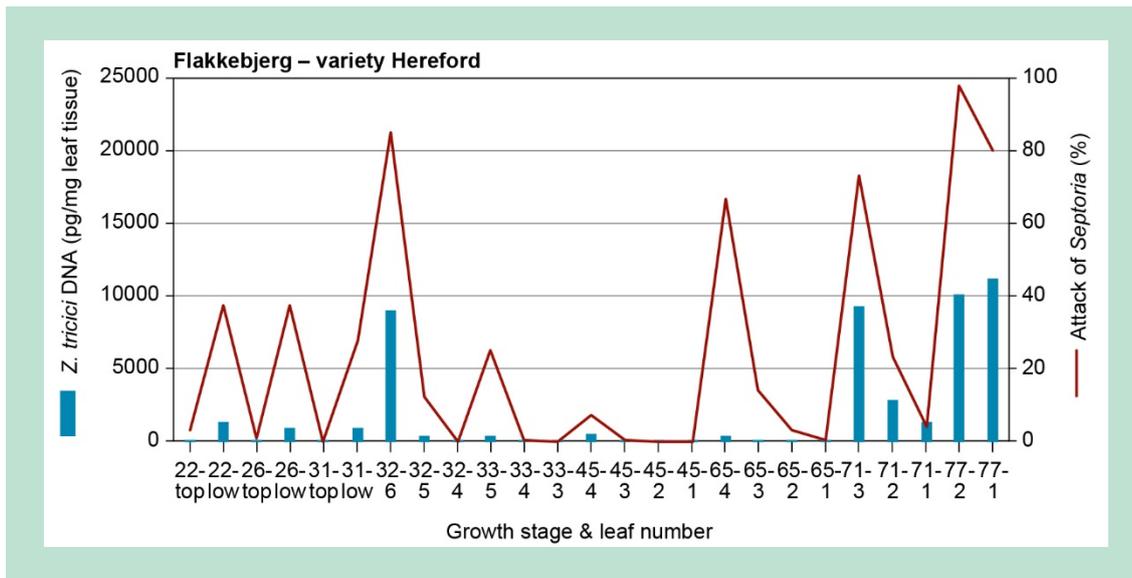


FIGURE 21. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Hereford. Data from Flakkebjerg. Hereford is susceptible to *Septoria* attacks. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

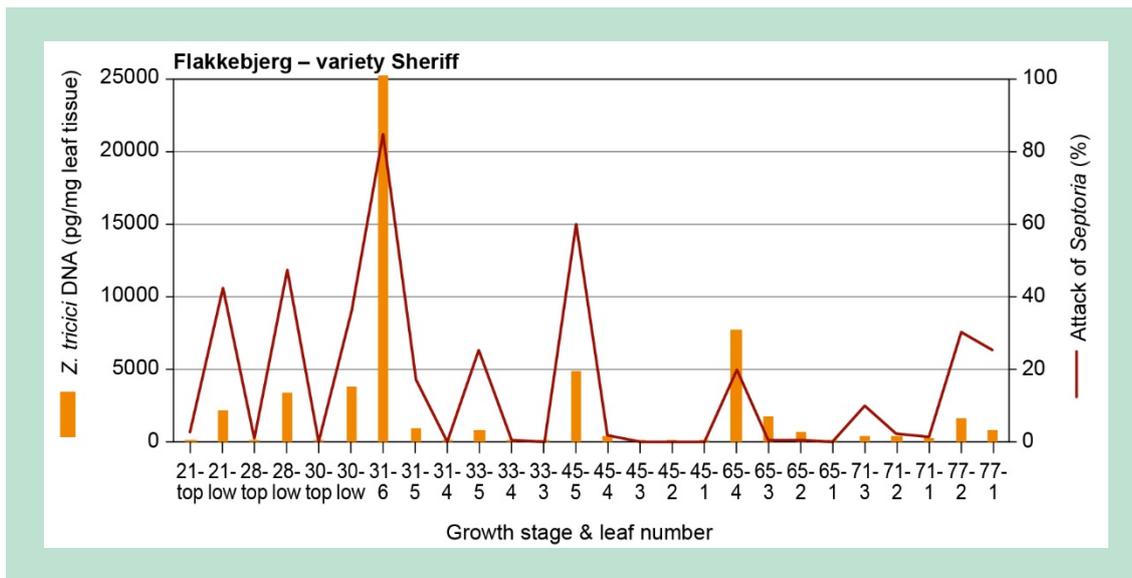


FIGURE 22. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Sheriff. Data from Flakkebjerg. Sheriff is less susceptible to *Septoria* attacks than Hereford. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

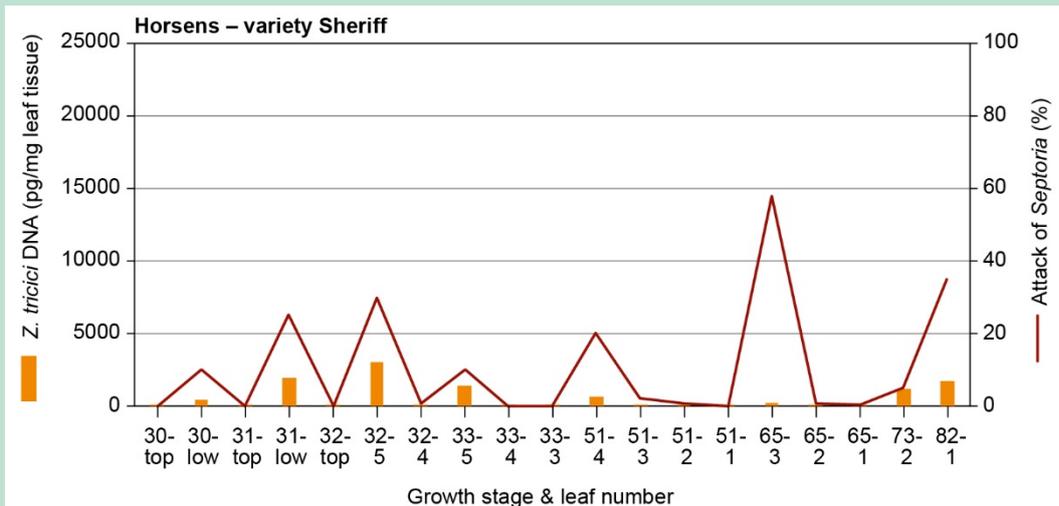


FIGURE 23. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Sheriff. Data from Horsens. Sheriff is less susceptible to *Septoria* attacks than Hereford. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

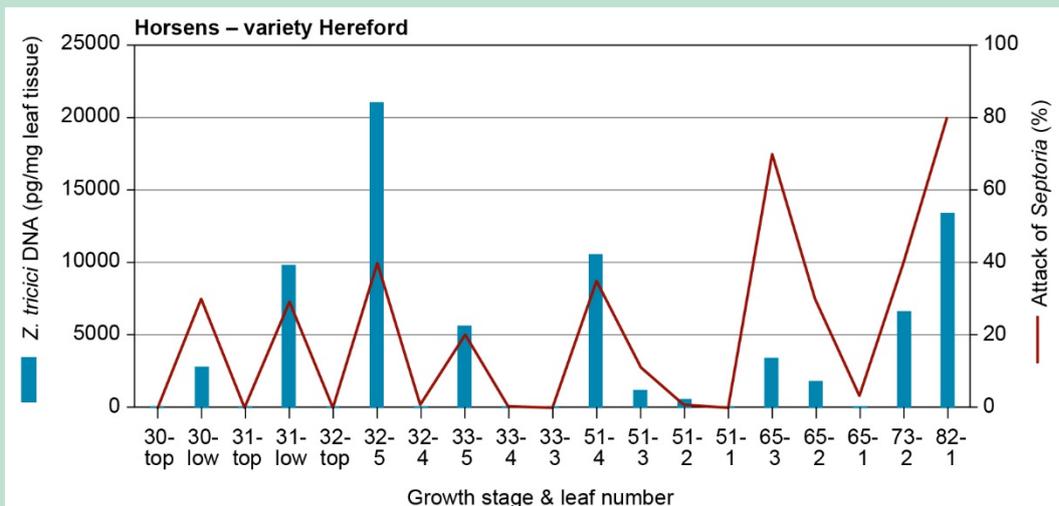


FIGURE 24. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Hereford. Data from Horsens. Hereford is susceptible to *Septoria* attacks. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

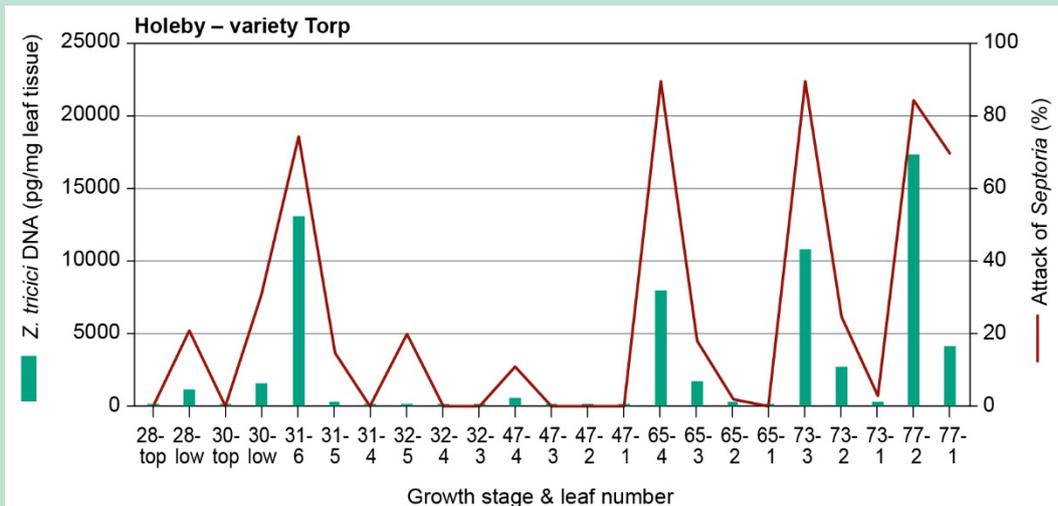


FIGURE 25. Link between DNA of *Z. tritici* and % attack of *Septoria* in the cultivar Torp. Data from Holeby. Torp is susceptible to *Septoria* attacks. “low” = Lower leaves with visible attack; “over” = top leaves without visible attack. The numbers 1, 2, 3, 4, 5 and 6 refer to the leaf layer, where the lowest number indicates the top leaf layer at each growth stage.

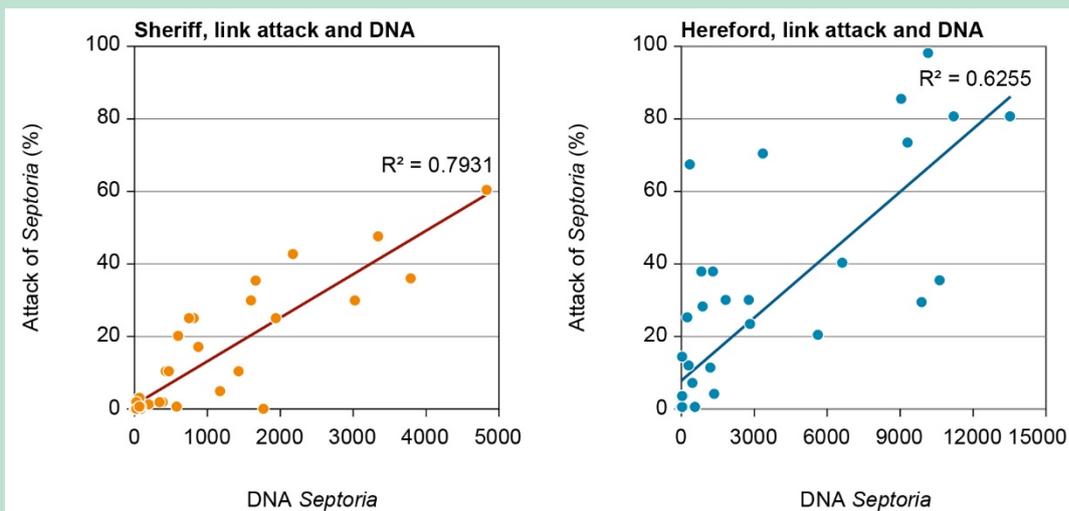


FIGURE 26. Correlation between DNA and visual assessments of *Septoria* in two cultivars from 2017.

The samples clearly showed the disease progress in the crop with increasing % attack step by step as new leaves develop. In approximately 22% of the samples categorized with 0% attack, minor levels of DNA were detected. Generally, Hereford was seen as the cultivar with most severe attack, and here the highest level of DNA was measured. The correlation between measured DNA and % attack of *Septoria* was only moderate ($R^2 = 0.6-0.8$) (Figures 20 and 26).

These data were used to calibrate the *Septoria* module of *SeptoriaSim*.

3.1.3 Use of results in SeptoriaSim

Originally, the Septoria module was calibrated to the observed Septoria cover observations from a dataset from 2003-2013 by aid of Monte-Carlo simulations (Bligaard, 2017). The parameters from this calibration were used as a starting point of the re-calibration to the data on Septoria cover in percent and the QPCR measurements of Septorial DNA collected in this project. The calibrated parameters were daily spore input rate, growth rate, spore production rate, rainfall limit for spore spreading, and humidity limit for fungal spore germination and survival on leaf surface. The calibration was carried out manually, i.e. one of the parameters was changed in the Septoria parameter input panel, the simulation was started, and the output was compared to the observations. This was done until it was assessed that the best possible fit had been obtained. This was first done using the observations from Flakkebjerg. The parameters from the calibration to Flakkebjerg data were used as a starting point for calibration to data from Holeby and Horsens. Hence, the calibration to the observations from Holeby and Horsens was in reality a fine-tuning and confirmation of the calibrated parameter values from Flakkebjerg. Comparisons between simulations and observations from 2017 are shown in the Figures 27-30.

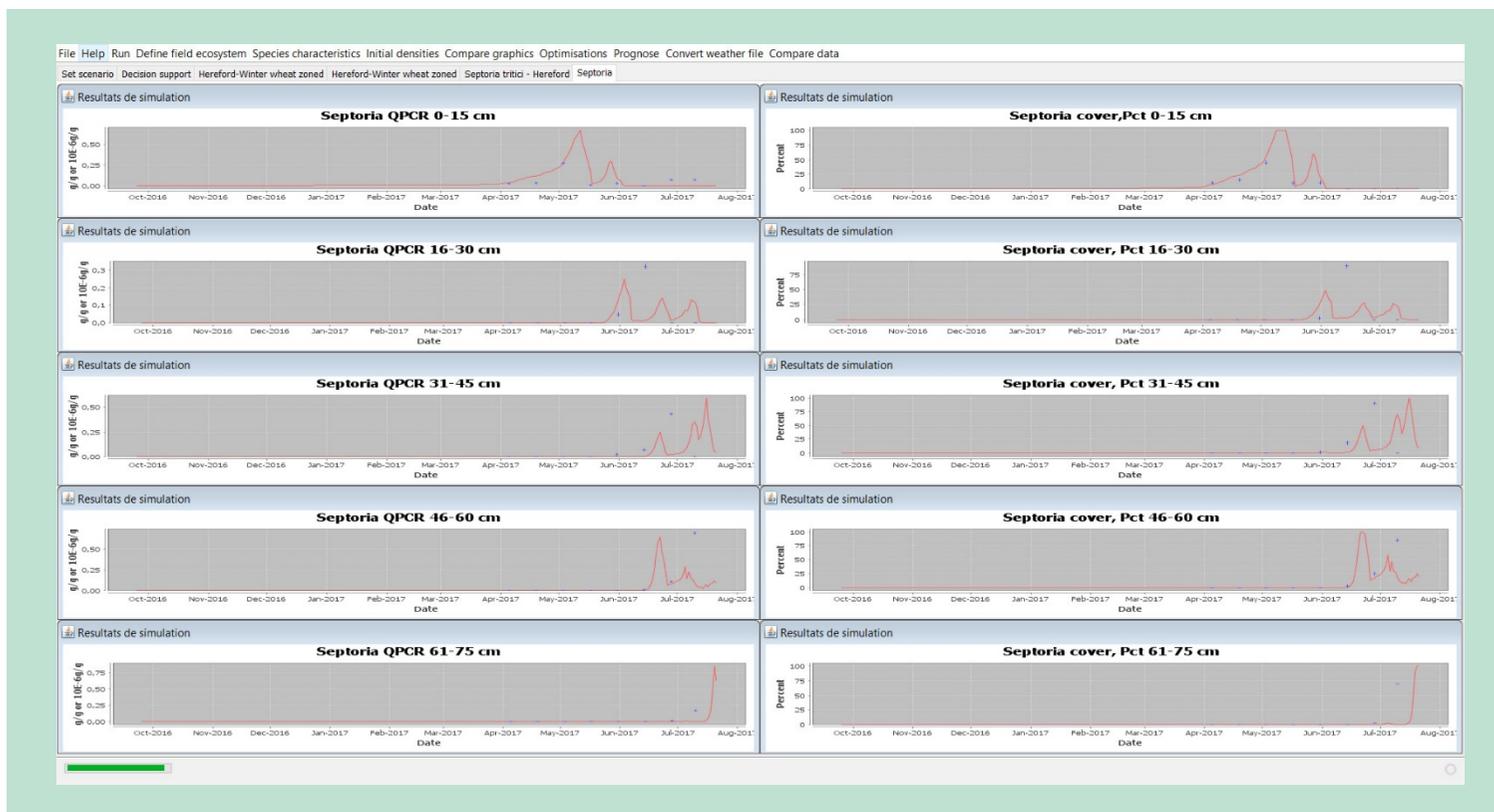


FIGURE 27. Comparison between simulated amount of Septoria and QPCR observations (left column) and simulated and observed Septoria cover (right column). The QPCR measurements have been multiplied by a scaling factor to fit to the same scale as the observations. Observations came from Holeby 2017 – variety Torp. Spore input was 750 spores/day. Screen dump from SeptoriaSim user interface.

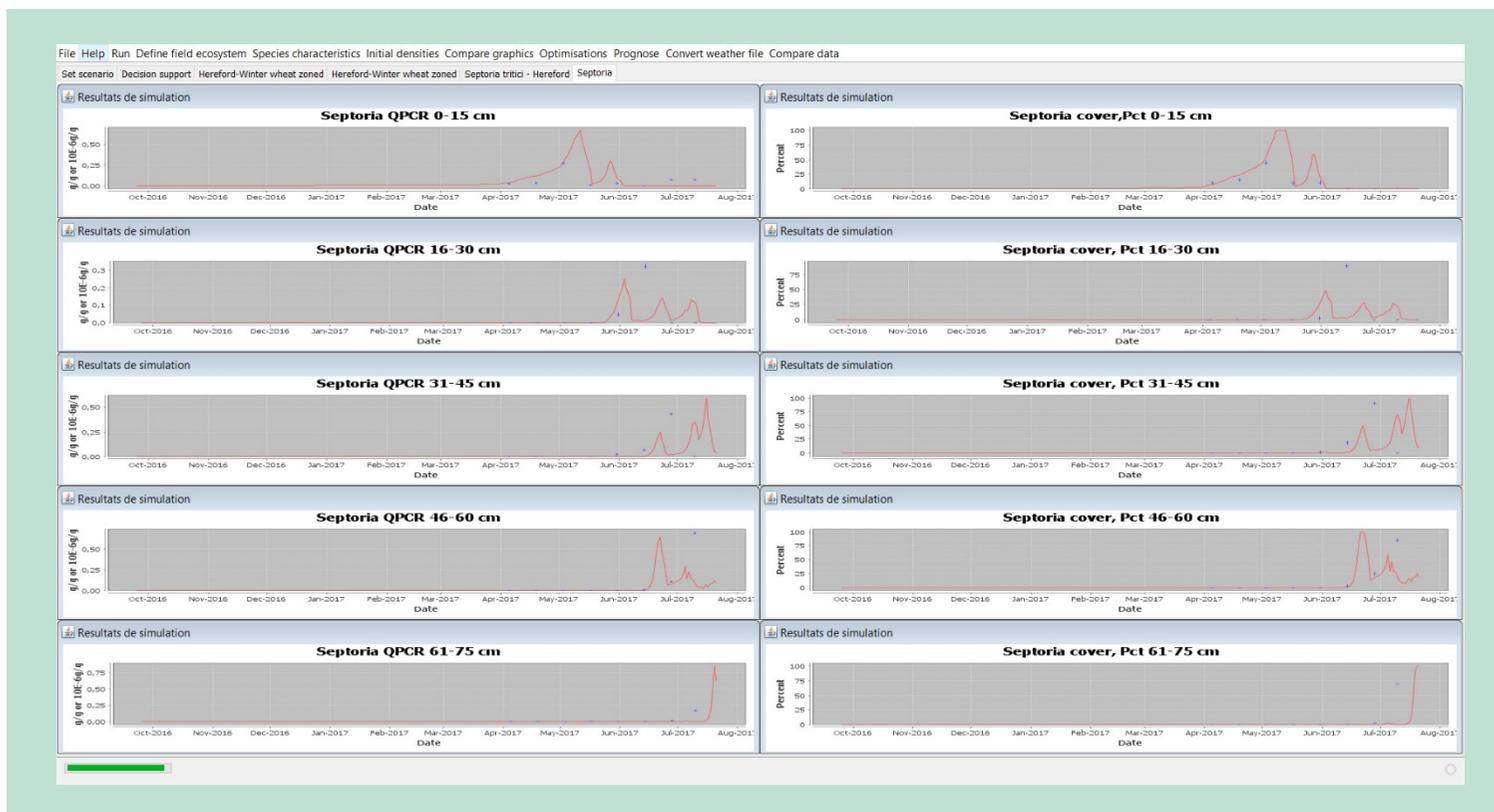


FIGURE 28. Comparison between simulated amount of Septoria and QPCR observations (left column) and simulated and observed Septoria cover (right column). The QPCR measurements have been multiplied by a scaling factor to fit to the same scale as the observations. Observations came from Flakkebjerg 2017 – variety Hereford. Spore input was 750 spores/day. Screen dump from SeptoriaSim user interface.

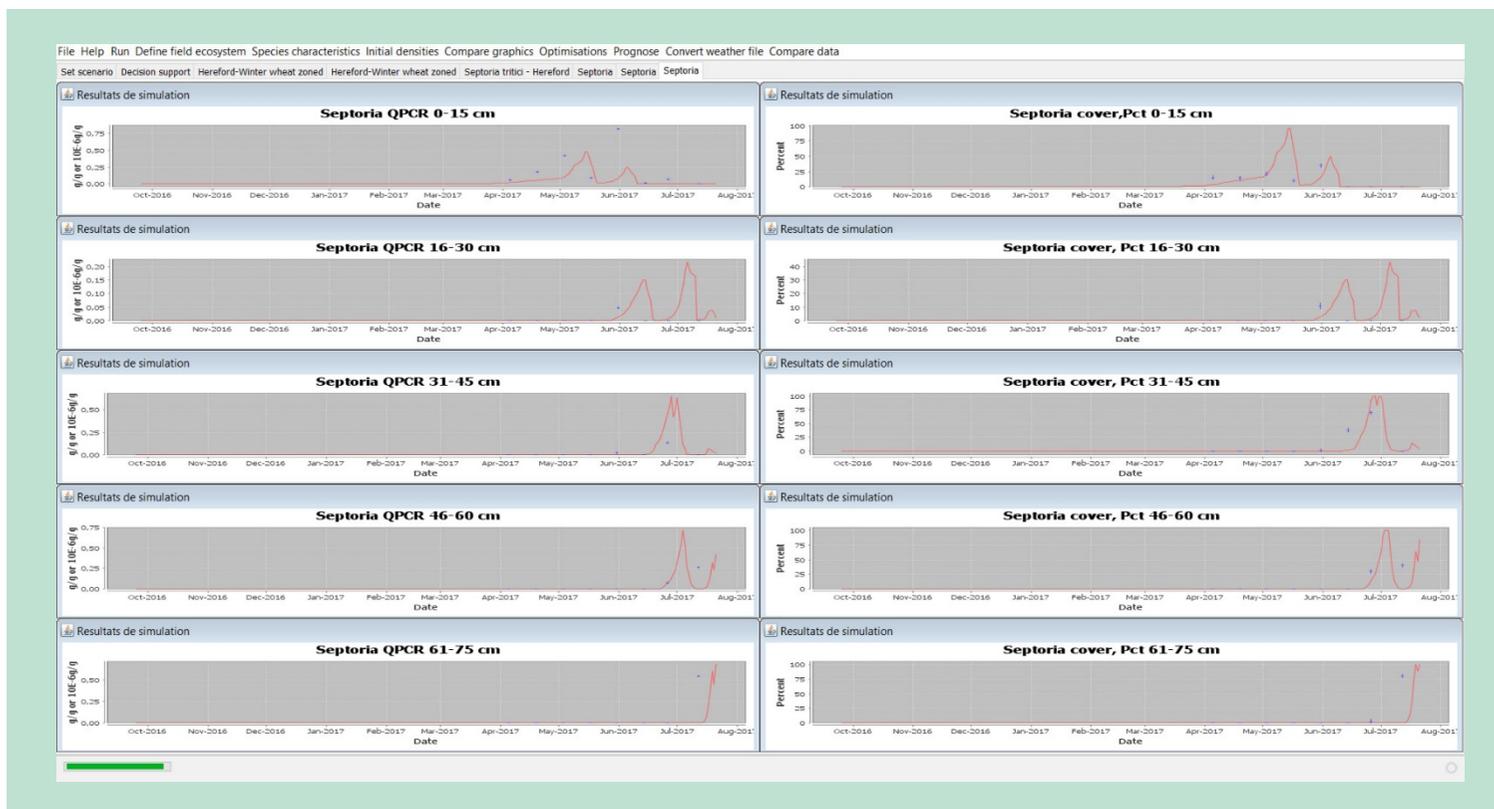


FIGURE 29. Comparison between simulated amount of Septoria and QPCR observations (left column) and simulated and observed Septoria cover (right column). The QPCR measurements have been multiplied by a scaling factor to fit to the same scale as the observations. Observations came from Horsens 2017 – variety Sheriff. Spore input was 500 spores/day. Screen dump from SeptoriaSim user interface.

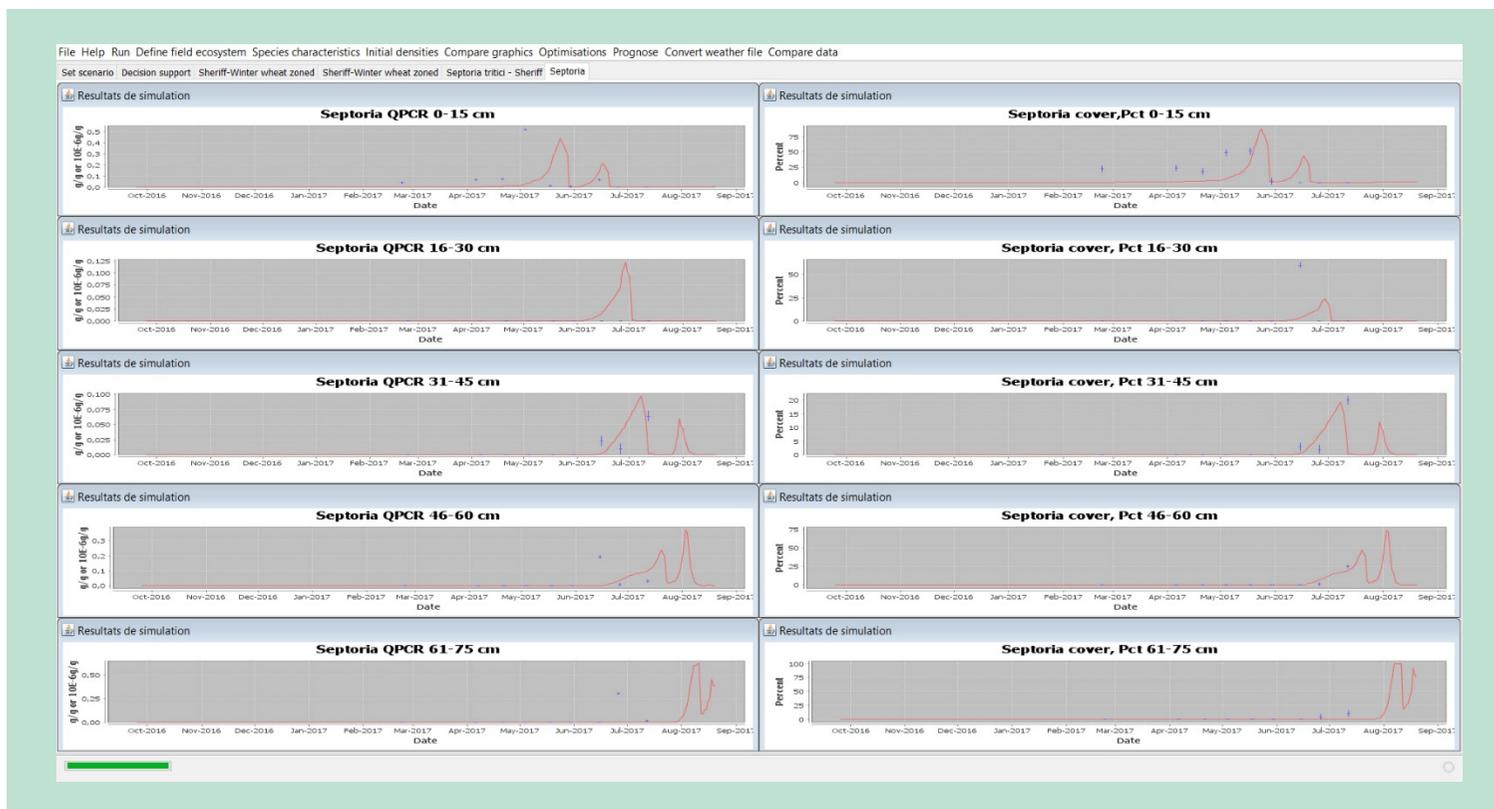


FIGURE 30. Comparison between simulated amount of Septoria and QPCR observations (left column) and simulated and observed Septoria cover (right column). The QPCR measurements have been multiplied by a scaling factor to fit to the same scale as the observations. Observations came from Flakkebjerg 2017 – variety Sheriff. Spore input was 500 spores/day. Screen dump from SeptoriaSim user interface.

3.2 Aphids

3.2.1 Field sampling program

The proportion of wheat straws infested with the three studied aphid species during the 2016 season is presented in Figure 31. Except for Borum 6 km, grain aphids by far outnumbered the other two species. The development of the grain aphid was rather uniform over the different locations and fields, with a maximum in late June, whereas the other two species differed more in development, both between locations (Borum, Viborg, Hammel) and between fields at a given location.

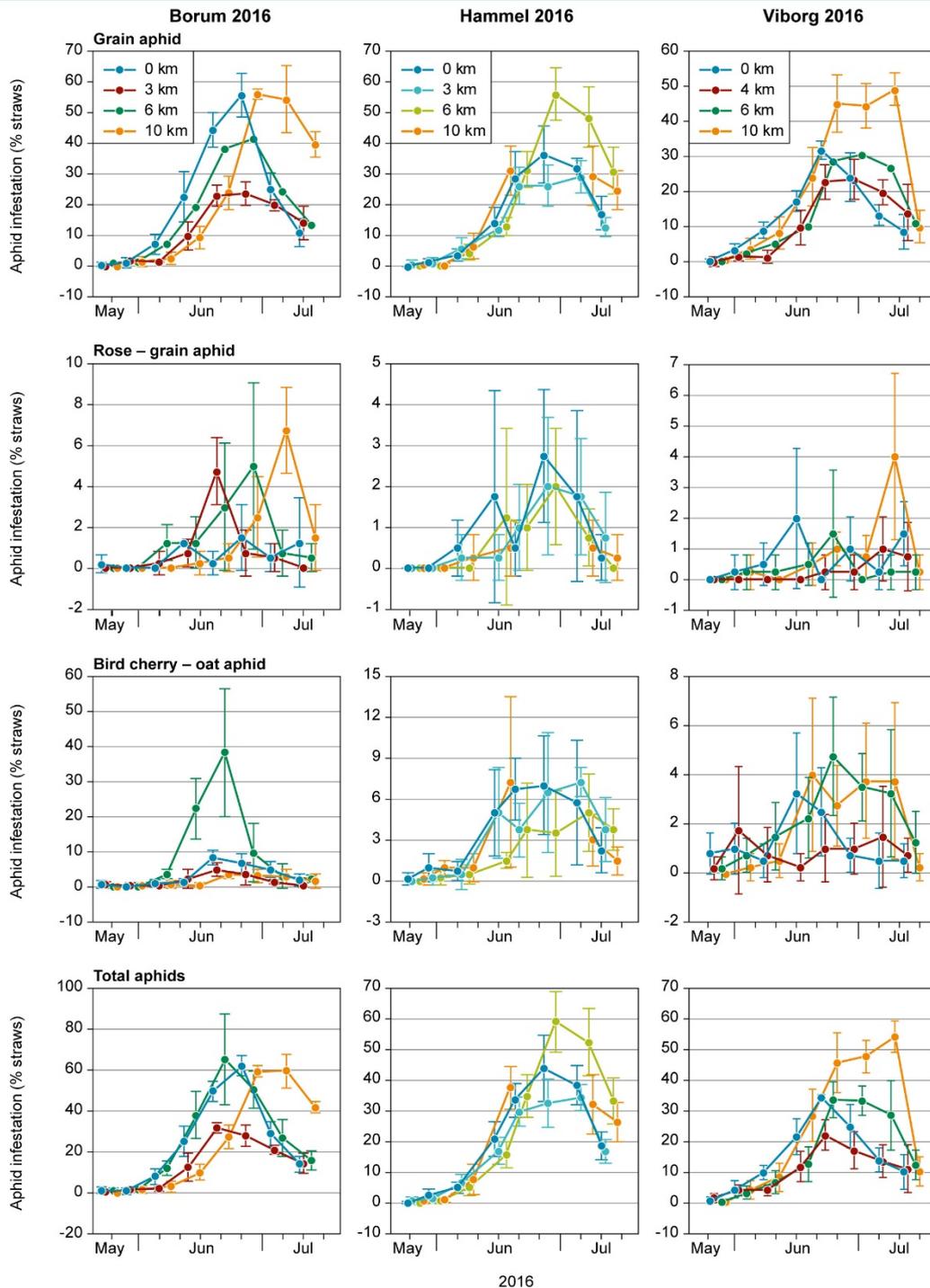


FIGURE 31. Proportion of inspected straws that were infested with the three aphid species in the unsprayed parts of the 12 study fields during the aphid season of 2016. “0”, “3”, “4”, “6” and “10” refer to the distance in km to the local Observation Web field. On every sampling date, 400 straws were inspected per field.

The following year, the grain aphid and the bird cherry – oat aphid occurred at comparable frequencies (Figure 32), whereas hardly any rose – grain aphids were found (not shown). In several fields, aphid infestation had not reached a climax on the last sampling dates, i.e. the very last days of June.

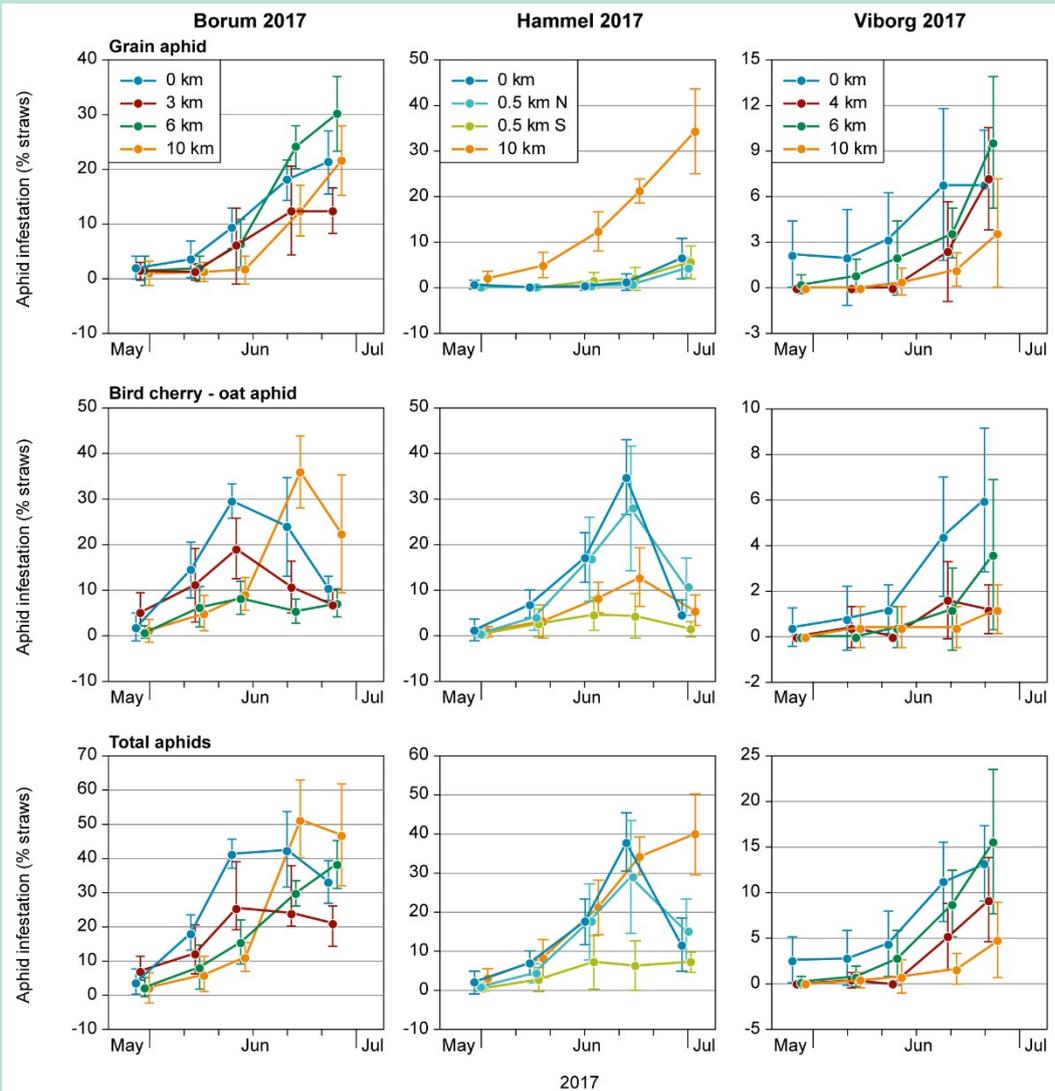


FIGURE 32. Percentage wheat straws infested with aphids in the unsprayed parts of the 12 study fields in the aphid season of 2017. On the first two sampling dates, 500 straws were inspected per field, whereas 250 straws were assessed the following dates.

3.2.2 Spatial variation

The deterministic part of the population growth model (eqn. 2, chapter 2.3.2.2) seemed to model the dynamics of aphid occurrence probabilities as a function of degree-days from June 1 2016 to July 1 2016 adequately when visually compared to the observed spatio-temporal mean occurrence probability data in the same period.

The posterior marginal distribution of the parameter that measures the effect of geographic distance on the spatial covariance, ρ_0 , is left-skewed towards the upper boundary and significantly larger than zero, and the site-specific initial occurrence probabilities are consequently positively correlated among the sites at the spatial scale of 10 km. However, the importance of this positive correlation for the among-site variation in aphid epidemics must be evaluated in relation to the estimated among-site variation in the initial occurrence probability as modelled by σ_0 , and population growth as modelled by σ_p .

It has been hypothesized that episodes of heavy rain may lead to decimation of the aphid population. However, if the observed rain records at the sites, which included several episodes of heavy rain, were manually scored according to severity and compared to the latent variables that model the among-site variation in population growth, ϵ_i , then there was no significant relationship between the heavy rain score and a relatively low population growth rate ($P = 0.89$).

The variation in aphid occurrence among sampling plots as a function of the geographical distance among plots at 30. Maj and 7. June 2017 is shown as a semivariogram (Figure 33). Generally, the variation among plots is relative low at the two sampling days, although the variation increases irregularly with time. There is a slight indication that the variation among plots increases with the distance among plots at the second sampling.

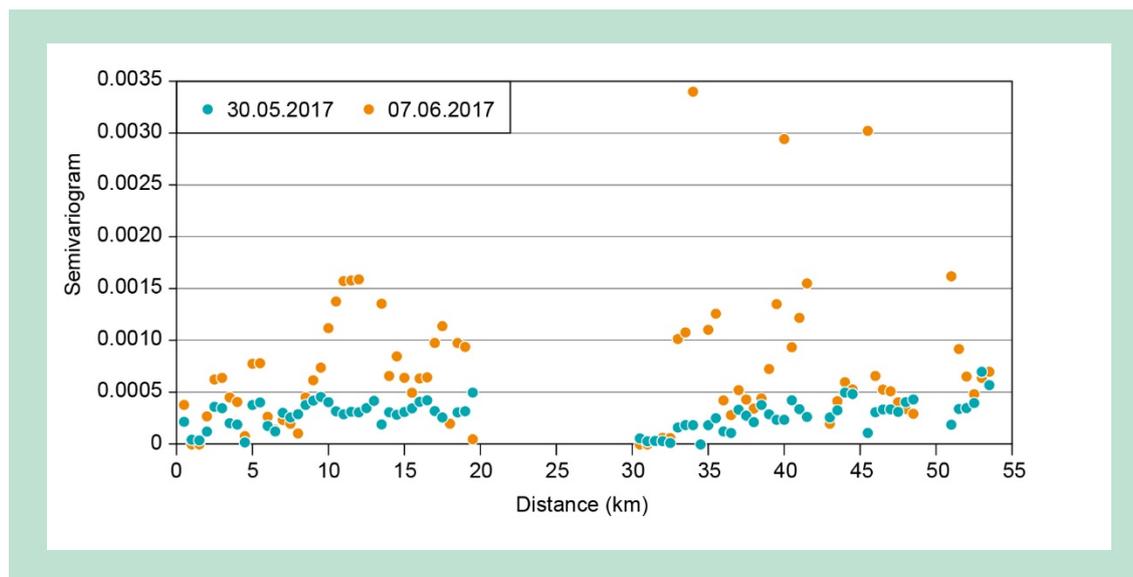


FIGURE 33. Semivariogram. The variation in aphid occurrence as a function of the geographical distance among sampling plots. Blue points: 30. Maj 2017, yellow points 7. June 2017.

The aphid population data from 2016 were used to calibrate the aphid module of the Septoria-Sim model. This was done by aid of a large number of simulations, where the simulation program choses a set of parameters randomly from intervals given from the user interface for each simulation. The parameter sets were then used as input to simulate the population development of an aphid species in each of the investigated 12 fields, and, throughout the simulations, the program made comparisons between observed and simulated values. At each observation date, the program calculated the squared difference between observed and simulated values, and these were summed during a growing season to obtain the sum of squared differences. The sums of squared differences were summed for all 12 fields to obtain a total sum of squared differences. By running a very large number of simulations with random choice of a number of important parameters, the combination of parameters giving the best fit to data was identified as the combination yielding the least total sum of squares. This procedure was carried out for all three aphid species.

The capacity of the aphid module of SeptoriaSim to simulate the observed aphid population data after the calibration was rather good when using the measured initial densities from each field (Figure 34). However, the simulations showed a better fit to the observed data if the average initial density from a cluster of fields was used as initial density for all fields in a cluster (Figure 35). The reason was most likely that the average value was a better estimate of the real initial density than the values from the single field.

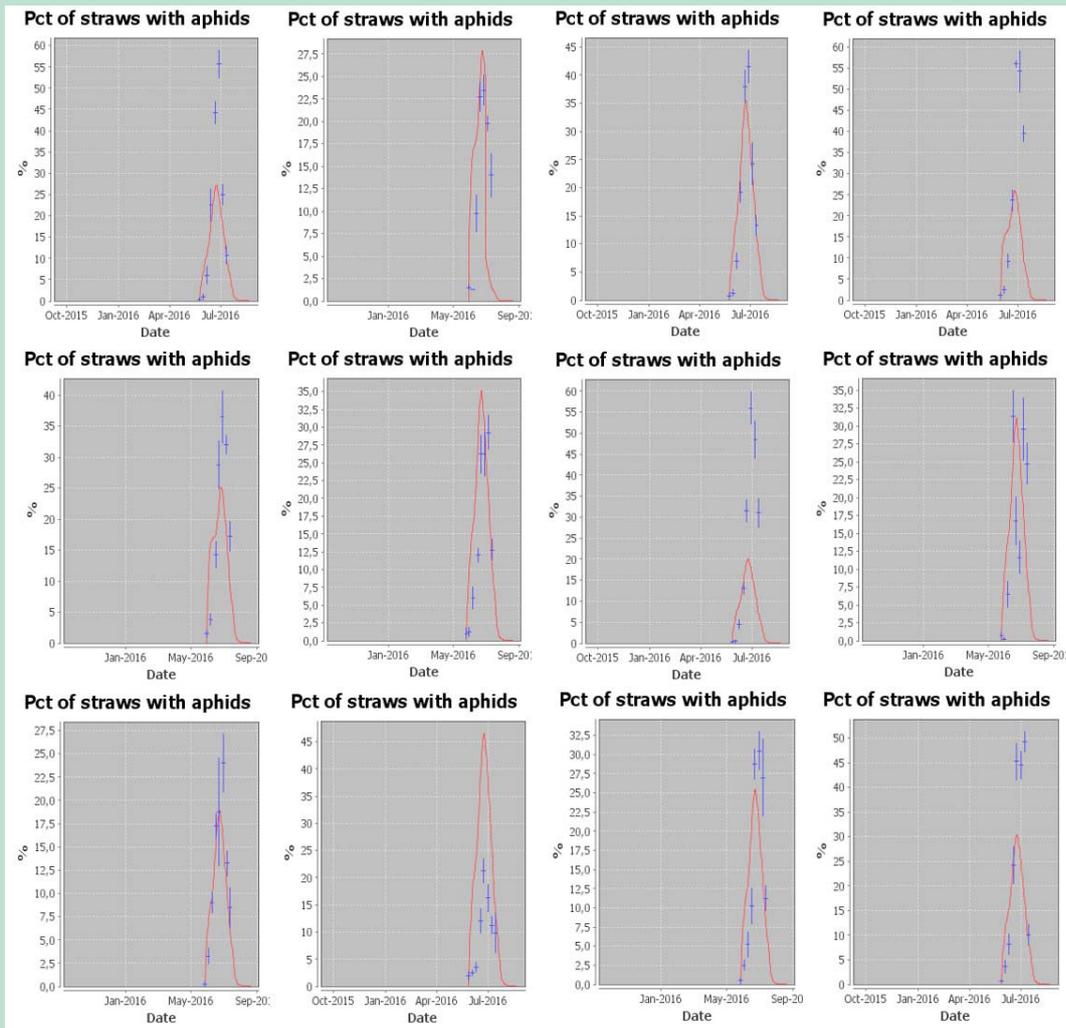


FIGURE 34. Comparisons between observed and simulated aphid densities when the observed initial density from the fields in question was used as input. The upper row is the Borum cluster, the middle row was from the Hammel cluster, and the bottom row was from the Viborg cluster. From left to the right, you find first 0 km, 3 km, 6 km and, finally, 10 km. Mind different scales on the y-axes. Screen dump from SeptoriaSim user interface.

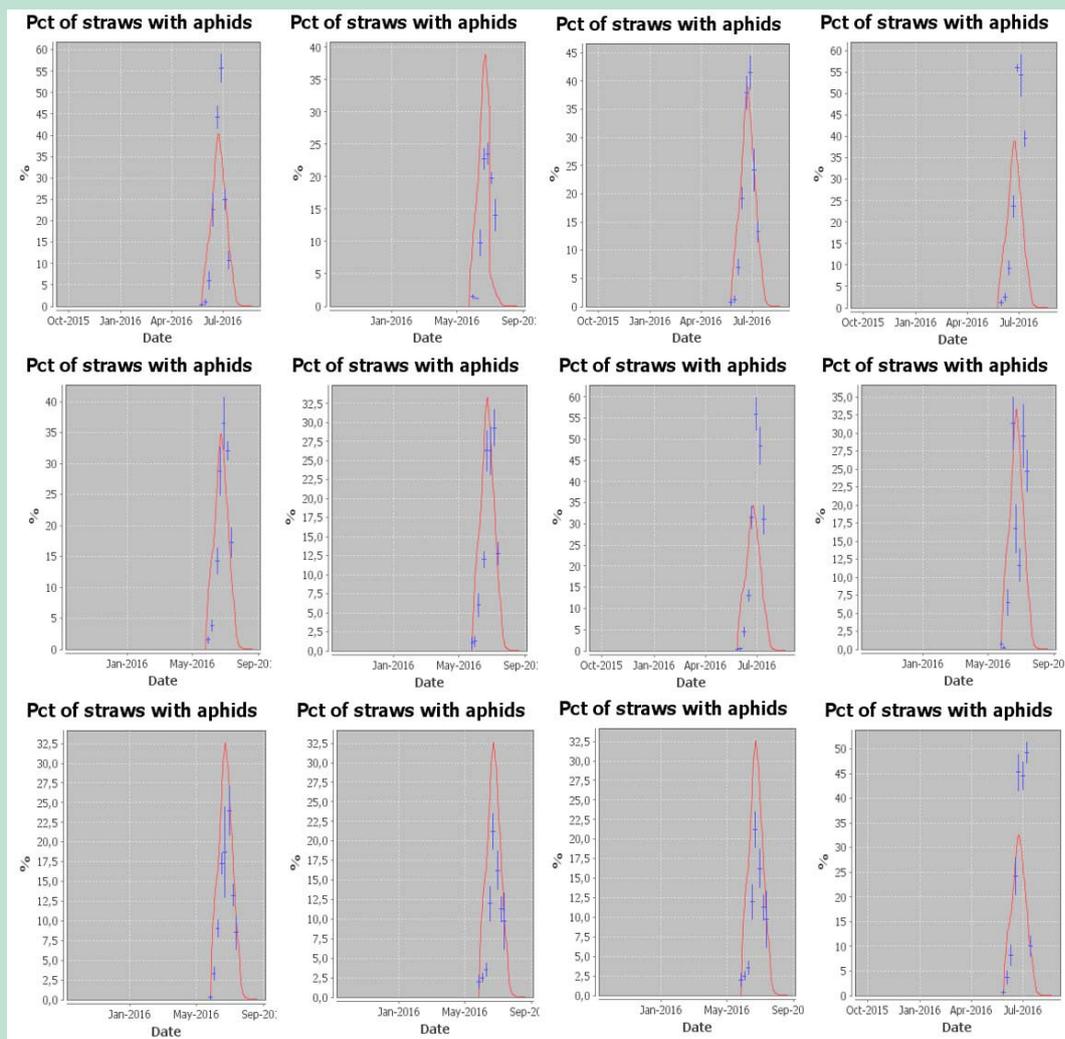


FIGURE 35. Comparisons between observed and simulated aphid densities when the observed initial density was the average initial density of all fields in a cluster from. The upper row is the Borum cluster, the middle row was from the Hammel cluster, and the bottom row was from the Viborg cluster. From left to the right you find first 0 km, 3 km, 6 km and, finally, 10 km. Mind different scales of the y-axes. Screen dump from SeptoriaSim user interface.

By comparing the simulations in Figure 34 and 35, we conclude that the best fit to observed data is achieved by using the average initial densities from a cluster as initial density of all fields in a cluster.

3.3 Growth analysis of winter wheat

3.3.1 Field data

The results (Figures 36–45) show that all leaves were found in the zone 0–15 cm until the rapid height growth started in early May. By the start of June, the wheat plants had reached a height that permitted a few leaves in the zone 61 – 75 cm. This demonstrates a very rapid height growth during May, which was also reflected in the results on the straws. The ears did not appear until late May or early June, and they were never seen in the zones below 45 cm. These comments apply to both varieties, Torp and Mariboss.

The growth analysis data were not tested for differences between the two varieties because the purpose of the analysis was to create data for calibration of SeptoriaSim.

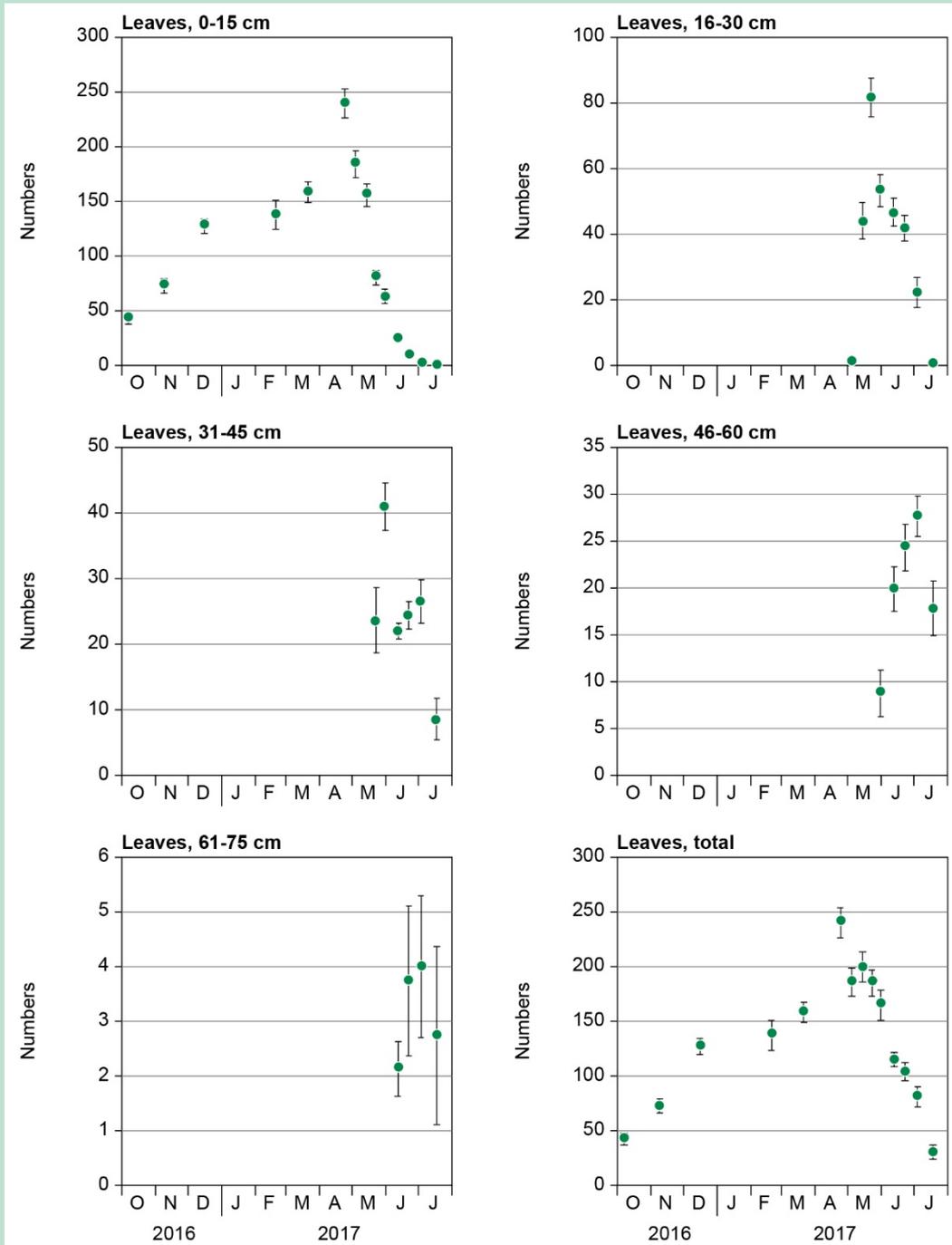


FIGURE 36. The number of leaves per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Torp.

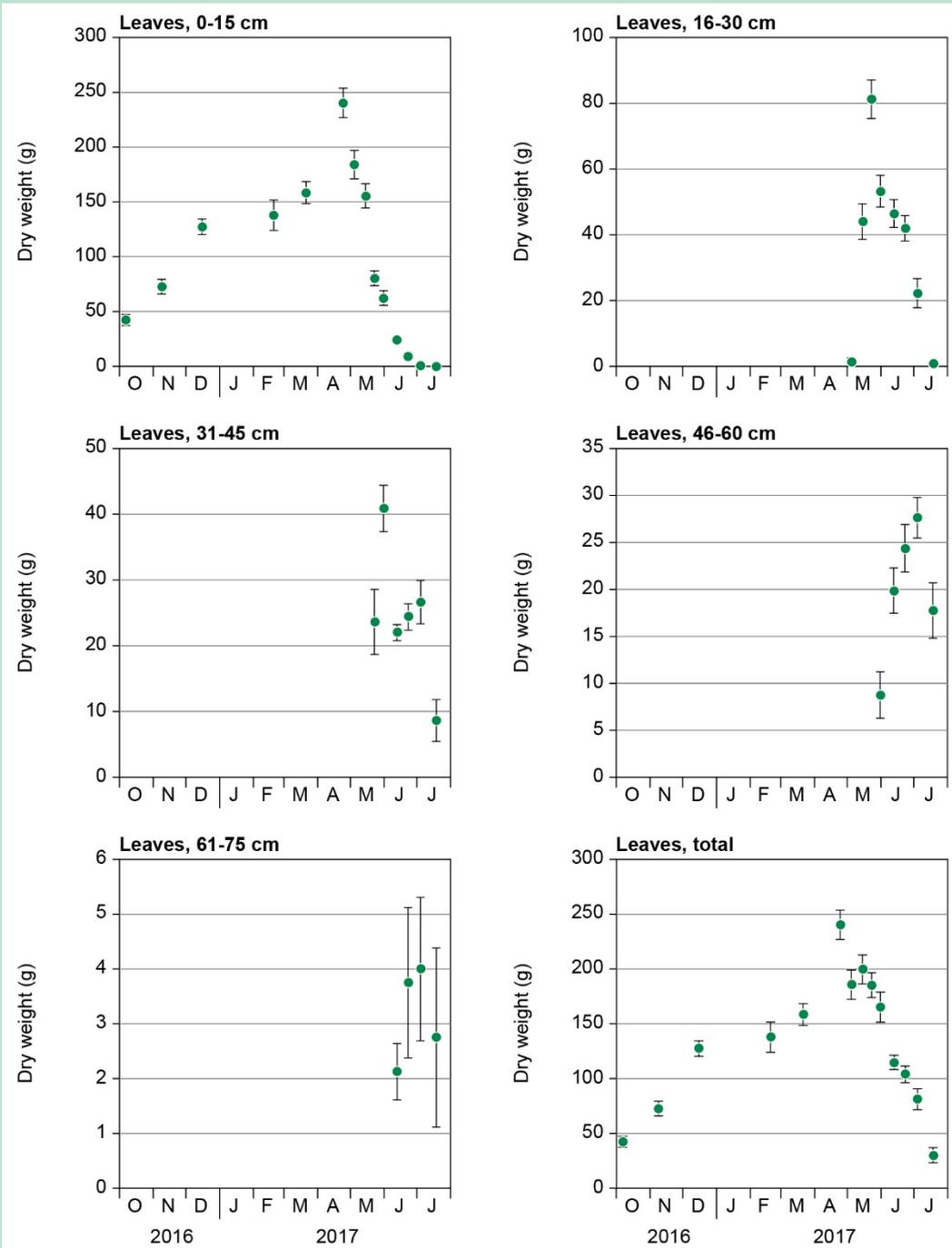


FIGURE 37. The dry weight of leaves per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Torp.

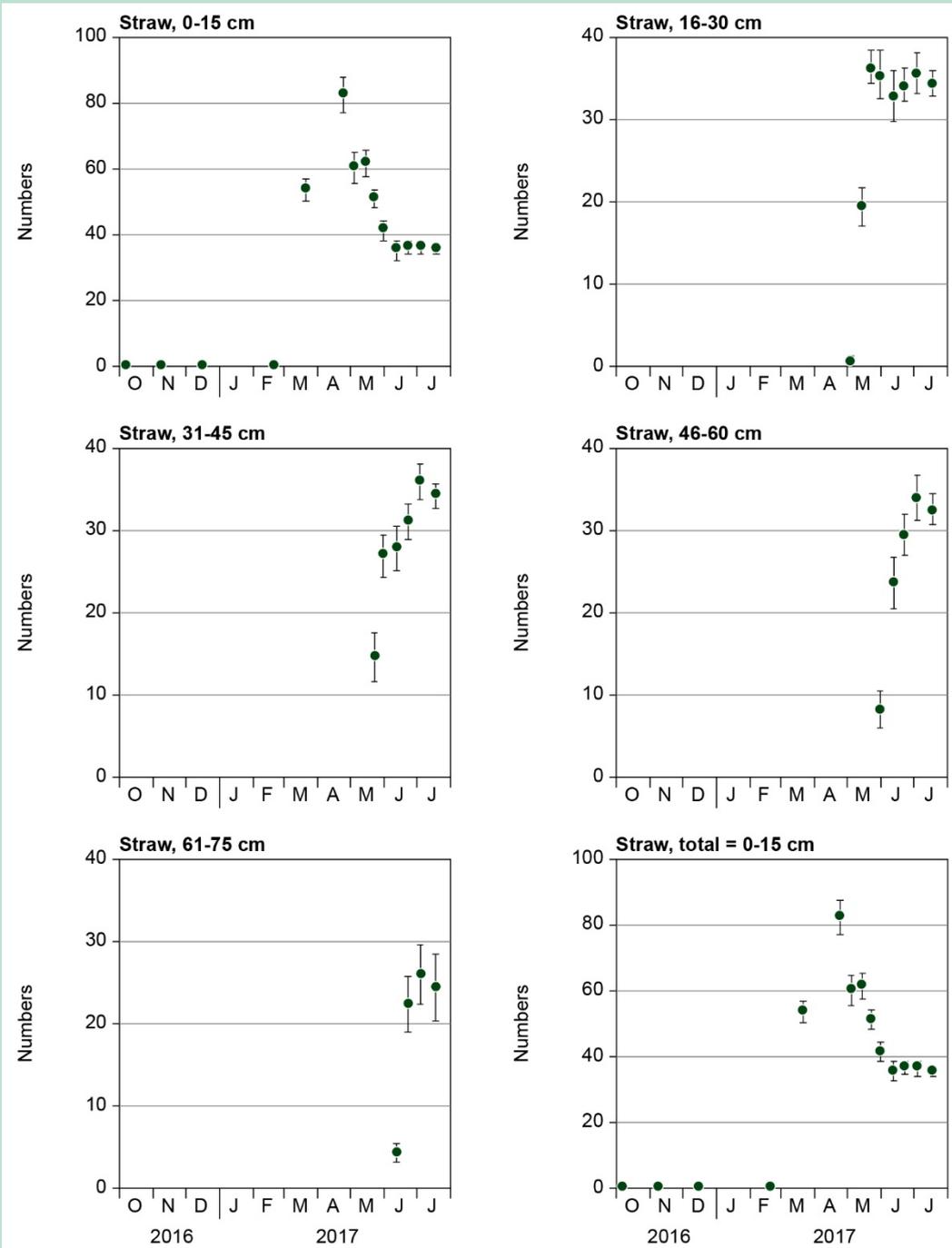


FIGURE 38. The number of straws per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Torp.

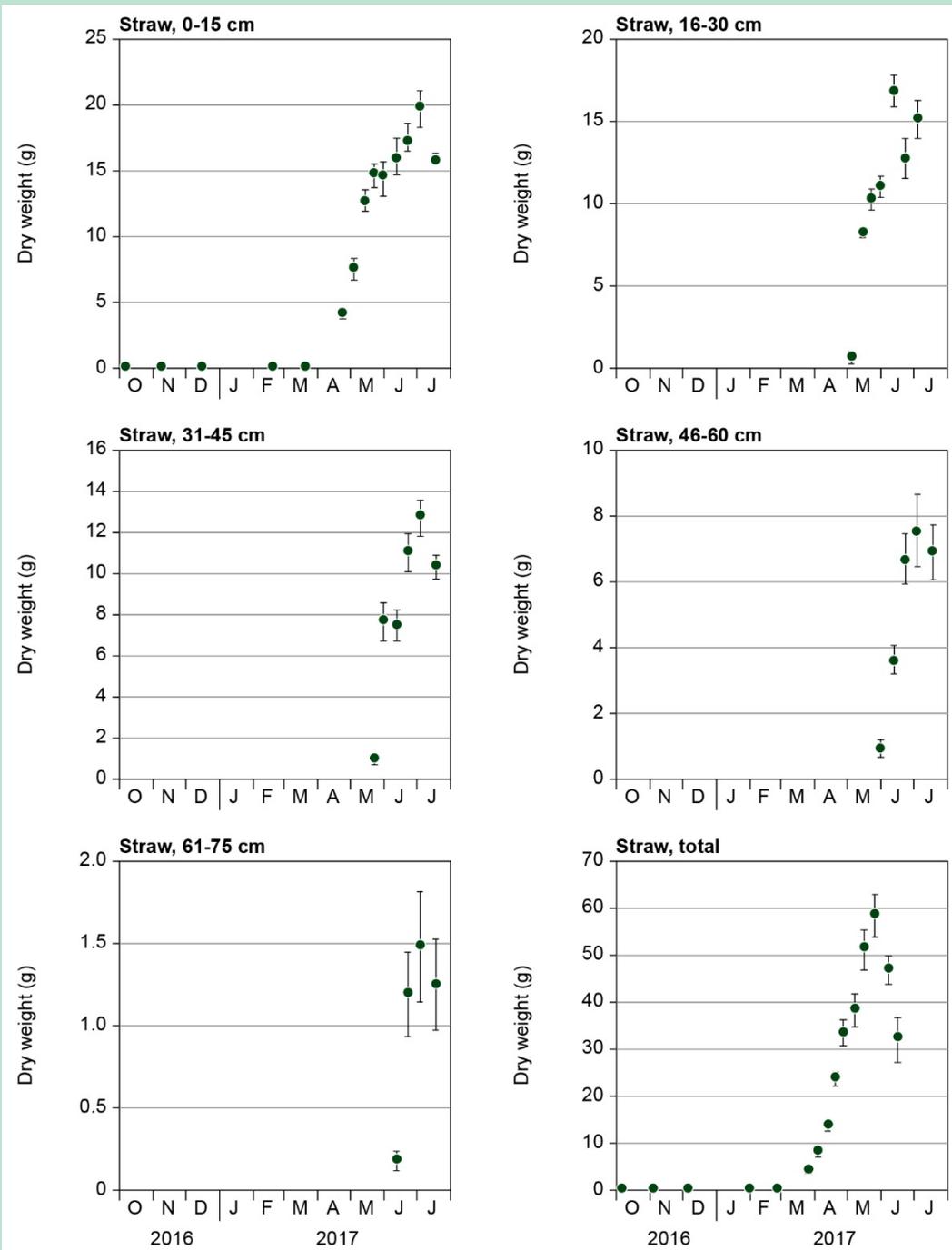


FIGURE 39. The dry weight of straws per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Torp.

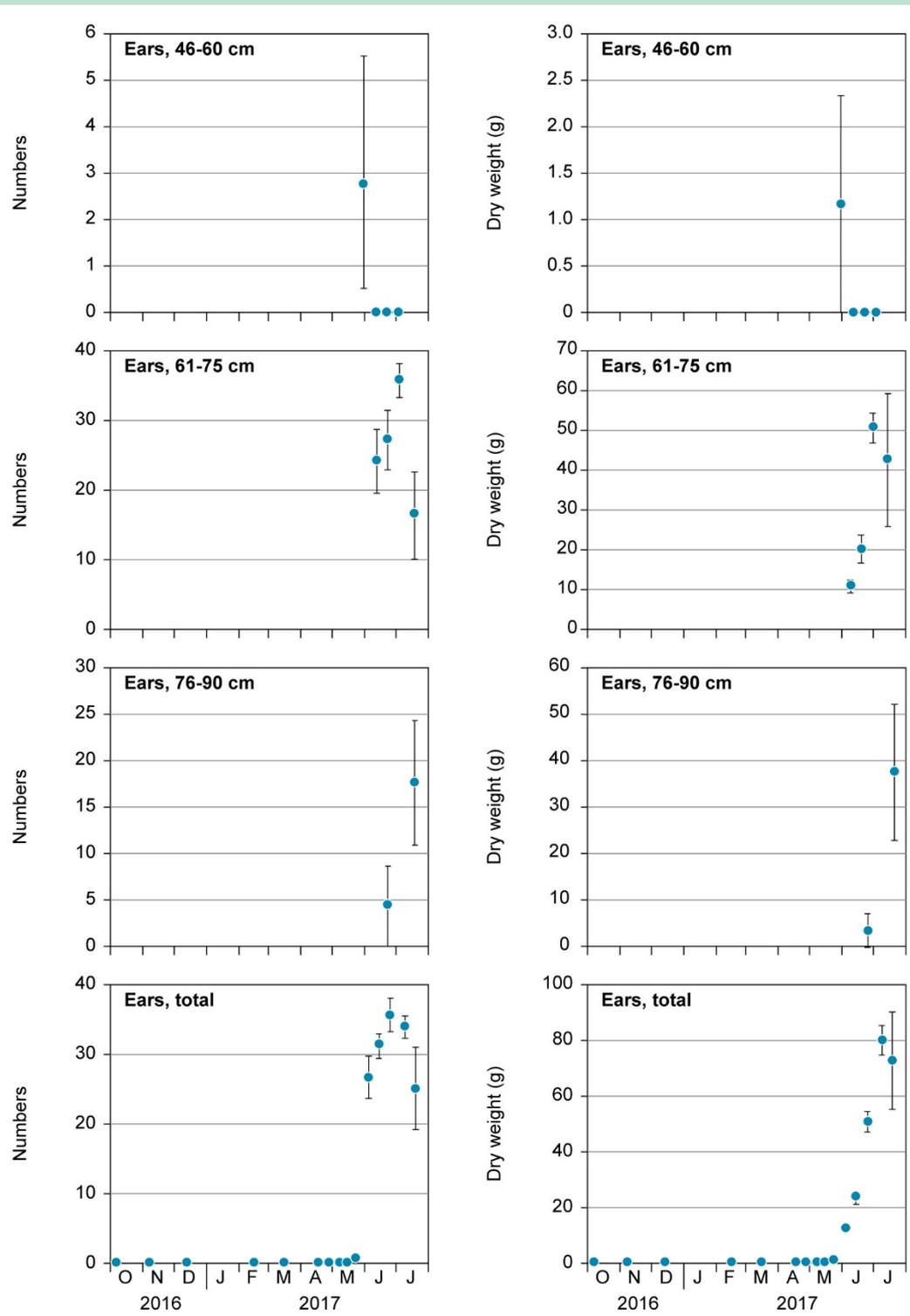


FIGURE 40. The numbers (left column) and dry weight (right column) of ears per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Torp.

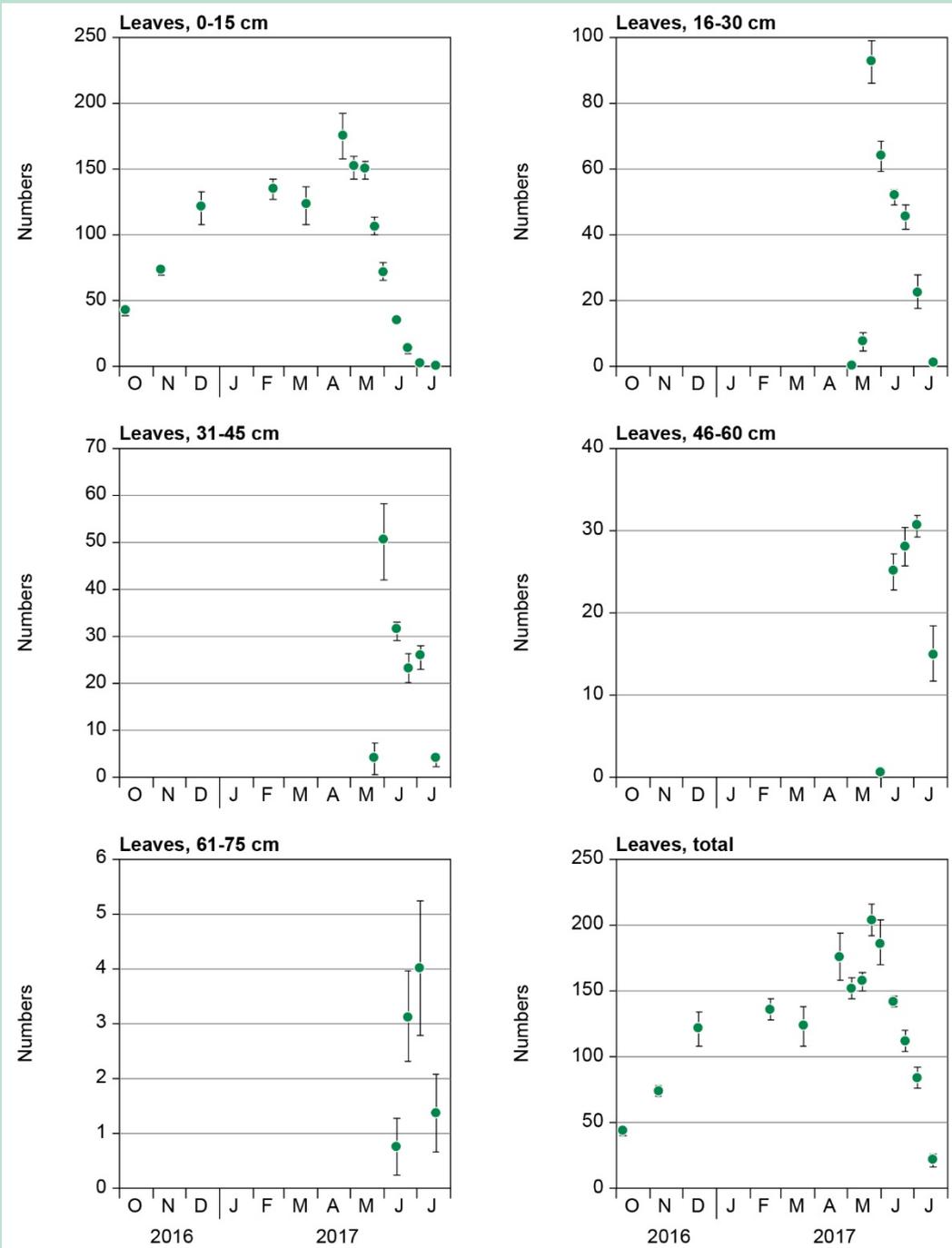


FIGURE 41. The number of leaves per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Mariboss.

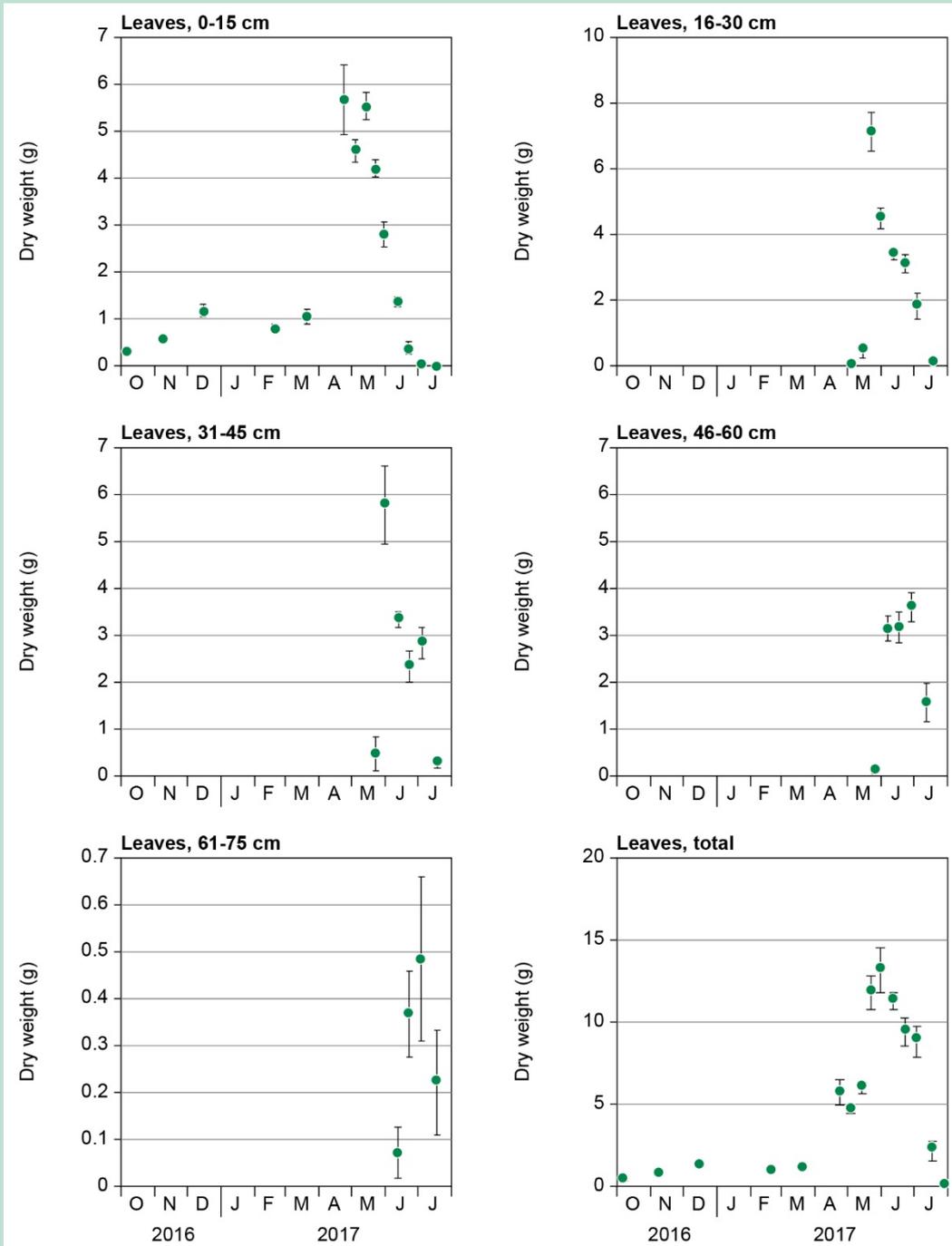


FIGURE 42. The dry weight of leaves per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Mariboss.

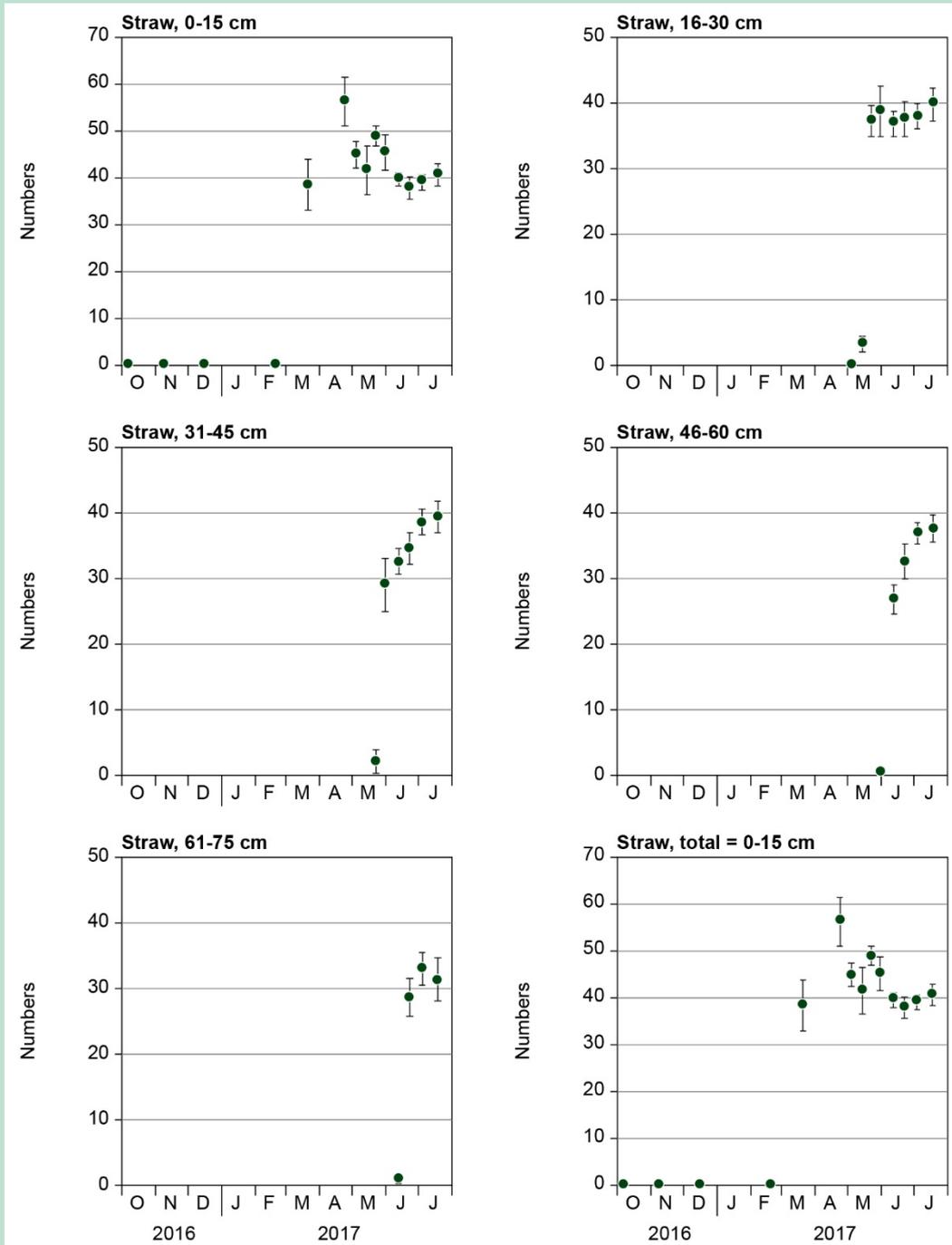


FIGURE 43. The number of straws per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Mariboss.

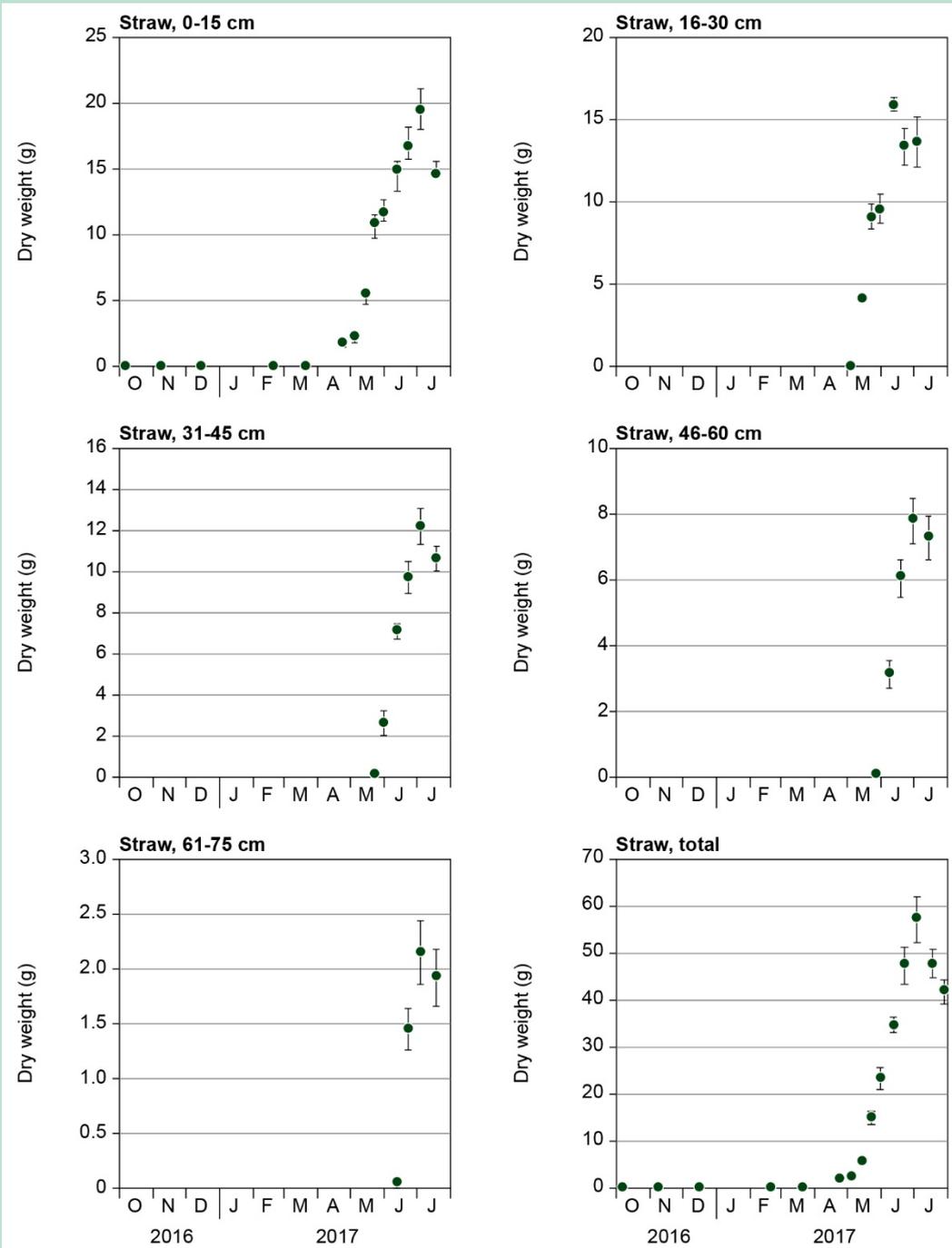


FIGURE 44. The dry weight of straws per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Mariboss.

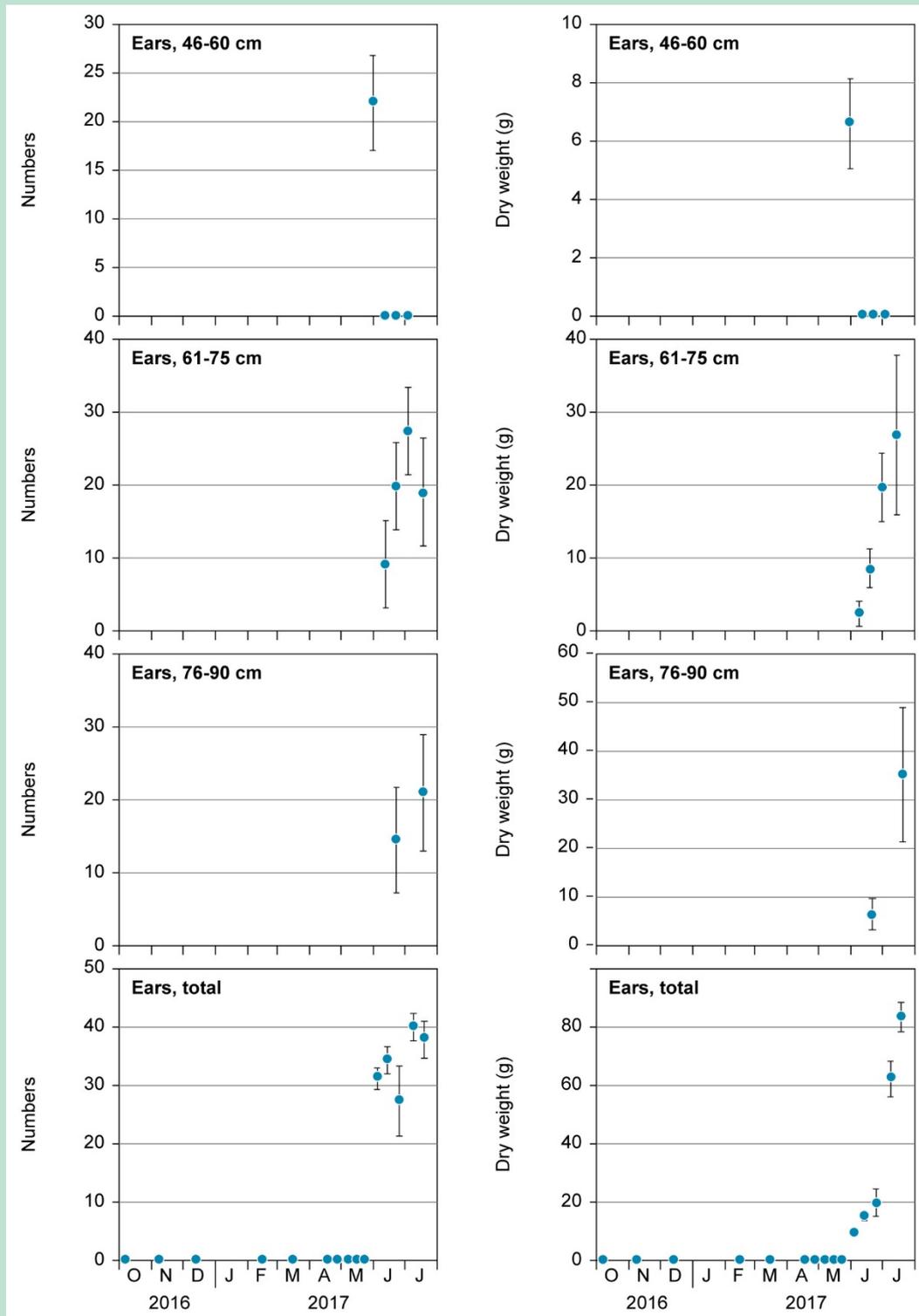


FIGURE 45. The numbers (left column) and dry weight (right column) of ears per 25 × 25 cm sample in different height zones during the growing season 2016-2017. Variety Mariboss.

The number of grains per ear and the mass of the grains (Table 2) differed between the two varieties, with Torp showing both larger numbers of grains and larger grain mass per ear than Mariboss.

TABLE 2. The number and biomass of grains of the two varieties Torp and Mariboss.

Variety	Number of grains per ear		Biomass of grains per ear	
	Average	SD	Average	SD
Torp	56.6	2.8	2.4	0.2
Mariboss	45.5	10.6	1.8	0.5

3.3.2 Winter wheat in SeptoriaSim

The winter wheat module of SeptoriaSim was originally calibrated in a two-step process to fit biomass growth data from 1992 and 1993 (Friis et al., 1995) and, then, to yield data from a historical dataset from 2003 to 2013. This calibration has been described by Bligaard et al. (2017). This calibration was not precise, as the data from 1992 and 1993 only covered the growth in total biomass from April to July, and the historical dataset only included sowing time, Septoria infestations, and yields. The latter dataset had the strength of consisting of data from around 150 field trials. However, the available datasets did not include information on the growth during autumn and winter, and they did not include information on the phenology of leaves, stems, and ears. Furthermore, they did not give any information on vertical distribution of biomass. This was found to be a weakness of the original version of SeptoriaSim, which was the reason why the detailed growth analysis was carried out in this project.

The growth analysis data shown in Figures 36-45 were used for comparisons with the simulated data from the original version of SeptoriaSim, and the comparison revealed a striking discrepancy between simulated and observed development in both leaves, tillers, and grains (Figures 46-48) for both varieties. This discrepancy strongly underlined the need for a re-calibration of the winter wheat module.

Calibration to the growth analysis data was done by: 1) changing input parameters such as the leaf creation rate, tillering rate, timing of tillering, bud creation rate, leaf creation rate, leaf growth rate, stem growth rate, and grain growth rate, 2) running the model, 3) comparing the graphical output from the simulation model with observed data and evaluating by eye whether the simulation fitted the observed data better. This was done manually and was stopped by the time the fit between observed and simulated values could not be improved further. The result was a much better fit between simulations and observations (Figures 49-54) for both varieties, Torp and Mariboss.

The winter wheat module with the original calibration was used in SeptoriaSim while running the simulations producing the warnings for the 2016 and 2017 trials.

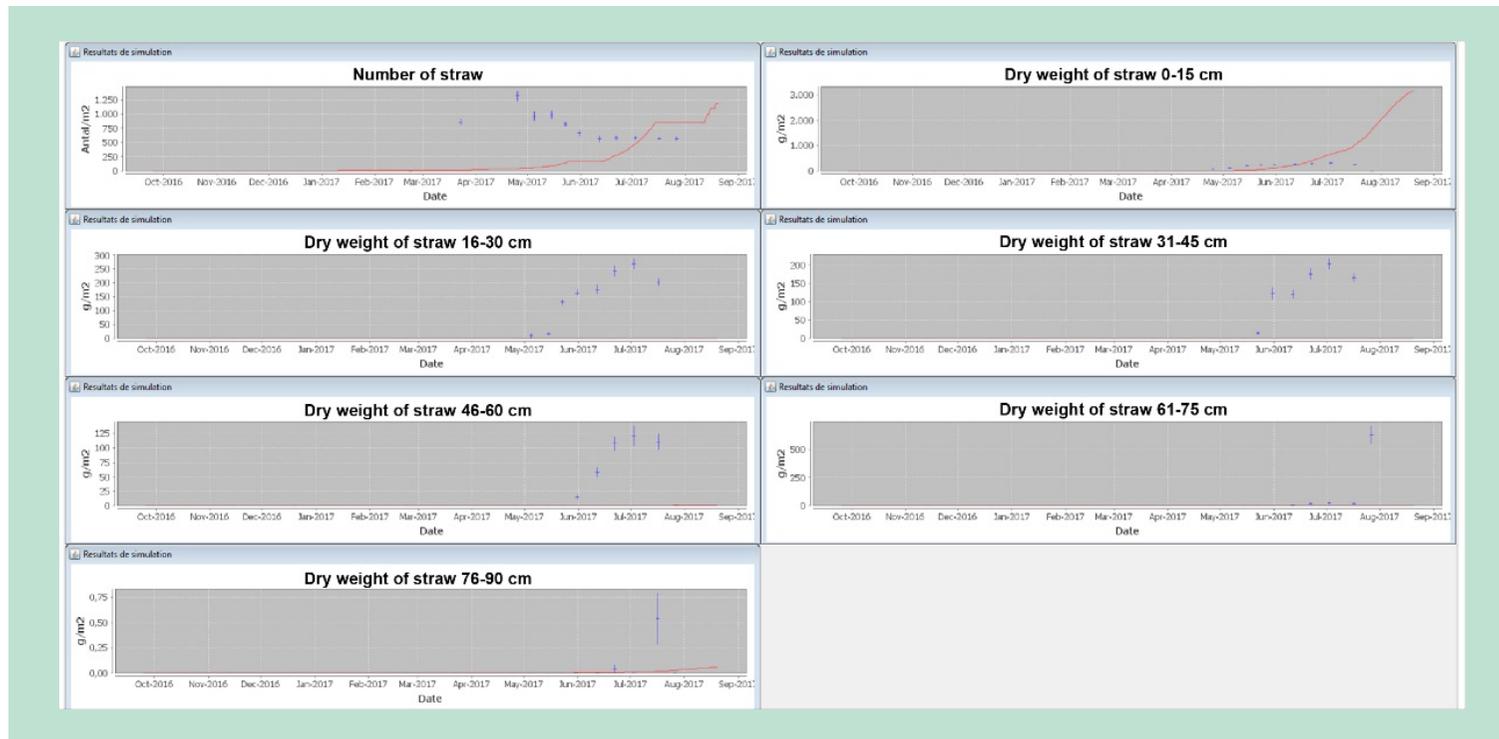


FIGURE 46. Comparison of simulated and observed values numbers of tillers per m², and dry weight of tillers in different height zones before calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

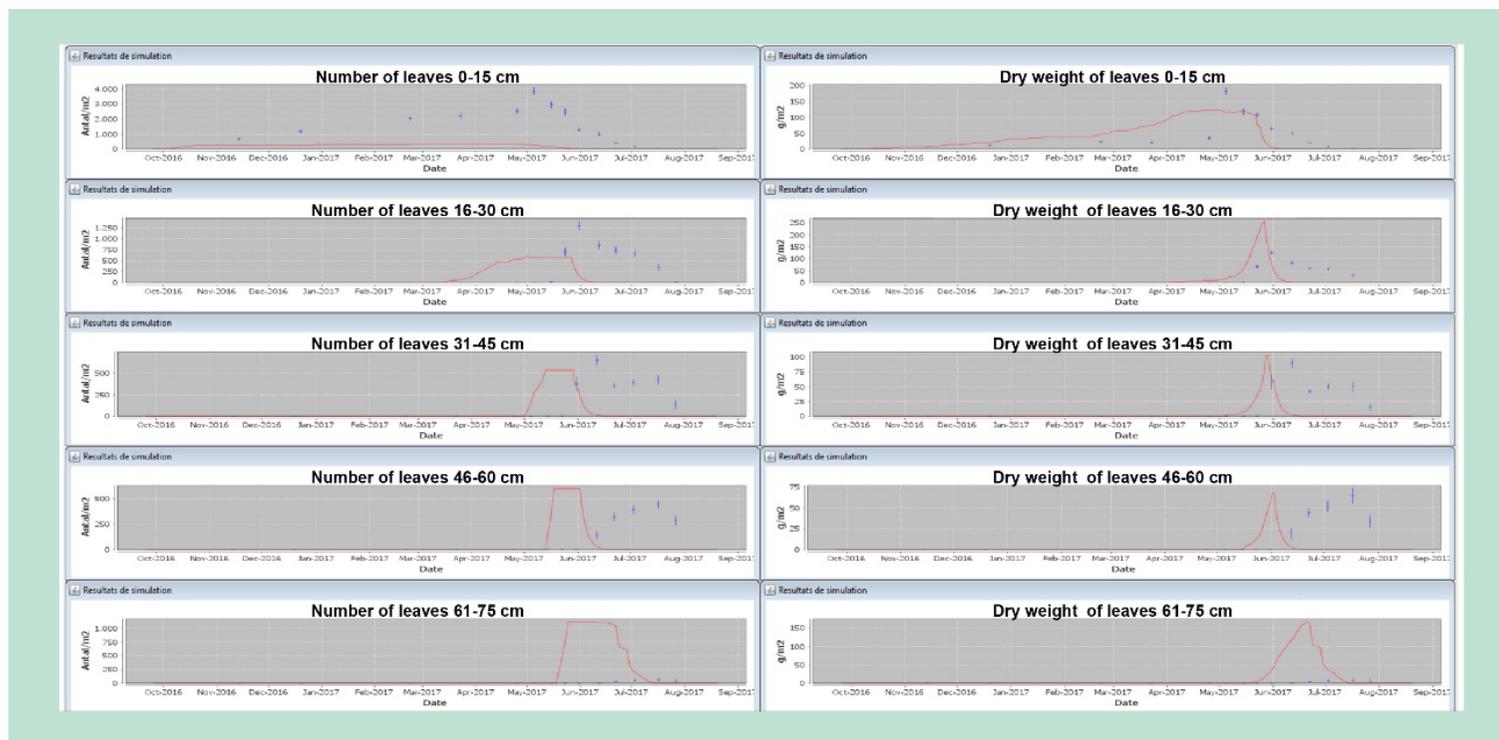


FIGURE 47. Comparison of simulated and observed values numbers and dry weight of leaves per m² in different height zones before calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

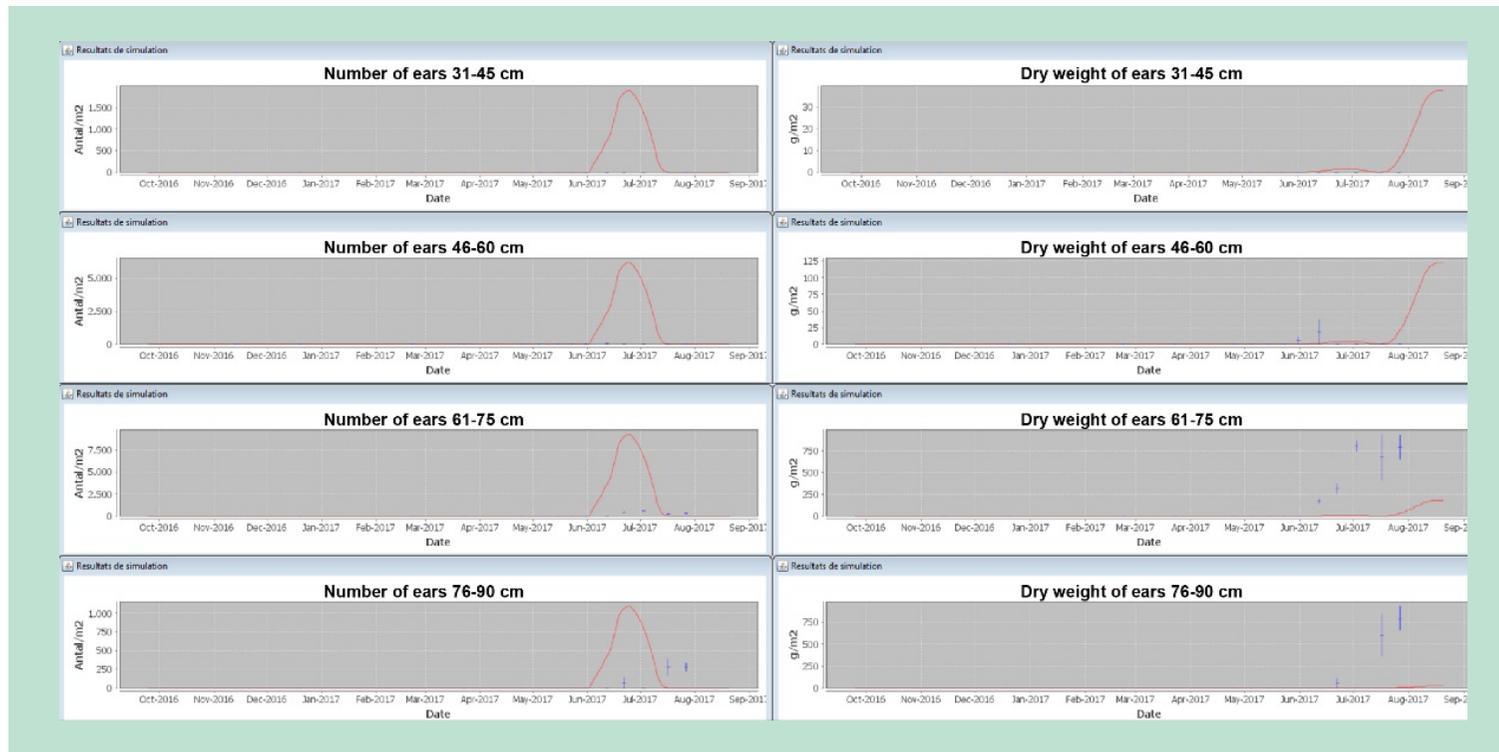


FIGURE 48. Comparison of simulated and observed values numbers and dry weight of leaves per m² in different height zones before calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

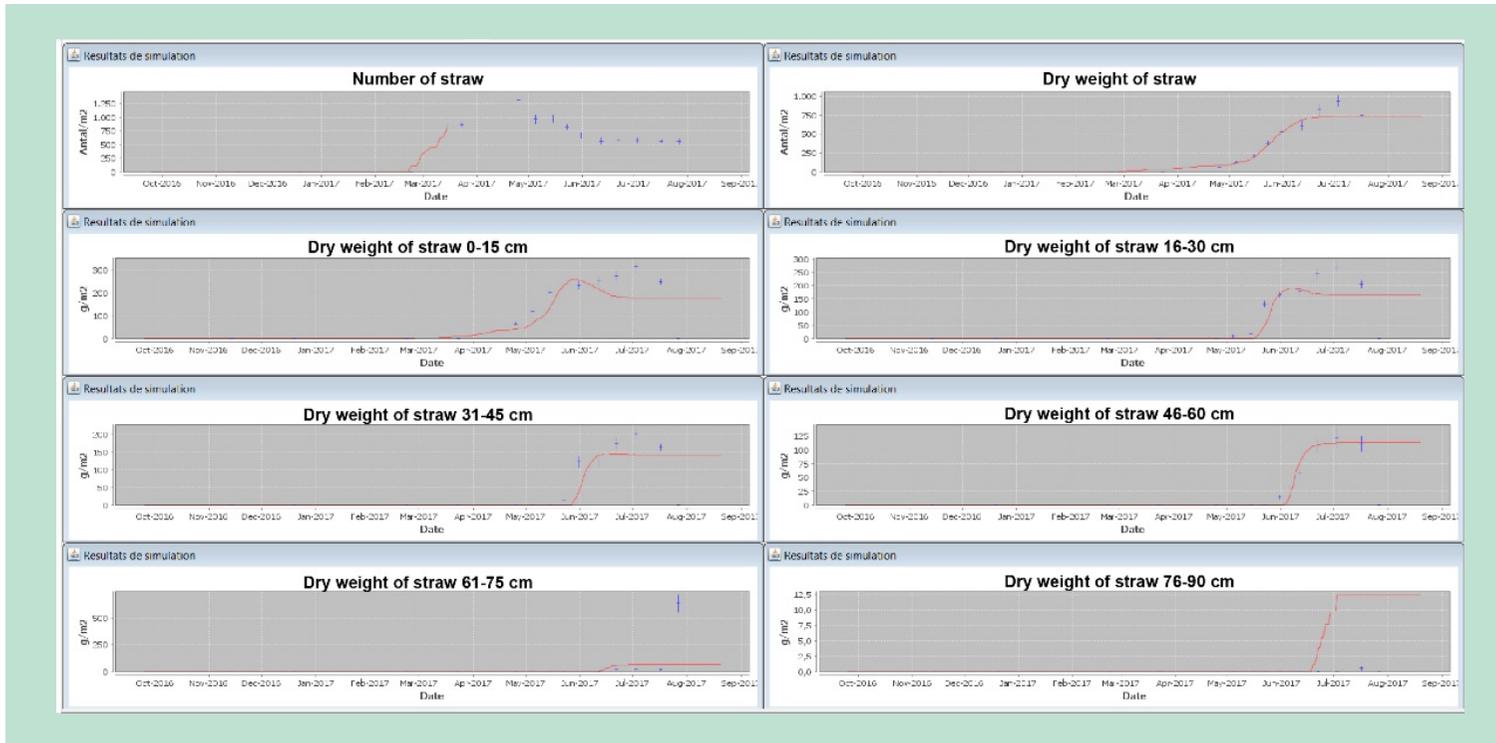


FIGURE 49. Comparison of simulated and observed values numbers of tillers per m², and dry weight of tillers in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.



FIGURE 50. Comparison of simulated and observed numbers of leaves per m² (left column), and dry weight of leaves (right column) in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

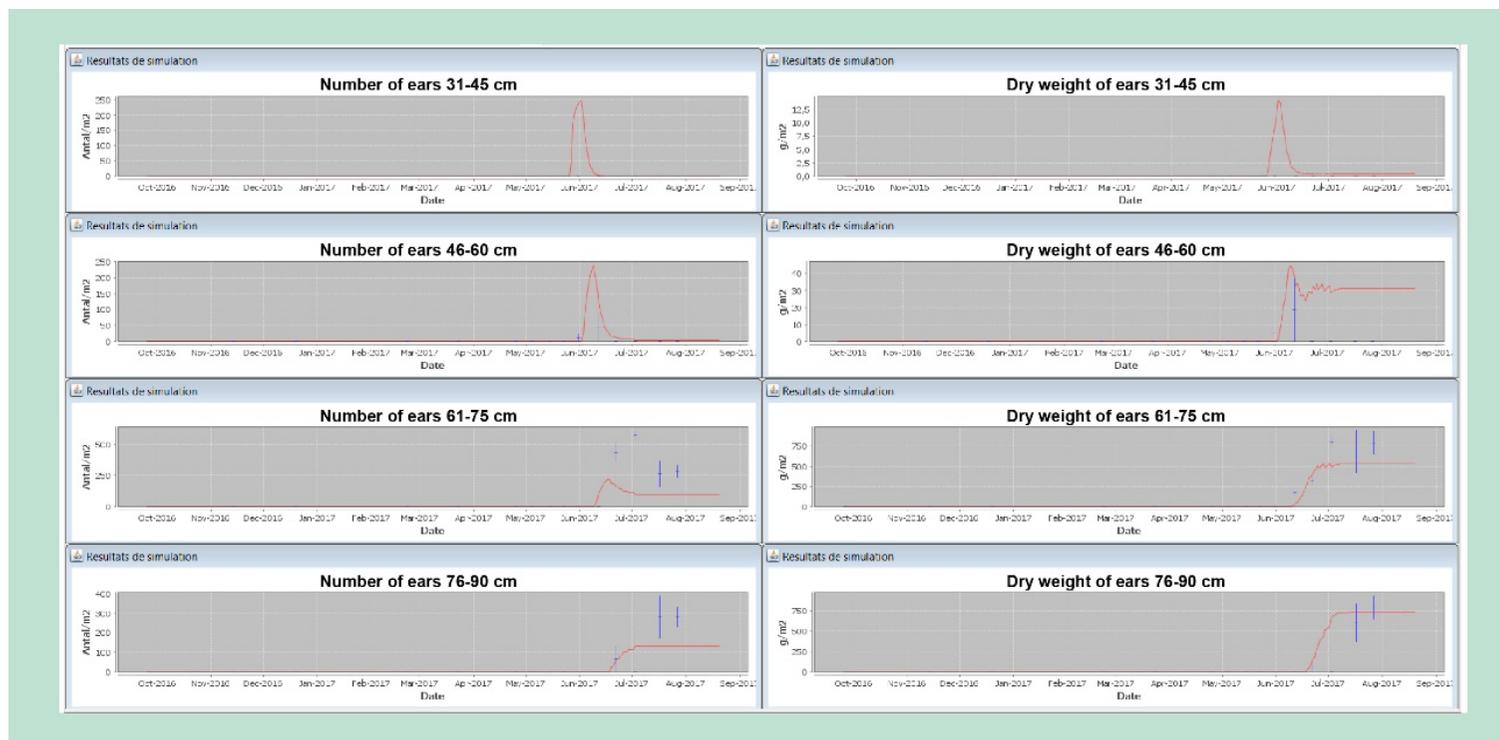


FIGURE 51. Comparison of simulated and observed numbers of ears per m² (left column), and dry weight of ears (right column) in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Torp. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

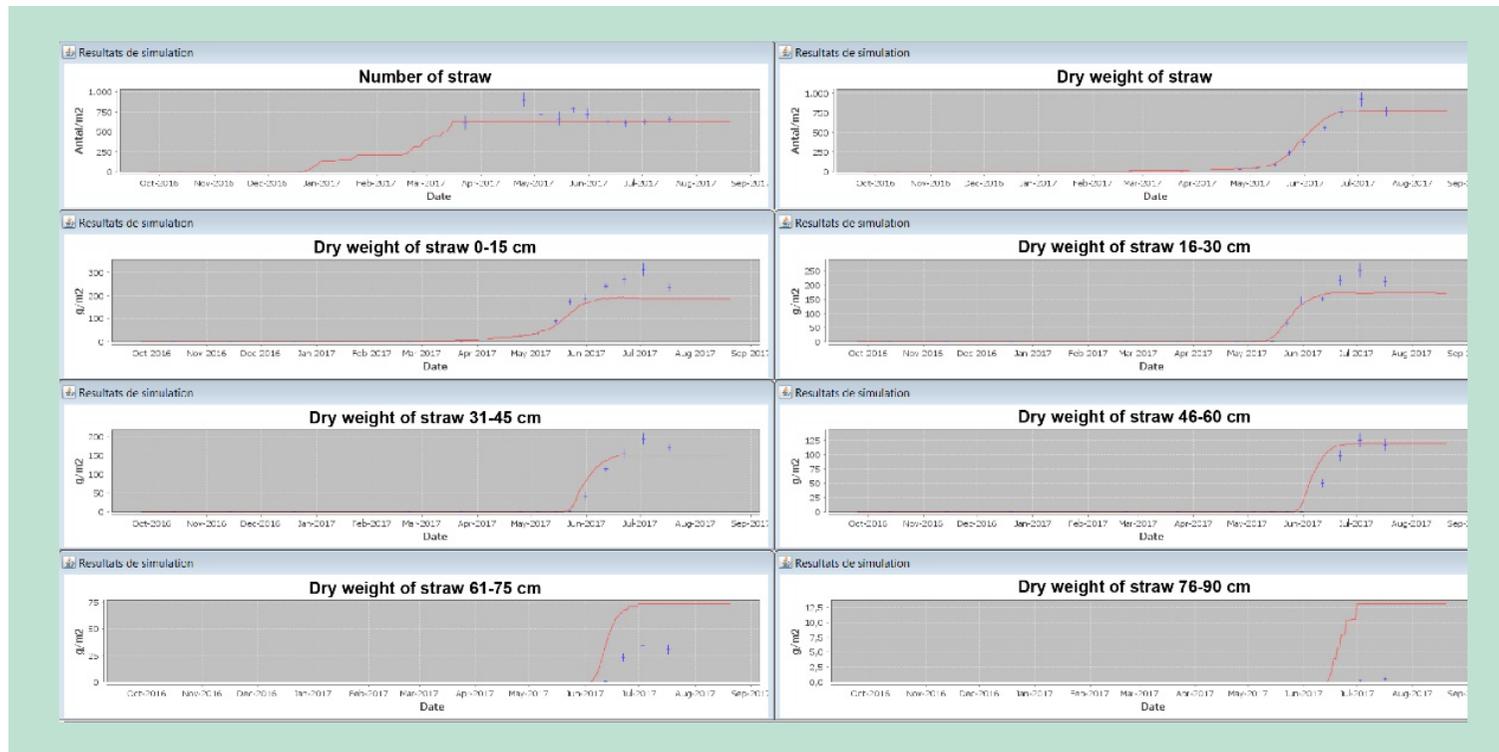


FIGURE 52. Comparison of simulated and observed values numbers of tillers per m², and dry weight of tillers in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Mariboss. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.



FIGURE 53. Comparison of simulated and observed numbers of leaves per m² (left column), and dry weight of leaves (right column) in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Mariboss. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

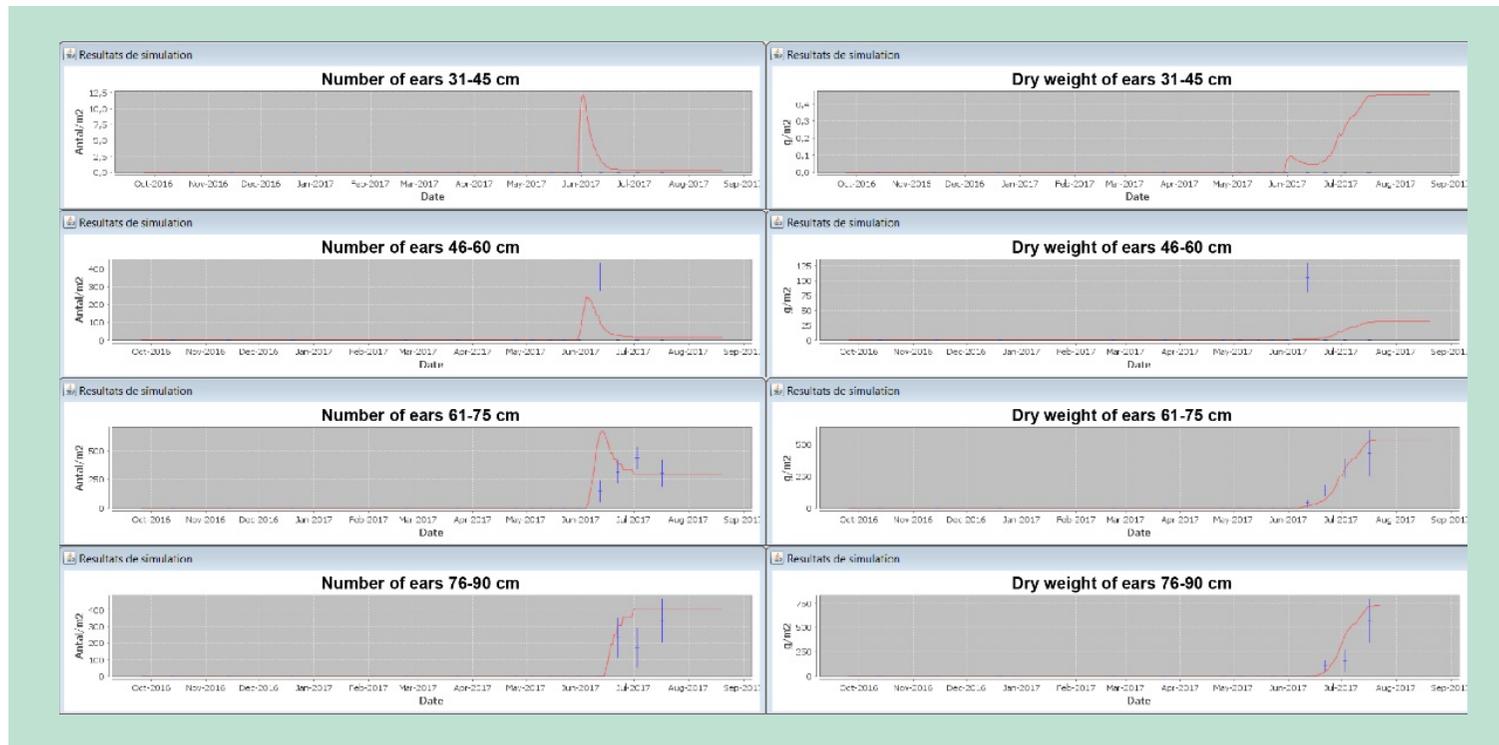


FIGURE 54. Comparison of simulated and observed numbers of ears per m^2 (left column), and dry weight of ears (right column) in different height zones after calibration to observed data from the growing season 2016-2017. The winter wheat variety was Mariboss. The observed values are the horizontal blue lines, the vertical blue lines are standard deviation, and the red line is the simulated values. Screen dump from SeptoriaSim user interface.

3.4 Decision support system trials

3.4.1 Flakkebjerg, Holeby, Horsens

Seven field trials testing three different models were carried out in 2016 and 2017, where the outcome of the models were compared with reference treatments applied up to three timings. In 2016, SeptoriaSim recommended an early treatment at all locations at first 10 days of May, and in Horsens a second treatment was recommended at the end of May. The humidity model (Model 2) recommended a treatment at all three localities following an event with 20 hours with 85% relative humidity in late May. Finally, CPO recommended one treatment in Horsens and Flakkebjerg, but none in Holeby (Table 3). This meant that SeptoriaSim, the Humidity model, and CPO on average released 1.33, 1.00, and 0.67 treatments in 2016, respectively.

In 2017, SeptoriaSim recommended one treatment at Horsens, but two treatments at the other two localities. The humidity model recommended two treatments at Flakkebjerg and Holeby, but three treatments in Horsens. CPO recommended three treatments in Flakkebjerg and Horsens, and only one in Holeby (Table 4). SeptoriaSim, the Humidity model, and CPO on average released 1.67, 2.33, and 2.33 treatments, respectively.

TABLE 3. Detailed dates for applications based on the three models SeptoriaSim, Humidity model, and Plant Protection On-line (PVO) in 2016.

2016	Flakkebjerg	Horsens	Holeby
SeptoriaSim	10 May	10 May + 7 June	4 May
Humidity model	26 May	26 May	27 May
CPO	24 May	26 May	None

TABLE 4. Detailed dates for applications based on the three models SeptoriaSim, Humidity model, and CPO in 2017.

2017	Flakkebjerg	Horsens	Holeby
SeptoriaSim	11 May + 23. May	26 May	9 May + 24 May
Humidity model	11 May + 8 th June	6 th May, 26 th May + 15 June	18 th May + 1 th June
CPO	3 th May + 23 th May + 8 th June (Sheriff only two last timings)	6 th May+ 26 th May + 15 June	23 th May

The yield responses in 2016 were lower than in 2017, which was in accordance with the general trends for the two seasons (Tables 5 and 6). Spraying had a significant positive impact on the yield in five of the seven experiments. Especially, the experiments with the susceptible varieties Nakskov and Hereford showed good positive responses. The experiments in Holeby 2016 and Horsens 2017 showed no significant effects on yield.

Spraying according to the models gave yield responses at about the same level as the standard treatments with triple application, except in Flakkebjerg variety Nakskov in 2016, where the triple application produced a slightly higher yield (Table 5). In 2017, following the recommendations of all models gave average yield responses between 3.0 and 19.8 hkg/ha (Table 6). In Horsens in the resistant variety Sheriff, following the recommendations of all three models (and the standard treatment schemes) gave only weak and insignificant increases, after low to

moderate attacks of Septoria. The trial with the resistant variety Sheriff at Flakkebjerg only responded half as much as the susceptible variety Hereford. The results confirmed that the input should be differentiated depending on the susceptibility of the variety. The resistant variety, Sheriff, was over treated in the trials in 2017, even though CPO only recommended two treatments in this cultivar, as the recommendations are delayed to first start at growth stage 37.

TABLE 5. Detailed yield data from the three validation trials carried out in 2016. Cost of treatments are specified and deducted from the yield in each treatment– providing a net yield.

	16300-1 (Flakkebjerg)						16300-2 (Horsens)			16300-3 (Holeby)			Average
	GS 32-33	GS 37-39	GS 55	Variety: Nakskov			Variety: Sheriff			Variety: Torp			
				Yield and increase hkg/ha	Cost hkg/ha	Net yield hkg/ha	Yield and increase hkg/ha	Cost hkg/ha	Net yield hkg/ha	Yield and increase hkg/ha	Cost hkg/ha	Net yield hkg/ha	Net yield hkg/ha
1. Untreated				80.6			90.4			89.3			
2. Bell 0.5	Bell 0.5	Bell 0.5	Bell 0.5	9.5	5.8	3.7	5.0	5.8	-0.8	-1.3	5.8	-7.1	-1.4
3.		Bell 0.5		2.9	2.9	6.0	4.9	2.9	2.0	-1.4	2.9	-4.3	1.23
4.		Bell 0.5	Bell 0.5	10.5	5.8	4.7	4.3	5.8	-1.5	0.1	5.8	-5.7	-0.83
5. Bell 0.5	Bell 0.5	Bell 0.5	Bell 0.5	10.8	8.7	2.1	4.8	8.7	-3.9	-1.4	8.7	-10.1	-3.97
6. SeptoriaSim				3.7	2.9	0.8	6.3	5.8	0.5	-2.1	2.9	-5.0	-1.23
7. Humidity model				7.3	2.9	4.4	3.9	2.9	1.0	0	2.9	-2.9	0.83
8. Crop protection online				8.2	2.9	5.3	4.5	2.9	1.6	-1.9	0	-1.9	1.67
LSD ₉₅				5.9			3.6			NS			

TABLE 6. Detailed yield data from the four validation trials carried out in 2017. Cost of treatments are specified and deducted from the yield in each treatment – providing a net yield.

			17300-1 (Flakkebjerg) Variety: Hereford			17300-2 (Flakkebjerg) Variety: Sheriff			
	GS 32-33	GS 37-39	GS 55	Yield and increase hkg/ha	Cost hkg/ha	Net yield hkg/ha	Yield and increase hkg/ha	Cost hkg/ha	Net yield hkg/ha
1. Untreated				84.0			94.6		
2.	Prosario 0.5	Bell 0.75		14.2	6.6	7.6	4.3	6.6	-2.3
3.		Bell 0.75		7.9	4.1	3.8	3.7	4.1	-0.4
4.		Bell 0.75	Prosaro 0.5	14.6	6.6	8	5.3	6.6	-1.3
5.		Bell 0.75	Prosaro 0.5	18.7	9.0	9.7	7.8	9.0	-1.2
6.		Bell 0.75	Prosaro 0.5	18.1	9.0	8.1	10.0	9.0	1.0
7.	Septoria Sim			16.8	6.6	10.2	7.9	6.6	1.3
8.	Humidity model			9.0	6.6	2.4	4.2	6.6	-2.4
9.	Crop protection online			19.8	9.0	10.8	8.2	6.6	1.6
	LSD ₉₅			5.0			5.0		

			17353-1 (Horsens) Variety: Sheriff			17353-2 (Holeby) Variety: Torp			
	GS 32-33	GS 37-39	GS 55	Yield and in-crease hkg/ha	Cost hkg/ha	Net yield hkg/ha	Yield and in-crease hkg/ha	Cost hkg/ha	Net yield hkg/ha
1. Untreated				94.0			114.2		
2.	Prosario 0.5	Bell 0.75		0.8	6.6	-5.8	12.7	6.6	6.1
3.		Bell 0.75		0.7	4.1	-3.4	9.9	4.1	5.8
4.		Bell 0.75	Prosaro 0.5	1.1	6.6	-5.5	15.8	6.6	9.2
5.		Bell 0.75	Prosaro 0.5	4.7	9.0	-4.3	17.0	9.0	8
6.		Bell 0.75	Prosaro 0.5	3.5	9.0	-5.5	17.9	9.0	7.9
10.	Septoria Sim			3.2	4.1	-0.9	13.0	6.6	6.4
11.	Humidity model			3.0	9.0	-6	17.6	6.6	11
12.	Crop protection online			3.0	9.0	-6	10.5	4.1	6.4
	LSD ₉₅			NS			6.2		

TABLE 7. Average net yields from the experiment in 2017 for all varieties and split on susceptible and resistant varieties.

				Average of susceptible varieties (Hereford and Torp)	Average of resistant variety (Sheriff)	Average of all varieties
GS 32-33	GS 37-39	GS 55		Net yield hkg/ha	Net yield hkg/ha	Net yield hkg/ha
1.Untreated				99.0	94.3	96.7
2.	Prosaro 0.5	Bell 0.75		6.85	-4.05	1.4
3.		Bell 0.75		4.80	-1.9	1.45
4.		Bell 0.75	Prosaro 0.5	8.60	-3.4	2.6
5.	Prosaro 0.5	Bell 0.75	Prosaro 0.5	8.85	-2.3	3.05
6.	Prosaro 0.5	Bell 0.75	Prosaro 0.5	8.00	-2.3	2.88
7.SeptoriaSim				8.30	0.2	4.25
8.Humidity model				6.70	-4.2	1.25
9.Crop protection online				8.60	-2.2	3.2

Concerning the net yield, which is most relevant for the farmers, all treatments showed very limited responses in 2016, ranging from averages of – 3.97 in the triple standard scheme to 1.67 following the recommendations of CPO (Table 5). Of the three models, CPO performed best in 2016, with the humidity model being second, but generally treating against Septoria in 2016 appeared not to increase net yields much.

In 2017, the average net yields ranged between averages of 1.25 hkg/ha using the humidity model to 4.25 hkg/ha following SeptoriaSim. Following CPO gave the second best net yield, 3.2 hkg/ha. Following both the standard treatments and the three models gave higher average net yields in 2017 than the best performing one in 2016.

Four out of the seven experiments showed generally negative or neutral net yield responses to fungicide treatments, while the only ones showing clear positive treatments were the experiments with susceptible varieties from Flakkebjerg (Nakskov in 2016, and Hereford in 2017) and the medium susceptible variety Torp in Holeby in 2017 (Table 7). Both experiments with Sheriff in 2017 showed generally negative net yields. However, using the models produced slightly increased net yields in Sheriff in all cases except in Flakkebjerg in 2017. Looking at the averages of the 2016 treatments (across varieties), CPO performed best, the humidity model was number two and SeptoriaSim number three. In 2017, SeptoriaSim performed best, with CPO being second and the humidity model third (Table 7). Generally, the net yields produced using the decision support systems were even with (susceptible varieties) or slightly higher (resistant variety) than the ones produced when treating according the standard schemes with three treatments. This especially applies to SeptoriaSim and CPO.

Concerning the number of treatments, SeptoriaSim recommended fewest treatments against Septoria in the trials in 2017 (average 1.80) and most treatments (average 1.33) in 2016. CPO behaved oppositely by triggering most fungicide applications in 2017 (average 2.25) and fewest in 2016 (0.67). Over the two years, SeptoriaSim and CPO triggered a very similar number of treatments (average 1.46 and 1.55 respectively), while the humidity model triggered 1.6.

3.4.2 Farmers' field tests

The results show low and non-significant increases in yield and 1000-grain weight in the trial plots (Table 8), and the Septoria treatment frequency was slightly lower in the trial plots than outside the trial plots. The aphid treatment frequency was the same in the trial plots as outside the trial plots.

TABLE 8. Results from the SeptoriaSim trials in farmers' fields.

Field	Trial plot				Outside trial plot			
	Yield (g)	1000-grain weight (g)	Septoria treatments (Dates or numbers)	Aphid treatments (Dates or numbers)	Yield (g)	1000-grain weight (g)	Septoria treatments (Dates or numbers)	Aphid treatments (Dates or numbers)
1	55.1 (2.5)	43.8 (2.0)	5/18, 6/3	No	63.3 (2.7)	41.8 (1.8)	2.0 (dates unknown)	No
2	68.4 (3.3)	38.1 (1.0)	5/18, 6/3	No	69.7 (3.1)	41.2 (0.7)	5/11, 6/10	No
3	78.2 (6.5)	44.8 (1.0)	5/16, 6/3	No	78.2 (6.5)	44.8 (1.0)	5/16, 6/3	No
4	72.8 (2.1)	50.2 (0.7)	5/18, 6/3	No	65.3 (2.7)	44.8 (1.0)	5/19, 6/15	No
5	76.0 (5.1)	41.0 (1.0)	5/21, 6/3	6/15	64.8 (2.6)	40.8 (1.1)	5/11, 6/2, 6/19	6/19
Avg.	70.1 (3.7)	43.6 (1.8)	2.0	0.2	68.3 (2.5)	42.7 (0.8)	2.2	0.2

Following the alerts from SeptoriaSim caused a slight decrease in Septoria treatments while keeping at least the same yield.

3.4.3 Relate to national average spray intensities

The general disease pressure in winter wheat in 2016 was moderate. In 2017, the attack was generally more severe, which is reflected in yield responses being 11.5 hkg/ha and 17.2 hkg/ha, respectively, for the two seasons (Figure 55) (Oversigt over landsforsøgene, 2017). This indicates that in 2016, 1-2 treatments were relevant at most sites, while, in 2017, 2-3 treatments would have been more appropriate. The experiences from different seasons confirm a major variation in Septoria risk across the country, which is also reflected in the Treatment Frequency Index level for fungicides across the country (Figure 56). Similar variation is seen for control of insecticides across the country, indicating that some regions, e.g. Langeland and Stevns, notoriously have a higher pressure of aphids than other regions (Figure 56).

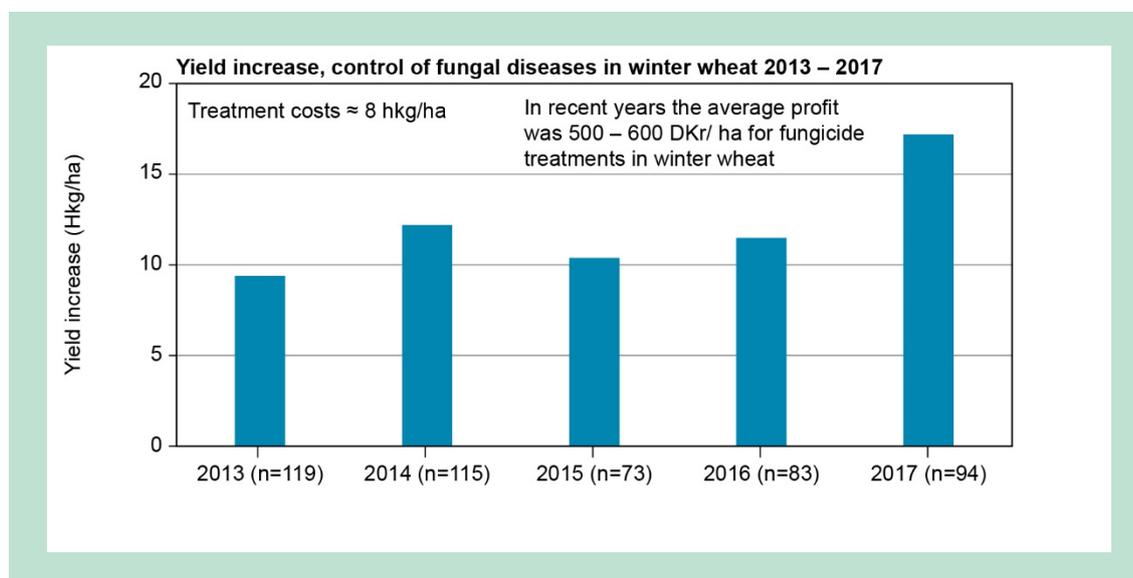


FIGURE 55. Average yield responses from disease control in winter wheat (Plantekongres, based on Ghita Nielsen 2018).

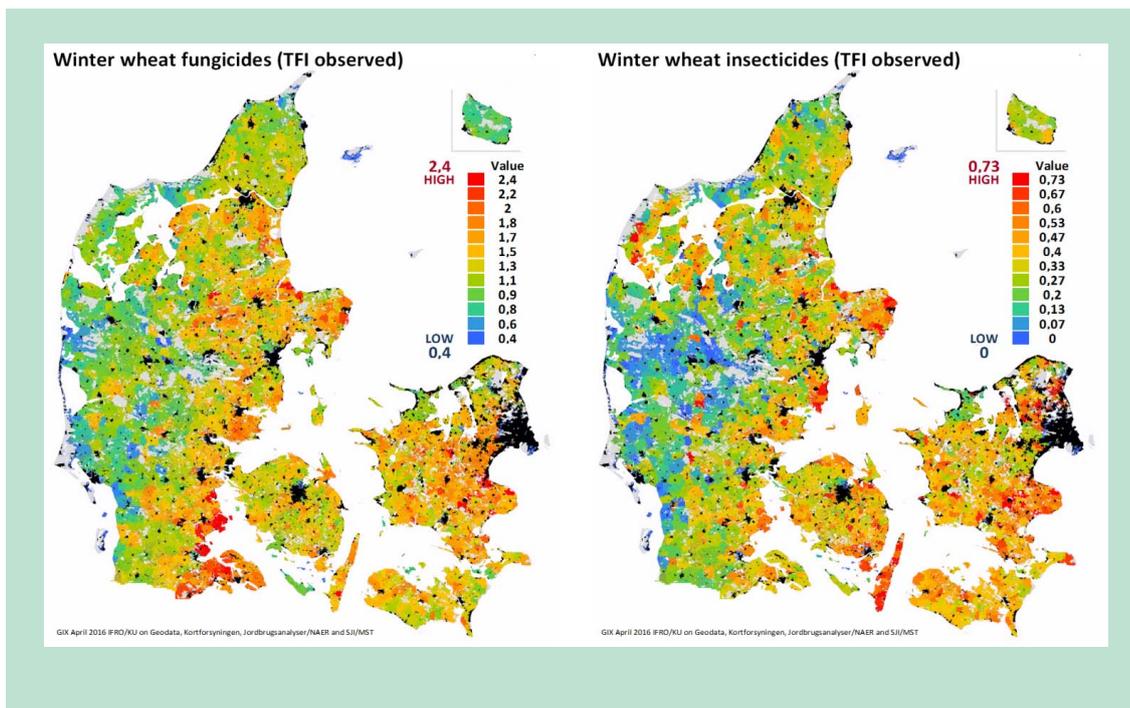


FIGURE 56. Treatment Frequency Index (TFI) values for fungicides and insecticides measured based on 4 years of farm spray reports (Ørum unpublished).

The Treatment Frequency Index is adjusted for dosages lower than labelled, and it does not show the number of applications per year. The number of fungicide applications has increased in Denmark since 2010 and has been above 2.5 since 2014 (Figure 57). Most fungicide applications in winter wheat are directed against *Septoria*, which means that the number of treatments released by all three models (*SeptoriaSim*, CPO, and the humidity model; 1.33, 1.00, 0.66, respectively) in 2016 was clearly below the national average number of fungicide treatments of about 2.6. The difference is smaller in 2017, where *SeptoriaSim*, CPO, and the humidity model released 1.66, 2.33, and 2.33 treatments, respectively, compared to the national average of around 2.7.

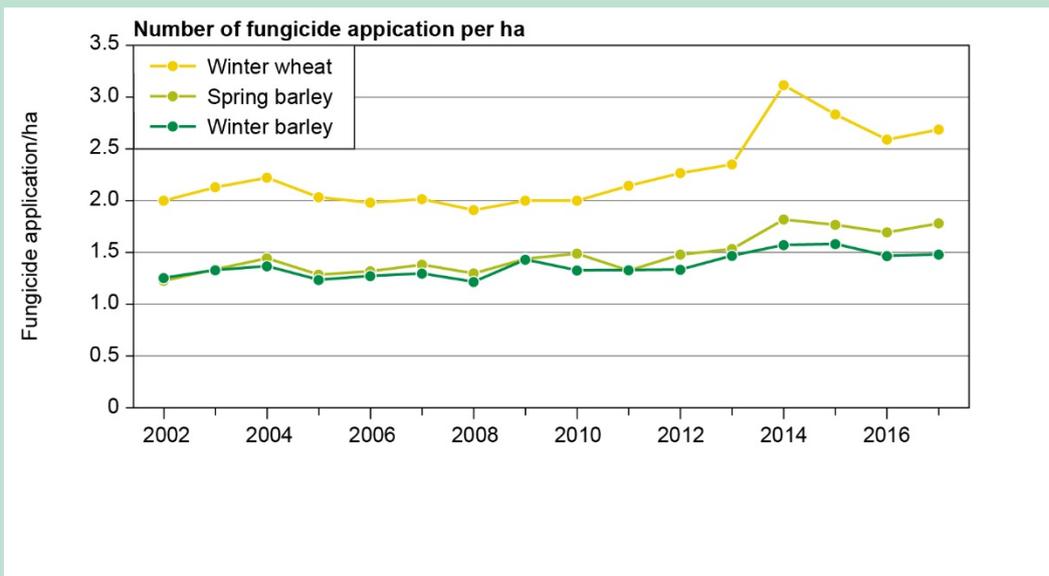


FIGURE 57. Average number of fungicide applications in Denmark from 2002 to 2017 in winter wheat, spring barley and winter barley (Based on Kleffmann-GmbH)

3.5 Recalibration of SeptoriaSim

3.5.1 SeptoriaSim projections – recalibrated model

After re-calibration of SeptoriaSim to the winter wheat growth analyses and to the observations of Septoria cover and DNA (QPCR-data), the procedure concerning emissions of warnings in May and June were repeated using the same weather files as originally used. In 2016, the new calibration would have triggered one application in Flakkebjerg, which is the same as the original version, and on almost the same date. In the resistant variety, Sheriff, the re-calibrated version would only have released one treatment in Horsens, which is one less than the original version. Only one treatment was triggered in Holeby on almost the same date using both calibrations (Table 9). Concerning the average number of treatments, the calibration reduced the figure from 1.33 to 1.00, which is clearly lower than the national average of about 2.6 (Figure 57)

TABLE 9. Dates for applications in original and re-calibrated versions of SeptoriaSim, Controlled experiments 2016.

2016	Flakkebjerg - Variety: Nakskov	Horsens - Variety: Sheriff	Holeby - Variety: Torp	Average treatments
SeptoriaSim original calibration	10 May	10 May + 7 June	4 May	1.33
SeptoriaSim re-calibrated	12 May	13 May	6 May	1.00

In the susceptible variety (Hereford) in 2017, the recalibration would have triggered only one treatment in Flakkebjerg compared to two with the original calibration, one treatment in Horsens (Table 10), which is equal to the original version, and two in Holeby, which is also similar to the result using the original version. The new version would, thus, have triggered an average treatment frequency of 1.5 in susceptible varieties in the controlled experiments in 2017, which is lower than the original version (2.0) and clearly lower than the national average (Figure 57).

TABLE 10. Dates for applications in original and re-calibrated versions of SeptoriaSim, Controlled experiments in 2017. Variety: susceptible to Septoria (Hereford, Torp).

2017	Flakkebjerg - Variety: Hereford	Holeby - Variety: Torp	Average treatments
SeptoriaSim original calibration	11 May + 23. May	9 May + 24 May	2.0
SeptoriaSim recalibrated	8 May	6 May + 27 may	1.5

In the resistant variety (Sheriff) in 2017, the recalibrated version released only one treatment in Flakkebjerg (Table 11), which is a reduction compared to the original version that released two treatments. In Horsens, the result with the re-calibrated version was almost the same as with the original one, and in Holeby, the recalibrated version released no treatments compared to two treatments with the original version. On average, the recalibrated version would have released only 1.0 treatments, which is a reduction from 1.5 when using the original version.

TABLE 11. Dates for applications in original and re-calibrated versions of SeptoriaSim, Controlled experiments in 2017. Variety: Good resistance to Septoria (Sheriff).

2017	Flakkebjerg	Horsens	Average treatments
SeptoriaSim original calibration	11 May + 23. May	26 May	1.5
SeptoriaSim recalibrated	6 May	28 May	1.0

Farmers' field tests

Variety: Torp

In the farmers' fields, the recalibrated version triggered first spray warnings against Septoria a little earlier than the original version, and no second treatments (Table 12). Concerning aphids, the recalibrated version released no applications, while the original calibrations released an application in one of the fields. Therefore, the use of the recalibrated version would have reduced the pesticide burden in the fields considerably. A comparison of the treatment frequency in the fields outside the trial plots with the treatment frequency indicated that the number of applications obtained by the re-calibrated version the difference would have been 1.4 lower.

TABLE 12. Dates for applications in original and re-calibrated versions of SeptoriaSim in farmers' fields.

2017	Field 1	Field 2	Field 3	Field 4	Field 5	Average treatments
SeptoriaSim original calibration, Septoria	18 May + 3 June	18 May + 3 June	16 May + 3 June	18 May + 3 June	21 May + 3 June	2.0
SeptoriaSim recalibrated, Septoria	14 May	14May	14 May	14 May	16 May	1.0
SeptoriaSim original calibration, Aphids	None	None	None	None	15 June	0.2
SeptoriaSim recalibrated, Aphids	None	None	None	None	19 June	0.2

4. Discussion

4.1 Decision support system trials

The overall objective of this project was to develop a combined decision support system for Septoria and aphids in winter wheat. This objective was fulfilled by improving the winter wheat and Septoria modules of SeptoriaSim and extend it with an aphid module. However, it was also an immediate objective to try out the system in field trials, which was done in controlled, randomized block experiments and in farmers' fields.

The results of the trials will here be discussed in relation to two endpoints: 1) the net yield of the treatment, i.e. the net revenue of the produced yield when the cost of the treatments has been subtracted, and 2) the number of treatments with fungicides and insecticides. The first endpoint is important for the farmers' economy, and the second endpoint is important for the environment and the Danish Environmental Protection Agency.

The trials did not provide significant results on whether the decision support systems produced better net yields than the standard schemes with three treatments or two late treatments. In some experiments, the decision support systems gave better yields than the standard schemes, and in other experiments, they showed the opposite result. The same conclusion was not reached by Bligaard et al. (2017) in similar experiments from 2014 and 2015, where following CPO gave lower net yields than standard schemes with two and three treatments, and the humidity model performed at about the same level in the variety Mariboss and better in Hereford. In Bligaard et al. (2017), SeptoriaSim was only tested in 2015, when this model was found to be the economically most attractive one. This means that overall the trials of Bligaard et al (2017) and this project reveal that SeptoriaSim performed best in two out of three years, but performed weakest in the third year. However, the year when SeptoriaSim performed weakest was in 2016, when the responses to treatments was generally weak. The humidity model performed best in 2014-2015 in Hereford, and it never produced negative net yields, came second in 2016, but was rather unstable in 2017. It is, therefore, not possible to decide which one of the models is economically most attractive, but SeptoriaSim gave the best net yields in both 2015 and 2017, which is two out three years.

Concerning the number of treatments, the results suggest that by using the decision support systems it was possible to reduce the number of treatments to about one in 2016 and about two in 2017. This means that decisions support systems can be used to reduce the number of treatments without losing net yields compared to the standard schemes with triple applications. In fact, in many cases the net yield was even positive compared to the triple standard applications. The results of the trials were not conclusive on which system reduced the number of treatments most, as CPO produced fewest treatments in 2016 and SeptoriaSim most. In 2017, this picture switched with SeptoriaSim releasing fewest treatments. The humidity model was second both years.

In the tests carried out in farmers' fields, the yield in the SeptoriaSim trial plots was not significantly higher than in the areas outside the trial plots, which was no surprise, as there were only slight differences in the spraying dates between the trial plots and the surrounding fields.

The results presented here for SeptoriaSim were made with the original calibration, and therefore the prognoses were made once again with the re-calibrated version, which built on investigations presented in this report. Using the recalibrated version of SeptoriaSim would have reduced the average number of treatments by 0.5 in the controlled experiments in both variety

types in 2017 and by 0.33 average in 2016. This means that the recalibrated version of SeptoriaSim clearly would have released fewer treatments than the originally used version, making it, environmentally speaking, clearly the most attractive model.

The recalibrated version was calibrated to results from 2016 and 2017 and could not be used in the trials. Therefore, it is not known whether the fewer treatments released by this version would have affected the net yield. However, in 2016 the recalibrated version removed the second treatment in Horsens, which was the trial where the two other models released only one treatment. The net yield of the trials with these two models and the standard scheme with only one treatment showed the highest net yields in this experiment. This indicates that one treatment might have been optimal. The recalibration also removed one treatment in Sheriff in Flakkebjerg in 2017. The other models also released two treatments in Sheriff in Flakkebjerg, and most standard treatment schemes produced negative net yields that give no indications of the impact of removing a treatment on the net yield. However, the treatments generally had low impact on the net yield in this experiment, which means that removing a treatment might not have changed things noticeably. Therefore, it is likely that removing the treatments, as suggested by the recalibrated version, would not have changed the net yield negatively.

The tests carried out in the farmers' fields showed slightly fewer Septoria treatments in the trial plots (2.0) than in the rest of the fields (2.2). Using the recalibrated version of SeptoriaSim, only 1.0 treatment would have been released in all fields. It is not possible to derive any strong conclusions concerning yields from the tests in the farmers' fields, but they pointed towards a slightly higher yield with slightly fewer treatments.

If the numbers of Septoria treatments released by the decision support systems are compared to the national average fungicide treatments (Figure 57), the decision support systems all released clearly fewer applications in both 2016 and 2017, with a large difference in 2016. This reduction would have been even larger if the recalibrated version based on the detailed investigations on growth of both winter wheat and Septoria presented in this report had been used.

Concerning aphids, SeptoriaSim did not release applications of insecticides in the controlled experiments, as the densities of aphids in these fields were very low in both 2016 and 2017. In the farmers' fields, SeptoriaSim released an application against aphids in one of the fields in mid-June, but could not justify a tank mixture of insecticide and fungicide, as a treatment against Septoria was not economically justified. This was not changed by using the recalibrated version of SeptoriaSim.

Comparing the performance of the decision support systems with the national Treatment Frequency Index is difficult, as the Treatment Frequency Index is blurred by different dosages. The national average treatment frequency of insecticides in winter wheat from 2012 to 2015 was 0.31 (Bekæmpelsesmiddelstatistikken 2012, 2013, 2014, and 2015), which is about 50% higher than seen in the trials in farmers' fields and much higher than seen in the controlled experiments, where no treatments against aphids were triggered. Furthermore, this Treatment Frequency Index may hide a number of applications with reduced dosages. This suggests that using SeptoriaSim can reduce the number of treatments against aphids in Denmark. Whether the same is the case for fungicides against Septoria is uncertain, but comparing the number of treatments released in the trials in this project with the national average treatments, it seems very likely. The latter conclusion, however, hinges on the assumption that the largest part of the fungicide applications in Danish winter wheat (Figure 57) is directed against Septoria.

4.2 New monitoring systems

One of the immediate objectives of the project was to develop new monitoring systems for the two pests Septoria and aphids. The measurements of the population development in 12 fields

in central Jutland were used to analyse the spatial variation in aphid densities, and the analysis of the results revealed that the aphid densities in late May and early June did not vary dramatically over a distance of up to 10 km. The practical consequence of this finding is that measuring the density thoroughly in one field seems to be a valid input density for simulations of the population development and damage during June and July for fields up to 10 km away. Therefore, farmers having their fields in a radius of 10 km around an aphid registration field do not need to count aphids in every winter wheat field, but can rely on counts from a central registration field. This not only accounts for assessments of aphid densities, it may be extended to involve forecasting aphid damage by simulation models such as the aphid module of SeptoriaSim. Axelsen et al. (2012) found that the request for valid input data concerning aphid densities was one of the obstacles for farmers' use of decision support systems to decide on whether to treat against aphids or not. Therefore, this result might be transformed into actions, making it attractive for farmers to use a decision support system on aphids.

Concerning the simulations of the population dynamics of Septoria in winter wheat and the damage it causes, simulations have shown a dependence on the daily input of spores. The dependence is, however, not crystal-clear, as the simulated value of treatments and number of treatments versus daily spore influx fluctuate considerably. The reason for these fluctuations in SeptoriaSim output is the complexity of the interactions in the model, where, for instance, delicate differences in the balance between biomass of roots, tillers, and leaves play a role for the uptake rate of nitrogen and the amount of photosynthates allocated to bud creation. Septoria damage affects the biomass of leaves and affects the balance between roots, leaves, tillers and reproductive organs. The impacts of disturbing this balance are often not linearly dependent on the disturbing factor and may cause the fluctuations. Consequently, evaluating the effects of increasing the daily input rate of Septoria spores must be done on trends, which clearly show that the spore input is important for the simulated Septoria damage to winter wheat. This means that monitoring the spores to be used as input to SeptoriaSim will improve the quality of the output from the system. It is seen in the spore trapping carried out in this project that during the growing season a relative constant flow of Septoria spores are released and available for infections. Depending on the locality, the trapping during important infection periods in May and June typically varied from 50-400 spores per day. The literature supports that these trapped spores are mainly ascospores and do not reflect the full picture of water spread pycnidia spores (Duvivier et al 2013), which generally are described as being the major source of inoculum during crop elongation. The level of Septoria spore influx varies considerably between Flakkebjerg, Holeby and Horsens, but whether spore catching, as measured in this project, matches the splash borne spores in the crop during rain and events with high humidity is not clarified. However, the trapped spores give very useful information, verifying that ascospores are very common, not only in the autumn as commonly described, but also during the growing season. The ascospores produced following reproduction play a major role in the fungi's ability to adapt to new cultivars with different resistance genes as well as adaptation to different fungicides.

If spore trapping are to be made a part of a future Septoria decision support system, it must be investigated whether the catching of ascospores links to the spread of pycnidia spores. In case of such a link, further investigations of the spatial variation could be relevant, as it was done for the aphids in this project. If the variation is limited over larger geographical ranges, it may be possible to carry out the costly spore trapping and quantification of spores at a regional scale and use the results as input for prognoses of Septoria damage and needs for control measures for larger geographical areas.

The activity in this project, following the development of disease as it moves up the crop, proved that DNA methods also have good ability to detect pre-symptomatic symptoms of Septoria attacks. This has previously been tested out using ELISA and other DNA methods (Fraaije et al. 1999). If these methods are to be incorporated as a part of a DSS system, the method

should be introduced as a field-based method, which makes it possible to detect early attack at field levels without needing to take samples into labs.

4.3 Spatial variation

One of the immediate objectives was to describe the spatial variation of aphids and develop stochastic spatial models that can be used to include uncertainties in warnings against aphids.

The site-specific initial occurrence probabilities were found to be positively correlated among the sites at the spatial scale of 10 km, but when the spatial variation in the initial occurrence probability was examined in more detail the following year, the spatial variation among plots in the beginning of the aphid epidemics did not seem to increase much with among-plot distance. Since the parameter that measures the effect of geographic distance on the spatial covariance, ρ_0 , also depends on the spatial variation in the following aphid epidemic, we tend to put more weight on the more detailed investigation in 2017 and conclude that our investigation suggests that there is limited spatial variation in the initial occurrence probability at the spatial scale of 10 km. Consequently, the overall results strengthen the working hypothesis that initial aphid population sizes and epidemics may be predicted in fields within a 10 km radius of the nearest aphid-monitoring site without imposing large uncertainties.

This result has large perspectives for the possibilities of designing an efficient monitoring system of aphid densities that can be used as the needed input for pesticide warnings or decision support systems. It would, for instance, be possible for farmers' associations to follow aphid infestations at a certain location and use the results from this location as input for decision support systems regarding aphid damage within a 10 km radius. The farmers' association could, then, issue warnings to their members within this area. We expect such an arrangement will critically reduce the needed time consumption for the individual farmer and that he, consequently, may be more inclined to use aphid controlling decision support systems.

The simple quadratic function used to model population growth as a function of degree-days performed adequately for modelling the growth in the aphid population from June 1, 2016, to July 1, 2016. However, the purpose of this simple quadratic model was only to model the deterministic part of the population growth in order to quantify the stochastic variation among sites, and the fitted quadratic model is not suitable for making actual predictions outside the domain of the collected aphid population data.

4.4 Stronger scientific foundation for warning models against the two pests

It was also an immediate objective of the project to provide a stronger scientific foundation for the warning systems in this project. The scientific investigations carried out in this project were mainly directed at improving the scientific foundation for SeptoriaSim, which is based on extensive biological knowledge on the involved organisms and the ecological interactions between them. Good simulations with SeptoriaSim also require a solid parameterization of parameters, such as growth and reproduction rates used to control the population growth of all organisms. This project has provided data on both the population growth of three species of aphids, the development of Septoria through QPCR analyses, and the growth of two varieties of winter wheat. These data have been used to parameterize SeptoriaSim, and the importance of this calibration was clearly demonstrated in the comparison between observed and simulated growth of winter wheat and Septoria before and after calibration to the collected data. Therefore, the reliability of the simulations carried out with SeptoriaSim has without doubt increased substantially with the calibration to the data collected in this project.

5. Conclusion

Using both decision support systems and the humidity model to time treatments against *Septoria* caused a lower number of fungicide treatments than actually seen in the national statistics (Figure 57), especially in 2016, but also in 2017. Further testing is needed to confirm and verify the potential of the systems in real use.

The capacity of *SeptoriaSim* to reduce the treatment frequency against aphids was not investigated thoroughly in this project. However, the treatment frequency in the controlled experiments and in the trials in farmers' fields suggested a strong reduction potential. The observed number of treatments was lower than both the Treatment Frequency Index for the particular areas (read from the map of Figure 56 to range between 0.2 and 0.4) and the average Treatment Frequency Index for the years 2012 to 2015 (0.31).

Consequently, considering the reduction potential in treatment frequencies against both *Septoria* and aphids in winter wheat without economic drawbacks, it seems very worthwhile to proceed to a more thorough testing in larger scale trials with farmers.

In this project, a large effort has been put into strengthening the scientific background for the decision support system *SeptoriaSim*, and the system has been shown to be capable of timing treatments against *Septoria* that generally gave net yields at the same level or higher than those of CPO without increasing the treatment frequencies. It may even reduce the treatment frequencies.

The potential for reducing the pesticide treatment frequency in winter wheat not only relies on *SeptoriaSim*/CPO/Humidity model, but also on the possibility of establishing regional assessments of input densities of aphids and *Septoria* spores. Such regional assessments may increase the willingness of farmers to use decision support systems to the benefit of both the farmers' economy and the environment.

6. Perspectives

6.1 Perspectives

This project has shown that a decision support system based on biological knowledge and projections of economic revenue of treatments may produce net yields of about the same size or better than the empirically based CPO. One important difference between the two decision support systems is that SeptoriaSim does not evaluate the need for control measures like CPO does. Instead, it gives predictions of the economic revenue of treating, and then it is up to the farmer to decide whether to treat or not. This decision may depend on the farmer's economic situation and his environmental attitude. This type of decision support system is an innovation, which has the capacity to avoid treatments that may be economically beneficial, but may only produce very low revenues that farmers do not regard worth the effort.

The model system of SeptoriaSim is a general model type that can be used for other crops and other pests. Thus, it will be relatively simple to add other winter wheat pests to the system, and a similar system can be established for oilseed rape and its insect pests without much effort.

6.2 Administrative perspectives

With one or more well-functioning decision support systems and the result that necessary input parameters can be applied for larger geographical areas, it is possible to qualify the decisions concerning pesticide treatments in winter wheat. The idea is to establish a system consisting of:

1. regional reporters who monitor the basic input of initial aphid densities and Septoria spore influx for SeptoriaSim
2. the reporters enter their results to a central database
3. a central service run SeptoriaSim weekly based on the input from the database and identify regional demands for control operations
4. the central service emits regional alerts to the farmers.

Such a system might help farmers make qualified decisions concerning Septoria and aphid control to the benefit of both farmers' economy and the environment

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Monitoring, warning, and decision support in winter wheat

The overall objective of this project was to develop a combined decision support system for *Septoria* and aphids in winter wheat by improving the simulation model *SeptoriaSim* and extending it with an aphid module. The work to fulfill this objective was split into four immediate objectives: 1) Develop new monitoring methods for the two pests, 2) Provide a stronger scientific foundation for warning models against *Septoria* and aphids, 3) Describe the spatial variation of aphids, and 3) Evaluate the reliability of the warning method/decision support tools in field trials.

Four types of investigations were carried out with the aim of both providing data for calibration of the model and enforcing the scientific background: 1) the background level of *Septoria* spores causing the initial infestation of winter wheat, 2) the growth of *Septoria* in the winter wheat leaves, 3) the population development of all three cereal aphids in winter wheat from late May to mid-July, 4) detailed growth analysis of winter wheat under field conditions.

This project has shown that a decision support system based on biological knowledge and projections of economic benefit of treatments may produce net yields equivalent to or better than the well-known empirically based CPO. One important difference between the two decision support systems is that *SeptoriaSim* gives predictions of the economic benefit of treating, and it is up to the farmer to decide whether to treat or not. This type of decision support system is an innovation that has the capacity to avoid treatments that may be economically beneficial, but not worth the effort for many farmers.

The project also showed that the use of decision support systems can be a tool to reduce the number of treatments against *Septoria*, and that *SeptoriaSim* can reduce the number of treatments against aphids.



The Danish Environmental
Protection Agency
Tolderundsvej 5
5000 Odense C

www.mst.dk