



QUANTITATIVE REAL-TIME PCR (qPCR) AND ELISA ASSAYS TO STUDY SUSCEPTIBILITY AND RESISTANCE TO APERGILLUS FLAVUS INFECTION AND AFLATOXIN ACCUMULATION IN SIX MAIZE LINES

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Introduction. Aspergillus flavus causes ear rot of maize, this infection has relevance because this fungi has the ability to produce aflatoxins, highly carcinogenic secondary metabolites which can cause serious health hazards to humans and domestic animals. Host resistance as a strategy for eliminating aflatoxin contamination of maize can be a viable approach leading to maize lines that could be safe and commercially useful.

The main objective of this research was to evaluate the resistance or susceptibility of six maize lines to infection and production of aflatoxin B1 by *A. flavus*.

Methods. In the present study 6 maize inbred lines were selected and classified on field studies as resistant or susceptible to infection by *A. flavus*. These lines were inoculated with a fungal spore suspension at the beginning of the silking through the channel of the stigmas and sampled the cob in 7-day intervals for a period of 49 days. The concentration (ppb) of AFB1 was determinate by ELISA technique. Quantification of *A. flavus* biomass was determined by quantitative real time polymerase chain reaction (qPCR).

Results. The maize lines tested showed a range of responses to inoculation with A. flavus, with aflatoxin measurements ranging from 1 to 208 ppb (Figure 1) and significant differences (P>0.05) in fungal biomass and infection coefficients that oscillated from 2 x10⁻⁵ to 2.8 x 10⁻² for resistant genotype line CML 495 and susceptible genotype line P502c1F9 respectively (Figures 2, 3).

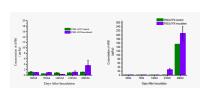


Figure 1. AFB1 accumulation in maize lines CML 495 and P502c1F9

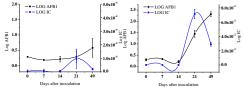


Figure 2. Correlation between fungal Biomass and AFB1 concentration in the maize lines CML 495 and P502c1F9.

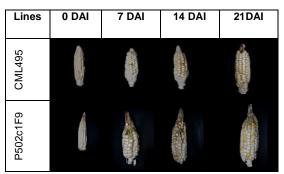


Figure 3. Maize cobs development and fungal infection after inoculation with *A. flavus*

Conclusions. qPCR using TaqMan allowed quantify infection determined both host and pathogen DNA from the same sample. The resistance and susceptibility was evaluated in two consecutive years, showing the same pattern of infection and AFB1 accumulation.

With this results was achieved to classify as resistant genotypes the maize lines CML495, DERRC2, and CML247 and susceptible genotype lines P502c1F9, DTPWC9-F76, CML 52, and CL-02510. More studies are needed to further explore the effects of defined host genes on colonization and contamination of maize by *A. flavus*.

References.

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2. Cleveland, T.E., Yu, J., Bhatnaghar, D., Chen, Y.Z., Brown, R.L., Chang, K.P., Cary, J.W., (2004).Toxicology 23; 345-380.





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