BIBLIOGRAPHY

TUMBAGA, MELCHOR B. MARCH 2013. Survey and Morphological Characterization of the Hyperparasite Associated with Coffee Leaf Rust (*Hemileia vastatrix*) in Arabica Growing Areas at La Trinidad, Benguet.

Adviser: Asuncion L. Nagpala, Ph.D.

ABSTRACT

The survey and collection of coffee leaves infected with rust were done in five Arabica coffee growing areas namely, Bektey, BSU main campus, Buyagan, Cruz, and Wangal, La Trinidad, Benguet from April to October 2012.

The study aimed to determine the percent parasitized rust lesions and parasitized uredospores and to characterize the hyperparasite associated with leaf rust based on morphological structures.

Survey results showed that rust infection in different sampling sites differed significantly with Bektey giving the highest mean infection of 89.93. The lowest rust infection with a mean of 19.00 was recorded from Buyagan. Percent parasitized rust lesions in all sampled area were comparable although the highest parasitized lesion of 61.72 % was recorded from sample coming from Buyagan.

The highest parasitized uredospores of 29.15% was recorded from BSU main campus samples while the lowest with a mean of 7.20% was noted from Buyagan.

In terms of the morphological structure, the hyperparasite had a conidia that is hyaline, ellipsoidal in shape and measured 2.86µm x 0.93µm with a conidiophores that is 40.3µm long bearing three to eight phialides measuring 8.9µm.

RESULTS AND DISCUSSION

Rust Lesions on Coffee Leaves from Different Sampling Site

Rust lesions. Table 1 shows the mean number of rust lesions recorded on the infected coffee leaves. Data shows that more number of rust lesions were counted on coffee leaf grown at Bektey with a mean of 89.93 and is significantly higher than the rust lesions found in Wangal with a mean of 19.00. Statistical analysis revealed significant differences of rust lesion on coffee leaves from the different sample sites.

Table1. Mean Rust Lesions of Infected Coffee Leaves

SOURCES OF SAMPLES	RUST LESIONS
Bektey	89.93[a]
BSU main campus	51.67[abc]
Buyagan	33.67[bc]
Cruz	67.67[ab]
Wangal	19.00[c]

Means with the same letters are not significantly different at 5% level DMRT.

Percent parasitized rust lesion. Table 2 shows the percent parasitized rust lesion wherein Wangal has the highest percentage of parasitism of 61.72 but not significantly different with means of the other sources. From the results, it appears that leaves with the highest leaf rust did not produce highest parasitized lesions. Figure 2 shows rust lesions parasitized with *Verticillium* sp.

Table 2. Mean Percent (%) of Parasitized Lesion.

SOURCES OF SAMPLES (%)	PERCENT PARASITIZED LESIONS
Bektey	23.07
BSU main campus	44.11
Buyagan	55.42
Cruz	36.59
Wangal	61.72

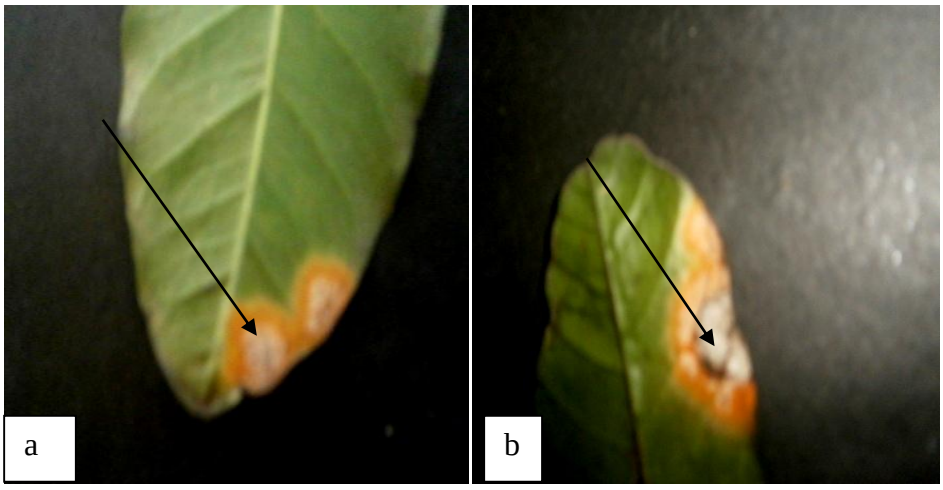

Figure 2. a and b parasitized rust lesion showing white growth of *Verticillium* pointed by arrows

Microscopic Observation

Microscopic count rust uredospores. Table 3 shows the mean of rust uredospores counted under the microscope. Results showed that coffee leaves from Wangal with the lowest rust also gave the lowest count of uredospores of 89.33 from coffee rust lesion. The highest count of uredospores of 179.67 was recorded from rust lesion collected from Bektey but did not differ on the number of uredospores from Buyagan, BSU Main campus, Cruz and Wangal.

Table 3. Mean Number of counted of Rust Uredospores from Rust Lesions Collected from Different Sites.

SOURCES OF SAMPLES	RUST UREDOSPORE
Bektey	179.67
BSU main campus	115.67
Buyagan	177.00
Cruz	172.00
Wangal	89.33

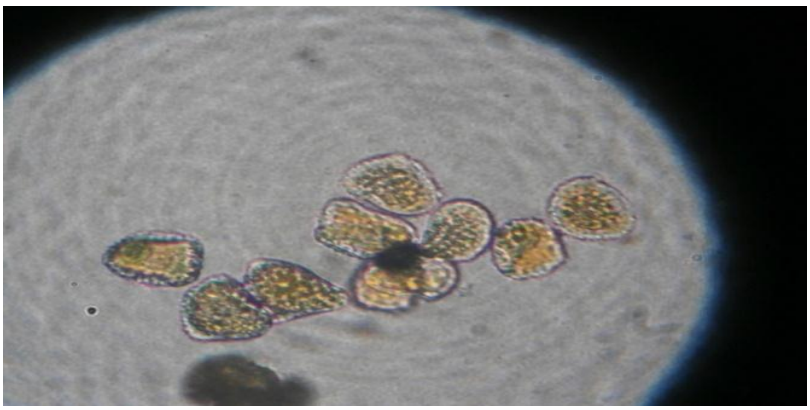

Figure 3. Uredospores of rust seen under the microscope (400X)

Percent (%) parasitized rust uredospores. Table 4 shows the percent parasitized uredospore where in the sample collected from BSU-compound had the highest percentage of uredospore that were parasitized by *Verticillium* with a mean of 29.15% and significantly differed other samples from the other area. Further observation revealed that rust lesions which gave the highest number of uredospores did not give highest percent parasitized uredospores.

Table 4. Mean Percent (%) of Parasitized Rust Uredospores.

SOURCES OF SAMPLES	PARASITIZED RUST UREDOSPORES (%)
Bektey	8.03[b]
BSU main campus	29.15[a]
Buyagan	7.20[b]
Cruz	11.71[b]
Wangal	12.32[b]

Means with the letters are not significantly different at 5% level DMRT.

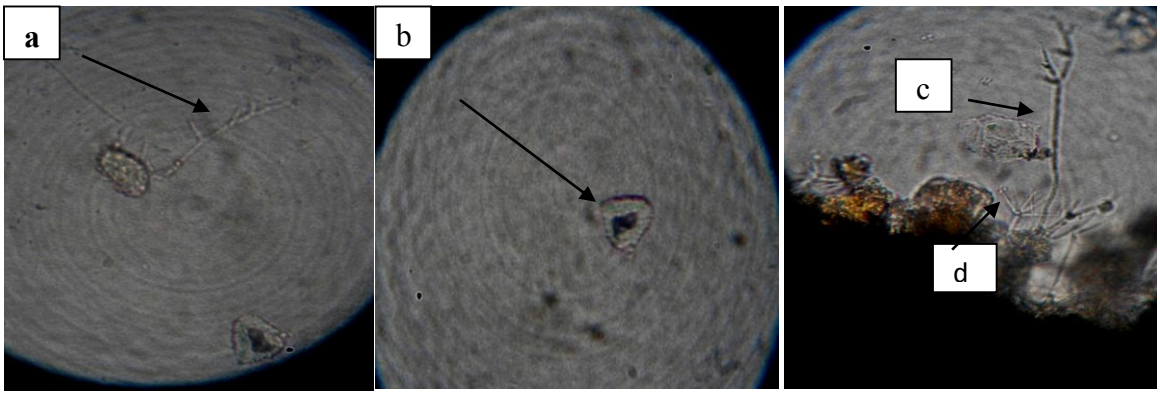

Figure 4.Microscopic observation of parasitized uredospore. a. conodiophore of *Verticillium* sp parasitizing uredospore b. Uredospore c. phialides bearing conidia (400x)

Morphological Structure of the *Verticillium* sp.

Table 5 summarizes the result of the microscopic observations. The conidia of the hyperparasite are ellipsoidal to cylindrical and hyaline. The conidia measures 2.86 µm x 0.93 µm. Phialide is 8.9 µm while the conidiophore is 3 µm long with three to eight phialides. The length of the Viegas (1939) identified *Verticillium lecanii* isolated from insect to have hyphae that is moderately branched, septate, smooth-walled, hyaline with 1.6-2.4 µm in diam. Conidiophores are tall, erect, septate, bearing phialides singly or in whorls of 2-8. Phialides is awl-shaped, hyaline, smooth measuring 14.3-22.2 × 1.6-2.4 µm. Conidia are produced at the tip of phialides, often forming a mucoid false head that are oval, ellipsoidal to shortly cylindrical measuring 3.2-4.8 × 1.6-2.0 µm. Chlamydospores are absent and teleomorph is unknown.

Table 5. Morphological Characteristics of the Hyperparasite *Verticillium sp*

CRITERIA	HYPERPARASITE	Viegas IDENTIFICATION (1939)
Shape of conidia	Ellipsoidal to cylindrical	Oval, ellipsoidal to shortly Cylindrical
Color of Conidia	Hyaline	Hyaline
Size of Conidia	2.86x.93µm	3.2-4.8(-6.4) × 1.6-2.0 µm
Phialides	8.9 µm 3 to 8 phialides per conidiophores	14.3-22.2x 1.6-2.4 µm
Conidiophore	40.3 µm, long	Tall, erect, septate, bearing phialides singly or whorl of 2-8

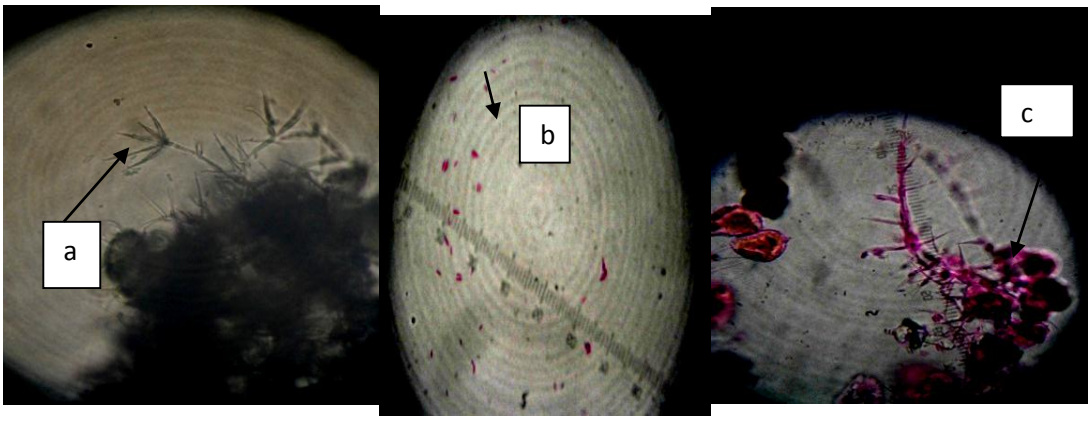

Figure 5. Hyperparasite structure a. phialides b.conidia(stained pink) pointed by an arrow c. conidiophore attached to the uredospores of coffee rust.

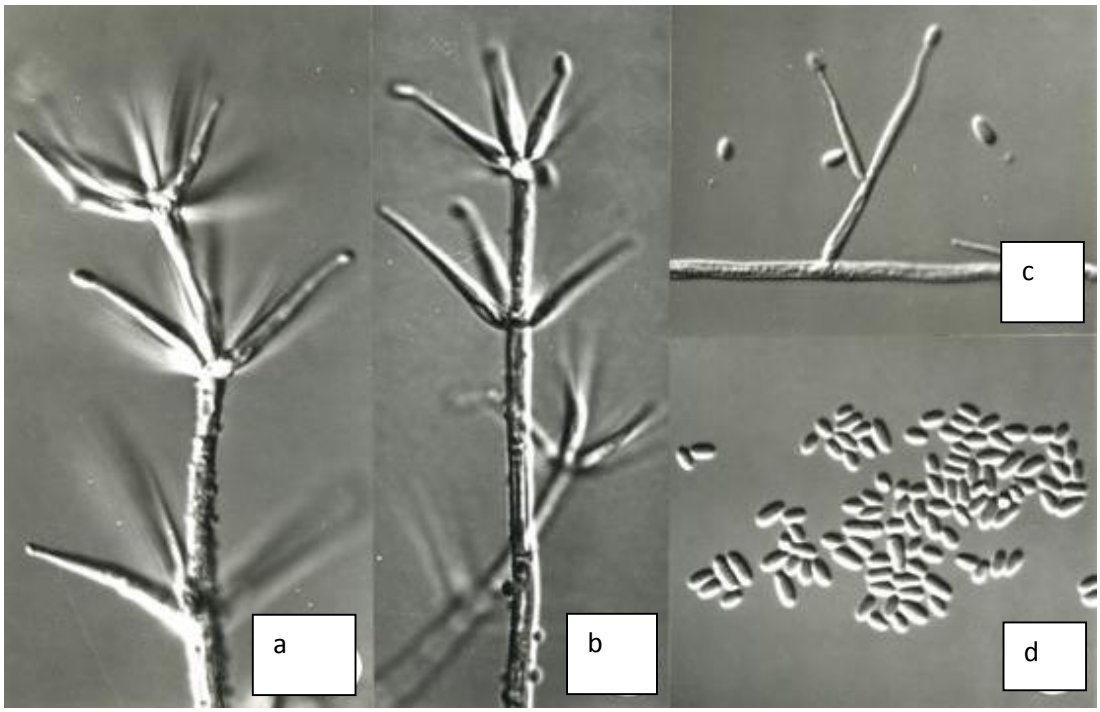

Figure 6. *Verticillium lecanii* a. and b. conidiophores bearing phialides, c. conidia attached to phialides d. conidia (Viegas, 1939) using electron microscope.

SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

Summary

The survey was conducted in five coffee growing areas in Bektey, BSU main campus, Buyagan, Cruz, and Wangal at La Trinidad, Benguet from April to October 2012. The study aimed to determine the distribution of coffee rust lesion, and those parasitized by the hyperparasite in different sampling sites, determine the parasitized uredospores and characterize the hyperparasite based on its morphological structures.

The study aimed to determine the percent parasitized rust lesions, parasitized uredospores and characterized the hyperparasite associated with leaf rust based on morphological structures.

Survey results showed that rust infection in different sampling sites differed significantly with Bektey giving the highest mean infection of 89.93. The lowest rust infection with a mean of 19.00 was recorded from Buyagan. Percent parasitized rust lesions in all sampled area were comparable although the highest parasitized lesion of 61.72 % was recorded from sample coming from Buyagan.

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In terms of the morphological structure, the hyperparasite has a conidia that is hyaline, ellipsoidal in shape and measures 2.86 µm x 0.93 µm with a conidiophore that is 40.3 µm long bearing three to eight phialides measuring 8.9 µm.

Conclusions

Coffee rust infection including the percent parasitize lesions differ in different sampling areas.

The hyperparasite associated with coffee rust based on the characterization is similar to *Verticillium lecanii* although measurements of conidia and phialides did not exactly fall within those characterized by Viegas in 1939.

Recommendations

1. Survey should be done during rainy and dry season and on different varieties to compare which of the existing varieties show higher parasitism by the hyperparasite

2. Explore an appropriate media for growing *Verticillium*.Identification of the species of the hyperparasite using electron microscope should be done.

3. Mass production of the hyperparasite and evaluation in greenhouse experiment should be done to determine its effectiveness.

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