Analyses of the pathogen and weather components of disease progress for modeling apple scab epidemics in integrated and organic production systems

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Summary: The pathogen and weather components of apple scab disease progress were analysed in a three-year study, in two environmental-friendly production systems (organic and integrated) on cvs. 'Idared', 'Jonica' and 'Mutsu'. Linear regression analyses of transformed disease incidence and severity data and "area under the disease progress curves" (AUDPC) were used for the analysis of the pathogen component. To evaluate the role of the weather component in apple scab epidemic, first, the weekly disease increase was determined at a certain week (*n*). Weekly disease increase was related to rainfall, relative humidity, Mills' wetness period, temperature and interaction between temperature and relative humidity. Five different periods were used in the analyses: i) week (*n*-1), ii) week *n*(*n*-1), iii) week (*n*-2), iv) week (*n*-1)(*n*-2) and v) week *n*(*n*-1)(*n*-2). In the analyses of the pathogen component, the best transformation function was the logistic one. Regression analyses showed that disease growth rates were higher for disease incidence and for the organic production system than for disease severity and for the integrated production system, respectively. Disease growth rates for leaf incidence were higher than fruit incidence on all the three cultivars. AUDPC values showed great differences in both leaf and fruit incidences among cultivars and between the two production systems. The results the of analyses of the weather component showed that the best relationships between disease increase and weather parameters were found for fruit incidence and leaf incidence in week (*n*-2) in the organic and integrated production systems, respectively. Results also demonstrated that in week *n*(*n*-1) temperature played a more important role in the fungus development than the water parameters (relative humidity, rainfall and leaf wetness). Consequently, infection process is significantly dependent on almost all weather parameters, but during the incubation period the most important weather parameter is the temperature. Results were compared with similar studies and biological interpretations of the analyses are discussed.

Key words: apple scab, weather parameters, mathematical functions, epidemiology, regression analyses, integrated and organic apple production

Introduction

Apple scab, caused by *Venturia inaequalis* (Cooke) G. Wint., attacks shoot, leaf and fruit, which results in fruit quality and yield loss in every year under wet weather conditions. According to its importance, many fungicide programs have been developed and more or less effectively used (*Sutton*, 1996). The extensive use of pesticides has been causing pollution of the environment and pesticide residues in the harvested crop. Therefore, environmental considerations have become increasingly significant and most European countries have changed their plant protection strategy. For these new strategies, guidelines have been established, and new control methods are applied in integrated and organic fruit production systems (*Vályi* et al., 1985, 1986; *Anonymus*, 1989; *Dickler*, 1990; *Dickler* & *Schäfermeyer*, 1991; *El Titi* et al., 1993; *Cross* & *Dickler*, 1994; *Miklay*, 1995; *Avilla*, 1996; and *Gonda*, 1995, 2000).

Although many factors have been investigated which affect apple scab infection (*Hirst* & *Stedman*, 1963; *Sutton* et al., 1976; *MacHardy*, 1996), there is little definitive documentation on the pathogen and the weather components of apple scab disease progress in environmentally-benign apple production systems. Consequently, such information is needed in order to design improved approaches to control scab in such growing systems.

In the case of the pathogen component, one method to investigate disease progress is that disease incidence and severity are assessed repeatedly in orchards during the growing season and disease progress curves are constructed. Such information can be quantified using mathematical models to summarize disease progress (*Rouse*, 1985; *Madden*, 1986; *Campbell* & *Madden*, 1990). Disease progress curves describing the seasonal development of apple scab have been constructed (*Analytis*, 1973; *Jeger*, 1981, 1984) and few studies have been made to quantify the structural elements of these curves (*Holb*, 2000, 2003; *Holb* et al., 2003). However, no attempt has been made to create multivariate analyses for disease progress data in order to model epidemics of apple scab.

In the case of the weather component, it has been known for a long time that there is a strong relationship between the weather parameters and the seasonal development of a plant pathogen. However, weather parameters play different roles in the disease development depending on the plant pathogen (*Jeger & Butt*, 1984). In the case of apple scab, temperature, leaf wetness and relative humidity are considered to be the most important factors affecting infection and epidemic progress of the disease. The relationship between weather parameters and development of apple scab was determined by *Mills* (1944) and *Mills & LaPlante* (1951). Nowadays, scab control strategies are mainly based on the Mills' table (*Mills & LaPlante*, 1951) or the revised Mills' table (*MacHardy & Gadoury*, 1989). In the last decades, several studies demonstrated that the Mills' criteria alone can not give sufficient information for the disease control (*Sutton* et al., 1976; *Scheer*, 1980; *MacHardy*, 1994; *Trapman*, 1994, *MacHardy*, 1996). Authors concluded that besides the Mills' criteria, other weather factors could also play an important role in the disease development. They suggested that a multivariate model for all weather parameters might give more accurate information for decision-making in scab control strategies.

The aim of this study was to analyze the pathogen and weather components of apple scab; and to compute mathematical functions for modeling apple scab epidemics in integrated and organic apple production systems.

Material and method

Orchard site and experimental layout

The study was conducted in an experimental apple orchard at Debrecen-Pallag, Hungary. The one ha orchard was divided into two experimental blocks. One of the blocks was treated according to the Hungarian integrated fruit production guidelines (*Anonymous*, 1995); and the other according to the Hungarian organic production guidelines (*Anonymous*, 1997). These guidelines have been applied since 1997, when the orchard was planted. The experimental orchard consisted of forty apple cultivars. The cultivars were planted in randomised blocks with five replicates in the experimental field. Each block consisted of seven trees, but observations were only made on the middle five trees of each plot. Single trees were used as observation units. The dwarf trees grafted on M26 rootstock were planted at a distance of 4 × 1.5 m and pruned to a spindle shape. Observations were made on cvs. 'Idared', 'Jonica', and 'Mutsu'.

Disease assessment

Disease assessments were made on leaves and fruits in 2001, 2002 and 2003. For leaves, five units were chosen at random, and for each unit, one-year-old lateral twigs were selected with 50 leaves. Each unit was tagged at the beginning of May, and the total number of healthy and diseased leaves was counted at each observation date. Twenty fruits were chosen at random for each observed tree at each observation date. Assessments were made on a weekly basis from the beginning of May until mid-October.

Disease measurement

Disease incidence and severity were calculated as the mean of three-year data set. Incidence of leaf and fruit were calculated separately as the number of diseased leaves and fruits divided by the selected number of leaves and fruits, respectively. To quantify the percentage of the total disease on leaf and fruit, disease severity grades were used according to *Croxall* et al. (1952a, 1952b). With the help of severity grades, severity equations were composed in order to get the percent of leaf and fruit severity.

The pathogen component

Mathematical functions and analyses of the pathogen component. The three-year combined data were used for the data analysis. Disease incidence and severity data (dependent variable, corresponding to '*y*' axis) were linearised based on transformation functions of *Analytis* (1973), *Hau & Kranz* (1977) and *Analytis* (1979). The transformation functions were:

i) logarithmic (10 based): $z = \log(x)$,
ii) exponential transformation: $z = \ln(x)$,
iii) Gompertz transformation: $z = (-\ln (-\ln (x))$,
iv) logistic transformation: $z = \ln (x / (1-x))$,
v) monomolecular transformation: $z = \ln (1 / (1 - (x)))$,
vi) Mitscherlich and Bertalanffy transformation: $z = \ln (1 / (1 - (x_i / x_{max})^{-2})))$.

Time (independent variable, corresponding to '*x*' axis) was used without transformation. Linear regression analyses were performed for all linearised dependent variables against non-transformed independent variables. The best regression equations were selected by the following criteria:

i) constants and coefficients with reasonably small standard error;
ii) P-value < 0.1;
iii) as high R^2 (coefficient of determination) as possible.

For analysing disease progression, only one transformation function was selected, which generally gave the best result for above selection criteria. Obtained linear regression equations were used to quantify the disease growth rate (k). Growth rates were obtained from slopes of linear regression equations over time (*Berger*, 1981).

Area under the disease progress curve (AUDPC). In order to make the analyses more accurate "area under the disease progress curve" was also calculated from the data points of fruit and leaf incidence based on *Naragajan & Muralidharan* (1995):

$$AUDPC = \sum_{i=n}^{n} \frac{(x_i + x_{i-f}) \bullet d}{2}$$

where x_i = disease incidence of fruit or leaf at the end of the week 'i', f = the number of successive evaluations of disease, d = interval between two evaluations.

The weather component

Sources of weather data set. For analysing the weather component of disease progress, the following weather data were recorded and calculated:

i) daily mean temperature (°C) recorded 1 m above ground,
ii) rainfall (mm) recorded with a standard pluviometer,
iii) daily mean relative humidity – RH (%) recorded 1.5 m above ground level,
iv) Mills' infection periods in hours calculated by the Mills' table.

Analysis of the weather component. To evaluate the role of the weather component in the apple scab epidemic, first, the weekly disease increase was determined at a certain week (n). Weekly disease increase was used as a dependent variable, corresponding to 'y' axis. Weekly mean temperature, relative humidity, rainfall, Mills' infection periods and interaction between temperature and relative humidity were used as independent variables, corresponding to 'x' axis. Weekly disease increase was related to rainfall, relative humidity, Mills' infection period, temperature, interaction between temperature and relative humidity and their combinations. Five different periods were used in the analyses:

i) week (n-1),
ii) week n(n-1),
iii) week (n-2),
iv) week (n-1)(n-2),
v) week n(n-1)(n-2).

Simple and multiple regression analyses were performed to relate disease increase to several weather parameters (*Sutton* et al., 1976). The aim was to find the best regression models, which explain the role of weather parameters in relation to disease increase in integrated and organic production systems. Selection criteria of a regression model were:

i) coefficients with positive sign in single and multiple regressions;
ii) constants and coefficients with a reasonably small standard error;
iii) P-value < 0.1 for each partial coefficient;
iv) as high coefficient of determination as possible.

Linear regression analyses were performed with a Genstat 5 statistical program package as the mean of the three-year data set.

Results

The pathogen component

Regression analysis. For this three-year study, the best transformation function was the logistic one. Values of intercept, slope, mean standard errors (MSE), coefficient of determination (R^2) and level of significance (F-test) of obtained linear regression equations are in *Table 1.*

Table 1 Linear regression analyses of disease progress of apple scab for fruit and leaf incidences on cvs. 'Idared', 'Jonica' and 'Mutsu' in integrated and organic apple production systems (Debrecen – Pallag, 2001–2003)

Cultivar	Intercept[a]	Slope[b]	MSE[c]	R^2 [d]	F-test[e]
		Organic production – fruit incidence			
Idared	–1.24±0.032	–0.003±0.002	0.536	28.3	ns
Jonica	–2.33±0.045	–0.006±0.002	0.319	69.5	**
Mutsu	–3.16±0.097	–0.007±0.003	0.425	44.7	*
		Organic production – leaf incidence			
Idared	–3.22±0.136	0.046±0.004	0.167	86.8	***
Jonica	–6.54±0.278	0.037±0.007	0.137	92.9	***
Mutsu	–5.35±0.211	0.039±0.008	0.111	94.7	***
		Integrated production – leaf incidence			
Idared	–3.25±0.567	0.018±0.004	0.257	78.5	**
Jonica	–5.46±0.346	0.026±0.005	0.186	82.7	**
Mutsu	–6.22±0.667	0.029±0.008	0.211	81.3	**

[a] Intercept is the constant of linear regression equations. Values are with standard errors.
[b] Slope value is the coefficient of linear regression equations and the disease growth rate *(k)* of disease progress. Values are with standard errors.
[c] MSE = mean standard error.
[d] R^2 = coefficient of determination.
[e] F-test = *** < 0.01, ** 0.01 – 0.05, * 0.05 – 0.1, ns > 0.1.

Fruit incidence and severity in the integrated production system were not analysed because of the very low values. Results of the regression analyses for leaf severity in both production systems and fruit severity for organic production system showed that the R^2 was generally low (below 0.5) and F-test was not significant except for leaf severity on 'Mutsu' at P = 0.1 (data not shown). For disease incidence, the R^2 showed generally high values (0.8) except for fruit incidence in the organic production system (*Table 1*). Leaf incidence for cv. 'Mutsu' showed the highest value of R^2 in the organic production system. Disease growth rate (k), obtained from the slope of linear regression equations, showed the highest values of leaf incidence for cvs. 'Idared', 'Jonica' and 'Mutsu' in the organic production system. Disease growth rates of fruit incidences were low and with negative signs on all the three cultivars.

Area under the disease progress curve (AUDPC). AUDPC values for fruit incidence were very low in the integrated production system for all the three cultivars (*Table 2*). In general, AUDPC was lower in the integrated than in the organic production system. AUDPC values for both leaf and fruit incidences showed great differences among cultivars. Cultivar differences in leaf incidences were the largest in the organic production system. AUDPC values of leaf incidence for the scab susceptible cultivars ('Jonica' and 'Mutsu') were ten times higher compared to cv. 'Idared'. Similarly to leaf incidences, great difference was found in the AUDPC values of fruit incidence in the organic production system. Cultivar differences in AUDPC values were smaller in the integrated than in the organic production system.

Table 2 Area under the disease progress curve (AUDPC) of apple scab for fruit and leaf incidences on cvs. 'Idared', 'Jonica' and 'Mutsu' in organic and integrated fruit production systems (Debrecen – Pallag, 2001–2003)

Cultivars	Integrated – leaf incidence	Integrated – fruit incidence	Organic – leaf incidence	Organic – fruit incidence
Idared	327.3 b[a]	0.1 b	1,221 b	115.1 b
Jonica	487.3 a	11.2 a	2,535 a	1,161.4 a
Mutsu	528.5 a	9.20 a	2,499 a	1,169.6 a
F-test[b]	***	**	***	***
SED (df =24)[c]	33.32	3.8	376.5	102.6
$LSD_{0.05}$	70.23	8.2	765.3	210.7

[a] Values within columns followed by different letters are significantly different.
[b] F-test = *** < 0.01, ** 0.01 –0.05, * 0.05 – 0.1, ns > 0.1.
[c] SED = standard errors of differences of mean values, df = degrees of freedom.

The weather component

The analyses of five different periods showed that the best relationships were found in week (n-2) and in week n(n-1) for all the three cultivars. In the case of week (n-2), the best relationships between disease increase and weather parameters were found for fruit incidence and leaf incidence in the organic and integrated production systems, respectively (*Table 3*).

Table 3 Significant relationships between disease increase of apple scab and weather parameters in week (n–2) in integrated and organic apple production systems for cvs. 'Idared', 'Jonica' and 'Mutsu' (Debrecen – Pallag, 2001–2003)

Weather parameters[a]	Coefficient sign[b]	R^{2c}
Organic production – cv. Idared – fruit incidence		
Mills	+	0.28*[d]
RH	+	0.19*
Temp + RH	+' +'	0.25*
Temp x RH	+	0.31*
Temp x RH + Mills	+' +'	0.35*
Temp x RH + RH	+' +'	0.33*
Integrated production – cv. Idared – leaf incidence		
Mills	+	0.58**
RH	+	0.42*
RH + Mills	+' +'	0.62**
Temp x RH +Temp	+' +'	0.61**
Temp x RH +Mills	+' +'	0.68**
Organic production – cv. Jonica – fruit incidence		
Mills	+	0.35**
Temp + RH	+' +'	0.26*
Temp x RH	+	0.29*
Temp x RH + Mills	+' +'	0.41**
Integrated production - cv. Jonica - leaf incidence		
RH	+	0.51**
Mills	+	0.65**
RH + Mills	+' +'	0.72**
Temp x RH +Temp	+' +'	0.62**
Temp x RH +Mills	+' +'	0.78**
Organic prodcution – cv. Mutsu – fruit incidence		
Mills	+	0.36**
Temp + RH	+' +'	0.24*
Temp x RH	+	0.28*
Temp x RH + Mills	+' +'	0.39**
Integrated prodcution – cv. Mutsu – leaf incidence		
RH	+	0.42**
Mills	+	0.63**
RH + Mills	+' +'	0.66**
Temp x RH +Temp	+' +'	0.59**
Temp x RH +Mills	+' +'	0.81***

[a] Mills=Mills' infection periods, Temp=Temperature, RH=Relative humidity.
[b] Signs of coefficient of linear regression equations are positive "+" or negative "–".
[c] R^2 = coefficient of determination.
[d] F-test = *** < 0.01, ** 0.01 – 0.05, * 0.05 – 0.1.

The relationships were better in the integrated production system compared to the organic one. In the organic production system, Mills' infection period (Mills), relative humidity (RH), temperature (Temp), interaction between relative humidity and temperature (Temp x RH), and their combinations gave significant relationship with fruit incidence in week (*n*–2) (*Table 3*). Relatively high correlations were found when two or three single weather parameters were related to disease increase in multiple regression analyses. The best relationships were the interactions among Mills' infection period, relative humidity and temperature for cv. Jonica ($R^2 = 0.41$ at $P = 0.044$).

In the integrated production system, significant relationships were found between leaf incidence and weather parameters for all the three cultivars (*Table 3*). The best relationships were the interactions among Mills' infection period, relative humidity and temperature in the case of cv. 'Mutsu' ($R^2 = 0.81$ at $P = 0.009$).

In the case of week *n*(*n*-1), leaf incidence gave good relationships with temperature at $P \leq 0.1$ in both the organic and integrated production systems (data not shown).

Discussion

The present study analysed the pathogen and weather components of apple scab disease progress on three apple cultivars in integrated and organic apple production systems.

In the case of the pathogen component of disease progression, we suggest those equations which were presented for leaf incidence on all the three cultivars in both the integrated and organic production systems (*Table 1*). These equations can greatly help to improve the model approach of cultivar-specific disease control programmes.

The most important parameter for analyses of disease progress is the disease growth rate (*k*). It was different for the production systems, cultivars and plant organs (*Table 1*). There is no scientific information about the disease growth rate of apple scab under different fungicide treatments, but results of *Gregory* et al. (1981), *Plaut & Berger* (1981), and *Rouse* et al. (1981) supported our findings on other diseases. They found that if epidemics began from lower levels of initial disease, then early disease progress was increasingly faster or the disease growth rate was increasingly higher. In this study, disease growth rate for leaf incidence on susceptible cultivars ('Jonica' and 'Mutsu') provided permanent disease progress with a high correlation. In contrast, disease growth rate for fruit incidence decreased disease development on a fruit. The reason for this is that some of the early-infested fruits had fallen and the ontogenic resistance of fruits steadily increased during the growing season. Consequently, the percentage of diseased fruits was somewhat lower through the summer and in early autumn, compared to the spring disease level.

The "area under the disease progress curve" showed great differences between the two production systems and among the three cultivars. *Plank* (1963), *Kranz* (1974) and *Jeger & Viljanen-Rollinson* (2001) found close correlations between the disease growth rate and the AUDPC for several diseases. In this study, we did not investigate the relationship between AUDPC and disease growth rate, but AUDPC showed more clear differences between the two plant protection systems and among the susceptibility of cultivars to apple scab than the disease growth rate. Therefore, AUDPC may be a better measure for cultivar susceptibility and one of the most important factors in modelling the disease progress of apple scab.

In this study, we determined the relationship between disease increase of apple scab and weather parameters in three consecutive years on three apple cultivars. For almost all weather parameters, the strongest relationships were found in week (*n*–2), which is two weeks before the symptoms appeared. At this time, the plant pathogen is in the infection and penetration stages of its life cycle. Therefore, it can be concluded that spore dissemination, spore germination, penetration and development of the first subcuticular hyphae are significantly dependent on almost all weather parameters. Results also demonstrated that temperature played the most important role in the fungus development in week *n*(*n*–1), which is the period of 14 days from infection until the appearance of symptoms. This period is the incubation period for *V. inaequalis,* which ranges from 10 to 15 days (*Tomerlin & Jones*, 1982). Consequently, the results suggested that the most important weather parameter is the temperature during the incubation period. In general, it can be stated that the best correlations between disease progress and weather parameters were always related with Mills' infection periods. These relationships demonstrated that interaction between weather parameters played a more important role in the disease increase than a single weather parameter. Therefore, the interactions among temperature, relative humidity and Mills' infection period are the most useful in modelling the weather component of disease progression of apple scab.

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