Report on Coffee Rust

By Dr. Mathias Oulé

Coffee rust is one of the most devastating diseases to affect coffee trees and is caused by the fungus *Hemileiavastatrix.* This disease, which affects the *Coffee arabica* tree, was long confined in Africa but has since spread to Asia even reaching South America in 1970. It is of concern to all coffee producing countries including Ivory Coast. Chemical control is still commonly used against coffee rust. Thus, a study was conducted on twenty (20) samples of coffee tree leaves suffering from coffee rust, taken from two zones in the south of Ivory Coast (Agboville and Adzopé), in order to evaluate the fungicidal activity of the PHMGH based disinfectant against the fungus responsible for coffee rust.

I- Preparation of the inoculum and the different concentrations of PHMGH

I-1. Preparation of the inoculum

10 g of leaves that were seriously infected with coffee rust were soaked in 90 ml of buffered peptone water. The suspension obtained was used as the test inoculum.

I-2. Preparation of the different concentrations of PHMGH

A range of 10 decreasing concentrations was prepared. The following formula was used to determine the necessary volumes of the PHMGH disinfectant needed to prepare the varying concentrations:

$$CiVi = CfVf \longrightarrow \boxed{Vi = \frac{CfVf}{Ci}} \qquad (1)$$

Where Ci = initial concentration of PHMGH (250 mg/mL)

Vi = initial volume of PHMGH (mL)

Cf = final concentration (mg/mL)

Vf = final volume (mL)

The different volumes of PHMGH calculated were added to a series of 10 tubes containing 10 ml of Sabouraud broth that were labeled from T_1 to T_{10}. The range of

dilutions corresponded to concentrations of 2.2 mg/mL, 1.9 mg/mL, 1.6 mg/mL, 1.3 mg/mL, 1 mg/mL, 0.7 mg/mL, 0.4 mg/mL, 0.1 mg/mL, 0.07 mg/mL and 0.04 mg/mL. Two tubes, labeled T_0 and T_C, containing 10 ml of Sabouraud broth were prepared and served as a negative control and growth control, respectively.

II- Determination of the MIC and the MFC

II-1. Determination of the MIC

The tubes labelled T_C and the tubes T_1 to T_{10} prepared previously were each inoculated with 1 mL of the test inoculum. The negative control, T_0, was not inoculated. The tubes were then incubated at 30°C for 72h. All tests were performed twice. The MIC was determined from the experimental tubes that showed no mold growth.

II-2. Determination of the MFC

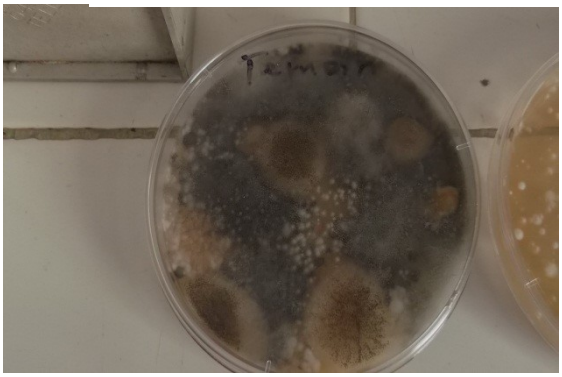
The MFC was determined from the experimental tubes used for the MIC test and showed no growth. After gently agitating the tubes, 0.1 ml was removed and plated on petris containing Sabouraud agar and chloramphenicol. The petris were incubated at 30°C for 72h. Growth was compared to a control, which consisted of a petri containing Sabouraud agar and chloramphenicol that was not inoculated. Please note that in order to see the growth of mold in a petri, 0.1 mL of the contents of tube T_C was inoculated on a petri containing Sabouraud agar and chloramphenicol.

III- Results

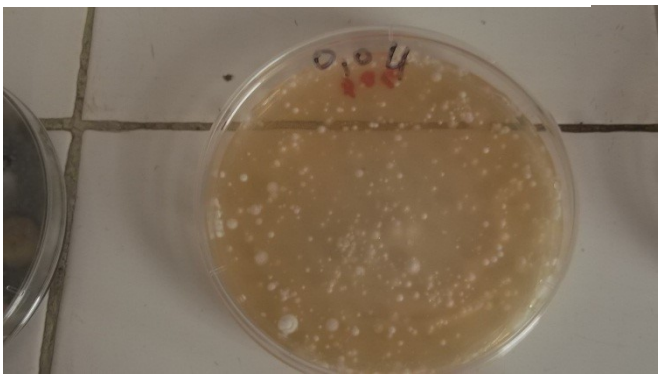
The evaluation of the antifungal activity of PHMGH by the determination of the MIC and MFC on the *in vitro* growth of fungi present on coffee leaves suffering from coffee rust revealed that the MIC and the MFC are the same and also the strong activity of PHMGH. In effect, the analysis of the experimental data that showed the growth of mold decreases as the concentration of PHMGH increases. The majority of mold is eliminated with a concentration of 0.04 mg/mL. However, after several experiments were carried out, one mold resisted all concentrations of PHMGH used. The latter is in

the form of round, curved white colonies on the Sabouraud and chloramphenicol petris (see photos in annex). Studies continue in order to determine the classification of the unknown fungus and to determine the time necessary for the microbicidal activity of PHMGH on the fungi present on the contaminated leaves.

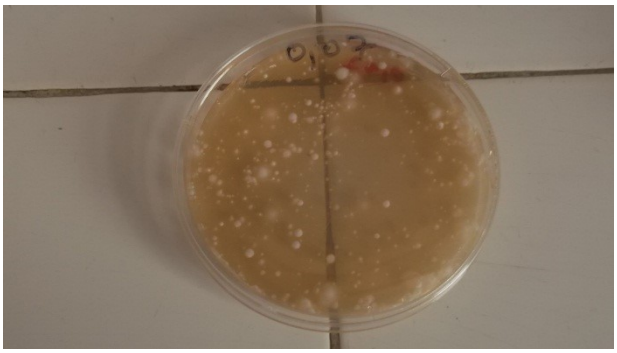
THE EFFECT OF PHMGH ON FUNGI ASSOCIATED WITH COFFEE RUST ON COFFEE TREE LEAVES



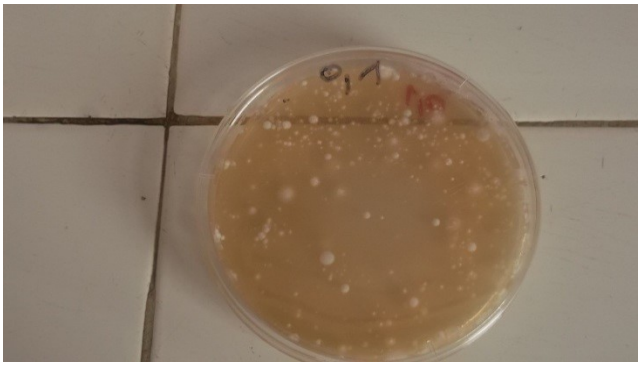
Control inoculated from T_C



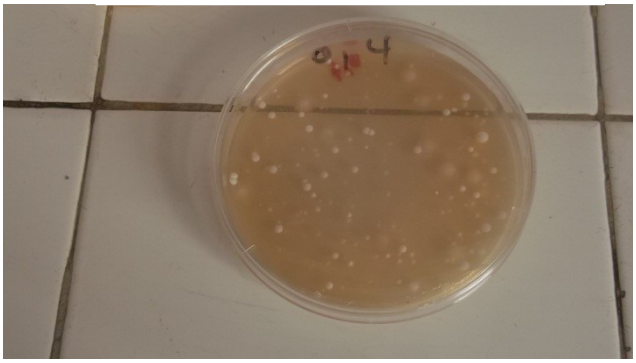
Concentration 0.04 mg/mL



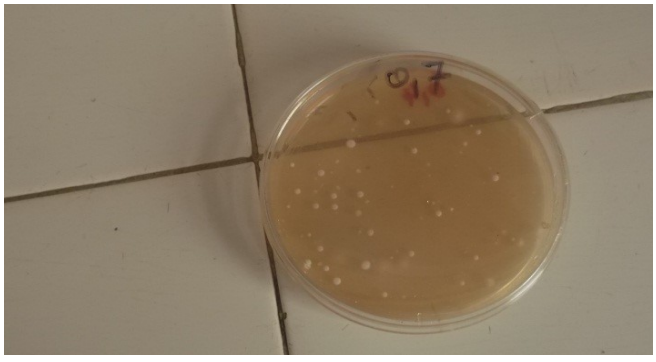
Concentration 0.07 mg/mL



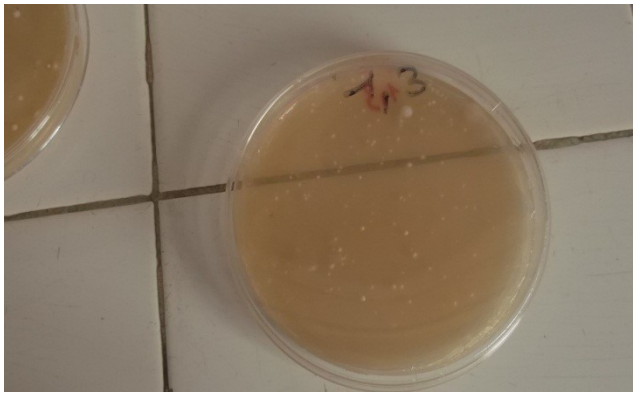
Concentration 0.1 mg/mL



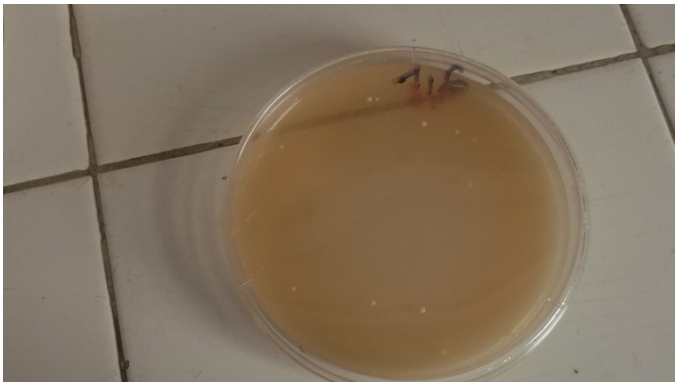
Concentration 0.4 mg/mL



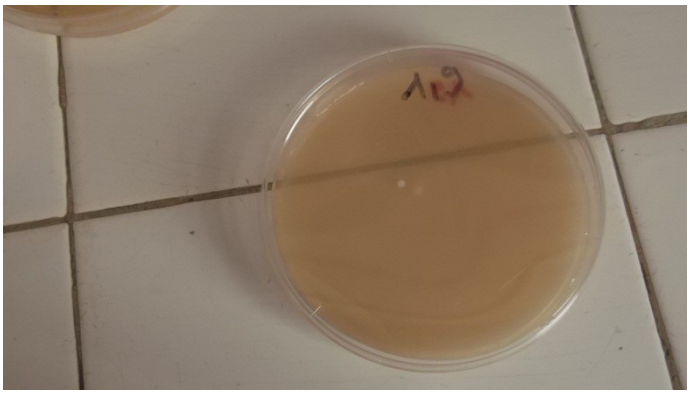
Concentration 0.7 mg/mL



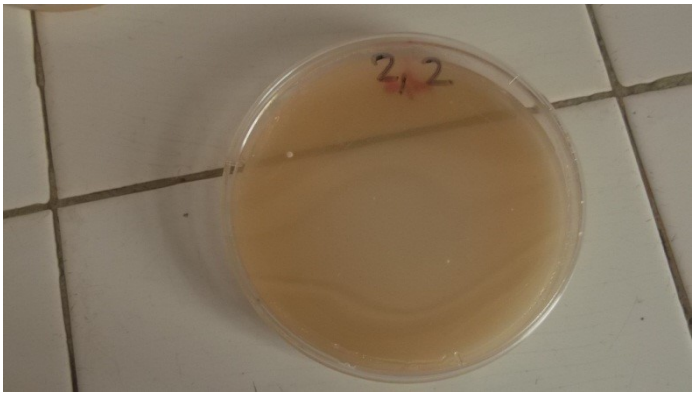
Concentration 1.3 mg/mL



Concentration 1.6 mg/ mL



Concentration 1.9 mg/mL



Concentration 2.2 mg/mL

PHOTOGRAPHS (TAKEN AT THE SAMPLE SITE) OF LEAVES INFECTED WITH COFFEE RUST