Stability Analysis of a Plant Pathogen Evolution Model

Narawitch Lertngim, Usa Humphries, Frank van den Bosch

Abstract: ***Stacking of multiple resistance genes in crop cultivars, pyramiding, is an effective way to control plant diseases. However, due to the evolution of the pathogen, resistance cultivar can be overcome. Breeders develop pyramids with more resistance genes to improve the durability of resistance. Since it is difficult to do field experiments on the evolution of the pathogen, mathematical models become an important tool to study the behavior of evolution of plant pathogen through stability analysis. A model for the evolution of pathogen is developed towards virulence against cultivar resistance genes. The aims of this research are to analyze the stability of the steady states of the model. The results from the analysis show that only the equilibrium point (0,0,0,1) was stable while the others were unstable. This stable equilibrium point is characterized by the strain virulent to both resistance genes. The other pathogen strains disappear from the population. Inthe future, the model can be used to determine the breeding strategic for plants which can delay the evolution of pathogen and prolong durability of pyramided plant.***

I. INTRODUCTION

Plant pests and pathogens are an important problem in agriculture. Loss from pests and pathogens are estimated to be up to 30% of crop product sale value annually and more than 10% annual of crop production costs is spent to control diseases [1]. Plant pest and pathogen resistance genes are widely used as an effective approach in damage control. Breeding for resistance has been done for decades [2]. However, the pest or pathogen often develops through mutation or another genetic which change the ability to feed or infect the plant that was previously resistant.

For example, BT crops are developed to carry an insecticidal protein derived from the bacterium Bacillus thuringiensis (Bt). BT resistant plants are grown over 22.4 million ha. worldwide and 13 million ha. in the United States in 2004 [3]. There are no reports about of crop failure after extensive usage of transgenic plants but there are concerns about evolution of resistance [4]. The strategies to prolong the useful life of BT resistance were explored and the most widely used approach is the refuge strategy which enables the sensitive pest strains to maintain a population in the refuge.

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The other method is pyramiding which stacking several different resistance genes into one cultivar. An approach first introduced by Watson and Singh 1953 [5]. These pyramided plants contain multiple genes which improved efficacy on the target pest and are assumed to have a longer useful life as it is more difficult for the pest to accumulate all resistance. Mutations needed to overcome the resistances. [6][7]. Nevertheless, pests can adapt to pyramids as well and therefore, study on evolution of plant pests and pathogens have become more important to improve crop production.

To study the adaptation of plant pathogens to crop resistance is complicated and requires considerable resources and time to do the genetical experiments. Mathematical models are thus useful tools in the study of adaptation. In this paper, the multiple strain population model from Fisher, 1930 is developed by adding mutations. Then, steady-state and stability analysis are used to study the behavior of plant pathogen populations. The aim of this research is to analyze the stability of the plant pathogen evolution model which will be useful to pest management.

II. PLANT PATHOGEN EVOLUTION MODEL

Plant pathogen is an organism which caused plant get infected. Since most plant pathogens are fungi or bacteria, the mathematical model describing the dynamic of pathogen populations is the exponential population growth model. The rate of change of each pathogen strain in the population depends on the number of individuals at time t and strain's per capita growth rate, r. a pathogen population adapting to two cultivar resistance genesis modeled, and therefore in the model presented, four genotypes of pathogen were modelled. [8].

There are five evolutionary forces for pathogen such as mutation, population size, gene flow, reproduction and mating system and natural selection [9]. Population size was included to the model as the density of each strain in the population, mutations were included in the model to study the evolution of pathogen since they are the driving factor behind the emergence of the new virulent pathogen strains. Evolution of pathogen possibly occurs both from avirulent to virulent strain and in opposite direction. Hence each pathogen strain can become any strain which may be more or less virulent. The concept of evolution is shown in the following diagram:

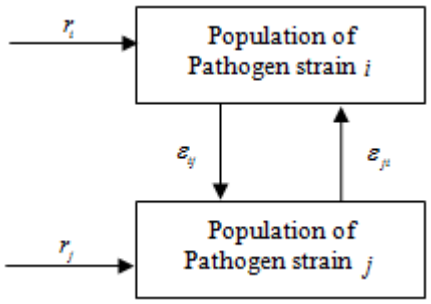


Fig. 1 Conceptual diagram of the plant pathogen evolution model

From the diagram, ε_{ij} represents mutation rate of pathogen strain i to strain j. Our multiple strain model has the form:

$$\left.\begin{aligned}
\frac{d\theta_1}{dt} &= (r_1-\bar{r})\theta_1+\varepsilon_{21}\theta_2+\varepsilon_{31}\theta_3+\varepsilon_{41}\theta_4-\varepsilon_1\theta_1\\
\frac{d\theta_2}{dt} &= (r_2-\bar{r})\theta_2+\varepsilon_{12}\theta_1+\varepsilon_{32}\theta_3+\varepsilon_{42}\theta_4-\varepsilon_2\theta_2\\
\frac{d\theta_3}{dt} &= (r_3-\bar{r})\theta_3+\varepsilon_{13}\theta_1+\varepsilon_{23}\theta_2+\varepsilon_{43}\theta_4-\varepsilon_3\theta_3\\
\frac{d\theta_4}{dt} &= (r_4-\bar{r})\theta_4+\varepsilon_{12}\theta_1+\varepsilon_{32}\theta_3+\varepsilon_{42}\theta_4-\varepsilon_2\theta_2
\end{aligned}\right\} \quad (1)$$

From the system (1), the mutation term with a positive sign represents mutations from other strains while a negative sign represents the mutations to other strains. There are four genotypes of pathogen in this model

1) θ_1 is the fraction of avirulent pathogen strain, meaning this pathogen strain is unable to infect a cultivar with resistance gene A or/and resistance gene B.

2) θ_2 is the fraction of the pathogen population virulent to gene A and avirulent to gene B

3) θ_3 is the fraction of the pathogen population avirulent to gene A and virulent to gene B

4) θ_4 is the fraction of the population virulent to both resistance genes.

The fitness of pathogen strain i (r_i) is given by logarithmic equation from Malthus, 1798 with including the fraction of the susceptible cultivar:

$$r_i = \frac{\ln R_0(1-x_i)}{\mu} \quad (2)$$

where R_0 is net-reproductive number, $1/\mu$is mean generation time of pathogen and x_i is the fraction of resistant cultivar affecting pathogen strain i.

The mean population fitness $(\bar{r})$ depends on the fraction of all strain and its dynamics is given by the differential equation:

$$\frac{d\bar{r}}{dt} = \sum_{i=1}^{4} r_i^2\theta_i-\bar{r}^2+\sum_{i=1}^{4} r_i\sum_{j=1,j\neq 1}^{4}\varepsilon_{ji}\theta_j-\sum_{i=1}^{4} r_i\varepsilon_i\theta_i \quad (3)$$

The system (1) and equation (3) are combined and becomes the mathematical model to study the evolution of plant pathogens. A steady-state and stability analysis is used to describe the behavior of plant pathogen evolution. The processes are described in the next section.

III. STABILITY ANALYSIS METHOD OF THE MODEL

An important characteristic of population models is the existence of one or more equilibrium points which leads to determine the steady state of the model. The method of stability analysis of linear ordinary differential equations system was applied to the model. Since the model is a nonlinear system, the method of linearization around a steady-state was introduced to transform the model to a linear system.

An equilibrium point of a set of differential equations is a solution that does not change in time. The rate of change of population at that point is equal to zero which mean each strain in the population has a constant density. To find the equilibrium points, all the derivatives in system (1) and (3) should be equal to zero, which becomes,

$$\left.\begin{aligned}
0 &= (r_1-\bar{r})\theta_1+\varepsilon_{21}\theta_2+\varepsilon_{31}\theta_3+\varepsilon_{41}\theta_4-\varepsilon_1\theta_1\\
0 &= (r_2-\bar{r})\theta_2+\varepsilon_{12}\theta_1+\varepsilon_{32}\theta_3+\varepsilon_{42}\theta_4-\varepsilon_2\theta_2\\
0 &= (r_3-\bar{r})\theta_3+\varepsilon_{13}\theta_1+\varepsilon_{23}\theta_2+\varepsilon_{43}\theta_4-\varepsilon_3\theta_3\\
0 &= (r_4-\bar{r})\theta_4+\varepsilon_{12}\theta_1+\varepsilon_{32}\theta_3+\varepsilon_{42}\theta_4-\varepsilon_2\theta_2
\end{aligned}\right\} \quad (4)$$

Since the mutation rate is very small, it has been ignored for consideration. The mean fitness rate is a function of all population variables, so the last equation can be ignored. From system (4) there are two cases to consider. Firstly, the density of the strain is zero, secondly the difference between growth rate and the mean fitness is zero. Since there are four equations, the total number of possible cases to find equilibrium points is 16. Then, considering each case and check that the candidate of steady-state also satisfies that the summation of all fractions must be equal to 1. So, there are only 4 equilibrium points which are shown in Table 1.

Then, the nonlinear system of differential equations is transformed to a linear system by using the Linearization method, a method for finding the linear approximation of nonlinear function of a system. The method can be extended to assess the local stability of an equilibrium point of nonlinear differential equation system. For simplicity, the model is written in matrix form and using a Taylor's series expansion to linearize the system, the higher order terms are ignored and only linear terms are collected. The linearized system can be written as:

$$\frac{d}{dt}\begin{bmatrix}\theta_1\\ \theta_2\\ \theta_3\\ \theta_4\end{bmatrix} \approx \begin{bmatrix}
r_1-2r_1\theta_1^*-r_2\theta_2^*-r_3\theta_3^*-r_4\theta_4^*-\varepsilon_1 & -r_2\theta_1^*+\varepsilon_{21} & -r_3\theta_1^*+\varepsilon_{31} & -r_4\theta_1^*+\varepsilon_{41}\\
-r_1\theta_2^*+\varepsilon_{12} & r_2-r_1\theta_1^*-2r_2\theta_2^*-r_3\theta_3^*-r_4\theta_4^*-\varepsilon_2 & -r_3\theta_2^*+\varepsilon_{32} & -r_4\theta_2^*+\varepsilon_{42}\\
-r_1\theta_3^*+\varepsilon_{13} & -r_2\theta_3^*+\varepsilon_{23} & r_3-r_1\theta_1^*-r_2\theta_2^*-2r_3\theta_3^*-r_4\theta_4^*-\varepsilon_3 & -r_4\theta_3^*+\varepsilon_{43}\\
-r_1\theta_4^*+\varepsilon_{14} & -r_2\theta_4^*+\varepsilon_{24} & -r_3\theta_4^*+\varepsilon_{34} & r_4-r_1\theta_1^*-r_2\theta_2^*-r_3\theta_3^*-2r_4\theta_4^*-\varepsilon_4
\end{bmatrix}\begin{bmatrix}\theta_1\\ \theta_2\\ \theta_3\\ \theta_4\end{bmatrix}+\vec{B} \quad (5)$$

where matrix B is nonhomogeneous matrix and the coefficient matrix which multiply with variables is called Jacobian matrix which denoted by J.

After that, stability of each equilibrium point is determined by considering the eigenvalues of its Jacobian matrix. The equilibrium point is substituted in the matrix then, the eigenvalues can be found from equation (6):

$$\det(J - \lambda I) \quad (6)$$

where λ is eigenvalues of Jacobian matrix.

The row operation method is used to transform the matrix to a triangular matrix, and calculate the characteristic equation. Finally, the eigenvalues can be calculated from solving the characteristic equation. If all eigenvalues are negative, the equilibrium point is locally asymptotically stable otherwise it is unstable. The fitness rate of each pathogen strain is always positive and the fitness rate of a virulent strain always higher than that of anavirulent strain. Hence, these assumptions are used to determine the stability of each equilibrium point and the results are shown in the Table 1.

IV. RESULTS

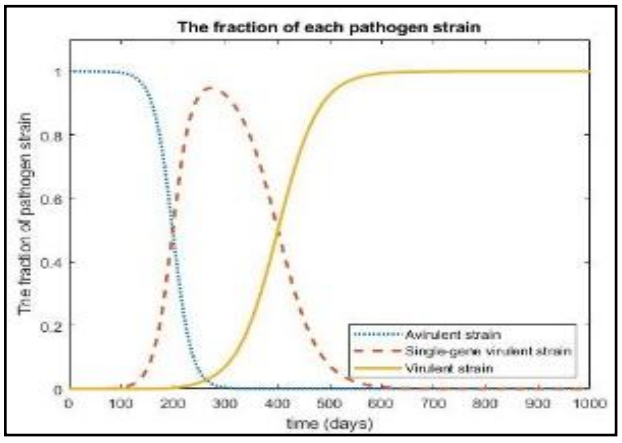
The stability analysis indicated that only one equilibrium point is stable, (0,0,0,1), while the other equilibrium points are unstable. The model is simulated using the classical Runge-Kutta method, RK4, with step size 0.01 which guarantee the stability of the numerical solution [10]. The mutation rate is defined as $10^{-6}$and 10^{-12} which depends on the number of mutant genes while the net-reproductive number and mean generation time are 20. The results show dynamic of three strains of pathogen population that is avirulent, single gene virulent to gene A and universal virulent pathogen strain. For the simulation, a single gene cultivar with resistant gene B is not considered so, the single-gene virulent to gene B strain is ignored.

For the initial condition of the system, supposing that there are three varieties, one is susceptible, the second is single-gene resistant and the third is the pyramided cultivar. These varieties are well mixed in the field. To explore the evolution of the plant pathogen strains and steady states of the model, MATLAB is used for model simulation. Next, using a susceptible cultivar fraction of 0.20, a fraction of 0.40 of both single-gene resistant gene A and pyramided cultivars. There are three main cases of simulation based on the initial pathogen strain in the field that is one, two and three types of pathogen, these three types of initial conditions are introduced to show that the model always has a stable steady-state at point (0,0,0,1).

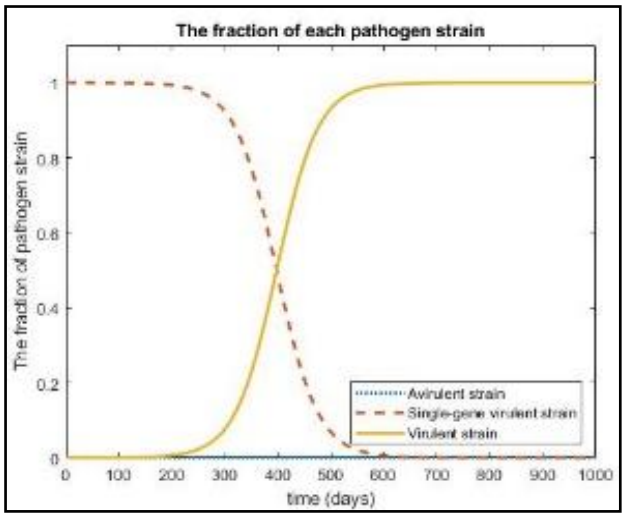
Table. 1 All four equilibrium points with their biological meaning their stability

Equilibrium point	Meaning	Stability
(0,0,0,1)	All pathogen in the field is virulent strain	Stable
(0,0,1,0)	All pathogen in the field is single-gene virulent to gene A strain	Unstable
(0,1,0,0)	All pathogen in the field is single-gene virulent to gene B strain	Unstable
(1,0,0,0)	All pathogen in the field is avirulent strain	Unstable

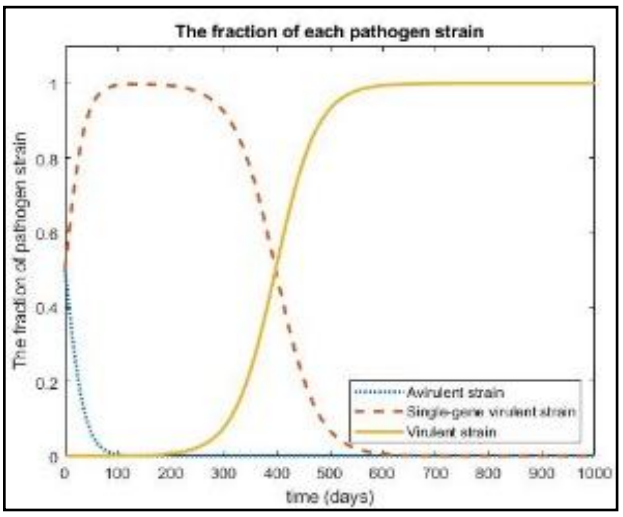
There are three cases of initial values with avirulent, single-gene virulent and both avirulent and single-gene virulent by assume fraction of each pathogen strain equal to 1 for the first two cases. For the third case, the fraction of the avirulent and single-gene stain are assumed to be 0.5. The simulation started from initial time until the system converged to steady state, the results of simulation are shown in Figure2:



(a)



(b)



(c)

Fig. 2 The fraction of each pathogen strain, avirulent (a), single-gene virulent (b), avirulent and single-gene virulent (c)

In Fig. 2 (a), all pathogens are avirulent at the start of the simulation then, due to the virulent strains emerging through mutations, the density of the avirulent strain starts to dramatically decrease after around 100 days and decrease to zero, meaning they cannot survive in the field compared to other strains. The single-gene virulent strains increase gradually at same time as the avirulent strain decreased. The peak value of the density of single resistant strains is almost 1 after which they start to decrease with the increase of the universal virulent strain. At the end of simulation, only the universal virulent pathogen strain remained in the field. This agrees with the stability analysis shown above. Using different initial conditions, single-gene virulent strain, Fig.2 (b), dominate in the field for 300 days before decreased and disappeared with lower rate when compared to the case of avirulent. Next, assuming that the fraction of the avirulent strain is 0.5, and the fraction of the single virulent strain is 0.5.in Fig. 2 (c). The avirulent strain dropped immediately after the start of the simulation with nearly the same rate of increasing of single-gene virulent pathogen strain. the single virulent strain is virtually equal to 1 day100 today 300 and then started to decrease when the universal virulent appeared to the field.

From all three cases of simulation the system eventually converged to the steady state which the entire pathogen population in the field of the universal virulent strain. In the cases where initially there is no universal virulent strain, mutations guaranteed that this strain eventually emerged in the population. In mathematical term, the steady state of the system occurred at the equilibrium point (0,0,0,1) which mean that all the entire pathogen population is of the universal virulent strain. Both interpretation in biological and mathematical meanings were similar [11].

V. CONCLUSIONS

Gene resistance is an effective and environmentally friendly way to protect crops but its durability is limited as the pathogen can gain virulence which breaking the resistance. Since obtaining experimental data is time and resource heavy, mathematical modeling plays the important roles to explore the behavior of pathogen evolution.

The stability analysis is used to investigate the steady states of the system. The only equilibrium point that is stable is that where the entire pathogen population is made up of only the universally virulent strain. In the biological words, avirulent strains cannot live in resistant field then, they take time to mutate and become more virulent strains. Finally, the most virulent strain will take over the field and dominate and the weaker strain will disappear.

In conclusion, the multiple strains model with evolution can be used to describe the behavior of pathogen evolution since the results of stability analysis is reasonable and related to biological meanings. In the future, other evolutionary forces of evolution should be included to the model to improve and give more reasonable results.

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