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Authors: Kanniah Rajasekaran, Ronald J. Sayler, Christine M. Sickler, Rajtilak Majumdar, Jesse Jaynes, Jeffrey W. Cary



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Control of *Aspergillus flavus* growth and aflatoxin production in transgenic maize kernels expressing a tachyplesin-derived synthetic peptide, AGM182

Kanniah Rajasekaran^{a*}, Ronald J. Saylor^b, Christine M. Sickler^a, Rajtilak Majumdar^a, Jesse Jaynes^c, Jeffrey W. Cary^a

^aFood and Feed Safety Research Unit, USDA-ARS, Southern Regional Research Center, New Orleans, LA 70124; ^bDepartment of Plant Pathology, University of Arkansas, Fayetteville, AR 72701; and ^cCollege of Agriculture, Environment and Nutrition Sciences, Tuskegee University, Tuskegee, AL 36088.

*kanniah.rajasekaran@ars.usda.gov

Highlights:

- Designed the synthetic peptide AGM182 modeled after the naturally occurring tachyplesin 1.
- AGM182 was five-times more effective in controlling *Aspergillus flavus* compared to tachyplesin 1.
- Transgenic maize plants expressing the synthetic peptide AGM182 were produced and advanced to third generation by selfing.
- Kernel Screening Assay showed significant reduction in fungal growth (72%) and spread inside transgenic kernels.
- Concomitant, significant reduction in aflatoxin levels (76-98%) was also achieved in transgenic kernels.

Abstract

Aspergillus flavus is an opportunistic, saprophytic fungus that infects maize and other fatty acid-rich food and feed crops and produces toxic and carcinogenic secondary metabolites known as aflatoxins. Contamination of maize with aflatoxin poses a serious threat to human health in addition to reducing the crop value leading to a substantial economic loss. Here we report designing a tachyplesin1-derived synthetic peptide AGM182 and testing its antifungal activity both *in vitro* and *in planta*. *In vitro* studies showed a five-fold increase in antifungal activity of AGM182 (vs. tachyplesin1) against *A. flavus*. Transgenic maize plants expressing AGM182 under maize *Ubiquitin-1* promoter were produced through *Agrobacterium*-mediated transformation. PCR products confirmed integration of the AGM182 gene, while RT-PCR of maize RNA confirmed the presence of AGM182 transcripts. Maize kernel screening assay using a highly aflatoxigenic *A. flavus* strain (AF70) showed up to 72% reduction in fungal growth in the transgenic AGM182 seeds compared to isogenic negative control seeds. Reduced fungal growth in the AGM182 transgenic seeds resulted in a significant reduction in aflatoxin levels (76-98%). The results presented here show the power of computational and synthetic biology to rationally design and synthesize an antimicrobial peptide against *A. flavus* that is effective in

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