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Deliverable 8.5

# Model on the durability of single and/or stacked disease tolerant traits for the Potato – *P. infestans* pathosystem

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## Abstract

The purpose of AMIGA DL 8.5 was to investigate options for more durable potato cultivation based on deployment of host resistance genes against potato late blight. Within the framework of AMIGA, host resistance is conveyed to potato cultivars using genetic modification but the approach and results described in this deliverable are just as valid for resistant potato cultivars produced using classical breeding techniques.

A modelling approach resulted in a multi-season, spatio-temporal model of the potato - *P. infestans* interaction including mutations and parallel, interacting, epidemics of (the newly emerged) pathogen races. Pathogen population diversity changes during the growing season due to mutation and selection producing new pathogen races. The ratio between pathogen races is transferred from the end of the season to the next to allow for progressive evolution. The size of the pathogen population is however forced through a population bottle neck between seasons as is the case in NE Europe and many other temperate regions around the world.

The model was used to investigate scenarios on the effect of single and stacked R genes, the effect of fungicides, the interaction between (stacking) R genes and fungicides and the effects of stepping stone landscapes in which mixed forms of host resistance, single R genes and stacks of 2, 3 and 4 R genes, are present.

The results show that single R genes are relatively easily overcome by *P. infestans*. Protectant fungicides were shown capable of counteracting the evolutionary development of *P. infestans* populations by nearly neutralizing (mutant) sporangia. Stacking 2 or more R genes presented a significant simulated evolutionary barrier that was difficult for the pathogen to overcome. The larger the stack, the more difficult to overcome. Also here protectant fungicides were shown able to significantly slow down pathogen adaptation.

Stepping stone landscapes were shown to be a highway for *P. infestans* “en route” to overcoming, potentially durable, larger stacks of R genes. These landscapes should thus be avoided at all cost when durability of potato production is the ultimate goal.

All in all, the results from this theoretical study support the approach taken in the IPM2.0 control strategy for potato late blight as described and tested in AMIGA DL’s 8.3, 8.4 and 8.6. Host resistance is a most valuable tool when striving for durable potato production. R genes are the vulnerable key components of the IPM2.0 strategy. Monitoring of the pathogen population in combination with a low fungicide input strategy to repair the shortcomings of host resistance when really necessary form the protective barriers around the R genes. When deployed correctly, the EU could enjoy the benefits of *P. infestans* R genes for decades, perhaps even more.

## Introduction

Potato late blight, one of the world’s most devastating plant diseases in potato and tomato, is caused by the oomycete *Phytophthora infestans*. In the past, potato late blight led to e.g. the Great Irish - and Continental Famine (Zadoks, 2008). Currently potato late blight still is the most important disease in potato cultivation, traditionally controlled by highly frequent (calendar based) fungicide applications (Cooke et al., 2011) supported by preventative cultural measures such as crop rotation, the use of healthy seeds and the timely destruction of primary sources of inoculum. Nevertheless potato late blight remains responsible for an estimated annual economic loss of M€ 1000 on the 6 Mha of potato grown in the EU (Haverkort et al., 2008).

Host resistance and subsequent cultivation of potato late blight resistant potato cultivars is the most (cost) effective and environmentally friendly way to control potato late blight (Schepers et al., 2009). Currently however, cultivation of resistant cultivars is very limited due to an overwhelming demand for a limited number of commercially successful but highly late blight susceptible cultivars. In addition, potato breeding is complex and time consuming (Rietman et al., 2012) mostly due to the tetraploid nature of the crop. It was also repeatedly shown that resistance gene (R gene) mediated host resistance was easily overcome by the highly adaptive pathogen *P. infestans* (e.g. Black et al., 1953, Fry 2008; Haas et al. 2009; McDonald and Linde 2002), especially if resistance is based on a single R gene. The origin of the adaptive capability of *P. infestans* was shown to reside in the *P. infestans* genome in combination with its high reproductive capacity (Haas & Kamoun et al., 2009). As a result, adaptation is “the Phytophthora infestans way of life” resulting in R genes being overcome (e.g. Black et al., 1953, Fry 2008; Haas et al. 2009; McDonald and Linde, 2002), resistance to active ingredients of fungicides (e.g. Dowley and O'Sullivan, 1981) and increased aggressiveness (e.g. Flier and Turkensteen, 1999). The net results are the often dramatic and sudden population changes such as those described by Drenth et al. (1993), Cooke et al. (2012) and Fry et al. (2013).

Recently developed GM technology overcomes some of the disadvantages of conventional breeding with respect to breeding for resistance to potato late blight: Stacking R genes in popular commercial potato varieties in a fraction of the time required for conventional breeding while the cultivar characteristics remain the same was successfully  demonstrated within the DuRPh project (Haverkort et al., 2016).

It is however to be expected that (large scale) cultivation of (GM) resistant potato cultivars leads to significant selection pressure on the pathogen population for virulent genotypes. Within the DuRPh project, a tailor-made potato late blight control strategy, based on resistant cultivars, aimed to mitigate the effects of selection pressure using a low input fungicide spray program (Haverkort et al., 2016). This strategy was also deployed within the AMIGA project (AMIGA deliverables 8.3, 8.4 and 8.6).

Durable deployment of host resistance genes is thus key to the success of a “next level” IPM control strategy for potato late blight. Durability however is a topic that cannot be easily explored experimentally since it involves larger temporal and spatial scales. For the purposes of the AMIGA project, a spatio-temporal simulation model of the *P. infestans* – potato interaction at landscape level was constructed to explore the effect of a range of realistic scenarios for R-gene deployment on durability of resistance and the control strategy.

# Materials and Methods

## Definitions and Spatial scales

A “region” is defined as a user-defined 2-by-2-km, or equivalent, area with potato cultivation in one or more fields of 1 ha each. “Landscape” is used more broadly as any assembly of potato and non-potato fields. “Host diversity” refers to the genetic composition of host (potato) populations. Pathogen population diversity refers to the genetic composition of the pathogen population. The 1 ha fields (patches) are the smallest spatial units simulated in the region. Host growth and late blight epidemics are assumed to occur homogeneously within fields but not between fields.

Hosts can be susceptible or resistant. Hosts may contain (combinations of) 0 – 4 different resistance genes. Intermediate forms of resistance (partial resistance, resulting in slower epidemics)  are not implemented in the current study. Pathogen races may contain 0 – 4 avirulence factors rendering them  pathogenic on hosts containing matching R genes. Thus, 16 different hosts and 16 different pathogen races can be created. Parallel epidemics are simulated for the various pathogen races on compatible hosts. Pathogen races are competing for leaf area on hosts patches compatible for them.

## Simulation platform

The BlightWorld simulator was developed using NetLogo. NetLogo is a free, open source, programmable modelling environment for simulating natural and social phenomena (Wilensky, 1999). BlightWorld is a SIR (Susceptible, Infectious, Removed) model representing key processes of the potato late blight epidemiology and population genetics at landscape level. BlightWorld is using a time step of 1 day.

## Weather

For the purpose of this study, the days are classified as suitable or unsuitable for epidemic development. Epidemics are allowed to develop during days suitable for epidemic development (blight days) whereas the epidemic is halted on unsuitable days (Skelsey et al., 2010). Analysis of relevant measured weather data from Eelde, the Netherlands, 1 April – 30 September, 1981 – 2010, revealed that blight days occurred from once every 10 days to once every three days during unconducive and conducive growing seasons respectively (not shown). In BlightWorld, Blight days can be assigned according to a list of measured weather data or as a user defined ratio between blight days and non-blight days.

## Host

Host resistance is governed by resistance genes. Each resistance gene recognizes a specific protein (effector) secreted by the pathogen. Recognition of 1 of these proteins is sufficient to initiate a hypersensitive response, killing the infecting pathogen and rendering the host resistant. Multiple host R genes thus recognize multiple effector proteins representing multiple triggers for the same hypersensitive response. In BlightWorld, the number of different R genes can be freely chosen but, for the present purposes, was set at a maximum of 4. A susceptible host does not contain any R genes (R0), resistant hosts may carry (combinations of) 1 – 4 R genes (R01, R02, ..R04, R012, R014, ..., …R01234). This results in 16 possible combinations of 0 – 4 R genes and thus potentially 16, user defined, different hosts.

**Pathogen**

*P. infestans* secretes a suite of hundreds of proteins into the host during the infection process. These proteins are called effectors since they have to perform infection related duties for the pathogen. R genes recognize specific proteins with initiation of the hypersensitive response as the result. Mutations in the gene responsible for production of an effector protein results in an altered effector protein which may evade recognition by the R gene. In that case, a new pathogen race is born, the hypersensitive response is not triggered anymore and the R gene involved is overcome. As a result, the host is susceptible to this new pathogen race. In BlightWorld, the basic/default pathogen race does not overcome any of the host R genes and can thus only infect host R0. Presence of altered effector genes/proteins (AVR factors) is represented by numbers corresponding to the R genes. AVR01 is necessary to overcome R01, AVR0123 is necessary to overcome R0123 etc etc.

The late blight epidemic is initiated from a user defined initial number of lesions equally divided over compatible host genotypes. Default setting: 100 lesions, 1 $cm^2$ each. Presence and quantity of pathogen races is user-defined. Late blight lesions are initiated as necrotic, no longer infectious (NLINF), tissue. NLINF tissue grows producing infectious tissue (INF) following a logistic growth curve at patch level. The epidemic proceeds on “blight days” and is halted (paused) on “non-blight days” (Skelsey et al., 2010). Only one single batch of sporangia is produced on each surface area unit of INF tissue. After 1 batch of sporangia has been produced, the INF tissue area becomes necrotic (NL-INF). Sporulation Density is user defined, default value $1x10^8$ sporangia/$m^2$. Sporangia are the dispersed over the world using the NetLogo “diffuse” directive which suffices for the aims of the present study. A standard escape fraction for sporangia from the canopy of 0.3 is assumed (Aylor et al., 2001). Survival of sporangia during atmospheric transport is modelled assuming a standard survival fraction of 0.1 (derived from Fry and Mizubuti 1998; Minogue and Fry 1981; Mizubuti et al. 2000). Dry deposition is modelled as described by Skelsey et al (2008).

Following deposition, sporangia are given the opportunity to infect using a user defined Infection Efficiency (IE, default value: 0.05 (e.g. Flier, W.G. and Turkensteen, L.J. 1999). Successful infections are given a surface area of 1 $cm^2$ each and enter a latent period with a user defined default length of 5 days. Following completion of the latent period, new lesions emerge as 1 $cm^2$ of NL-INF tissue each which is added to the pool of NL-INF tissue already present. This completes the life cycle.

**Mutations**

Adaptation is “the Phytophthora infestans way of life”. Adaptation is based on changes in the genome, by a multitude of mechanisms, followed by random effects (e.g. founder effects) and selection pressure. Direct measurements of the mutation-frequency for effector genes (avirulent → virulent), although commonly observed at phenotypic level, are however not available. Some of the factors playing a role in the generation and establishment of new virulent pathogen races include: the size of the *P. infestans* population, the mutation frequency (avirulent → virulent), fitness of the new genotypes, possibilities for establishment (acreage of the previously resistant potato cultivar) and competition with non-virulent pathogen races.

Since current knowledge on mutations occurring in *P. infestans* is limited, we looked at other micro-organisms where similar processes take place (Kondrashov and Kondrashov, 2010). Spontaneous mutations are expressed as the frequency a random genomic base pare is replaced with another base pare (single nucleotide polymorphism) during one (1) reproductive cycle.

The mutation frequency is however influenced by many factors e.g.:

- Amplification of the mutation frequency.
- The mutation frequency may differ between species and even between different locations on the genome.
- The mutation frequency is co-regulated by other genes which may vary between species, between individuals and even within individuals.
- A variety of repair mechanisms may be active. Activity may depend on type of mutation and location on the genome.

Kondrashov and Kondrashov (2010) list a number of methods to estimate mutation frequencies. Based on research with *Drosophilae melanogaster*, *Sacheromyces servisiae*, *E. coli* and *M. tuberculosis*, they estimate the natural mutation frequency per nucleotide and including repair mechanisms in the $1x10^{-7}$ to $1x10^{-9}$ range with a most likely frequency around $1x10^{-8}$.

In the model, only single step mutations were included. It was calculated that for a mutation frequency of 1x10-8 a  double mutation (leading to virulence to 2 R genes in 1 step) is highly unlikely to occur and survive on the spatial scale of the Netherlands (see paragraph below). To overcome combinations of R genes in the BlightWorld model, *P infestans* thus has to rely on stepwise mutations, 1 R gene at a time.

If every potato crop in the Netherlands (150000 ha) is completely destroyed by potato late blight race 0, this would only generate 1.5E5 x 4E4 x 1E8 x 1E-16 = 60 mutants compatible with to 2 R genes. The chance that one or more of these mutant spores is able to survive and infect is minute. In addition and fortunately, only a tiny fraction of the Dutch potato acreage is annually destroyed by late blight.

BlightWorld therefore includes the possibility to adjust the mutation frequency avirulent → virulent within the range $1x10^{-6}$ to $1x10^{-12}$. In the model $1x10^{-6}$ is used as default value for the current purposes.

Scenarios

The useful life-span of host resistance is influenced by a complex and interacting set of processes and variables. The BlightWorld simulator was used to explore the effect of a range of realistic scenarios for R-gene deployment on durability of resistance and the control strategy.

A landscape with 100% susceptible potatoes was used as a reference scenario. Host resistance was introduced in several forms: 50% of the landscape was assigned to a potato cultivar containing 1 R gene, 2 R genes, 3 R genes or 4 R genes. This was done with or without additional protectant or curative fungicide applications. Also, a landscape containing 20% susceptible potato (R0), 20% R1, 20% R12, 20% R123 and 20% R1234 was simulated. In contrast to the previous scenarios with just 1 type of host resistance, here, the various forms of resistance present in the same landscape may serve as stepping stones for the pathogen to overcome increasingly complex forms of host resistance step by step.

Simulations were run for 10 consecutive years except for the “stepping stone scenario” which was run for 30 years. The pathogen population always started off as 100% race 0 (only compatible with susceptible potato). The response variable of choice was the pathogen population diversity: the fractions of the various virulent pathogen genotypes. The pathogen population diversity was kept between consecutive years within a scenario to allow for progressive evolution. Default parameter settings were chosen to simulate worst case scenarios and prompt responses from the system: Mutation frequency: $1x10^{-6}$, Weather: always suitable for epidemic development, Fraction potato in the landscape: 20%, Start of the growing season: Julian date 120, Start of the epidemic: Julian date 150 (just before the crop reaches maximum ground coverage), end of the season: Julian date 240.

In between seasons, the pathogen population experiences a bottle neck. Next year’s epidemic was therefore initiated by distributing 100 $1cm^2$ lesions (NL-INF) mirroring the final composition of the pathogen population in the previous season. To allow for random effects, founder effects etc., the product of the *P. infestans* genotype fraction and the 100 initial lesions was used to draw the final number of initial lesions for each pathogen genotype (race) from a poisson distribution.

**Fungicides**

Fungicides may, or may not, be applied (users choice) as part of the scenarios. Protectant fungicides are distinguished from curative fungicides. Protectant fungicides reduce the infection efficiency of sporangia by a user defined percentage (default: 99%). Curative fungicides reduce the infection efficiency of sporangia by a user defined percentage (default: 99%) **AND** the expansion of necrotic tissue ( = production of infectious tissue (INF)) by a user defined percentage (default: 50%). The default value was chosen based on experts opinions saying that the efficacy of a curative application varies from 99% - 0% based on the timing of the application relative to the time of infection. 99% effect is reached when applied immediately following infection, 0% effect is reached when applied 24 hours or more after infection. The default value of 50% was chosen as the average of both extremes.

## Landscape generation

In BlightWorld, the user defines the settings for landscape: The size of the world, the fraction potato in “the world”, the average field size (in ha), host diversity (presence and quantity of the different hosts (R genes) and the start date and end date of the growing season (Julian dates). The model randomly generates a landscape containing clustered potato patches (fields)  representing these settings. As the landscape is, to some extent, random, position of fields vary from year to year.

# Results

## BlightWorld general look and performance

A typical BlightWorld “world” is given in Figure 1. A new world is randomly generated, based on the landscape and crop parameters set by the user, at the start of each “growing season”.

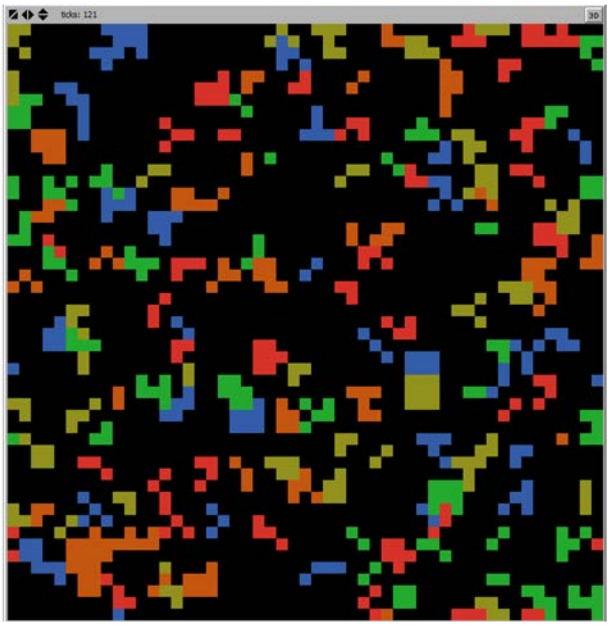


**Figure 1.** Result from "BlightWorld": a 5 x 5 km world containing 25% potato equally distributed over potato cultivars R0 (red), R1 (orange), R12 (green), R123 (bright green) and R1234 (blue).

The result of a P. infestans race 0 epidemic in a landscape containing 20% susceptible potato (R0) is given in **Error! Reference source not found.**.

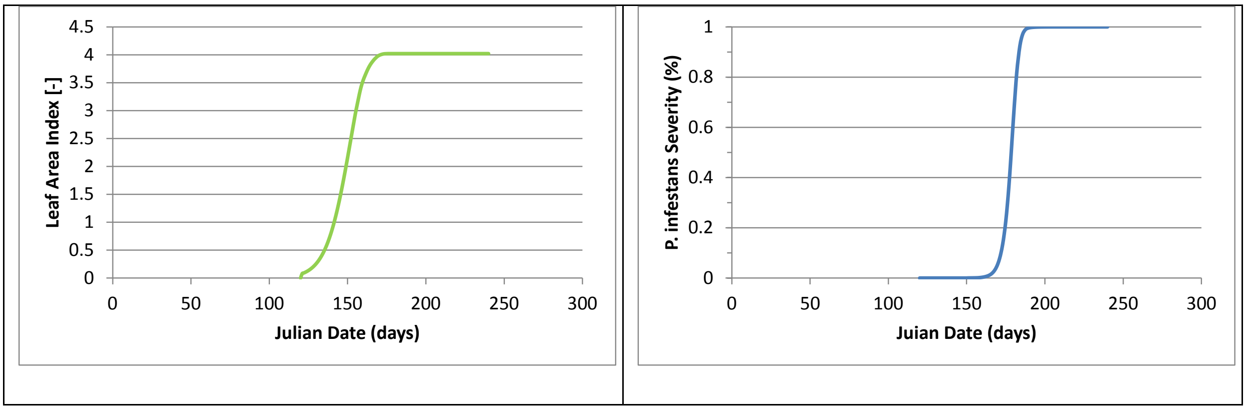


**Figure 2.** Growth curve of a potato crop in BlightWorld (left) and the result of a *P. infestans* race-0 epidemic (right, average severity per potato patch).

## Scenario's

### Stacking of resistance genes

The following results are based on scenarios where resistance genes may or may not be used in the landscape. In addition, R genes may or may not be stacked (Haverkort et al., 2008, 2016). Simulation runs were carried out for landscapes containing potatoes that were: 100% susceptible R0 (reference scenario), 50% R0 + 50% R1, 50% R0 + 50% R12, 50% R0 + 50% R123 or 50% R0 + 50% R1234. The pathogen population always started out as 100% race 0 (compatible to R0 only). Scenarios were run for 10 consecutive years. The pathogen population diversity was kept between years. Fungicides were not used. The results are given in Figure 3.

The results show that, in absence of fungicides and under these simulated weather conditions, the 100% susceptible landscape is always completely destroyed. This is equivalent with practical experience with susceptible, unsprayed crops under high disease pressure in the Netherlands and elsewhere. When 1 R gene is introduced, 58% of all potatoes in the landscape are destroyed in year 1: 50% R0 plus 8% R1. A mutation in pathogen race 0 renders part of the pathogen population compatible with the newly introduced resistant potato cultivar. This new race 01 claims about 13% of the population after year 1. In following years it quickly becomes dominant due to its ability to infect both potato cultivars grown. Race 01 stabilizes at approximately 85% of the population since it does not have any other competitive advantage over race 0 apart from its extended host range. From year 2 onwards again all potatoes in the landscape are destroyed making the newly introduced resistant cultivar useless.

When 2 or more r genes, stacked in the same potato cultivar, are introduced in the absence of fungicides, apparently, the evolutionary hurdle is too big to overcome, even for *P. infestans*. Year after year, only the susceptible potatoes in the landscape are destroyed whereas the resistant potatoes escape infection. Apparently, the numbers of double mutants built up on potato R0 every season are too low to survive the bottle neck the pathogen population experiences every winter. In absence of a winter event, undoubtedly, multiple mutations would arise in sequence and settle in the population.

Although the general trends will remain the same, this result is likely to be sensitive for changes in the mutation frequency, size of the “world” (= total production of sporangia) and the dispersal capability of *P. infestans* sporangia.

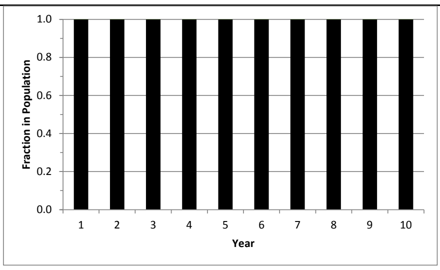
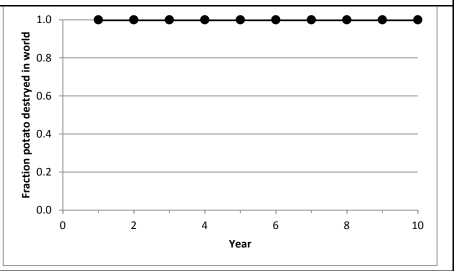
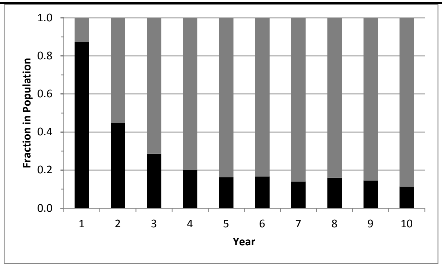
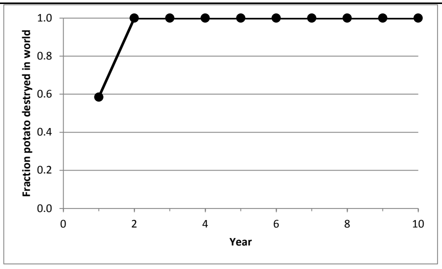
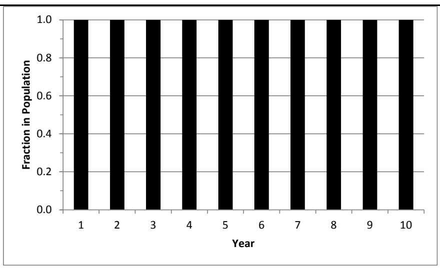
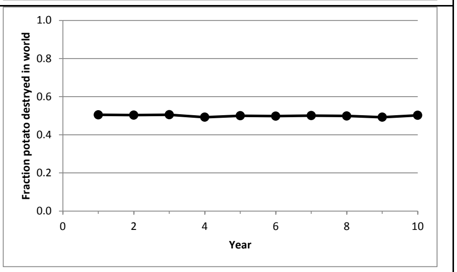
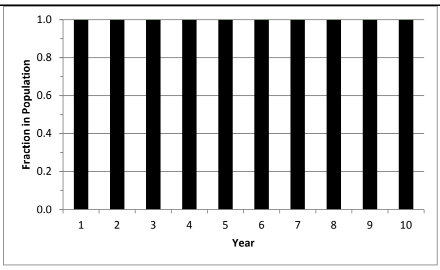
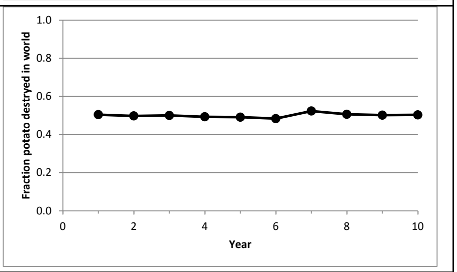
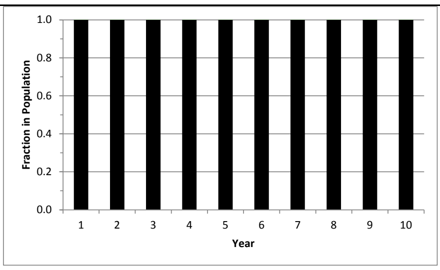
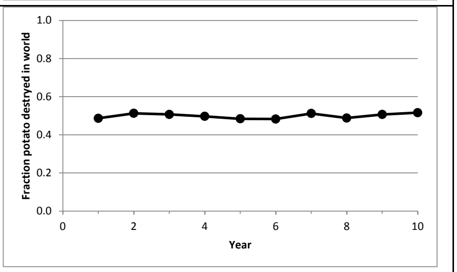
| Host diversity | Pathogen population diversity | Potato destroyed in landscape |
|---|---|---|
| 100% R0 |  |  |
| 50% R0 +<br>50% R1 |  |  |
| 50% R0 +<br>50% R12 |  |  |
| 50% R0 +<br>50% R123 |  |  |
| 50% R0 +<br>50% R1234 |  |  |

**Figure 3.** The effect of stacking of R genes on pathogen population diversity and the fraction potato destroyed in the landscape. Pathogen races: race 0: ■, race 01: ▨.

## Fungicides

### *Protectant fungicides*

Protectant fungicides typically are not taken up by the plant  and are thus only active on the outside of a potato plant. Sporangia landing on the crop come into direct contact with the active ingredient and  are highly likely to die. Within BlightWorld, a 99% efficacy was assumed against sporangia.

The results (Figure 4) show that 100% of the potatoes are destroyed every year in the 100% susceptible landscape. This is due to the way primary infections are introduced: 100 lesions are distributed over the landscape. Protectant fungicides will be active against the sporangia produced by the lesions but they will not affect expansion of necrotic tissue since this is driven by growing mycelium inside the foliar potato tissue. Although this is a little bit at odds with reality, the foliage is modelled as 1 continuous  sheet, practical experience confirms it is close to impossible to control established late blight infections with protectant fungicides only.

When 1 or more resistance genes are introduced, protectant fungicides are sufficiently effective against mutant spores to avoid build-up of new R gene compatible pathogen races. This is an indication R genes can be protected from being overcome by applying only protectant fungicides.

The AMIGA potato field trials in Ireland and the Netherlands successfully used this phenomenon in the IPM2.0 control strategy (AMIGA deliverables 8.3 and 8.6). Combined with the results from stacking R genes this indicates that single R gene containing resistant cultivars can be protected from being overcome using protectant fungicides. Multiple R gene containing resistant cultivars do not have to be sprayed at all.

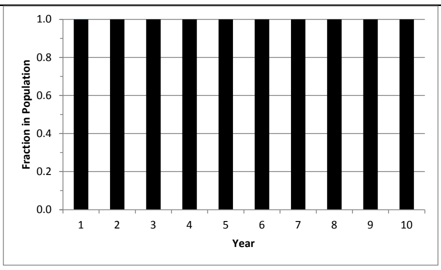
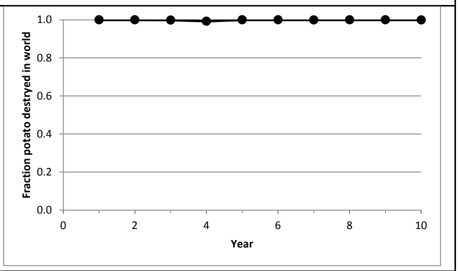
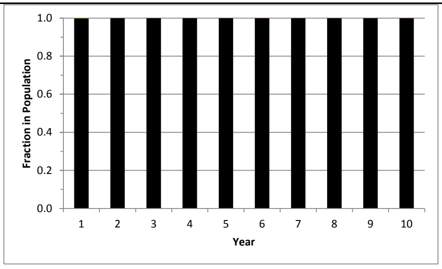
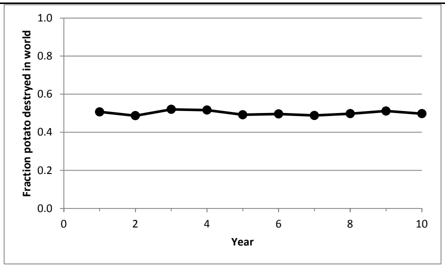
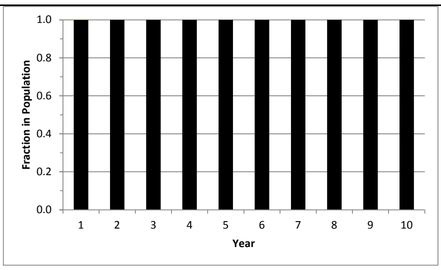
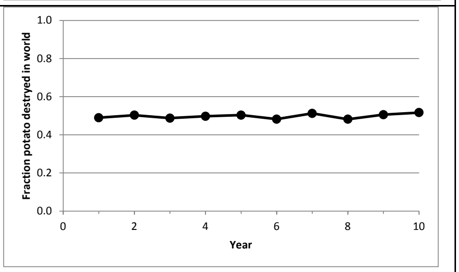
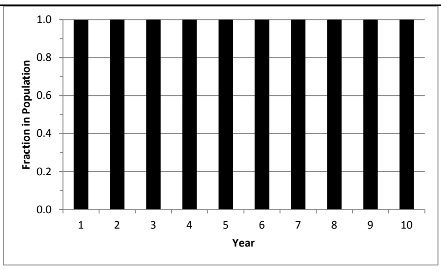
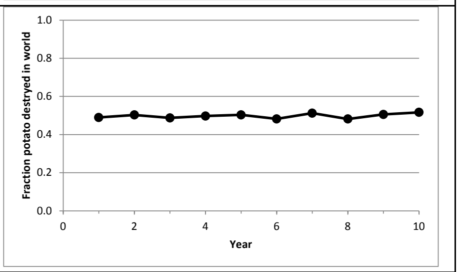
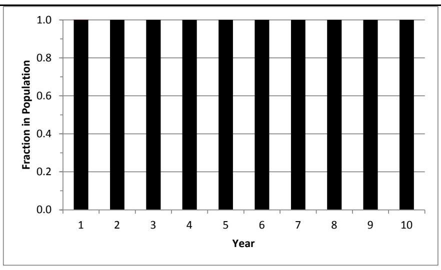
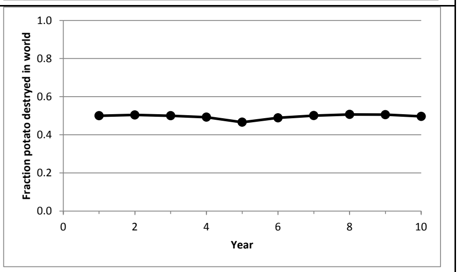
| Host diversity | Pathogen population diversity | Potato destroyed in landscape |
|---|---|---|
| 100% R0 |  |  |
| 50% R0 +<br>50% R1 |  |  |
| 50% R0 +<br>50% R12 |  |  |
| 50% R0 +<br>50% R123 |  |  |
| 50% R0 +<br>50% R1234 |  |  |

**Figure 4.** The effect of **protectant fungicides** on pathogen population diversity and the fraction potato destroyed in the landscape. Protectant fungicides reduce the number of infections from spores by 99%. Pathogen races: race 0: ■, race 01: ■.

### *Curative fungicides*

Curative fungicides combine a protectant capability with a curative effect, reducing expansion of necrotic tissue (production of infectious tissue). Similar to protectant fungicides, a 99% efficacy against sporangia was assumed combined with a 50% efficacy against mycelial colonisation of foliar tissue.

The results (Figure 5) show that the level of infection in the 100% susceptible landscape is indeed reduced and stabilizes around 50% potatoes destroyed. When 1 or more resistance genes are introduced, new R gene-compatible pathogen races do not emerge as described for protectant fungicides above. In addition, the level of infection of the susceptible half of the landscape is slightly reduced and stabilises around 35% of the landscape. For the susceptible potato by itself this means around 30% is not infected.

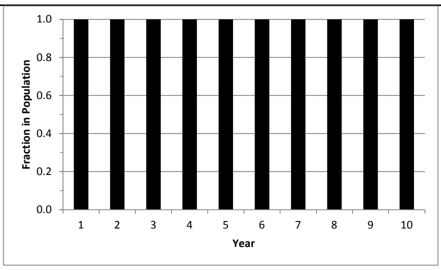
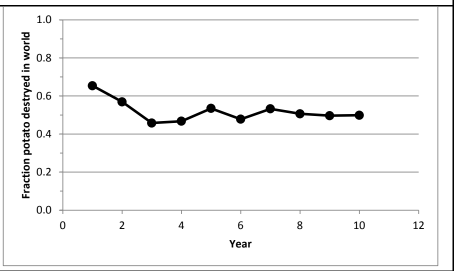
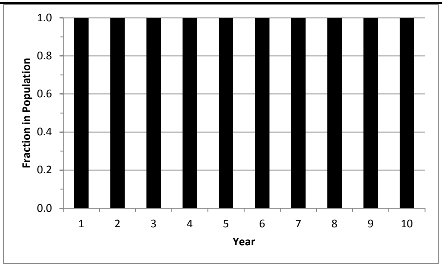
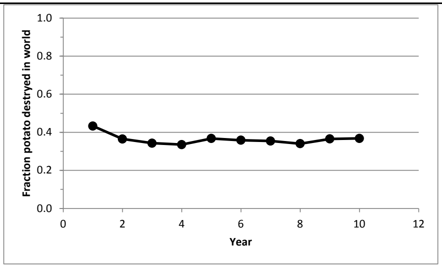
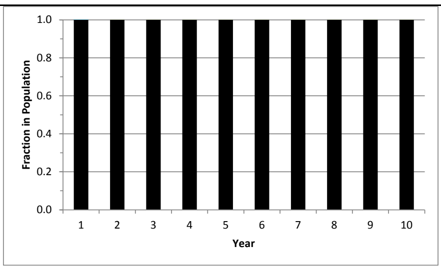
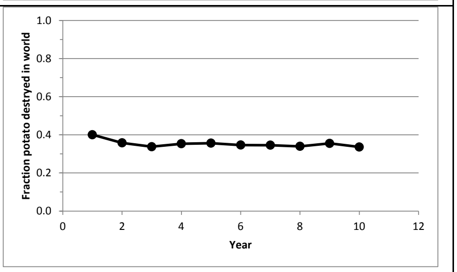
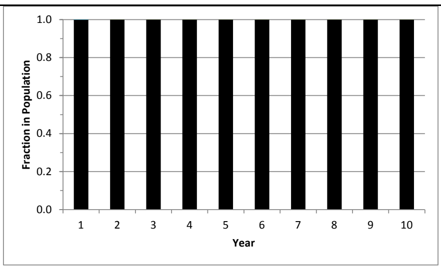
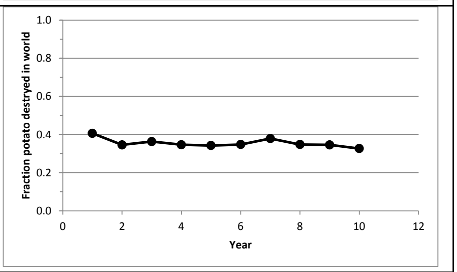
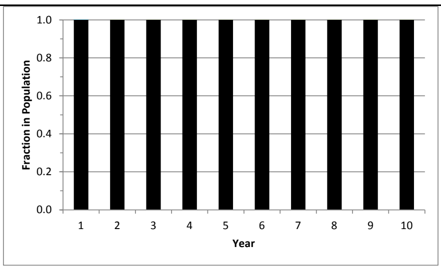
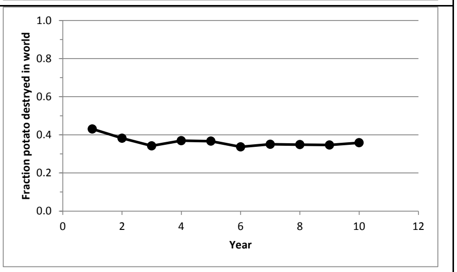
| Host diversity | Pathogen population diversity | Potato destroyed in landscape |
|---|---|---|
| 100% R0 |  |  |
| 50% R0 +<br>50% R1 |  |  |
| 50% R0 +<br>50% R12 |  |  |
| 50% R0 +<br>50% R123 |  |  |
| 50% R0 +<br>50% R1234 |  |  |

**Figure 5.** The effect of **curative fungicides** on pathogen population diversity and the fraction potato destroyed in the landscape. Curative fungicides reduce the number of infection from spores by 99% and the expansion of necrotic tissue (producing infectious tissue) by 50%. Pathogen races: race 0: ■, race 01: ▨.

## R genes as stepping stones

In the real world, potato breeders are striving for potato late blight resistant potatoes based on stacked R genes. The intermediate products, containing single R genes or stacks with R genes already overcome, are however also introduced. These various forms of simpler host resistance, present in the same landscape, may serve as stepping stones for the pathogen to overcome increasingly complex forms of host resistance step by step. This was investigated in a 30 year scenario, with and without protectant fungicides, where the landscape contained 20% of R0, R1, R12, R123 and R1234 respectively. The pathogen population started out as 100% race 0.

The results are shown in Figure 6. From the results it is clear that the single R gene is overcome first, followed, one R gene at a time, by more and more complex forms of host resistance. This therefore seems to be a quick and effective way to overcome potentially durable forms of host resistance. "Stepping stone landscapes" should thus be avoided at almost all cost.

Timely application of protectant fungicides, as shown above, results in neutralisation of the *P. infestans* evolutionary capacity. New R gene compatible races, although they do emerge, do not become a significant part of the population due to the combined effect of protectant fungicides and the population bottle neck experienced in winter.

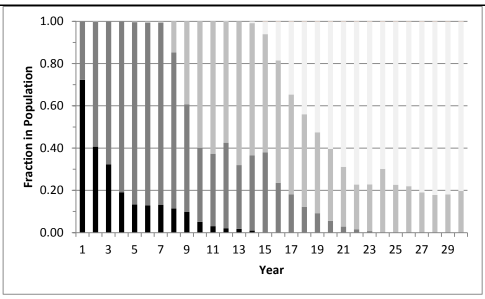
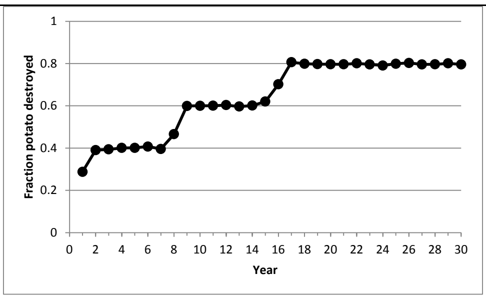
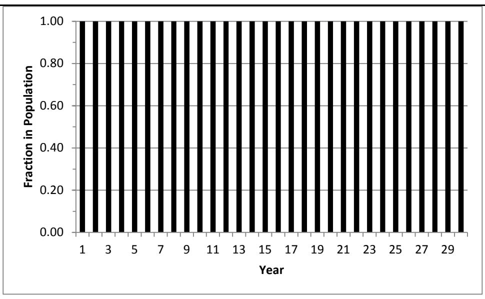
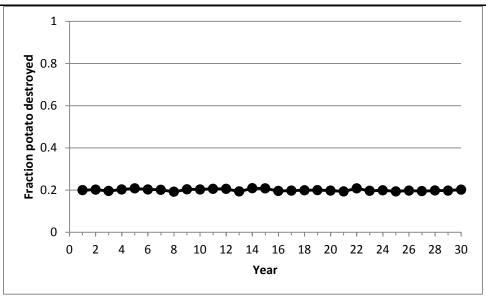
| Host diversity | Pathogen population diversity | Potato destroyed in landscape |
|---|---|---|
| 20% R0, +<br>20% R1, +<br>20% R12, +<br>20% R123, +<br>20% R1234 |  |  |
| 20% R0, +<br>20% R1, +<br>20% R12, +<br>20% R123, +<br>20% R1234<br>+<br>Protectant fungicides |  |  |

**Figure 6.** The effect of providing R genes as stepping stones in the landscape with or without application of additional protectant fungicides. Pathogen races: race 0: ■, race 01: ■.

## Discussion

The purpose of AMIGA DL 8.5 was to investigate options for more durable potato cultivation based on deployment of host resistance genes against potato late blight. Fungicide applications against potato late blight are regarded the main component of the environmental side effects of potato production. Enhanced durability can be achieved through an IPM2.0 control strategy for potato late blight based on host resistance and supported by pathogen population monitoring and low input fungicide applications when strictly necessary. This strategy allows for a reduction of the fungicide input of 80% or more (see also AMIGA DL's 8.3 and 8.6). The more durable the forms of host resistance deployed, the more durable the potato cultivation.

A modelling approach using NetLogo resulted in a regional, multi-season, spatio-temporal model of the potato - *P. infestans* interaction including mutations and thus parallel, interacting, epidemics of (the newly emerged) pathogen races. Pathogen population diversity thus develops during the growing season as influenced by the potato cultivars (R genes) grown and the fungicides applied. Mutation and selection thus dictates pathogen population diversity as influenced by the landscape and management practises. Progressive evolution, including multiple growing seasons is made possible by transferring pathogen population composition from one season to the next. The size of the pathogen population is however forced through a “winter population bottle-neck” as is the case in Northern Europe and many other temperate regions around the world.

The model was used to investigate scenarios on the effect of single and stacked R genes, the effect of fungicides, the interaction between (stacking) R genes and fungicides and the effects of stepping stone landscapes in which mixed forms of host resistance, single R genes and stacks of 2, 3 and 4 R genes, are present.

R gene deployment options using single – or stacked resistance genes in absence or presence of protectant or curative fungicides were investigated using a modelling approach. In addition, the effect of landscapes containing both, simple and complex forms of host resistance was investigated. In this scenario, the simpler forms of host resistance may serve as stepping stones for *P. infestans* to overcome more complex forms of host resistance. For this purpose, a spatio-temporal simulation model of the *P. infestans* – potato interaction at landscape level (BlightWorld) was constructed.

The results show that, under late blight conducive weahter conditions, susceptible potato landscapes in absence of fungicides are always completely destroyed. Single R gene resistance yields some relief although only for a short time, 1 year, due to quick adaptation of the pathogen population. The new R gene compatible pathogen race quickly becomes dominant in the pathogen population rendering the recently introduced resistant cultivars useless. This result is supported by a wealth of real world observations, e.g. Black et al., 1953.

Introduction of potato cultivars containing 2 or more stacked R genes presents a significant evolutionary hurdle, even for *P. infestans*. The combined effect of R genes and the population bottle neck experienced in winter (or a host free period) is sufficient to prevent the stack of R genes being overcome. In absence of population bottle neck due to winter or a host free period, the pathogen population is more likely to overcome the simpler forms of stacked R genes. When sufficient numbers of progeny are allowed to be produced, more rare, double mutations may also occur. In this respect, *P. infestans* population genetics is very much a numbers game.

Two types of fungicides were distinguished and introduced: protectant fungicides which knock out spores and curative fungicides knock out spores and reduce the rate of colonization by mycelium. Due to the way primary infections were introduced, 100 lesions are distributed over the landscape, protectant fungicides did not result in acceptable control levels for susceptible crops. In the real world, protectant fungicides, when applied at the correct moments in time, are very capable of preventing infections by killing incoming spores. Protectant fungicides are however not very active against already established infections (see e.g the Euroblight fungicide table) which is confirmed by the current simulation results.

When 1 or more resistance genes are introduced, protectant fungicides are sufficiently effective against mutant spores to avoid build-up of new R gene compatible pathogen races. This is a strong indication R genes can be protected from being overcome by applying only protectant fungicides. In addition, protectant fungicides generally are less damaging to the environment than curative fungicides.

The AMIGA potato field trials in Ireland and the Netherlands successfully used this phenomenon in the IPM2.0 control strategy (AMIGA deliverables 8.3 and 8.6). Single R gene containing resistant cultivars were successfully protected from being overcome using protectant fungicides. Along the same line of thought, multiple R gene containing resistant cultivars do not have to be sprayed at all as long as two or more R genes are fully functional/not overcome.

From the results on the “stepping stone scenarios” it can be concluded that *P. infestans* is capable of efficiently overcoming stacks of R genes when the single R gene components and smaller stacks are provided in the landscape. When striving for durable potato production, “stepping stone landscapes” should thus be avoided at all cost.

All in all, from the results described in AMIGA DL’s 8.3, 8.4,.8.5 and 8.6 it can be concluded that much more durable forms of potato late blight control are realistic. The necessary components are currently available although they can be improved upon with respect to availability of stacked R genes, e.g. using cisgenic modification (Jacobsen and Schouten, 2007). The risk of R genes being overcome can be mitigated by stacking R genes, monitoring of pathogen population development with respect to virulence to R genes deployed and, when strictly necessary, a low input fungicide spray strategy or replacement of the R gene stacks used by new efficient ones. Host resistance thus is a most valuable tool when truly striving for durable potato production. When deployed correctly however, the current results indicate that the EU could enjoy the benefits of *P. infestans* R genes for decades, perhaps even more.

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# Appendix 1: Netlogo code of the BlightWorld simulator

When copied to NetLogo, the text below will align.

```
;*******************************************************************************************************************************
; BlightWorld.nlogo                                                              ;
; A NetLogo SIR model for Phytophthora infestans epidemics in a 2D "potato region" sized ? x ? km            ;
; in a grid of 100 x 100 grid cells. Grid cells are 100 x 100m (1ha)                                    ;
; Simulations run over multiple - independent - years with a time step of 1 day.                               ;
; Regions are made up of potato fields (host) and non-potato fields (non host).                                 ;
; Potato fields may contain various potato cultivars, 1 cultivar per field                                   ;
; differing in R-gene content. Susceptible cultivars do not contain R-genes.                                   ;
; Resistant cultivars contain 1 - 4 different R-genes                                                 ;
; P. infestans starts out as avirulent on all R-genes (population 100% Race-0).                              ;
; Mutations occur during epidemic development generating functional mutants virulent to 1 or more R-genes.          ;
;                                                                       ;
; The setup of the region with respect to cultivars is user defined. The P. infestans population starts out as 100% Race-0   ;
; but virulent mutants are kept in the population between seasons when sufficient in number.                    ;
;                                                                       ;
; Geert Kessel, Francine Pacilly and Mark Kramer, 2014 & 2015                                       ;
; Geert.Kessel@WUR.NL  Francine.Pacilly@WUR.NL  Mark.Kramer@WUR.NL                                     ;
;                                                                       ;
; +31 (0)317 480814                                                              ;
; Copyrights: Geert Kessel,  Plant Research International, Wageningen UR                              ;
; P.O.Box 16, 6700AA, Wageningen, the Netherlands                                               ;
;*******************************************************************************************************************************
; V20160622

; ---------------------------------------------- Initialisation ---------------------------------------------------------------
; ----------------------------------------------------------------------------------------------------------------------------
breed [pathogens pathogen]                    ; Turtle pathogens/pathogen
breed [fields    field   ]                    ; Turtle fields/field

pathogens-own
[
  p-race                                      ; P. infestans genotype (race)
  phase                                       ; Phase of the life cycle (0 = spore, 1 = L1, 2 = L2, ..., 5 = L5, 6 = Inf = Infectious, 7 = NLInf = No Longer Infectious)
  quantity                                    ; Quantity in which each of the stages above is present on this patch. # for spores, % leaf area for the others.
]

fields-own
[
  t-fieldnr                                   ; Unique number identifying a field (turtle)
  t-fieldsize                                 ; Size of the field (ha or patches)
  t-h-type                                    ; Potato cultivar grown on this field (t-h-type = R-gene content)
]

patches-own
[
  field                                       ; Holds a patch number which is that fields "leader" If a patch has not been assigned to a field, this holds "nobody"
  potato                                      ; potato on this patch? (yes (1)  or no (0))
  p-fieldnr                                   ; field number assigned to this patch (= turtle ID sitting on patch)
```

```
  p-fieldsize
  p-h-type
  emergence
  tub-init
  maxlai-day
  end-tub
  tuber-gr
  init-lai
  IE
  lai
  max-lai
  yield

  tmp-conc ; for diffuse                          ; Temporary concentration used in calc. of difusion
  old-conc ; for wind                            ; Old concentration used in clac of wind dispersal
  new-conc ; for wind                             ; New concentration used in calc of wind dispersal
  mobile-spores
  immobile-spores
  quantities                                     ; List per patch and p-types containing quantities per phase
  newvalues                                       ; List per patch and p-types containing newvalues per time step
  severity                                      ; Severity per patch, sum of quantities for all p-types present

  q                                           ; Helper variable used in procedure advance epidemic
  r                                           ; Helper variable used in procedure advance epidemic
  s                                           ; Helper variable used in procedure advance epidemic
 ]

 globals                                        ; Global variables
 [
  base-types                                     ; "base-types" list which contains the base spore types (single virulences to be combined in the "spore types")
  types                                        ; " types" list containing basic names of host or pathogen types, used to generate h-types (host types) and p-types (pathogen types)
  h-types                                       ; "h-types" host types; list containg the " names" of the potato cultivar as defined by their R-gene content
  p-types                                       ; "p-types" pathogen types; text vector containg the "names" of the pathogen race as defined by their AVR-gene content
  nrfields                                     ; The desired number of potato fields
  nrpotatopatches                                  ; Nr of potato pathces i the world.
  cv-count                                      ; list containing the patch counts for each potato cultivar
  primaryfoci                                    ; Number of potato patches with a primary P. infestans infection

  ; Counters
  season-counter                                   ; Counter for the number of growing seasons (years)

  ; lists
  cv-fractions-list                               ; list containing the fractions of all potato cultivars in the landscape
  host-fieldnr-list                                ; list containing the nr of fields for each cultivar
  cv-colors-list                                ; list containing the patch colors of all potato cultivars in the landscape
  sum-cv-fractions-list
  avr-fractions-list                              ; list containing the fractions for all relevant P. infestans avr's combinations present in the population
  avr-fieldnr-list                               ; list containing the number of patches infected by the different P. infestans races at the start of the season (primary infections)
  transitions                                   ; list of lists: 1st index, index of original in sp-types elements. 2nd index: list of types to which a (direct) transition is possible

  emergence-list                                   ; List containing the Julian emergence dates for the various cultivars
  tub-init-list                                 ; List containing the tuber initiation day for each cultivar
  maxlai-day-list                                 ; List containing the day at which max LAI is reached for each cultivar
```

```
 end-tub-list                                  ; List containing the end day for tuber formation for each cultivar
 tuber-gr-list                                 ; List containing the infection efficiency for each cultivar
 init-lai-list                                ; List containing the tuber growth rate for each cultivar
 max-lai-list                                  ; List containing the initial LAI for each cultivar
 IE-list                                      ; List containing the infection efficiency for each cultivar
 rgr-list                                     ; List containing the epidemic relative growth rate for each cultivar
 SD-list                                      ; List containing the Sporulation Density for each cultivar
 prim-lesion-size                               ; Size of a lesion after infection by a spore and the latent period has passed [m2]
 cvcount                                       ; List containing the measured number of patches for each of the potato cultivars in the landscape
 cvfrac                                       ; List containing the measured fraction of the potato cultivars in the landscape
 fung-eff                                      ; Protectant fungicide efficacy: kill rate of spores
 cur-eff                                      ; Curative fungicide efficacy: reduction of production of sporulating tissue (lesion expansion).
 phyto-day
 ; Escape and Deposition of spores
 escape-fraction                                ; [-] Fraction of spores present in Canopy escaping the canopy to be dispersed in the atmosphere
 surv-frac                                     ; [-] Fraction of spores surviving atmospheric dispersal
 DE                                           ; [-] Deposition Efficiency
 DE-max                                        ; [-] Maximum Deposition Efficiency
 k-dep                                        ; [-] Deposition coefficient, gives DE of 0.95 around LAI = 3
 SPORE                                         ; Fase nr of spores. During infection the pathogen goes through a number of phases, 0 - NL-INF
 LAT-PER                                       ; Latent Period, length of the latent period (days) equal to NL-INF - 2
 INF                                          ; Fase nr of infectious tissue (NL-INF - 1)
 NL-INF                                        ; Phase nr of No Longer Infectious tissue

 totlai                                       ; Sum of LAI for the world
 cvareas                                       ; List with relative leaf areas per h-type
 overall-severity                               ; Sum of all diseased leaf area for the world
 world-severity
 p-type-area                                    ; List: Total area occupied per p-type for the world
 p-type-frac                                   ; Fractions diseased leaf area occupied by the different p-types

; From the Interface
; frac-potato_                                  ; The desired fraction of potato in the landscape (from interface)
; Primary-Foci_                                  ; The average (!) nr of primary foci requested to start the season.
                                             ; Used to draw the true nr of primary foci from a poisson distribution with average (lamda) = Primary-Foci
; Protectants_                                  ; Protectant fungicides are used (on) or not (off)
; Curatives_                                    ; Curative fungicides are used

; Mutation-Rate_                                  ; Mutation rate per locus (AVR)
; Primary-lesions_                                ; Average number of primary (initial) lesions per infected patch
]

; ---------------------------------------------------------------------------------------------------------------------------
; -------------------------------------- Set parameters --------------------------------------------------------------------
to set-parameters
 ; v20151512
 ; Create and set the necessary basic paramters, lists etc. Only runs once at the start of a run.
 ; -------------------------------------------------------------------------------------------------------------------------
 ; General

 set base-types [ "1" "2" "3" "4" ]                ; Lists representing the R genes and Avr genes present/relevant in/to the system.
 set types     [ "0"         ]                  ; Initialisation of list used for pathogen virulence spectra and host R gene content:
                                             ; To be completed/filled by " create-types"  procedure
```

```

  ; ------------------------------------------------------------------------------------------------------------------------
  ; Host

  ; Landscape level
  ;                   [R0    R01   R02   R012  R03   R013  R023  R0123 R04   R014  R024  R0124 R034  R0134 R0234 R01234]  ; Possible R-gene content / R-gene combinations
  set cv-fractions-list [0.5   0.5   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   ]  ; Fractions of cultivars in the landscape
  set cv-colors-list    [15    24    25    43    26    44    45    64    27    46    47    65    48    66    67    105   ]  ; Color numbers for each R-gene combination
                                                                                                                          ; 0 R-genes: Red; 1 R-gene: shades of Orange, 2 R-genes: shades of yellow; 3 R-genes: shades of green; 4 R-genes: Blue

  ; Cultivar Specific Parameters
  set emergence-list    [90    90    90    90    90    90    90    90    90    90    90    90    90    90    90    90    ]  ; Average Julian emergence date
  set tub-init-list     [21    21    21    21    21    21    21    21    21    21    21    21    21    21    21    21    ]  ; Tuber initiation day, in days after emergence
  set maxlai-day-list   [54    54    54    54    54    54    54    54    54    54    54    54    54    54    54    54    ]  ; Day at which maximum LAI is reached in days after tuber initiation
  set end-tub-list      [90    90    90    90    90    90    90    90    90    90    90    90    90    90    90    90    ]  ; End day for tuber filling in days after tuber initiation
  set tuber-gr-list     [600   600   600   600   600   600   600   600   600   600   600   600   600   600   600   600   ]  ; Maximum Tuber growth rate [UNIT??????]
  set init-lai-list     [0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 0.069 ]  ; Set the initial lai after emergence [-] from Spitters (1989); 0.0155m^2 foliage on 0.225m^2 soil.
  set max-lai-list      [5     5     5     5     5     5     5     5     5     5     5     5     5     5     5     5     ]  ; Set the maximum lai [-]

  ; Pathogen race (p-race) specific parameters for virulent races on the respective
  set IE-list           [0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  0.05  ]  ; Set the Infection Eficiency for this cultivar (fraction succesfull spores) [-] for compatible (!) interactions
  set rgr-list          [0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   0.2   ]  ; Set the cultivar specific maximum relative growth rate for the epidemic (apparent infection rate [-])
  set SD-list           [1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   1E8   ]  ; Set the cultivar specific maximum Sporulation Density (sporangia/m2)



  ; ------------------------------------------------------------------------------------------------------------------------
  ; Pathogen
  ; Create the life cycle phases a pathogen is going through based on the (user defined) length of the latent period:
  set SPORE          0                                                                                                        ; DO NOT CHANGE! Spores are in phase 0
  set LAT-PER        5                                                                                                        ; Length of the latent period in days. User defined!
  set NL-INF         (LAT-PER + 2)                                                                                            ; Phase number for No Longer INFectious (necrotic) tissue
  set INF            (NL-INF - 1)                                                                                             ; Phase number for INFectious tissue
                                                                                                                              ; (first (0) = spore, 1 = L1 (latent day 1), 2 = L2, ..., last-1 = Inf (infectious), last = NLInf (No Longer Infectious)
  set prim-lesion-size  0.0001                                                                                                ; Size of a primary lesion immediately after the latent period (m2). 1 cm2 = 0.0001 m2

  set avr-fractions-list [1.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   0.0   ]  ; P. infestans Avr composition (races) at population level (fractions)
                                                                                                                              ; Infection is regulated through a gene for gene relationship.
                                                                                                                              ; The pathogen needs matching Avr genes to all R-genes present in the host to infect.
end

; ---------------------------------------- Setup ---------------------------------------------------------------------------------
to setup                                                  ; Setup Procedure
  ; v20151512

  ; General setup
  clear-all                                               ; Start with a clean slate

  ; Random runs or exact replications?
  if not keep-seed?                                       ; Test for setting of the "keep-seed?" switch
  [                                                       ; "keep-seed?" = on:    KEEP random seed which gives exact reproducible results
    set seed-for-random new-seed                          ; "keep-seed?" = off:   do NOT KEEP the random seed which gives results initiated with a different random seed including random variation.
  ]
  random-seed seed-for-random                             ; Sets the seed of the pseudo-random number generator to (the integer part) variable "seed-for-random"
```

```

ifelse not Protectants_
[
 set fung-eff 0
]
[
 set fung-eff 0.99
]

ifelse not Curatives_
 [
  set cur-eff 0
 ]
 [
  set cur-eff 0.99
 ]

; Call set up Procedures
set-parameters                          ; Call procedure "set-parameters"
create-types                           ; Call Procedure " create-types" This procedure expands the list already contained in the default "types" to contain
                                  ; all possible combinations of the "base-types" for P. infestans (virulence) and potato (R-genes)
set h-types types                       ; Host Types: Create list for all possible R-gene contents for Potato cultivars from "types" list (copy of "types")
set p-types types                       ; Pathogen types: Create list for all possible Avr contents for P. infestans from "types" list (copy of "types")
create-transitions                      ; Create a list of lists with possible transitions due to mutation: 1st index, index of original in sp-types
                                  ; elements: list of types to which a (direct) transition is possible
place-pathogens


; Error checking
; Host parameters
if precision (sum cv-fractions-list) 1 != 1.0           ; Check on sum of fractions for all cultivars, sum should be 1.
[                                 ; Abort if this sum is not equal to 1 and ask for user modification.
 error "The sum of the fractions of cultivars in the landscape is NOT equal to 1, please adjust and run again"
]
; Do some more checks (To Do)
; ...

; Pathogen parameters
if precision (sum avr-fractions-list) 1 != 1.0          ; Check on sum of P. infestans Avr fractions, sum should be 1.
[                                 ; Abort if this sum is not equal to 1 and ask for user modification.
 error "The sum of the P. infestans Avr-fractions in the landscape is NOT equal to 1, please adjust and run again"
]
; Do some more checks (To Do)
; ...


; Time keeping & counters

set season-counter 1                       ; Set the (growing) season counter to 1 at the start of the simulation run.

reset-ticks                          ; Reset the ticks to zero
tick-advance start-season_                  ; Forward the ticks to the start of the season.
                                  ; Ticks serve as "day counter" within seasons.
```

```
 write-header
 write-summary
end

; ----------------------------------------------------------------------------------------------------------------------------
; ----------------------------------------------------------------------------------------------------------------------------


; ------------------------------------------ Go ------------------------------------------------------------------------------
to go
 start-new-season                                          ; Start a new Season if the Julian date = 90 (approximately 1 April)
 grow-foliage                                            ; Call procedure grow-foliage
 grow-tubers                                             ; Call procedure grow-tubers

 if ticks = Start-day-epidemic_
 [
  start-epidemic                                         ; Intialise the epidemic
 ]

 if ticks >= Start-day-epidemic_
 [
  select-phyto-day                                        ; Call procedure select-phyto-day
  advance-epidemic                                         ; Call procedure advance-epidemic
  produce-spores                                          ; Call procedure produce-spores
  foreach reverse p-types                                   ; Call procedure do-mutations
  [                                                    ;  - reverse to avoid double mutations
   do-mutations ?
  ]
  disperse-spores                                        ; Call procedure disperse-spores
  summarize-epidemic                                        ; Call summarize-epidemic procedure
 ]
 summarize-landscape                                        ; Call summarize landscape procedure
 end-season                                              ; Call end-season procedure
 tick                                                 ; Advance tick by 1 (day)
 recolor
 write-summary
 type "Season: " type season-counter type " Day: " type ticks
 type " avr-fractions-list: " print avr-fractions-list
end
; ----------------------------------------------------------------------------------------------------------------------------

; ----------------------------------------------- Compound Procedures ------------------------------------------------------
; ----------------------------------------------- Start a New Season ------------------------------------------------------
to start-new-season
; v20150813
; Procedure to reset timers, the landscape and the Phytophthora population at the beginning of a new growing season
if ticks = start-season_
 [
  establish-fields                                        ; Run Procedure to establish 1 patch sized potato fields in the landscape
  place-pathogens                                          ; Run Procedure to place 1 pathogen turtle of each p-type on each patch
  plant-potato                                           ; Run Procedure to plant potato on the 1 patch sized fields
  assign-cultivars                                        ; Run Procedure to assign a cultivar (R-gene combination) to each potato field
  grow-fields                                            ; Run Procedure to grow 1 patch sized potato containing fields to the required average size (mean-fieldsize_)
```

```
    load-cultivar-variables                            ; Run Procedure to load the appropriate cultivar variables to the patches
  ]
end
; ------------------------------------------------------------------------------------------------------------------------

; ------------------------------------------------- End a Season ----------------------------------------------------------
to end-season
  ; Mixed Turtle & Patch procedure to end and summarize the growing season
  if ticks = end-season_
  [
    ; Summarize the season in tables, lists and graphs

    ; Adjust or reset timer & counters for the new season.
    set season-counter season-counter + 1
    reset-ticks
    tick-advance start-season_ - 1

    ; Remove turtles from the world
    ask fields [die]
    ask pathogens [die]


    ; Reset Patch variables
    ask patches
    [
      set potato          0
      set emergence       0
      set lai             0
      set maxlai-day      0
      set max-lai         0
      set p-fieldsize     0
      set yield           0
      set tub-init        0
      set end-tub         0
      set tuber-gr        0
      set init-lai        0
      set IE              0
      set severity        0
      set p-fieldnr       0
      set p-h-type        0
      set pcolor          black
      set tmp-conc        0
      set old-conc        0
      set new-conc        0
      set mobile-spores   0
      set immobile-spores 0
      set Overall-severity 0
      set world-severity  0
      set totlai          0
    ]
  ]
end
; ------------------------------------------------------------------------------------------------------------------------
```

```
;-------------------------------- Individual Procedures ----------------------------------------------------------------------
;------------------------------------------------------------------------------------------------------------------------------
;------------------------------------------------------------------------------------------------------------------------------
to create-types
; v20150327
; Global procedure to create a list of "types" (Global); all possible combinations of 1, 2, 3, .. etc as given in "base types"

  foreach base-types                                ; Geneste "for loop"over (1) "base-types" (global) en (2) "tmp-types" (global)
  [
    let base-type ?                                 ; local variable " base-type"
    let tmp-types types
    foreach tmp-types
    [
      let tmp-type ?                                ; local variable "tmp-type"
      let new-type (word tmp-type base-type)              ; "word" concatenates the inputs together and reports the result as a string.
      set types lput new-type types                 ; Add (lput) "new-type" to the end of the "types" list and reports the new list as "types".
    ]
  ]
end
;------------------------------------------------------------------------------------------------------------------------------

to place-pathogens

  ; Place a pathogen turtle for each type * phase combination on each patch.
  foreach p-types
  [                                                 ; "for loop" over the various "pathogen types"
    let tmp-type ?
    foreach n-values (NL-INF + 1) [?]
    [
      let tmp-phase ?
      ask patches
      [
        sprout-pathogens 1                          ; Assigns one (1) turtle of each race * phase" combination to each patch
        [
          hide-turtle
          set p-race tmp-type
          set quantity 0
          set phase tmp-phase
        ]
      ]
    ]
  ]
end

;------------------------------------------ Establish Single Patch Potato Fields ---------------------------------------------
to establish-fields
; v20150916
; Turtle procedure to create single-patch "fields" (Turtles) in an agricultural landscape.
; This procedure runs 1x per year cycle at the start of the growing season.
; The result serves as input for the "plant-potato"procedure.
```

```
 set host-fieldnr-list [ ]                                   ; Create an empty list host-fieldnr-list to contain the nr of fields for each host type

 let nrflds                                                  ; Calculate the number of potato fields required
   round(((world-width * world-height) * frac-potato_)
     / mean-fieldsize_)

 foreach (n-values length cv-fractions-list [?])
 [
   let i ?
   let tempnr round((item i cv-fractions-list) * nrflds)          ; Calculate the number of fields for each cultivar
   set host-fieldnr-list  lput tempnr host-fieldnr-list           ; Add this number to a list
 ]

 set nrfields sum host-fieldnr-list                               ; Calculate the total/resulting number of fields

 create-fields nrfields                                      ; Create "nrfields" fields (breed/turtles) in the landscape
 [
   setxy (random-xcor) (random-ycor)                              ; Randomly select an X and Y coordinate
   while                                                     ; Keep some away from established fields or fields that are too close
   [
     any? other fields-here or                               ; Check 1) whether a field is present on the selected coordinates
     any? other fields in-radius sqrt(mean-fieldsize_)            ; Check 2) whether a field is present within radius sqrt(mean-fieldsize) of the selected coordinates
   ]
   [
     setxy (random-xcor) (random-ycor)                            ; If 1 or 2 are true, select a new random set of coordinates
   ]
   set t-fieldnr [who] of self                               ; Give the turtle (field) a unique number
   hide-turtle                                               ; Hide the turtle from view
 ]
;  ask fields                                                ; Give the fields a color for checking purposes
;   [
;     ask patch-here
;     [
;       set pcolor red
;     ]
;   ]
 ; print count fields                                        ; For checking purposes
end
;-------------------------------------------------------------------------------------------------------------------------------

;-------------------------------------------------- Plant Potato -------------------------------------------------------------
to plant-potato
; v20150916
; Turtle procedure to "plant potatoes" on the already established 1 patch fields

 ask fields                                                  ; Ask the fields (sized to 1 patch at this point)
 [
  ask patch-here                                             ; to set "potato patch property" for patch underneath to "1" (potato present)
  [
    set potato 1
    ;set pcolor brown                                         ; Give potato fields a color for checking purposes
    set p-fieldnr [t-fieldnr] of myself                          ; Assign the unique turtle field-number to the patch underneath the field.
  ]
```

```
 ]
 ask fields with [potato = 0]
 [
   die
 ]
end
;----------------------------------------------------------------------------------------------------------------------------

;---------------------------------------- Assign Potato Cultivars -------------------------------------------------------------
to assign-cultivars
; v20151006
; Turtle Procedure assigning potato cultivars to single patch fields such that the required fractions for each cultivar in the landscape (cv-fractions-list) are met.

ask fields [set t-h-type ""]                                    ; Assign a dummy t-h-type to all fields

foreach (n-values length cv-fractions-list [?])                    ; Create and loop through a list of numbers  0 - "length cv-fractions-list"
[
  let i ?                                                    ; Good Programming Practice to replace the "?"
  let targetnr  (item i host-fieldnr-list)                         ; local variable "number" representing the number of fields for the cultivar under consideration
  let freefields fields with [t-h-type = ""]
  let tmp-type (item i types)                                    ; local variable "tmp-type" representing the cultivar type (R-gene content)  under consideration

  ask n-of targetnr freefields [set t-h-type tmp-type]
  ask fields with [t-h-type = tmp-type]
  [
    ask patch-here
    [
      ;set pcolor item i cv-colors-list                              ; set the patch color (pcolor) to the appropriate pcolor for this cultivar
      set p-h-type [t-h-type] of myself                             ; Match the patch "p-h-type" to the field "t-h-type"
    ]
  ]
]
end
;------------------------------------------------------------------------------------------------------------------------------

;--------------------------------------------------  Grow Fields ----------------------------------------------------------------
to grow-fields
; v20151006
; Patch procedure to grow the 1 patch sized potato containing fields to the required average size "mean-fieldsize_" from the interface
set nrpotatopatches
  round((world-width * world-height) * frac-potato_)

foreach (n-values length cv-fractions-list [?])                    ; Create and loop through a list of numbers  0 - "length cv-fractions-list"
[
  let i ?
  let frac (item i cv-fractions-list)                              ; Read the required fraction for this cultivar in the landscape
  let targetnr round(frac * nrpotatopatches)                        ; Calculate the required nr of patches per cultivar derived from the fraction above

   while
   [ count patches with [p-h-type = item i types] < targetnr]
   [
     ask one-of patches with [p-h-type = item i types and count fields-here > 0]
     [
```

```
    let target one-of neighbors with [potato = 0]
    if target != nobody
    [
     ask target
     [
      set potato 1
      set p-fieldnr [p-fieldnr] of myself
      set p-h-type [p-h-type] of myself
      ;set pcolor item i cv-colors-list
     ]
    ]
   ]
  ]
]
end
;-------------------------------------------------------------------------------------------------------------------------------

; -------------------------------------------------- Load cultivar specific parameters ---------------------------------------
to load-cultivar-variables
;  v20150821
;  Patch Procedure loading the cultivar specific growth, yield and resistance parameters

foreach (n-values length h-types [?])                              ; Create and loop through a list of numbers  0 - "h-types"
[
 let i ?
 ask patches with [(potato = 1) and (p-h-type = item i h-types)]
 [
  set emergence int random-normal (item i emergence-list) 5.5       ; set the julian emergence date (integer) including a random factor
                                                             ; command: "random-normal mean standard-deviation"
  set tub-init   (emergence  + item i tub-init-list  )             ; Set the julian start-day for tuber filling
  set maxlai-day (tub-init   + item i maxlai-day-list)             ; Set the julian day at which the maximum lai is reached
  set max-lai    (            item i max-lai-list   )             ; Set the maximum value for the leaf area index (lai)
  set end-tub    (tub-init   + item i end-tub-list   )             ; Set the julian end-day for tuber filling
  set tuber-gr   (            item i tuber-gr-list  )             ; Set the maximum tuber growth rate [UNIT?]
  set init-lai   (            item i init-lai-list  )             ; Set the initial lai after emergence [-] from Spitters (1989); 0.0155m^2 foliage on 0.225m^2 soil.
  set IE         (            item i IE-list       )             ; Set the Infection Eficiency for this cultivar (fraction succesfull spores) [-]

  ; Initialise the Leaf Area Index (LAI) and Severity (% destroyed foliage) per patch
  set lai                          init-lai               ; Initialise the lai with init-lai
  set severity                          0               ; Severity: Patch specific sum of fraction destroyed foliage (LAI) by the combined P. infestans genotypes.
  set yield                            0               ; Yield: Patch specific tuber yield (kg/ha)
 ]
]
end


; ------------------------------------------------- Grow Potato Foliage -------------------------------------------------------
to grow-foliage
; v20160607
; Patch procedure simulating cultivar specific crop growth (LAI)

 ask patches with [potato = 1 and ticks > emergence and severity < 100]
 [
```

```
    ifelse (ticks <= maxlai-day)
    [
      set lai lai + (0.15 * (lai * (1 - severity)) * (1 - (lai / max-lai) ^ 0.8))        ; Growth phase, growth limited by severity (fraction dead tissue)
    ]
    [
;;      set lai lai - (0.10 * lai * (1 - (lai / max-lai)))             ; Senescence phase
    ]
  ]
;  ask patches with [potato = 1]
;  [
;    set pcolor scale-color ([pcolor] of self) lai 10 0
;  ]
end


; ------------------------------------------------- Grow Potato Tubers -  -----------------------------------------------------
to grow-tubers
; Calculate tuber growth per day since tuber initiation untill crop senescence.
; Tuber growth rate is determined by the max-tuber growth rate (tuber-gr) and reduced by light interception and infection level (severity).
  ask patches with
  [
    potato = 1
    and ticks > tub-init
    and ticks < end-tub
  ]
  [
    set yield yield + tuber-gr * (1 - (2 * severity / 100)) * (1 - exp(-0.7 * lai))
  ]
end

;-----------------------------------------   Start the epidemic    -------------------------------------------------------------
to start-epidemic
; v20151012
; Patch Procedure to randomly start a low level P. infestans epidemic in the landscape.
; First year: only P. infestans genotype Avr0 starts the epidemic.
; Later years: the end of season Avr population composition (Avr fractions present) from the previous year
; is used as a starting point.

foreach (n-values length avr-fractions-list [?])                    ; Loop over a list of numbers  0 - "length avr-fractions-list"
[
  let i ?                                                    ; Good Programming Practice to replace the "?"
  let targetnr
    ((item i avr-fractions-list) * primary-lesions_)                ; Local variable "targetnr" representing the initial number lesions for the pathogen race and season under consideration

  let available-patches patches with
  [(potato = 1) and (is-subset p-h-type item i p-types)]             ; Select available patches: patches with potato and a compatible host type for the



  ifelse targetnr > 0                                         ; We have > 0 lesions to assign to patches
  [
    ifelse targetnr < count available-patches                       ; We have more patches than lesions
    [
      ask n-of targetnr available-patches
```

```
   [
   ask pathogens-here with
    [p-race = item i p-types and phase = NL-INF]       ; Initialise the last phase NL-INF (No longer infectious area) with the quantity required
    [
     set quantity                         ; Set quantity of phase NL-INF for the P. infestans genotype under consideration to fraction necrotic including some random variation
     (prim-lesion-size / lai)
    ]
   ]
  ]
  [
   ask available-patches                      ; more lesions than patches
   [
    let lesionsperpatch targetnr / count available-patches   ; Calculate the average number of lesions to be assigned to each available patch.
    ask pathogens-here with
    [p-race = item i p-types and phase = NL-INF]       ; Initialise the last phase NL-INF (No longer infectious area) with the quantity required
    [
     let primary-lesions random-poisson (lesionsperpatch)    ; Assign the number of lesions per patch with some random variation
     set quantity                         ; Set quantity of phase NL-INF for the P. infestans genotype under consideration to fraction necrotic including some random variation
     ((primary-lesions * prim-lesion-size) / lai)
    ]
   ]
  ]
 ]
 [                                   ; targetnr = 0. No lesions to distribute over patches.
  ask available-patches
  [
   ask pathogens-here with
   [p-race = item i p-types and phase = NL-INF]        ; Initialise the last phase NL-INF (No longer infectious area) with the quantity required
   [
   set quantity   0                        ; Set quantity of phase NL-INF for the P. infestans genotype under consideration to fraction necrotic including some random variation
   ]
  ]
 ]
]
end

;--------------------------------------------   Produce spores   ------------------------------------------------------------
to produce-spores
; v20151027
; Patch procedure looping over the pathogen races and calculating the sporulating area and numbers of spores (sporangia) produced.

if (phyto-day = 1)                          ; Produce spores only when the weather is suitable for epidemic development
[
 foreach (n-values length p-types [?])              ; Create and loop over a list of numbers  0 - "length avr-fractions-list"
 [
  let i ?                             ; Loop over p-race's (pathogen races)
  ask patches with
   [(potato = 1)]
   [
    ask pathogens-here with
     [p-race = item i p-types and phase = NL-INF]      ; Ask pathogen with p-race under consideration and phase = NLINF
     [
      let inf-area                       ; Calculate fraction of sporulating/infectious foliage (INF) based on NLINF
```

```
        quantity * (item i rgr-list) * (1 - quantity)
                                     ; area using a logistic growth equation: dN/dt = rN ((K-N)/K). K = 1, r = Apparent-Infection-Rate

      ask pathogens-here with
       [p-race = item i p-types and phase = SPORE]       ; Calculate the spores produced on sporulating leaf area (inf) and assign to quantity p-race phase 0 (spores)
       [
        set quantity inf-area * lai * (item i SD-list)
        ; print quantity
       ]
     ]
   ]
 ]
]
end

;--------------------------------------------  Escape from Canopy  ------------------------------------------------------------
to escape
; v20151217
; Patch procedure calculating fraction of spores escaping from the canopy.
; Spores have to escape the canopy before they can be dispersed through the atmosphere
; A crude approximation is derived from Aylor, Fry, Mayton and Andrade-Piedra 2001
; Phytopathology 91: 1189 - 1196
; In a better approximation, the escape fraction depends on the settling velocity (vs), u* (U-star) and Fx(LAI). NOT IMPLEMENTED.

 set escape-fraction 0.3

end

;--------------------------------------------  Disperse spores  ------------------------------------------------------------
to disperse-spores
; v20160608
; Procedure dispersing spores (per p-type) by diffusion and wind dispersal,
; survival during transport and deposition on a neighbouring crop.

 foreach p-types
 [
  let spores pathogens with [p-race = ? and phase = SPORE]      ; Selected pathogen turtle (p-race and phase) as local variable "spores"
  escape                              ; 1) call procedure "escape"
  diffuse-spores   spores                   ; 2) call procedure "diffuse-spores" incl. escape, diffusion and move-by-wind
  survive-dispersal spores                   ; 3) call procedure "survive-dispersal"
  deposit-spores   spores                   ; 4) call procedure "deposit-spores"
 ]

 ;show-selected-type-conc                      ; Call display procedure for spore dispersal
end

;--------------------------------------------  Advance Epidemic by 1 time step (1 day)  -------------------------------------
to advance-epidemic
; v20160603
; Turtle procedure to advance the epidemic by 1 time step (1 day)
 if (phyto-day = 1 )
 [
  ask pathogens
```

```
[
  set phase phase + 1                              ; Age phase by 1 time step, keep quantities intact
]                                                  ; (This removes phase 0 "SPORE" and adds phase NL-INF + 1)

let i -1
foreach p-types                                    ; Loop over pathogen p-types
[
  let p-type ?
  set i i + 1
  let rgr-list-i item i rgr-list
  let IE-list-i item i IE-list
  let p-type-pathogens pathogens with [p-race = p-type]

  ; Store current values for quantities phases INF and NL-INF
  ask p-type-pathogens with [phase = INF]               ; Store quantity for phase = INF = quantity old final latent phase
  [ set q quantity  ]

  ask p-type-pathogens with [phase = NL-INF]            ; Store quantity for phase = NL-INF = quantity old INF
  [ set r quantity  ]

  ask p-type-pathogens with [phase = (NL-INF + 1)]      ; Store  quantity for phase = (NL-INF + 1) = quantity old NL-INF
  [ set s quantity  ]

  ; Calculate new values for NL-INF
  ask p-type-pathogens with [phase = NL-INF]
  [
    set quantity r + s                                  ; New quantity for phase NL-INF = quantity old NL-INF + quantity old INF
  ]

  ; Calculate new values for INF
  ask p-type-pathogens with [phase = INF]
  [
    set quantity (q + ((r + s) * rgr-list-i)) * (1 - severity)     ; Calculate new quantity for phase = INF: (emerging lesions + growth from NL-INF) * logistic correction for room to grow.
    set quantity quantity * (1 - cur-eff)                ; Effect of Curative fungicides
  ]

  ; The Infection process
  ask p-type-pathogens with [phase = SPORE + 1]          ; Calculate new quantity for phase = 1 (latent infection day 1): (spores * IE * primary lesion size)
  [
    ifelse (is-subset p-h-type p-race) and (phyto-day = 1)       ; Test whether the pathogen is compatible on the R-genes "grown"on this patch. (p-h-type is subset of p-race) and the weather is suitable for infection.
    [                                                   ; If true
      set quantity                                      ; Calculate the relative area with new lesions based on quantity of spores, IE and size of primary lesions
      (quantity * (IE-list-i) * prim-lesion-size) / lai
      set quantity quantity * (1 - severity) * (1 - fung-eff)       ; Correct for spores on already infected tissue and the effect of fungicides - if used -
    ]
    [
      set quantity 0                                    ; If False
    ]
  ]
]

; Tidy up and reset for a new day
ask pathogens with [phase = (NL-INF + 1)]               ; RE-use the extra phase created above as the new phase "SPORE"
```

```
    [
      set phase SPORE
      set quantity 0                                  ; Re-establish phase SPORE (0) and set quantity to 0, ready for re-use
    ]
  ]
;  ; Display the result
;  ask pathogens with [phase = NL-INF and quantity > 0]
;  [
;  set pcolor scale-color brown quantity 1 0
;  ]
end
;----------------------------------------  Summarize landscape  ------------------------------------------------------------
to summarize-landscape
; Procedure summarizing the basic characteristics of the landscape for checking purposes
; Number of patches with potato
; Fraction of each cultivar in the landscape
; Area of each cultivar in the landscape
  set cvcount []
  set cvfrac  []
  set cvareas []

  foreach h-types
  [
    let tmp-type ?
    let tmpcount     count patches with [p-h-type = tmp-type]
    let potatocount  count patches with [potato = 1]
    let tmparea      sum [lai] of patches with [p-h-type = tmp-type]
    let tmpfrac      tmpcount / potatocount

    set cvcount      lput tmpcount cvcount
    set cvfrac       lput tmpfrac  cvfrac
    set cvareas      lput tmparea  cvareas
  ]
end



; --------------------------------------  Summarize Epidemic  ------------------------------------------------------------
to summarize-epidemic
; v20160607
; Patch procedure to summarize the late blight epidemic

  ; 1. Per patch:
  ask patches [set severity 0]                          ; Reset the severity value for each patch
  ask pathogens with [phase = NL-INF]
  [
    set severity severity + quantity                    ; Total Severity per patch [0 - 1] as the sum of all quantities at phase NL-INF
  ]

;  ask pathogens with [phase = NL-INF and quantity > 0]
;  [
;  set pcolor scale-color brown quantity 1 0
;  ]
```

```
 ; 2. For the World:
 set totlai sum [lai] of patches                                   ; Sum of LAI for the world
 set overall-severity sum [(severity * lai)] of patches              ; Sum of all diseased leaf area for the world
 ifelse totlai > 0
 [
  set world-severity overall-severity / totlai                      ; Fraction diseased of all potato in the world
 ]
 [
  set world-severity 0
 ]

 set p-type-area []                                              ; Area per p-type for the world.
 set p-type-frac []
 foreach p-types
 [
  let tmp-type ?
  let tmpquant sum [(quantity * lai)]
    of pathogens with [p-race = tmp-type and phase = NL-INF]       ; Total realtive leaf area occupied by p-type under consideration.
  let tmpfrac 0
  if overall-severity > 0 [
   set tmpfrac (tmpquant / overall-severity)
  ]
  set p-type-area lput tmpquant p-type-area                        ; Collect areas per p-type in list.
  set p-type-frac lput tmpfrac  p-type-frac                        ; Collect fractions of p-types in pathogen population in list.
  set avr-fractions-list p-type-frac                               ; Update the avr-fractions-list to the current situation
 ]

end


;----------------------------------------- Spore dispersal by diffusion and wind dispersal -----------------------------------
to diffuse-spores [spores]
; v20151216
; Patch procedure for dispersal of spores by diffusion and wind dispersal.

 ask spores
 [
  set mobile-spores   (quantity * escape-fraction)                 ; Calculate nr of spores available for dispersal (escaping the canopy) (patches own)
  set immobile-spores (quantity * (1 - escape-fraction))           ; Calculate nr of spores remaining in the canopy (patches own)
 ]

 ; Disperse by diffusion
 repeat 50                                                ; According to "Santiago" TO BE OPTIMIZED
 [
  diffuse mobile-spores diffusion-constant_                        ; Diffuse the mobile spores on each patch, save in "mobile-spores"
 ]

 ; Disperse by wind
 ;move-by-wind                                             ; Call procedure "move by wind" as a 2nd step in de dispersal process
                                                  ; Use the "mobile-spores" result after diffusion as input, save result
                                                  ; by updating "mobile-spores"
```

```
  ; Calculate the final result: quantity of spores per patch and p-type after diffusion and wind dispersal
  ask spores
  [
    set quantity immobile-spores + mobile-spores                    ; Update turtle property "quantity" (of spores) with remaining spores + incoming spores
  ]
end
;------------------------------------------------ Wind dispersal ------------------------------------------------------------
to move-by-wind
; v20160608
; Procedure for dispersal by wind

  ask patches
  [
    set new-conc 0
    set old-conc mobile-spores                              ; Copy for easier code
  ]

  ask patches                                               ; just move everything -- add spreading over multiple patches...
  [                                                         ; to do: process fractional wind-x and wind-y
    let my-conc old-conc                                    ; Copy for easier code
    ask patch-at wind-x wind-y                              ; Move spores to patch at (x,y) from myself
    [
      set new-conc new-conc + my-conc
    ]
  ]

  ask patches
  [
    set mobile-spores new-conc                              ; Update "mobile-spores" with the result of "move-by-wind"
  ]
end

;------------------------------------------------ Survive dispersal ---------------------------------------------------------
to survive-dispersal [spores]
; v20151216
; Patch procedure calculating the quantity of surviving spores after dispersal/non dispersal
; To be improved

  set surv-frac 0.1

  ask spores
  [
    set quantity quantity * surv-frac                       ; Dispersed and non-dispersed spores are assumed to survive equally well
  ]
end

;--------------------------------------------   Deposit spores   ------------------------------------------------------------
to deposit-spores [spores]
; v20151120
; Patch procedure to calculate spore deposition on the crop after dispersal using the deposition efficiency (DE)

  set DE-max       0.1                                  ; Maximum deposition efficiency
  set k-dep        1                                    ; Coefficient, gives DE of 0.95 x DE-max at LAI = 3
```

```
                                                ; and DE = DE-max at LAI = 5
  ask patches with [potato = 1]
  [
    set DE DE-max * (1 - exp(-1 * k-dep * lai))          ; Effect of leaf area index (lai)
    set DE DE * (1 - severity)                          ; Fraction spores landing on healthy tissue.
  ]

  ask spores
  [set quantity quantity * DE  ]                      ; Calculate the number of deposited spores on the canopy
end

; ------------------------------------------ Reporter: Transition possible ? -----------------------------------------------
to-report transition-possible? [type1 type2]
; v20160610 Mark's original
; Counts the number of common members between type1 and type2 and selects those with 1 additional member.

  let added 0                                           ; will be number of base-types in type2 not in type1
  let deleted 0                                         ; will be number of base-types in type1 not in type2
  foreach base-types                                      ; check occurrences of all base-types in type1 and type2
  [
    let base-type ?
    ifelse member? base-type type2
    [
      if not member? base-type type1
      [
        set added added + 1
      ]
    ]
    [
      if member? base-type type1
      [
        set deleted deleted + 1
      ]
    ]
  ]
  ; transition only when one base-type is added and none deleted
  report added = 1 and deleted = 0
end

; ------------------------------------------ Create list of lists with of possible transitions ------------------------------
to create-transitions
; v20160610

  set transitions []

  foreach p-types
  [
    let original ?
    let targets []

    foreach p-types
    [
      let target ?
```

```
      if transition-possible? original target
      [
        set targets lput target targets
      ]
    ]
    set transitions lput targets transitions
  ]
end

;-------------------------------------------------------- Do mutations according to Garp ------------------------------------
to do-mutations [spore-type]
; v20160610

  let i position spore-type types
  let targets item i transitions
  let n-targets length targets

  ask pathogens with [phase = SPORE and p-race = spore-type]
  [
    let mutants round (quantity * Mutation-Rate_)
    set quantity quantity - n-targets * mutants
    foreach targets
    [
      let target ?                                         ; type that gets increased in current cell
      ask pathogens-here with [phase = SPORE and p-race = target]
      [
        set quantity quantity + mutants                            ; actually only one per target
      ]
    ]
  ]
end
;---------------------------------------   Select Phytophthora day    --------------------------------------------------------
to select-phyto-day
  set phyto-day one-of [1 1]
end

;---------------------------------------------   Report Subset   ------------------------------------------------------------
to-report is-subset [a b] ; relative to types!
; Reporter to test whether subset a of "types" is a subset of subset "b"of types.

if (not is-string? a) or (not is-string? b) [report false]
let result a != true
foreach types
[
  let a-type ?
  if member? a-type a
  [
    if not member? a-type b
    [
      set result false
    ]
  ]
]
```

```
report result
end

; ----------------------------------------- Display the selected spore type --------------------------------------------------
to show-selected-type-conc
; v20151120
; Procedure to display spore dispersal for a selected type
  ifelse member? selected-type types
  [
    ask pathogens with
      [p-race = selected-type and phase = 0]
      [
        set pcolor scale-color turquoise quantity 0 1
      ]
  ]
  [
    ask patches [ set pcolor black ]
  ]
end

; ----------------------------------------- Calculate a safe mean without error messages -------------------------------------
to-report safe-mean [data]
  ifelse length data = 0
  [
    report 0
  ]
  [
    report mean data
  ]
end

; ----------------------------------------- Recolor the world according to Garp ---------------------------------------------
to recolor
  with-local-randomness
  [
    ifelse how-to-color = "lai"
    [                                          ; true ifelse 1
      ask patches
      [
        set pcolor scale-color green lai 0 8
      ]
    ]

    [                                          ; false ifelse 1
      ifelse how-to-color = "severity"
      [                                        ; true ifelse 2
        ask patches
        [
          set pcolor scale-color brown severity 0 1
        ]
      ]
      [                                        ; false ifelse 2
        ifelse how-to-color = "Potato cultivar"
```

```
    [
     foreach (n-values length h-types [?])
     [
      let i ?
      ask patches with [p-h-type = item i h-types]
      [
       ifelse severity = 0
       [
        set pcolor item i  cv-colors-list                ; set the patch color (pcolor) to the appropriate pcolor for this cultivar
       ]
       [
        set pcolor scale-color brown severity 0 1
       ]
      ]
     ]
    ]
    [ ; Do Nothing
    ]
   ]
  ]

 ]






;to recolor
;  with-local-randomness
;  [
;   ifelse how-to-color = "lai"
;   [
;
;   ]
;   [
;    ifelse how-to-color = "severity"
;    [
;     ask patches
;     [
;      set pcolor scale-color brown severity 0 1
;     ]
;    ]
;   ]
;   [
;    ifelse how-to-color = "potato cultivar"
;    [
;     foreach (n-values length cv-fractions-list [?])          ; Create and loop through a list of numbers  0 - "length cv-fractions-list"
;     [
;      let i ?                              ; Good Programming Practice to replace the "?"
;       ask patches with p-h-type = item i h-types
;       [
;        set pcolor item i cv-colors-list                ; set the patch color (pcolor) to the appropriate pcolor for this cultivar
```

```
;       ask patches with  severity > 0
;       [
;        set pcolor scale-color brown severity 0 1
;       ]
;      ]
;    ]
;   ]
;   [ ; default do nothing
;   ]
;  ]
; ]
end

; ------------------------------------------ Write the header to the result file ----------------------------------------------
to write-header
 if log-filename != "" [
;  if file-exists? log-filename [file-delete log-filename]
  file-open log-filename
  file-type "Season,Ticks,Mutation Rate,Cultivars,Overall-Severity,world-severity,Total-LAI"
  foreach n-values length h-types [?] [
   file-type ",LAI ras " file-type ?
  ]
  foreach n-values length p-types [?]
  [
   file-type ",Inf area p-race " file-type ?
  ]
  foreach n-values length p-types [?]
  [
   file-type ",Spores p-race " file-type ?
  ]
  foreach n-values length p-types [?]
  [
   file-type ",NL-INF p-race " file-type ?
  ]

  file-type ",avr-fractions-list"
  file-type ",seed:" file-print seed-for-random
  file-close
 ]
end

; ------------------------------------------ To write summary data of the results ---------------------------------------------
to write-summary
; v20160608
; Procedure to generate summary output data in a data file
 if log-filename != "" [
  file-open log-filename
  with-local-randomness
  [
   file-type season-counter
   file-type ","
   file-type ticks
   file-type ","
```

```
      file-type Mutation-rate_
      file-type ","
      file-type cv-fractions-list
      file-type ","
      file-type overall-severity
      file-type ","
      file-type world-severity
      file-type ","
      file-type totlai

      ; Average LAI per host type (potato cultivar)
      foreach h-types
      [
        let h-type ?
        file-type ","
        file-type safe-mean [lai] of patches with [potato = 1 and p-h-type = h-type]
      ]

      ; Absolute sporulating area in the world
      foreach p-types
      [
        let p-type ?
        file-type ","
        file-type sum [quantity * lai] of pathogens with [p-race = p-type and phase = INF]
      ]

      ; Absolute number of spores in the world per p-race
      foreach p-types
      [
        let p-type ?
        file-type ","
        file-type sum [quantity * lai] of pathogens with [p-race = p-type and phase = SPORE]
      ]

      ; Fraction occupied leaf area per p-race
      foreach p-types
      [
        let p-type ?
        file-type ","
        file-type safe-mean [quantity] of pathogens with [p-race = p-type and phase = NL-INF]
      ]

      ; Current avr-fractions-list
      file-type ","
      file-type avr-fractions-list
      file-type ","
      file-print ""
    ]
    file-close
  ]
end
```