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Influence of Quince Rootstocks on Entomosporium Leaf Spot (*Entomosporium mespili*) Susceptibility in European Pear cv. Abate Fetel

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Abstract: Entomosporium leaf spot (ELS) is caused by the fungus *Fabraea maculata* (anamorph: *Entomosporium mespili*) and affects most pear cultivars and quince rootstocks in Brazil. The aim of this study was to characterize the effect of Adams, EMA and EMC quince rootstocks on ELS in European pear cultivar "Abate Fetel" in Southern Brazil, during the 2009/2010, 2010/2011 and 2011/2012 growing season. The incidence and severity of disease was quantified weekly in 100 randomly leaves distributed in four medium-height branches per plant with eight replications. Disease progress curves of ELS were constructed and the epidemics compared according to: (1) the beginning of symptoms appearance (BSA); (2) the time to reach the maximum disease incidence and severity (TRMDI and TRMDS); (3) area under the incidence and severity disease progress curve (AUIDPC and AUSDPC). The data were analyzed by linear regression and adjusted for three empirical models: Logistic, Monomolecular and Gompertz. The Abate Fetel cultivar under all rootstocks evaluated was susceptible to *E. mespili*. However, there were significant differences in ELS intensity among rootstocks evaluated. The highest ELS intensities were observed in combinations with EMA and Adams quince rootstock. Abate Fetel cultivar grafted on EMC quince rootstock showed all epidemiological variables results significantly different when compared with EMA quince rootstock. EMC quince rootstock induced late resistance compared with the other considered rootstocks. The Logistic model was the most appropriates to describe the ELS progress of Abate Fetel cultivar under all rootstocks evaluated in the edafoclimatic conditions of Southern Brazil, during the 2009/2010, 2010/2011 and 2011/2012 growing season.

Key words: Epidemic, epidemiological models, *Entomosporium mespili*, *Pyrus communis*.

1. Introduction

In 2011/2012 pear (*Pyrus communis* L.) production in Brazil was only around 20 thousand tons. There is potential for expansion of pear cultivation, especially in Southern Brazil where climatic and soil conditions are favorable, and this could represent an opportunity for diversification of temperate climate fruit production. However, the limited availability of pear cultivars and rootstocks adapted to different regions are some of the limiting factors for economic production [1].

Worldwide, most commercial pear cultivars are grafted on to *Pyrus* or *Cydonia* rootstocks. In Brazil, most of the currently cultivated pear orchards are grafted on to *P. calleryana* (Decne.) rootstock, which supports high plant vigor but require on average six to seven years before fruit production begins [2, 3]. Plant vigor is a good measure of plant potential and is estimated by parameters including plant height, canopy volume and fertility index [4-7]. However, high vigor delays the fruit set and creates difficulties in some management practices, hindering expansion of pear cultivation [8].

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The development of quince (*C. oblonga* Mill.) rootstock clones has improved pear cultivation in Europe, USA, South Africa and Brazil, as grafted trees are characterized by less vigor and earlier fruit production [8-11]. The clonal quince rootstocks East Malling (EM) were developed in the 1920s at the East Mailing Research Station in England. These rootstocks, including EMA and EMC, are easy to propagate and have become the preferred option for pear cultivation in southern Europe [12, 13]. EMC rootstock has the lowest vigor, supporting small pear plants [12]. Adams rootstock, like EMA, is a quince clonal selection of the D'Angers type, selected in 1965 in Belgian and still widely used in Belgium, Netherland and Italy [13, 14]. According to Thompson and Morgan [15], the rootstocks EMA, Adams and EMC are considered semi-vigorous (3-4 m), semi-dwarfing (2.4-3 m) and dwarfing (2-2.4 m), respectively.

The Abate Fetel pear is one of the most important cultivars worldwide and it is being introduced into Brazil because of its high fruit quality, high planting density, heavy fruit production and long cold storage qualities [7-16]. However, the Abate Fetel cultivar is susceptible to Entomosporium leaf spot (ELS). ELS is caused by *Entomosporium mespili* (DC ex Duby) Sacc—(teleomorfo: *Fabraea maculata* Lév. G.F. Atk). The disease has a worldwide distribution, with highest incidence in Europe, Australia, Canada and USA. It is considered to be one of the most important diseases of European pears. In South America, ELS has already been reported in Paraguay and Brazil [17]. Currently, ELS is considered the main disease in pear cultivars in Southern Brazil, causing severe defoliation [17, 18].

The literature includes many references to the influence of rootstocks over important pear production parameters [3, 7, 10, 19]. However, there are no reports on the effects of quince rootstocks grafted with Abate Fetel cultivar in Southern Brazil regarding disease susceptibility. The choice of quince rootstock variety is based on plant vigor and early fruiting, not on the effects on ELS susceptibility. Therefore, it is important to ascertain the influence of quince rootstocks clones on ELS susceptibility in European pear. The objective of this study was to characterize the effect of Adams, EMA and EMC quince rootstocks on ELS in "Abate Fetel" cultivar of European pear in Southern Brazil, during the 2009/2010, 2010/2011 and 2011/2012 growing season.

2. Materials and Methods

The experiments were carried out in a commercial European pear Abate Fetel cultivar orchard located in Lages municipality, State of Santa Catarina, Southern Brazil (27°48′S, 50°19′W), at an altitude of 960 m above sea level, during the growing season of 2009/2010, 2010/2011 and 2011/2012. The climate of the region is humid mesothermic (Cfb) according to the Köppen classification. Higher precipitation occurs from October to March and during this period, the average monthly rainfall was around 167 mm. The rainfall, relative humidity and minimum, average and maximum temperature data recorded at Lages/SC municipality during the period of evaluation and were available from the Santa Catarina Hydrology and Environmental Resources Center–Epagri (Fig. 1). The soil is a cambisol with high values of clay (513 g/kg) and organic matter (75 g/kg). The orchard consisted of three year-old of European pears Abate Fetel cultivar grafted on to quince rootstocks EMA, EMC and Adams, trained to a slender spindle at a high density with a spacing of 3.0 m × 1.0 m. Trees were approximately 2.5 m tall.

The experiment followed a completely randomized design with eight replications, using five plants per experimental plot. The middle portion of each tree was assessed for the incidence and severity of ELS during the 2009/2010, 2010/2011 and 2011/2012 growing season. Under natural inoculum, the incidence and severity of ELS was evaluated weekly from the beginning of symptom appearance, random sampling of 25 young leaves per branch on four medium-height (approximately 1.25 m) branches of

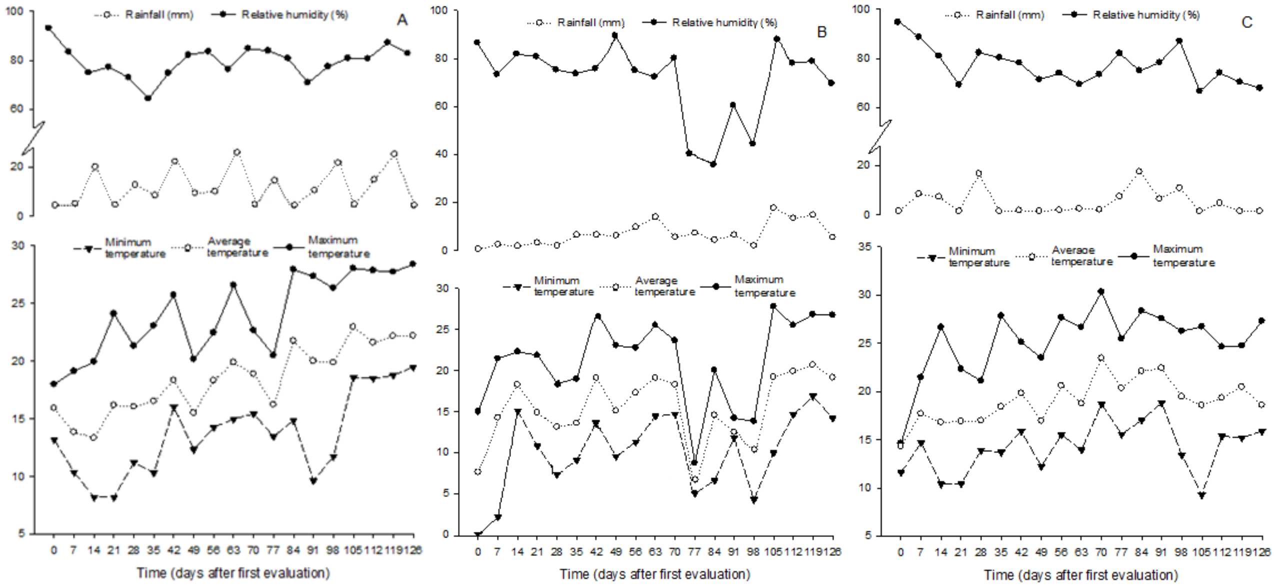

Fig. 1 Rainfall (mm/day), relative humidity (%) and minimum, average and maximum temperatures (°C) recorded at Lages/SC municipality, Southern Brazil from the first symptoms appearance of the 2009/2010 (A), 2010/2011 (B) and 2011/2012 (C) growing seasons.

each tree during nine weeks. The incidence of the ELS was defined as the number of leaves with ELS symptoms divided by the total number of leaves evaluated. The ELS severity was assigned to an infection class adapted from Ref. [20] based on the following scale: 0 (no lesions), 1 (one to five lesions), 2 (six to 25 lesions), and 3 (more than 25 lesions). Mean disease severity of each plot was calculated using the following formula:

$$S = \sum_{n=1}^{N} I_n / 3 \cdot N$$

where, S is the index of relative disease severity (from 0 to 1); I_n is the disease severity class of each *n*th leaf; N is the total number of leaves assessed; and 3 is the maximum level of severity.

The disease progress curves were constructed and the epidemics were compared in all evaluated growing seasons, using four epidemiological measures: the beginning of symptom appearance (BSA), the time to reach the maximum disease incidence and severity (TRMDI and TRMDS) and the area under the incidence and severity disease progress curve (AUDIPC and AUDSPC). The AUDPC was calculated using the following formula: AUDPC = Σ ((Yi + Yi + 1)/2)(ti + 1 – ti), where, Y = disease intensity (incidence and severity), t = time and i = number of evaluation during the time. This area represented the trapezoidal integration value of severity [21].

The incidence and severity progression data were adjusted to three empirical models, Monomolecular ($y = 1 - (1 - y_0)\exp(-rt)$), Logístico ($y = 1/(1 + ((1/y_0) - 1)\exp(-rt))$ and de Gompertz ($y = \exp(-(-\ln(y_0))\exp(-rt))$), where, y = incidence or severity (in proportion from 0 to 1) over time t, y_0 = initial disease level and r = disease increment rate for each empirical model. The quality-adjusted model was verified by the coefficient of determination value (R^2) and by the residual error [22]. The estimate parameters models selected were compared between cultivars by t test ($P < 0.05$) [23]. Statistical analysis system (SAS®) version 9.1 was used for all the data analysis.

3. Results and Discussion

The average temperature, relative humidity and sum of rainfall during September to January of 2009/2010 (18.4 °C, 79.6% and 791.3 mm), 2010/2011 (15.8 °C, 71.9% and 668.2 mm) and 2011/2012 (19.1 °C, 82.7%

and 544.7 mm) period of evaluation were 17.7 °C, 78.1% and 668.1 mm, respectively (Fig. 1). These conditions were considered favorable for the development of ELS infection because of the humid temperate climate and frequent spring rains. The favorable environmental conditions for *E. mespili* infection are temperatures between 14 °C and 28 °C with optimum of 20 °C, and rainfall above 30 mm [24].

The first symptoms of ELS were observed in the first half of October in the 2009/2010 and 2011/2012 growing season and in the first half of November in the 2010/2011 (Table 1). The interval of the beginning of symptoms occurrence and greatest ELS intensity can be explained by the different climatic conditions observed in 2009/2010 and 2011/2012 growing season when compared with 2010/2011. The later occurrence of symptoms in 2010/2011 was probably due to the low temperature (minimum, average and maximum), low relative humidity and low rainfall of the growing season, when compared with the climatic conditions of the 2009/2010 and 2011/2012 growing season (Fig.1). All combinations of Abate Fetel cultivar on quince rootstocks Adams, EMA and EMC were susceptible to ELS. However, there were significant differences in susceptibility between quince rootstocks (Table 1).

The epidemiological variables are presented in Table 2. The frequency of infected leaves on most of the Abate Fetel/rootstock combinations was generally high, indicating a lack of immunity. The principal variable utilized to differentiate the susceptibility of rootstock combinations is the disease progress curve. The frequencies of infected leaves on most of the Abate Fetel combinations grafted trees were generally high, indicating a lack of immunity. The shapes of the progress curves for ELS incidence and severity for all combinations were similar in all evaluated growing seasons. The incidence of ELS was greater for the Abate Fetel/EMA combination than for the Abate Fetel/Adams and Abate Fetel/EMC in all growing seasons (Fig. 2). There was no significant difference between the Abate Fetel/Adams and Abate Fetel/EMC combinations (Fig. 2). The incidence and severity curves

Table 1 Beginning of symptom appearance (BSA), time to reach the maximum disease incidence and severity (TRMDI and TRMDS), and area under the incidence and severity disease progress curve (AUIDPC and AUSDPC) of the Entomosporium leaf spot in Abate Fetel cultivar grafted under quince rootstocks EMA, Adams and EMC (cultivar/rootstock) during 2009/2010, 2010/2011 and 2011/2012 growing seasons in Lages/SC municipality, Southern Brazil.

Growing seasons	BSA (days)	TRMDI (days)	TRMDS (days)	AUIDPC[1]	AUSDPC[1]
2009/2010					
Abate Fetel—Quince EMA	15.7 b*	47.2 b	39.3 b	789.2 a	23.1 a
Abate Fetel—Quince Adams	21.0 b	61.2 a	49.0 b	484.3 b	9.88 b
Abate Fetel—Quince EMC	32.3 a	69.1 a	58.6 a	246.7 c	4.49 c
C.V. (%)	22.3	18.5	19.1	25.8	280
Means	23	59.2	49	506.7	12.5
2010/2011					
Abate Fetel—Quince EMA	19.4 b	42.2 b	41.3 b	769.8 a	17.7 a
Abate Fetel—Quince Adams	23.3 b	60.9 a	47.5 b	437.5 b	7.42 b
Abate Fetel—Quince EMC	29.7 a	66.7 a	55.9 a	277.6 c	3.94 c
C.V.(%)	20.9	24.1	20.3	28.6	22.0
Means	24.1	56.6	48.23	494.96	9.68
2011/2012					
Abate Fetel—Quince EMA	17.5 b	51.6 b	41.1 b	680.0 a	11.5 a
Abate Fetel—Quince Adams	25.5 b	63.0 a	63.8 a	355.4 b	7.57 b
Abate Fetel—Quince EMC	30.6 a	65.6 a	69.1 a	203.5 c	4.29 c
C.V. (%)	21.3	17.1	11	24.1	26.6
Means	24.5	60	58	412.3	7.85

[1] Area calculated by trapezoidal integration value according to Campbell and Madden (1990);
[2] Coefficient of variation; *Means followed by the same letter in the same column are not significantly different (*t*-test, $P < 0.05$).

Table 2 Residual error (Error) and coefficient of determination (R^2) adjusted by Monomolecular, Logistic and Gompertz models to the Entomosporium leaf spot incidence and severity in Abate Fetel cultivar grafted under rootstocks EMA, Adams and EMC (cultivar/rootstock) during 2009/2010, 2010/2011 and 2011/2012 growing seasons in Lages/SC municipality, Southern Brazil.

Cultivar/rootstock	Monomolecular[1]		Logistic[2]		Gompertz[3]	
	Error	R^2	Error	R^2	Error	R^2
Incidence 2009/2010						
Abate Fetel-EMA	1.08	0.98	3.02	0.87	1.01	0.98
Abate Fetel-Adams	3.43	0.77	0.17	0.99	2.46	0.88
Abate Fetel-EMC	2.62	0.82	0.09	0.99	1.91	0.90
Incidence 2010/2011						
Abate Fetel-EMA	2.72	0.89	0.19	0.99	1.79	0.95
Abate Fetel-Adams	3.36	0.75	0.16	0.99	2.32	0.88
Abate Fetel-EMC	2.24	0.81	0.06	0.99	1.63	0.90
Severity 2009/2010						
Abate Fetel-EMA	0.45	0.86	0.01	1.00	0.36	0.90
Abate Fetel-Adams	0.35	0.86	0.003	1.00	0.28	0.91
Abate Fetel-EMC	0.45	0.66	0.001	1.00	0.37	0.77
Severity 2010/2011						
Abate Fetel-EMA	1.10	0.73	0.010	0.99	0.85	0.83
Abate Fetel-Adams	0.37	0.86	0.003	1.00	0.30	0.91
Abate Fetel-EMC	0.30	0.77	0.001	1.00	0.25	0.84
Incidence 2011/2012						
Abate Fetel-EMA	1.77	0.81	0.23	0.93	1.71	0.94
Abate Fetel-Adams	2.61	0.95	0.19	0.90	2.66	0.98
Abate Fetel-EMC	2.04	0.79	0.11	0.97	1.21	0.83
Severity 2011/2012						
Abate Fetel-EMA	0.93	0.65	0.014	1.00	0.52	0.93
Abate Fetel-Adams	0.52	0.82	0.002	0.99	0.36	0.85
Abate Fetel-EMC	0.38	0.69	0.003	1.00	0.31	0.96

[1]Monomolecular $y = 1 - (1 - y_0)\exp(-rt)$, [2]Logistic $y = 1/(1 + ((1/y_0) - 1)\exp(-rt)$ and [3]Gompertz $y = \exp(-(-\ln(y_0))\exp(-rt)$, where y = incidence or severity in proportion of 0 to 1 in time t and, y_0 = initial level of disease and r = disease increment rate for each empirical model.

showed a significant increase from the 10th day after first evaluation (DAFE) which was associated with the climatic conditions and pre-existing inoculum (initial inoculum) and infection rate. The expression of the disease incidence and severity through an epidemiological variable quantification is very important to describe the disease progress the relationship with the climatic condition, the forecasts model validation, and the integrate management application [21].

The Abate Fetel/EMA combination showed significant differences in the timing of the symptoms appearance (BSA) in all three evaluated growing seasons when compared with the Abate Fetel/Adams and Abate Fetel/EMC combinations (Table 1). In general, the BSA in Abate Fetel/EMA combination started between 15-19th DAFE, while in Abate Fetel/Adams and Abate Fetel/EMC started between 21-25th and 29-32th DAFE in all three growing season evaluated, respectively. However, there was no significant difference between Abate Fetel/Adams and Abate Fetel/EMA in all three growing season evaluated.

The time to reach the maximum disease intensity and severity (TRMDI and TRMDS), and area underthe incidence and severity disease progress curve (AUIDPC and AUSDPC) exhibited significant differences between the Abate Fetel/EMA combination and ELS susceptibility in all three

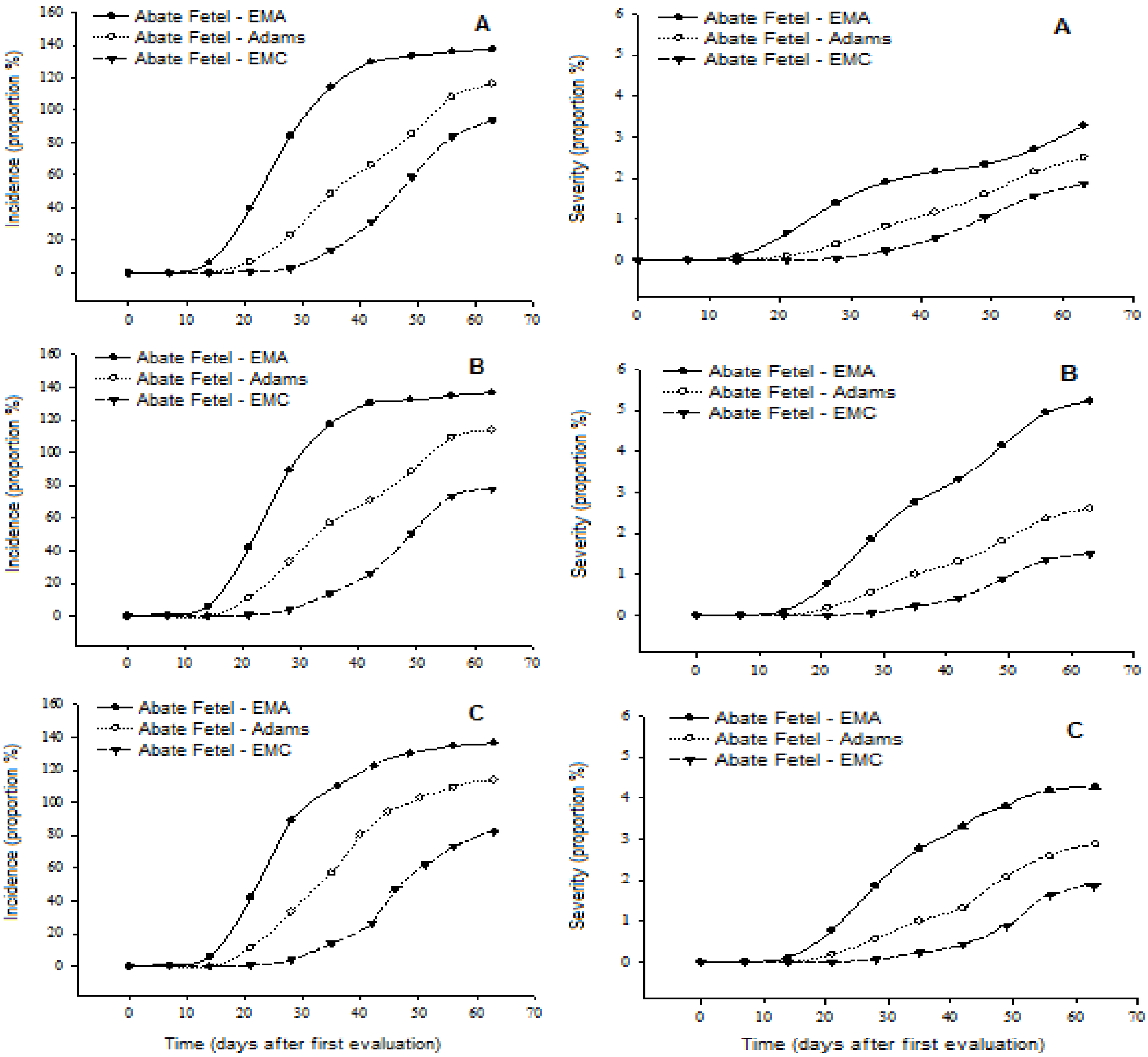

Fig. 2 Progress curve of the incidence and severity of Entomosporium leaf spot for each combination of cultivar and rootstock evaluated during 2009/2010 (A), 2010/2011 (B) and 2011/2012 (C) growing season in Lages/SC municipality, Southern Brazil.

The time to reach the maximum disease intensity and severity (TRMDI and TRMDS), and area under the incidence and severity disease progress curve (AUIDPC and AUSDPC) exhibited significant differences between the Abate Fetel/EMA combination and ELS susceptibility in all three growing season evaluated (Table 1). The temperature associated with rainfall directly influenced the ELS intensity (Fig. 2). The temperatures were higher in the 2009/2010 and 2011/2012 growing season than in the 2010/2011 growing season (Fig. 1). This situation might be the principal factor in the anticipation of the BSA, TRMDI and TRMDS and the higher AUIDPC and AUSDPC in the susceptible combinations of Abate Fetel/EMA and Abate Fetel/Adams.

The AUIDPC and AUSDPC of the ELS in Abate Fetel/EMA were also significantly higher when compared with others combinations in all growing seasons evaluated (Table 1). These data could be associated with many morphological features induced by EMA rootstock in the Abate Fetel cultivar including high vigor [25] and high canopy growth [6, 7]. In the Refs. [15] and [25], the EMA rootstock is considered more vigorous than Adams (10% to 20%) and EMC (~30%) [12]. These features could contribute to the high ELS intensity, characterized by the highest values of the epidemiological variables BSA, TRMDI, TRMDS, AUIDPC and AUSDPC (Table 1). Additionally, the EMA quince rootstock is less precocious than Adams and EMC quince rootstocks [26], induces high productive efficiency and adapted to high plant density of 1,500 to 2,000 plants/ha [12-27]. These features, together with high plant density adaptation can promote the emergence of favorable microclimates for pathogen infection and increase the development of many diseases in annual and perennial crops [28, 29]. Also, the dissemination and multiplication rate of the pathogens is related to the distance, accessibility and/or connectivity between resource organs, (i.e., direct contacts between individual plants) as well as other physiological parameters that are also affected by plant density [29]. High humidity and temperatures between 20 °C to 25 °C are considerated favorable for the development of ELS infection [23]. Therefore, the internal canopy plant microclimate is the most important factor in the increase in disease severity [30].

The Adams and EMC quince rootstocks can be good options for grafting of the Abate Fetel cultivar because they showed a significant difference among TRMDI, TRMDS, AUIDPC and AUSDPC when compared with EMA quince rootstock. The TRMDI, TRMDS, AUIDPC and AUSDPC are considerated consistent epidemiological variable for evaluation of cultivars diseases susceptibility [18]. However, the choice between Adams and EMC quince rootstocks will depend on many regional and economic factors associated with plant vigor, early production, fruit productivity, productive efficiency [25], fruit set, fruit weight [31, 32], propagating facility, relation between rot system and soil depth and plant density [33].

An appropriate empirical model which describes disease progress curves can be selected from analysis of disease progression through time. The selection used in this study aimed to estimate variables that can be used to compare epidemics [22]. The logistic model allowed the best data adjustment based on the coefficient of determination (R^2) and residual error (Table 2). According to the this model, the velocity of disease increase was proportional to the amount of pre-existing inoculum (initial inoculum) and infection rate [34] and might well be utilized for polycyclic diseases pathosystems [22]. However, the logistic model adjustment for the disease severity was justified because the field severity rate was quantified from healthy leaves inside of area with high disease incidence (Fig. 1). In this case, the high inoculum determined the model adjustment which is one of the Logistic model characteristic.

The velocity of the ELS development was different in the Abate Fetel cultivar grafted in all rootstocks evaluated and consequently the ELS susceptibility degree was also different. Based on the disease progress rate (r), the disease development was faster in the Abate Fetel grafted in EMA quince rootstock than in all others rootstocks combinations evaluated. The Abate Fetel/EMC combination was the most resistant to ELS when the BSA, TAMID, TAMSD, AACPID and AACPSD was taken into account as differentiated variable.

The favorable climatic and biological conditions for disease development and pathogen infection are very important and fundamental for disease control strategies. The epidemic development of ELS within the growth cycle of the pear is determined by the initial amount of ELS and the rate at which the ELS increase, described by the apparent infection rate. To reduce the severity of ELS produced by such polycyclic disease, the reproductive rate of the pathogen must be decreased by chemical control.

4. Conclusions

The EMA quince rootstock induces greater ELS susceptibility in Abate Fetel cultivar when compared with Adams and EMC quince rootstocks. The EMC quince rootstock induced late resistance to Abate Fetel cultivar compared with the other considerated rootstocks when BSA, TAMID, TAMSD, AACPID and AACPSD were taken into account as differentiated variable. The logistic epidemiological model was the most appropriates to describe the ELS progress on Abate Fetel cultivar under quince Adams, EMA and EMC rootstocks in the edafoclimatic conditions of Southern Brazil, during the period of September to January of 2009/10, 2010/11 and 2011/12 growing season.

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