Protocol of evaluation of Metalosate copper in the control of *Lasiodiplodia theobromae* avocado

1. METHODOLOGY

The experiment was carried in a greenhouse of the facilities of the Department of health vegetable of the University National San Luis Gonzaga. The conditions of temperature were between 24 - 28 ° C and 60 - 70% wetness relative.

It was used plant in pot of about 1 year age of the grow Hass grafted about pattern Zutano. For the inoculations, an isolated was used of *L. theobromae*: the 115 (isolated of a plant of avocado from of Chincha). In the test is used the listed products in the Table 1.

Table 1. Treatments staff to the control of *l. theobromae* in inoculations artificial in avocado.

Dosage	Type of application	Number of de applications	Interval between entre applications	
Treatments		**Applications preventive**		
T0. Control Aplic. preventive				
T1. Metalosate copper	1,0 L/ha	Leaf	02	10 days
T2. Met Cu + Met B	1,0 L/ha + 0,5 L/ha	Leaf	02	10 days
T3. Metalosate copper	4,0 L/ha	Drench	02	10 days
T0. Control Aplic Curative		**Applications Curative**		
T1. Metalosate copper	2,0 L/ha	leaf	02	10 days
T2. Met Cu + Met B	2 L/ha + 0,5 L/ha	leaf	02	10 days
T3. Metalosate copper	4,0 L/ha	Drench	02	10 days

Apply by spray leaf and via drench of agreement to the set on the treatment the product. For the applications leaf, previously be held the calculation of spending of water, and finally was applied a volume average of 120 ml of stock fungicide by plant, using a backpack spray

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hydraulic of pressure

previous. For applications via drench be calculated previously the volume of hold of water. The applications be did by packaging graduates, applying 0.4 L of the suspension per plant.

Strategies of assessment efficacy of products

A. Strategies preventive:

For East test was used groups of ten plant of avocado by every treatment (dose of Metalosate copper). Two applications were made consecutive of each treatment in intervals of 10 days between applications. In the Group of plants witness was applied on the same intervals with water single.

Five days after of the last application, was inoculated the plant of every treatment with the isolated of *L. thebromae* . The plant was inoculated on the part more broadband of the outbreak on the area of the variety. The goal of these inoculations was to evaluate the effect of the products evaluated in applications before the infection, and also like affected the applications leaf or via drench in the area of injury caused by the pathogen in the variety.

B. Strategies curative:

Groups of plant of every treatment was inoculated previously with the isolated of *L. thebromae*. Five days after of the inoculation was made two applications consecutive in intervals of 10 days between applications to the various treatment. Similar to the described, the plant was inoculated on the area of the variety, and was carried out applications of plants via leaf.

Process of inoculation of plants

The bark of the area of the range was disinfected superficially with alcohol ethyl to the 96 °. Of this area was extracted a disc of bark by a punch sterile of 8 mm of diameter. On the wound be placed in contact direct with the cambium exposed, a disc of PDA the similar diameter colonized with the mycelium of the isolated of *l thebromae*, and the wound it was plugged with the disc of bark extracted. The area of inoculation it was damped with drops of water sterile, it was wrapped with a tape of parafilm for protect the area inoculated of the dewatering (**Figure 1)**.

Evaluation of results

Three weeks after of the inoculation, it was removed carefully the bark of every plant inoculated around of the area of the wound. The area of the injury resulting it was featured on a blade of plastic transparent, and was transferred to a leaf of paper in white, and it was scanned **(Figure 2).** By the program Assess (American Phytopathological Society, St. Paul MN), it was

quantified the area of the injury of every treatment by the analysis of image. The size of the area of the wound of inoculation was removed of the area total obtained for calculate the size real of the injury. For confirm that the injuries were caused by *l. theobromae,* have made planting of the affected tissue in middle PDA's to the least two plants of every treatment.

Additionally, have included the values averages of the percentage of control every treatment. This data was calculated by the following formula:

% Control = 100 - [(area of injury x 100) / area of injury of the witness].

Analysis statistical

The results was expressed like area of injury in cm² by treatment; it was put special emphasis on the areas injury developed the area of the range in both strategies of application carried out. The areas of injury of the factors in study is submitted to an analysis of the variance (ANOVA), setting the level of significance in $P < 0,05$.

Given that the registered values not had a distribution normal, was hold a transformation of the data original to $\sqrt{x}$ or $\sqrt{x + 1}$ for stabilize the variance. With these value is held a test of separation of average using the analysis of Fisher of the minimum difference significant (LSD). The program statistical used for the analysis was SAS 9.0.

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Figure 1. Process of inoculation of avocado plants with L. theobromae in greenhouse. Inoculations in the area of the pattern and the variety of avocado

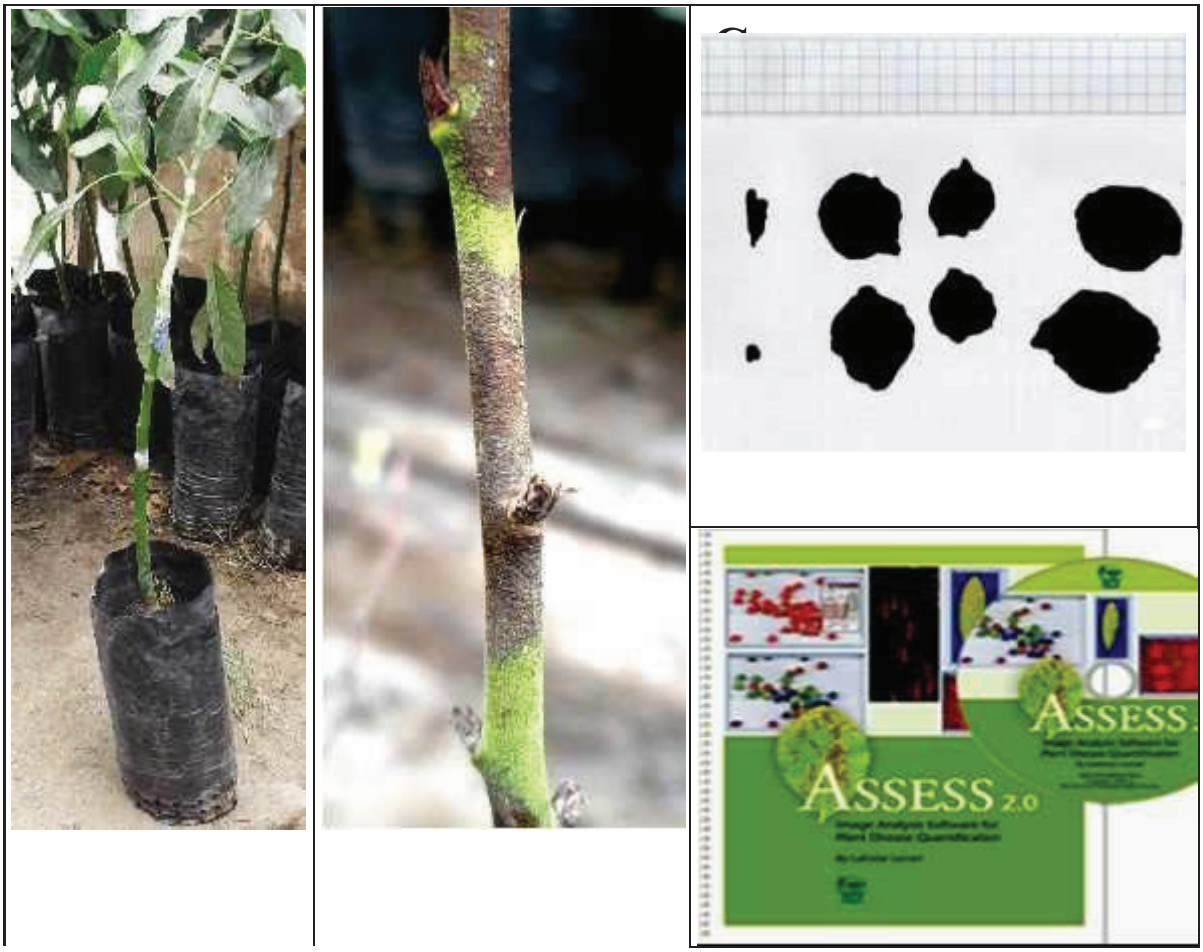
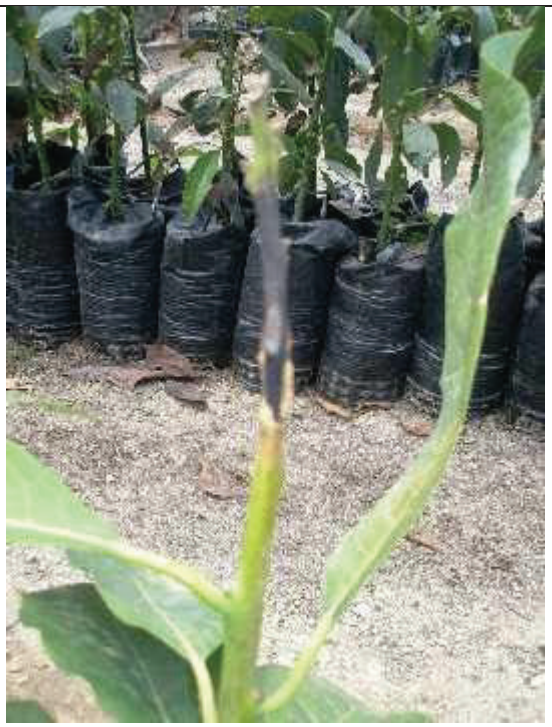


Figure **2. Procedure for the assessment of the area of the injury to - b.** Area of the injury in plant of avocado**. b.** Area of the injury paper **c.** processing program of images digital

2. RESULTS



3a

Figure 3b. Area of injury registered in inoculations on the area of the range (in arrows) in the treatment Witness. Note the outbreak leaf affected by the advance of the injury e early of sporulation (3b).

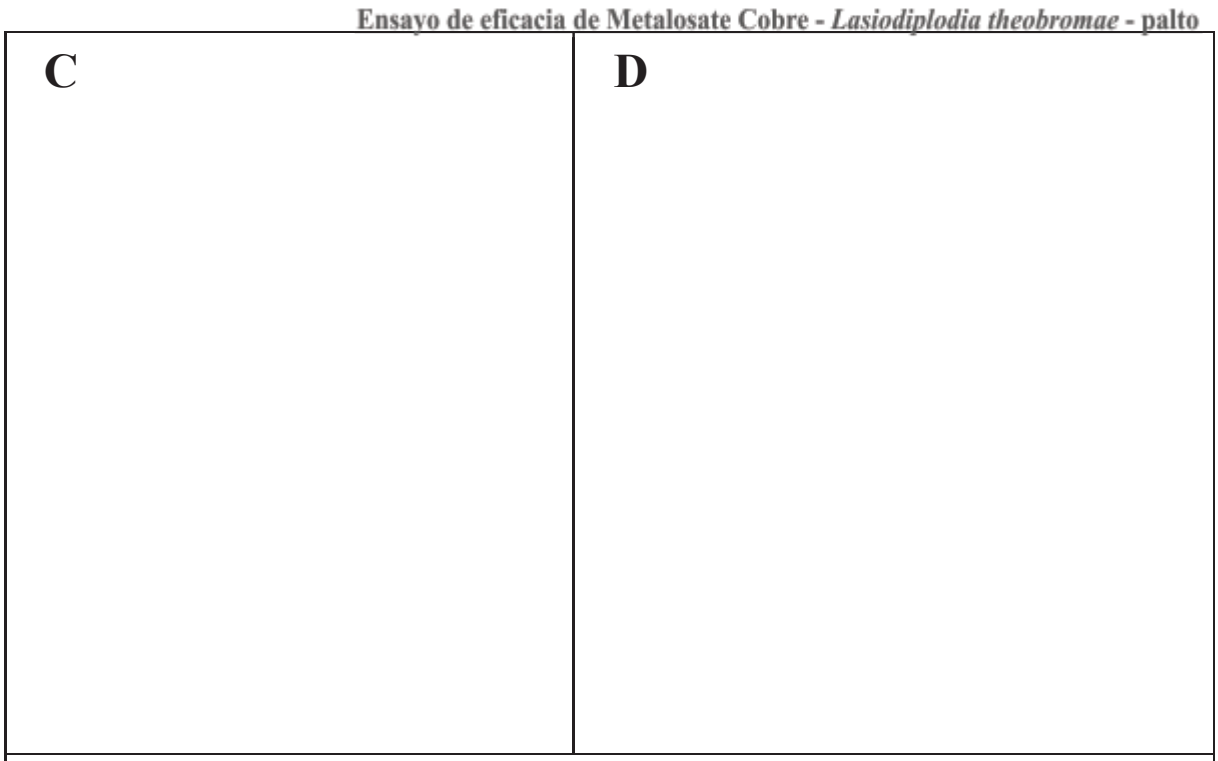


Figure **3 c and 3d. Strategies healing.** Area of injury registered in inoculations on the area of the variety: T1 (Metalosate copper: 2.0 L / ha) (3 c) and T2 (Metalosate copper: 2.0 l / has + Metalosate Boron 0.5 L / ha) (3d). Note the advance of the injury starting of the point of inoculation (in arrows).

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E

F

Figure **3e and 3f. Strategies preventive.** Area of injury registered in inoculations on the area of the variety: T1 (Metalosate copper: 1.0 L / ha) (3 c) and T2 (Metalosate copper: 1.0 l / ha + Metalosate Boron 0.5 L / ha) (3d). Note the advance of the injury starting of the point of inoculation (in arrows).

A. Strategies preventive

Table 2. Effect of the applications leaf and via drench of metalosate copper on the area of injury by *l. theobromae* in avocado plants var. Hass.

Evaluation of the preventive strategies

Factor		Value of *P* [w] Area of injury in cm²	
Effects main Treatment		<0,0001 (**)	
Treatments	**Method of Application**	**Area of injury (cm²)**	
T0. Control Aplic.		11,4[x] 3,5[y]	a[z]
T1. Metalosate copper: 1,0 L/ha	Foliar	4,1 2,3	b
T2. Met Cu + Met B: 1,0 L/ha + 0,5 L/ha	Foliar	3,5 2,1	b
T3. Metalosate copper: 4,0 L/ha	Drench	0,5 1,2	c
	Average	**4,9 2,3**	
	Coef. Variability	19,2%	

[w] Values of *P*> 0.05 not are significantly several of remember to the test of the Minimum difference significant (LSD). (**) High significance, (NS) not there significance. [x] Each data it's average of 10 values (plants), and makes reference to the area of injury in cm² developed starting of the area of inoculation by infection by *Lasiodiplodia theobromae*; and data original converted to √x + 1, [z] Numbers in columns followed by the in letter not are significantly various of agreement to the test of LSD.

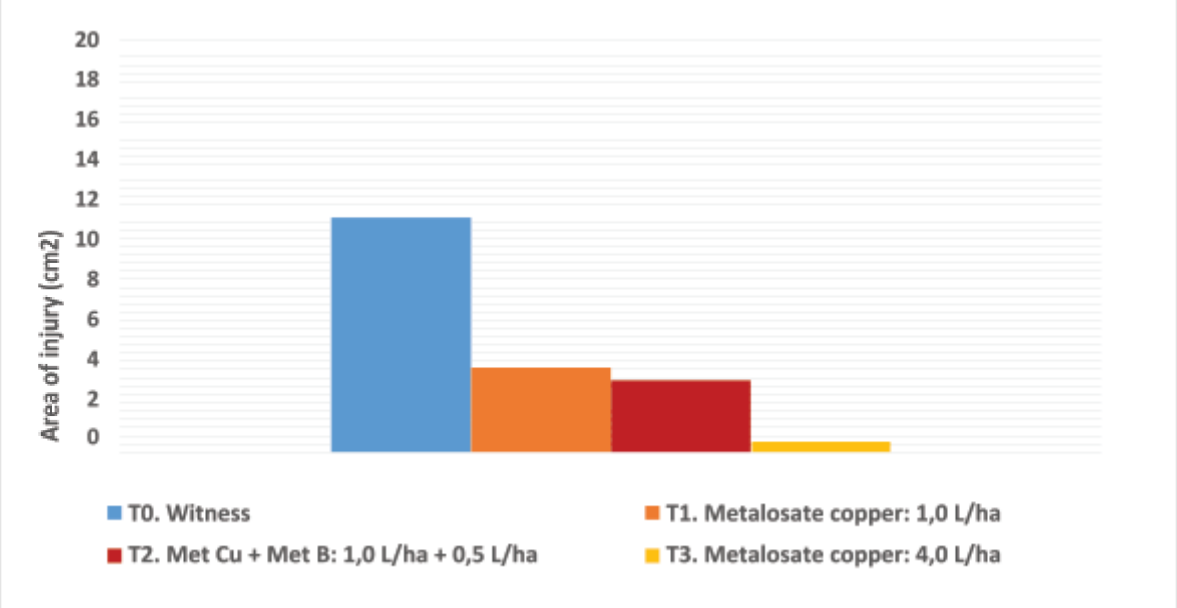

Figure 4. Influence of the applications leaf and via drench on the injury (area of injury in cm²) caused by *Lasiodiplodia theobromae* plant of avocado.

B. Strategies curative

Table 3. Effect of the applications leaf and via drench of metalosate copper on the area of injury by *l. theobromae* in avocado plants var. Hass.

Evaluation of the strategies healing.

Factor		Value of *P* [w] Area of injury in cm²	
Effects main			
Treatment		<0,0001 (**)	
Treatments		**Application**	**Area of injury (cm²)**
T0. Control Aplic.		13,1[x]	3,6[y] a[z]
T1. Metalosate copper: 2,0 L/ha	leaf	9,3	3,0 a
T2. Met Cu + Met B: 2,0 L/ha + 0,5 L/ha	leaf	9,9	3,1 a
T3. Metalosate copper: 4,0 L/ha	Drench	3,5	1,9 b
	Average	**9,0**	**2,9**
	Coef. Variability	11,6%	

[w] Values of *P*> 0.05 not are significantly several of remember to the test of the Minimum difference significant (LSD). (**) High significance, (NS) not there significance. [x] Each data it's average of 10 values (plants), and makes reference to the area of injury in in cm² developed starting of the area of inoculation by infection by *Lasiodiplodia theobromae*; [and] data original converted to √x, [z] Numbers in columns followed by the in letter not are significantly various of agreement to the test of LSD..

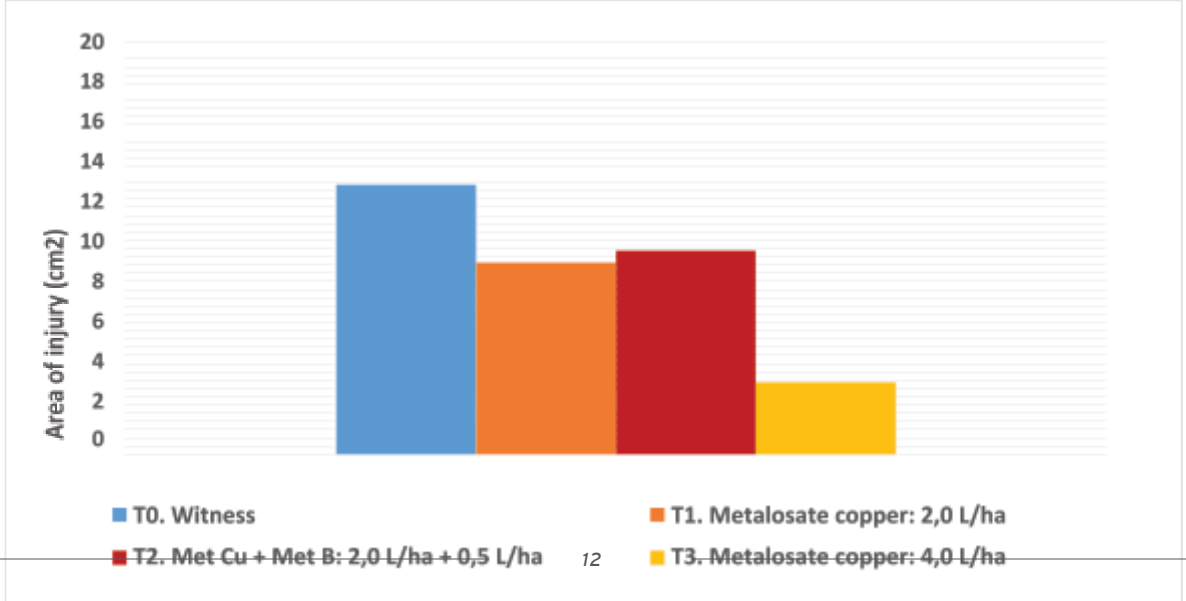

Figure **5.** Influence of the applications leaf on the injury (area of injury in cm^2) caused by *Lasiodiplodia theobromae* in plants avocado.

Table 4. Percentage of control of different dose applications leaf and via drench of Metalosate copper front to infection by *Lasiodiplodia theobromae* in avocado.

Assessment of the efficiency of the applications preventive.

Treatments	Type of Application	% Control
T1. Metalosate copper: 1,0 L/ha	leaf	64,0 [z]
T2. Met Cu + Met B: 1,0 L/ha + 0,5 L/ha	leaf	69,3
T3. Metalosate copper: 4,0 L/ha	Drench	95,6

[z] Each data it's the result of the conversion of the formula: % control = 100 - [(Area of injury x 100) / Area of injury [of the witness].

Table 5. Percentage of control of different dose applications leaf of Metalosate copper front to infection by *Lasiodiplodia theobromae* in avocado.

Assessment of the efficiency of the curative applications.

Treatments	Type of Application	% Control
T1. Metalosate copper: 2,0 L/ha	leaf	29,0[z]
T2. Met Cu + Met B: 2,0 L/ha + 0,5 L/ha	leaf	24,4
T3. Metalosate copper: 4,0 L/ha	Drench	73,3

[z] Each data it's the result of the conversion of the formula: % control = 100 - [(Area of injury x 100) / Area of injury [of the witness].

3. CONCLUSIONS

1. It was evaluated two strategies of application of Metalosate copper on the culture of avocado, preventive and curative. The results obtained indicate that best results were obtained by the applications preventive. The average of area of injury generated by *L theobromae*, in, were considerably minor in preventive applications compared with the areas of injury registered for the curative applications.
2. In the preventive applications, all the dose evaluated of Metalosate copper decreased the areas of injury *l. theobromae* **(Table 2).** In General terms have not been recorded differences statistics significant between the treatment that carried out via leaf) (treatments T1 and T2); regardless of, the best treatment of the evaluated was the treatment T3 (Metalosate copper 4.0 L / has), who exercised the best efficiency on the control the injury by the pathogen. The

percentage of control

obtained are consistent with the analysis statistical **(table 4)**, and indicates that greater percentage of control was obtained with treatment T3.

3. On the curative applications, all the dose evaluated of Metalosate copper decreased the areas of injury by *L. theobromae* **(table 3).** Without However, not be recorded differences significant between treatments that was made via leaf (treatments T1) (and T2) with the treatment T0 (witness without application). In this test, was determined that statistically the best treatment was the T3 (Metalosate copper 4.0) (L / ha). The percentage of control obtained are consistent with the analysis statistical **(table 5),** e indicates that the more percentage control was obtained treatment T3.
4. The higher dosage applied, or the type of application (via (drench) could have influenced the treatment T3 have obtained the best result.
5. Given that, the product Metalosate copper not is a synthetic fungicide, but nevertheless the preventive and curative applications managed limit the infection on the area variety by *Lasiodiplodia*, evidence of an effect promoter of defense of plants.

It is all how I have to inform,

Kind regards

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