# COMPARISON AND VALIDATION OF DIAGRAMMATIC SCALES FOR BROWN EYE SPOTS IN COFFEE TREE LEAVES

Comparação e validação de escalas diagramáticas para cercosporiose em folhas de cafeeiro

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#### ABSTRACT

A diagrammatic scale with six levels (0.1-3.0; 3.1-6.0; 6.1-12.0; 12.1-18.0;18.1-30.0; 30.1-49.0%) was developed, compared, and evaluated along with two other scales to measure the severity of brown eye spots in coffee trees leaves. The scale was designed based on two others already in use in order to increase the efficiency of evaluation and for estimation values to approach as close as possible to their actual values. Two evaluations were performed using each of the three diagrammatic scales and one was performed without a diagrammatic scale, in seven day intervals. Using the proposed scale, the evaluators demonstrated better precision levels, accuracy, reproducibility, and repeatability in the estimations, when compared to the evaluators who did not use the diagrammatic scale, or who used existing scales. The proposed diagrammatic scale provided a reliable estimation to evaluate brown eye spot severity on coffee tree leaves.

Index terms: Cercospora coffeicola, pathometry, Coffea arabica.

#### **RESUMO**

Foi desenvolvida, comparada e avaliada com outras duas escalas uma escala diagramática com seis níveis (0,1-3,0; 3,1-6,0; 6,1-12,0; 12,1-18,0; 18,1-30,0; 30,1-49,0%), para medir a severidade da cercosporiose em folhas de cafeeiro. A escala foi construída baseada em outras duas já existentes, procurando aumentar a eficiência da avaliação e aproximar os valores estimados o mais próximo dos valores reais. Foram realizadas duas avaliações com a utilização de cada uma das três escalas diagramáticas e uma avaliação sem o seu auxílio, em intervalos de sete dias. Com o emprego da escala proposta, os avaliadores apresentaram melhores níveis de precisão, acurácia, reprodutibilidade e repetibilidade nas estimativas, quando comparados aos avaliadores que não utilizaram a escala diagramática, ou que utilizaram as escalas existentes. A escala diagramática proposta demonstrou fornecer uma estimativa confiável para avaliar a severidade da cercosporiose em folhas de cafeeiro.

Termos para indexação: Cercospora coffeicola, patometria, Coffea arabica.

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## INTRODUCTION

Brown eye spots, in which the etiological agent is the necrotrophic fungi *Cercospora coffeicola* Berkeley and Cooke, is found in the majority of coffee (*Coffea arabica* L.) growing regions of Brazil, causing injuries in the leaves and fruits (TALAMINI et al., 2003; SANTOS et al., 2008; PEREIRA et al., 2011). A period of foliar wetness from 6-12am is favorable to the progress of the disease (FERNANDES et al., 1991) and the optimum temperature for the growth and germination of the fungi is 24 to 30 °C (ECHANDI, 1959). Coffee farms which are established in environments of extreme insolation can present greater disease severity, because the brown eye spot fungi type produces the cercosporina toxin, activated by light (DAUB et al., 2005). The disease symptoms in leaves are circular in format, with a dark brown colored spot encircled by a light

yellow halo, sometimes with concentric rings. In response to the presence of brown eye spots on the leaves, the plant produces ethylene which can cause intense defoliation, creating quantitative losses, reducing the yield and productivity of the culture, even in reduced severity. Because of this it becomes extremely important to evaluate the intensity of the disease, with great accuracy and precision, even when it occurs in short intervals and with low amounts of severity.

The evaluation of brown eye spots in coffee tree leaves has been carried out with the aid of two diagrammatic scales, the Fernandes scale (1988) and the Oliveira et al. (2001) scale. Based on these two scales, improvements were made with the addition of real photographs and shorter intervals, and the consideration of both the superior and inferior limits of the disease, which must correspond, respectively, to the

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maximum and minimum amount of the disease found in the field (NUTTER JUNIOR; SCHULTZ, 1995). The representation of the symptoms must also be considered, and must be as close as possible to those observed in the plant at the various levels of disease severity. Therefore, the limits of visual acuity must be considered, following the Weber-Fechner law of stimulus, as described in previous work on scale elaboration (BELASQUE JUNIOR et al., 2005; ANGELOTTI et al., 2008; SALGADO et al., 2009; SUSSEL et al., 2009).

In this way, the main objective of the present work was to elaborate upon, compare, and validate a diagrammatic scale to perfect the evaluation of the severity of brown eye spots in coffee tree leaves.

## MATERIAL AND METHODS

Coffee leaves with disease symptoms in different levels of severity have been collected in the field, in naturally infected plants and as well as in vegetation houses, from plants inoculated with *C. coffeicola*. The leaves have been photographed with a deep white background, using a digital camera. With the aid of the Image Tool® 2002 program, the percentage of necrotic area of each photographed leaf was determined. Based on the visual acuity law of Weber-Fechner, and on the maximum and minimum levels of severity observed, a diagrammatic scale with six levels of severity was developed, considering the form and distribution of the observed lesions more frequently. After establishing the percentages of the disease to be represented in the scale, real leaf images were used that had the presence of lesions of brown eye spots in the creation of the scale.

In the validation and comparison of the scale, 50 images of coffee tree leaves with different levels of severity (SPÓSITO et al., 2004; MICHEREFF et al., 2006; ANGELOTTI et al., 2008) of brown eye spots, were surveyed by seven evaluators without experience in quantification of diseases. Each evaluator received a copy of each diagrammatic scale to estimate the severity of the disease. These images have been inserted randomly in individual slides for visualization in Microsoft® PowerPoint® 2000. In the first evaluation, seven evaluators attributed notes to the presented leaves, without the aid of the scale. After seven days, the evaluators conducted the second evaluation, where they used one of the diagrammatic scales for the first time. To evaluate the estimated repeatability of this scale, a second evaluation was carried out seven days later, with the same evaluators. This procedure was continued with all three diagrammatic scales totaling seven evaluations, in the following order: Oliveira et al. (2001), Fernandes (1988) and the new scale.

The accuracy and precision of each evaluator was determined through linear regression analysis, with real

severity achieved with the aid of the Image tool® program v 3.0. The accuracy of estimation of each evaluator, as well as the group of evaluators, was determined by test t applied to the intercept of linear regression (a), to verify the hypothesis Ho: b = 1 (y= a+bx), and to the angular coefficient of the straight line (b), to test the hypothesis Ho: b = 1 (y = a + bx), to 5% probability level (P=0.05). Intercept values significantly different from 0 (zero) indicate overestimation (>0) or underestimation (<0) of the actual severity at low levels of disease intensity. Although the values of the angular coefficient of the straight line that deviate significantly from 1 (one), indicate overestimated (>1) or underestimated (<1) surveying of the actual severity in all the intensity levels of the disease (NUTTER JUNIOR; SCHULTZ, 1995). The estimated repeatability by the same evaluator was determined using these same parameters obtained in regression where the first evaluation was compared to the second (NUTTER JUNIOR, et al., 1993). The precision of estimation was determined by the coefficient of determination of the regression (R<sup>2</sup>), by the variance of absolute errors and by estimate repeatability, determined by the regression of the second evaluation compared to the first same sample

The reproducibility of the scale was evaluated using the coefficient of determination of the linear regressions between the estimated severities of the different evaluators, combined in pairs (NUTTER JUNIOR et al., 1993; NUTTER JUNIOR; SCHULTZ, 1995). The regression analyses were accomplished using the SAS v 8.1 program and the other calculations in Microsoft® Excel® 2000.

## RESULTS AND DISCUSSION

The maximum value of severity of brown eye spots in coffee tree leaves observed in the field was 49.0%, due to the coalescence of lesions and the minimum of 0.2%. The diagrammatic scale was elaborated with six levels of severity to quantify brown eye spots in coffee tree leaves, with the intervals of 0.1-3.0; 3.1-6.0; 6.1-12.0; 12.1-18.0; 18.1-30.0; 30.1-49.0% of diseased leaf area (Figure 1). Values of severity higher than 49.0% have not been included in the scale, because they have not been observed in the field due to leaf senescence. In the diagrammatic scale developed by Fernandes (1988), values of 6, 12, 18, 25 and 50% were used for different leaf wetness sizes (10, 20, 30, 40 cm<sup>2</sup>). However, since the scale was represented by figures, it was not validated and the intermediate severity levels of the disease which must be in accordance with the limitation of human visual acuity, as defined by the "Weber-Fechner law of stimulus" were not followed.

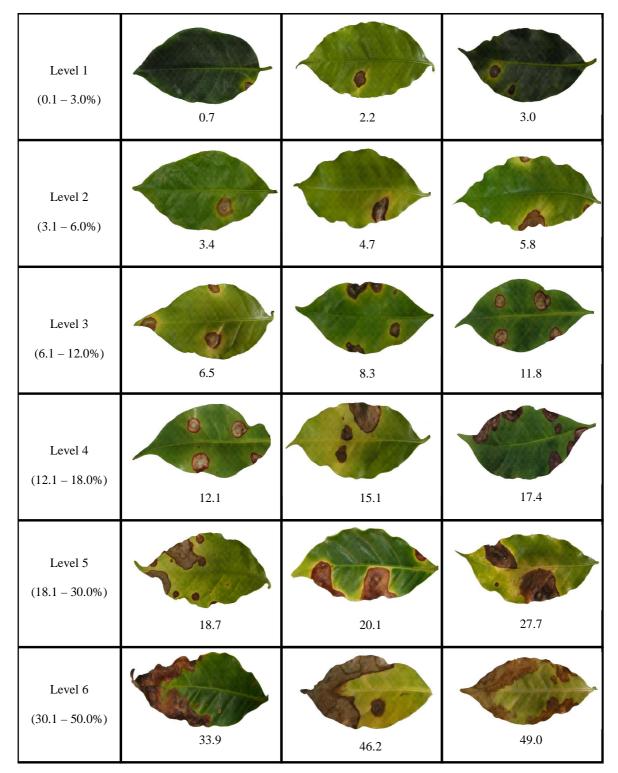


Figure 1 – Diagrammatic scale of the severity of brown eye spot (*Cercospora coffeicola*) in coffee tree leaves (*Coffea arabica* L.).

Oliveira et al. (2001) considered percentage intervals obeying this law, using severity values of 0, 3-6, 6-12 and 12-25%. The scale was validated, however it does not present all the severity levels of brown eye spots in coffee tree leaves and presents great intervals of percentage of the disease severity.

It was apparent that with the use of the diagrammatic scale, all the evaluators provided more accurate values of intercept equal zero for the straight line regression between actual and estimated severity. Without the use of the scale, generalized overestimation of the brown eye spot severity occurred. The angular coefficient of the straight line did not differ significantly from 1 in six of the seven evaluators (Table 1A, B and C). For the Fernandes (1988) and Oliveira et al. (2001) scales, the values of the angular coefficient were often significantly different by 1, indicating the presence of systematic deviation in all the intensity disease levels, with a tendency to overestimate (Table 1B). It is different for the proposed diagrammatic scale though, as only two evaluators (28.6%) presented the angular coefficient of the straight line values significantly different from 1 (Table 1C).

The overestimation of disease severity levels without the use of the diagrammatic scales, as verified for brown eye spots in coffee tree leaves, has also been reported by some authors in other pathosystems (SPÓSITO et al., 2004; MICHEREFF et al., 2006; SALGADO et al., 2009). Also, the propensity of the evaluators to overestimate the severity of the disease with the use of the diagrammatic scale resembles effects noticed in studies involving the validation of diagrammatic scales (LEITE; AMORIM, 2002; SPÓSITO et al., 2004).

The precision of estimation without the aid of the scale was bigger when compared to the diagrammatic scales of Fernandes (1988) and Oliveira et al. (2004), with  $(R^2)$  the coefficient of determination between 0.60 to 0.89 and average of 0.79 (Table 1A, B).

The distribution of residue assessments carried out without the aid of the diagrammatic scale presented estimates with absolute errors varying between 18.16 and 19.22 (Figure 2A). Using the scale of Fernandes (1988) and Oliveira et al. (2001), all the evaluators had low precision in estimates of R² with the average between the two evaluations of 0.75 and 0.68, respectively (Table 1A, B). The coefficient of determination varied between 0.38 and 0.94 for the evaluations with the scale of Fernandes (1988), with an average of 0.76 and 0.74 respectively for the first and the second evaluation. Using the scale of Oliveira et al. (2001), the coefficient of determination varied between 0.08 and 0.89, with averages of 0.66 and 0.70 respectively for the first and second evaluations. However, the evaluators improved the precision of their estimations with the use of the proposed scale, with coefficient

of determination varying between 0.76 the 0.95 for the first evaluation (Table 1C), and between 0.72 and 0.95 for the second evaluation, with averages of 0.87 and 0.85 respectively approaching the R<sup>2</sup> values of the validation of other scales (HALFELD-VIEIRA; NECHET, 2006; SUSSEL et al., 2009).

There was a decrease in the absolute error of the estimations with the aid of the diagrammatic scale proposed, where the points of residuals were well distributed throughout when compared to the values of absolute error of obtained estimates without the use of the proposed scale (Figure 2). With the use of the proposed diagrammatic scale, the absolute error did not present the tendency pattern, or have the propensity, to overestimate or subestimate the severity, and the same ones varied on the first and second evaluation when using the proposed scale between -14.11 and 14.07, and -19.11 and 16.31, respectively (Figure 2). The diagrammatic scales of Fernandes (1988) and Oliveira et al. (2001) also reduced the values of absolute errors of the estimates when compared to the estimates without the use of the scale.

However, in both scales and both repetitions, for severity values above 40%, the values of the absolute errors were higher than in other levels and when compared to the proposed diagrammatic scale, with subestimation occurring constantly.

The absolute errors of the estimates for the diagrammatic scale of Fernandes (1988) varied in the first and the second evaluation between -24.11 and 14.22, and -24.11 and 20.22, respectively. Yet for the scale of Oliveira et al. (2001) the absolute errors of the estimates varied on the first and the second evaluation between -29.11 and 14.19, and -29.11 and 12.89, respectively (Figure 2).

The majority of the absolute errors of the evaluators who used the proposed scale was less than 10%, and was considered good according to the criteria used in some validation studies of diagrammatic scales (MICHEREFF et al., 2006; SUSSEL et al., 2009), however this can be improved with the training of the evaluators (NUTTER JUNIOR; SCHULTZ, 1995; ANGELOTTI et al., 2008).

The evaluators presented good repeatability on the estimates of severity of brown eye spots in coffee tree leaves using the proposed diagrammatic scale, because the average amount of variation in the first explained evaluation compared to the second evaluation was of 90% (Table 2C). Between the two evaluations, only for one evaluator (6) the value of the intercept was not significantly different from zero, due to the tendency of evaluators to either overestimate or to underestimate the severity in the second of the proposed scales. However, in no evaluators were the values of the angular coefficient of the straight line significantly different from 1 (Table 2C).

Table 1 – Intercept (a), angular coefficient of the straight line (b) coefficient of determination ( $R^2$ ) of the equations of simple regression line relating visual estimation of the brown eye spot ( $Cercospora\ coffeicola$ ) of coffee tree leaves ( $Coffea\ arabica\ L$ .) to the actual severity, without the aid of the diagrammatic scale: (A) of Fernandes (1988), (B) of Oliveira et al. (2001) e (C) proposed scale, using the program Image Tool®.

# A – Scale of Fernandes (1988)

	11	Without Scale -			With Scale							
Evaluator	V				st evaluation	n	2 <sup>nd</sup> evaluation					
	а	b	$R^{2**}$	а	b	$R^{2^{**}}$	а	b	$R^{2**}$			
1	6.13	0.98	0.81	0.83*	1.03	0.94	1.58	0.75*	0.86			
2	3.19	0.84	0.89	2.97	$0.79^{*}$	0.84	3.01	0.85	0.90			
3	8.62	$0.64^{*}$	0.60	5.97	$0.56^{*}$	0.63	7.25	$0.66^{*}$	0.72			
4	2.68	0.85	0.78	2.82	0.87	0.79	2.79	0.85	0.77			
5	5.56	0.84	0.77	4.93	0.88	0.70	6.26	$0.41^{*}$	0.38			
6	5.99	0.89	0.85	3.82	0.88	0.91	6.55	$0.49^{*}$	0.85			
7	2.12	0.90	0.86	5.40	$0.54^{*}$	0.53	6.09	0.85	0.74			
Average		•	0.79	•		0.76			0.74			

## B - Scale of Oliveira et al. (2001)

	v	Without scale -			With scale							
Evaluator	V				st evaluation		2 <sup>nd</sup> evaluation					
	а	b	$R^{2**}$	а	b	$R^{2^{**}}$	а	b	R <sup>2**</sup>			
1	6.13	0.98	0.81	3.70	$0.58^{*}$	0.75	2.72	0.64*	0.83			
2	3.19	0.84	0.89	5.06	$0.54^{*}$	0.72	2.60	$0.77^{*}$	0.87			
3	8.62	$0.64^{*}$	0.60	4.75	$0.48^{*}$	0.73	2.44	$0.79^{*}$	0.87			
4	2.68	0.85	0.78	4.43	$0.86^{*}$	0.75	$3.26^{*}$	0.98	0.65			
5	5.56	0.84	0.77	12.09	-0.28*	0.08	11.28	-0.28*	0.10			
6	5.99	0.89	0.85	2.41	$0.46^{*}$	0.89	4.47	$0.52^{*}$	0.83			
7	2.12	0.90	0.86	4.10	$0.48^{*}$	0.66	4.44	0.83	0.74			
Average	,		0.79			0.66			0.70			

## C - Proposed Scale

	Without scale -			With scale							
Evaluator				1	1 <sup>st</sup> evaluation			2 <sup>nd</sup> evaluation			
	а	b	$R^{2^{**}}$	а	b	$R^{2**}$	а	b	$R^{2**}$		
1	6.13	0.98	0.81	1.35	0.87	0.92	0.28*	1.01	0.95		
2	3.19	0.84	0.89	1.77	0.88	0.90	1.75	$0.79^{*}$	0.90		
3	8.62	$0.64^{*}$	0.60	4.90	0.75*	0.81	4.28	0.82	0.86		
4	2.68	0.85	0.78	$1.41^*$	0.93	0.9	$1.84^{*}$	0.90	0.79		
5	5.56	0.84	0.77	$1.74^{*}$	0.92	0.76	$1.67^{*}$	0.86	0.76		
6	5.99	0.89	0.85	$0.46^{*}$	0.97	0.95	1.59	0.97	0.94		
7	2.12	0.90	0.86	2.73	0.83	0.83	4.29	0.98	0.72		
Average			0.79			0.87			0.85		

<sup>\*</sup>Asterisk represents hypothesis situations where null hypothesis (a=0 or b=1) was rejected by the test t (P=0.05). \*\*Asterisk represents significant situations with 1% probability for the test t (P<0.01).

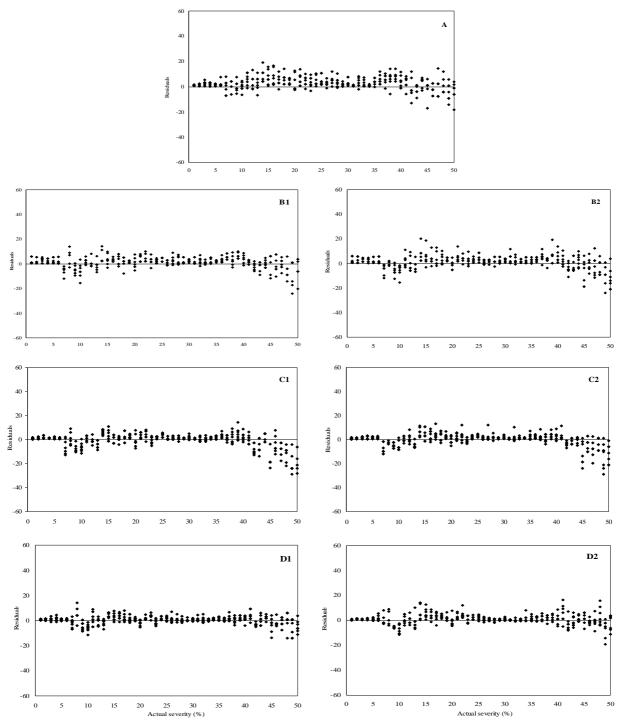


Figure 2 – Distribution of the residuals (estimated severity – actual severity) of the brown eye spot estimation in coffee tree leaves carried through without the aid of diagrammatic scale (A), with the aid of the diagrammatic scale of Fernandes (1988) in two evaluations (B1 and B2), with the aid of the diagrammatic scale of Oliveira et al. (2001) in two evaluations (C1 and C2) and with the aid of the proposed diagrammatic scale in two evaluations (D1 and D2). Points represent the 50 estimations of each evaluator.

Table 2 – Intercept (a), angular coefficient of the straight line (b) and coefficient of determination ( $R^2$ ) of the equations of simple regression line relating the second to the first estimation of brown eye spot ( $Cercospora\ coffeicola$ ) in coffee tree leaves ( $Coffea\ arabica\ L$ .) by the same evaluator, with the aid of the diagrammatic scales: (A) of Fernandes (1988), (B) of Oliveira et al. (2001) e (C) proposed scale.

A – Scale of Fernandes (1988)			
Evaluator	а	b	$R^{2^{**}}$
1	1.16*	0.72*	0.88
2	$1.42^{*}$	0.95	0.83
3	3.18	0.93	0.72
4	$0.47^{*}$	0.94	0.91
5	3.26	0.51*	0.65
6	5.30	$0.49^{*}$	0.74
7	4.06	1.02	0.59
Average			0.76
B – Scale of Oliveira et al. (2001)			
Evaluator	а	b	$R^{2^{**}}$
1	0.83*	0.9	0.72
2	-1.21*	1.13	0.75
3	-0.92*	1.21*	0.65
4	-1.93 <sup>*</sup>	1.16	0.87
5	$0.77^{*}$	0.85	0.86
6	1.97	1.10	0.88
7	$1.78^*$	1.27	0.61
Average			0.76
C – Proposed Scale			
Evaluator	а	b	$R^{2^{**}}$
1	-0.79 <sup>*</sup>	1.12	0.95
2	$0.54^{*}$	0.86	0.93
3	-0.01*	1.02	0.91
4	$0.95^*$	0.92	0.81
5	$0.19^{*}$	0.93	0.97
6	1.26	0.99	0.97
7	$2.24^*$	1.09	0.74
Average			0.90

<sup>\*</sup>Asterisk represents hypothesis situations where null hypothesis (a=0 or b=1) was rejected by the test t (P=0.05). \*\*Asterisk represents significant situations with 1% probability for the test t (P<0.01).

With the use of the diagrammatic scales of Fernandes (1988) and Oliveira et al. (2001), low repeatability was noticed when compared to the proposed scale because the average amount of variation in the first evaluation explained the second evaluation, and in both the cases, was 76% (Table 2, B). Between the two evaluations, three evaluators (1, 2 and 4) presented intercept values significantly different from zero for the Fernandes scale (1988), while for the Oliveira et al. (2001) scale, only evaluator (6) did not obtain the intercept value significantly different from zero. For the angular coefficient of the straight line, significantly different values

of 1 for three evaluators (1, 5 and 6) was observed using the scale of Fernandes (1988) and only one evaluator (3) for scale of Oliveira et al. (2001) (Table 2B).

The reproducibility of the severity evaluations without the use of diagrammatic scale was low, because the linear regression among the estimated severities of seven evaluators produced coefficient of determination varying from 43 and 91%, with 68% being average.

With the use of the proposed diagrammatic scale the evaluations presented good reproducibility, because in both evaluations the values of the coefficient of determination were raised, varying between 57 and 94%, with an average of 79% in both evaluations (Table 3). On the other hand, with the use of the diagrammatic scales of Fernandes (1988) and Oliveira et al. (2001), lower reproducibility was noticed because the linear regression between estimated severities of seven evaluators produced coefficients of determination varying between 22 and 86%, with an average of 62% for the scale of Fernandes (1988) and varying between 42 and 91%, with an average of 70% for the scale of Oliveira et al. (2001) (Table 3).

In this way, the reproducibility of the evaluations with the use of proposed diagrammatic scale was considered good, according to the literature criteria (ANGELOTTI et al., 2008; SALGADO et al., 2009; SUSSEL et al., 2009).

The establishment of an accurate, reproducible and standardized scale as a possible reference is desirable, as standardizing the assessment methodology of diseases allows comparisons to be made between different institutions and locations (MADDEN et al., 2007; SUSSEL et al., 2009).

Table 3 – The coefficient of determination ( $R^2$ ) of equations of linear regression related to the estimates of brown eye spot ( $Cercospora\ coffeicola$ ) in coffee tree leaves ( $Coffea\ arabica\ L$ .) between evaluators without the aid of diagrammatic scale and with the aid of the diagrammatic scales of Fernandes (1988), Oliveira et al. (2001) and with the proposed scale.

Without Scale							_						
Evaluator	2	3	4	5	6	7	-						
1	0.73	0.58	0.85	0.91	0.77	0.71							
2		0.61	0.62	0.68	0.86	0.84							
3			0.43	0.49	0.61	0.51							
4				0.83	0.57	0.65							
5					0.63	0.67							
6						0.82							
Scale of Ferna	indes (19	88)											
Evaluator			1 <sup>st</sup> eva	luation					2 <sup>nd</sup> eva	luation			
Evaluator	2	3	4	5	6	7	2	3	4	5	6	7	
1	0.80	0.61	0.83	0.74	0.86	0.60	0.86	0.66	0.73	0.38	0.76	0.65	
2		0.60	0.63	0.59	0.68	0.53		0.73	0.72	0.31	0.79	0.73	
3			0.52	0.47	0.57	0.47			0.53	0.22	0.81	0.6	
4				0.82	0.76	0.54				0.69	0.66	0.56	
5					0.72	0.53					0.34	0.24	
6						0.49						0.64	
Scale of Olive	ira et al.	(2001)											
Evaluator		1 <sup>st</sup> evaluation						2 <sup>nd</sup> evaluation					
Evaluator	2	3	4	5	6	7	2	3	4	5	6	7	
1	0.72	0.7	0.62	0.56	0.75	0.60	0.83	0.91	0.42	0.75	0.83	0.63	
2		0.69	0.59	0.73	0.83	0.70		0.91	0.63	0.71	0.82	0.67	
3			0.72	0.69	0.69	0.62			0.49	0.70	0.78	0.66	
4				0.75	0.71	0.57				0.72	0.58	0.58	
5					0.74	0.62					0.81	0.73	
6						0.77						0.71	
Proposed Scal	e												
Evaluator			1 <sup>st</sup> eva	luation					2 <sup>nd</sup> eva	luation			
Evaluator	2	3	4	5	6	7	2	3	4	5	6	7	
1	0.88	0.86	0.87	0.78	0.93	0.77	0.93	0.83	0.83	0.82	0.94	0.78	
2		0.80	0.88	0.69	0.90	0.78		0.84	0.75	0.69	0.93	0.75	
3			0.78	0.71	0.79	0.75			0.69	0.65	0.88	0.73	
4				0.75	0.83	0.74				0.84	0.68	0.57	
5					0.79	0.74					0.71	0.62	
6						0.81						0.80	

Ciênc. agrotec., Lavras, v. 35, n. 6, p. 1067-1076, nov./dez., 2011

## **CONCLUSIONS**

The proposed diagrammatic scale to evaluate the severity of brown eye leaf spots in coffee tree leaves, when compared to the Fernandes (1988) one and to the Oliveira et al. (2001), showed improvement in the levels of accuracy, precision and measurement of reproducibility.

The proposed diagrammatic scale provided good estimations of the intensity of the disease, according to the evaluated variables.

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