

BEST PRACTICE

Ecofriendly bioherbicide approach for *Striga* control

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Root parasitic weeds of the genus *Striga* are a significant constraint to cereal and cowpea production in sub-Saharan Africa. They can cause total crop losses particularly during drought, in infertile soils and cereal monocropping. *Striga*

causes annual losses of US\$7 billion and affects incomes, food security, and nourishment of over 100 million people mostly in sub-Saharan Africa.

Each *Striga* plant can produce thousands of seeds, viable for over

10 years. Their intimate interaction with different host plants prevents the development of a silver bullet control technology that subsistence farmers can adopt. Hence, it is widely accepted that an integrated approach to *Striga* management is required for which biocontrol represents a crucial component.

Bioherbicide innovation

A bioherbicide is a plant pathogen used as a weed biocontrol agent (BCA), which is applied at sufficient rates to rapidly cause a disease epidemic that kills or severely suppresses the target weed. The use of biocontrol technology to manage *Striga* is a desirable control method as it is environmentally friendly, safe to farmers and crop consumers, specific to the target host, and has the potential to be



Maize/Sorghum field infested with *Striga hermonthica* (pink flowers) in Bauchi State, Nigeria. Photo by A. Elzein, IITA.

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economically viable. In addition, biological control also assists in the development of a balance of nature, the creation of more biodiversity, and sustaining of complex ecological interactions.

Since the early 1990s, a series of intensive disease surveys in many countries of sub-Saharan Africa has evaluated hundreds of microorganisms for their pathogenicity and virulence against *Striga*. *Fusarium oxysporum* Schlecht isolates have been the most promising. However, the discovery of a highly effective pathogen is only one step in the process of developing bioherbicides, for which the inoculum mass production, formulation, delivery, and storage ability must be optimized, and the mode of action, host specificity, and biosafety evaluated and fully understood.

The most widely studied and used fungal isolate that met all requirements for a potential bioherbicide for *Striga* is *F. oxysporum* Schlecht f. sp. *strigae* Elzein et Thines (isolates Foxy2 and PSM197). These are highly virulent, attack *Striga* in all growth stages—from seed to germination,

from seedling to flowering shoot; protect the current crop yield; and prevent seed formation and dispersal.

F. oxysporum f. sp. *strigae* is highly host-specific to the genus *Striga*, and does not produce any known mycotoxic compounds. Thus, its use does not pose health risks to farmers, input suppliers, traders or consumers or threaten crops or the environment. Its unique DNA constitution differs from other forms of *F. oxysporum* deposited in GenBank (www.ncbi.nlm.nih.gov/Genbank), known to cause crop diseases. Indeed, this ensures its biosafety and greatly facilitates its wider application and use as a bioherbicide.

In addition techniques for massive production of inoculum of *F. oxysporum* f. sp. *strigae* was optimized based on simple and low-cost methods and using inexpensive agricultural by-products available in sub-Saharan Africa. The chlamydospores produced by this fungus have the advantage of being able to survive extreme environmental events while still remaining viable. This is an important feature required for a BCA suited to hot and dry climatic conditions of

cereal production in sub-Saharan Africa, and to produce stable, durable, and pathogenic propagules.

Extensive research by the University of Hohenheim (UH, Germany), IITA (Benin), McGill University (Canada), and Institute for Agricultural Research - Ahmadu Bello University (Nigeria), has enhanced application of *F. oxysporum* f. sp. *strigae*, its formulation into bioherbicide products, and its delivery for practical field application. The *Striga* bioherbicide contains the *Striga* host-specific *F. oxysporum* f. sp. *strigae*, applied in massive doses to create a high infection and disease level to kill or severely suppress *Striga*.

Promotion in West Africa

The bioherbicide is a component of the IITA-led project, Achieving sustainable *Striga* control for poor farmers in Africa, funded by the Bill & Melinda Gates Foundation to intensively promote technologies to combat *Striga* in sub-Saharan Africa. The project will validate the potential of the bioherbicide seed treatment technology across major *Striga*-infested agroecological zones and maize-based farming systems, while also confirming the

biosafety and developing molecular detection tools. Here are the highlights of the results:

Technology

validation: Several multilocation trials were conducted under natural and artificial *Striga* infestation across two agroecological zones in northern Nigeria to evaluate the efficacy of *Striga* bioherbicide (*F. oxysporum* f. sp. *strigae*). The inoculum produced by UH and SUET seed company was delivered as a film-coat on maize seeds (see below). The application of the bioherbicide technology in combination with *Striga* resistant maize reduced *Striga* emergence by 73% and 39%, compared to the susceptible and resistant controls, respectively, and prevented 81% and 58% of emerged *Striga* plants from reaching flowering and 56% and 42% of the maize

plants from attack by *Striga* (see next page). The combination of bioherbicide with *Striga* susceptible variety significantly reduced *Striga* emergence by 53%, resulting in 42% reduction in number of flowering plants and in 21% increase in grain yield compared to the susceptible control.

In addition, disease symptoms were recorded on emerged *Striga* plants parasitizing maize plants coated by the bioherbicide. The reduction in *Striga* emergence across maize varieties indicates the effectiveness of the bioherbicide to attack seeds under the soil surface. The synergistic effect of the bioherbicide technology combined with the *Striga* resistant maize is expected to reduce the *Striga* seedbank and thus the impact of *Striga* on subsequent maize crops.

Biosafety: To further ensure the safety of *Striga* BCA and to demonstrate and increase awareness among farmers, regulatory authorities, and stakeholders, a wide host range study was carried out using 25 crops in collaboration with IAR-ABU and the Nigerian Plant Quarantine Service (NPQS) under field and greenhouse conditions in Nigeria. Results revealed that none of the test plants showed any infection by the biocontrol agent both in the field and greenhouse, and no detrimental growth effects were measured or visual losses to plant health recorded in any of the inoculated crops tested, i.e., inoculation with the *Striga* BCA did not cause any delay in emergence, and a decrease in plant height, plant vigor, chlorophyll content per leaf, shoot fresh and dry weight.



The delivery of *Striga* bioherbicide as a film coating onto resistant maize seeds using a professional seed treatment technology: Maize film-coated seeds with chlamydo spores of *F. oxysporum* f. sp. *strigae* isolate PSM197 (1, red), isolate Foxy2 (2, yellowish), with *Arabic gum* (3, green); and untreated control (4, yellow). Photo by A. Elzein, IITA.

Hence, the Nigerian regulatory authorities (NPQS, NAFDAC) and other stakeholders were satisfied and confident that no disease was produced on plants other than *Striga* by the BCAs and that it is safe to use. In addition, a mycotoxin produced by *Striga* bioherbicide *F. oxysporum* f. sp. *strigae* was analyzed and evaluated by our project partner, the University of Stellenbosch in South Africa. An evaluation of existing isolates of *F. oxysporum* f. sp. *strigae* does not produce well-known mycotoxins (e.g., Fumonisin and Moniliformin) that pose a threat to animal or human health. This finding further confirms the safety of this bioherbicide.

Molecular detection

tools: Development of a monitoring tool specific to the *Striga* bioherbicide is important to certify inoculum quality, monitor the presence and persistence of the BCA in soils, and validate its

Effect of the bioherbicide on Striga emergence at Gar-Alkali in Bauchi state in 2012: Bioherbicide was applied as a film-coat on maize seeds using Arabic gum as adhesive carrier and professional seed coating technology. Photos by A. Elzein, IITA.



Untreated maize susceptible variety - control.



Untreated maize resistant variety - control.



Bioherbicide combined with the maize resistant variety.



Typical disease symptoms on *Striga* shoots caused by the bioherbicide coated on maize seeds at Gar-Alkaleri in Bauchi state in 2012: the first infection occurred on the tip of the oldest leaves, which turned black and curved; the symptoms later on were extended to younger leaves and growing points as well which were wilted and changed to black. Photo by A. Elzein, IITA.

environmental biosafety. UH is developing a monitoring tool.

The AFLP fingerprinting technique was successfully used in developing a primer pair capable of differentiating the *F. oxysporum* f. sp. *strigae* group from other *Fusarium* species. In addition, the monitoring tool has shown a high specificity for isolate Foxy2 and was used to

monitor its spread and persistence in rhizobox experiments under different management practices using Kenyan soils. This promising result provides a proper baseline to further the existing primer set.

Bioherbicide + pesticide technology:

The novel combination and integration of the bioherbicide technology plus imazapyr herbicide for *Striga* control with pesticides in a single-dose seed treatment to control fungal pests offers farmers with maize seed that is able to achieve its yield potential. The use of each technology (BCA or imazapyr) has been shown to be effective when applied independently using seed coating techniques, but have not been integrated.

The compatibility of *Striga* BCAs with different pesticides (herbicides and fungicides with insecticide components) was studied in vitro in the laboratory. *Striga* BCAs showed excellent compatibility with imazapyr (a herbicide seed coating used in combination with IR maize to control *Striga*), Metsulfuron Methyl (MSM) (a herbicide seed coating developed

by DuPont to control *Striga* in sorghum), and glyphosate (an intensively used herbicide). A similar result was also achieved with the commonly used seed treatment fungicides at the recommended application doses. Accordingly, doses and complementary seed coating protocols for the three compatible technologies (BCA, herbicide, and fungicides) have been developed and IR maize seeds were successfully coated with a single-dose seed treatment of BCA inoculums and imazapyr. The results showed that imazapyr did not interfere with the BCA during seed coating, with BCA growth and sporulation after coating, and with IR maize seed germination. Seeds of IR maize varieties can thus be coated with the herbicide and the BCA and then fungicide and delivered to farmers using the same input pathway. Screenhouse and field trials are being carried out to generate data on the combined efficacy of the applied technologies. The demonstrated compatibility of *Striga* BCA with the different pesticides that contain a wide range of active

ingredients indicate that the combination and delivery of the *Striga* bioherbicide technology with a large number of pesticide products is possible. These findings are expected to provide a triple action seed coating package for direct control of *Striga* and fungal diseases of maize in sub-Saharan Africa.

Suitability to African farming systems

Our strategy for scaling-up the bioherbicide innovation is based on using technology appropriate to Africa to ensure that sustained production of the bioherbicide is feasible at a cost affordable to African small-scale farmers. The seed-coating treatment requires significantly less inoculums, establishes the BCA in the cereal rhizosphere, i.e., the infection site of *Striga*, and provides a simple, practical, cost-effective delivery system for adoption by input suppliers to subsistence farmers. Arabic gum as a coating material has been shown to increase the rate of mycelia development and enhance BCA sporulation. Its availability in sub-Saharan Africa at a low price is an additional economic advantage. A

commercial seed coating process, developed and optimized at UH with SUET Seed Company in Germany, is being transferred and adapted at IITA, Ibadan, to be used as an experimental production unit for capacity building and as a model for eventual transfer of seed treatment technology to the private sector after validation.

Outlook

One unique advantage of this bioherbicide is that the ability of *Striga* to become resistant to it is virtually unknown as a consequence of the suite of enzymes and secondary metabolites that the BCA produce to become pathogenic and virulent against the target (*Striga*). Hence after validation, delivering the bioherbicide technology in combination with resistant maize or with the herbicide imazapyr is expected to increase efficacy in controlling *Striga*. Bioherbicide and other compatible technologies have different modes and sites of action against *Striga*, and in a combination they will have a much greater chance of reducing the potential risk of development of resistance to a single technology (resistant

varieties or herbicides) used separately and repeatedly.

The potential delivery of coated seeds of resistant maize with bioherbicide in one package to farmers using the same input pathway will reduce transaction and application costs and enhances the economic feasibility and adoptability of the technologies. Similarly, compatibility of BCA with imazapyr and fungicides allow seed coating of IR-maize with bioherbicide, imazapyr, and fungicides with a single-dose seed coating application.

Future plans

Currently, large-scale field testing is ongoing and is being implemented to further validate bioherbicide efficacy across two agroecological zones where the common scenarios for maize infestation by *Striga* in northern Nigeria are represented. For understanding of farmers' preferences and perceptions, socioeconomic analysis and cost-benefit analysis of bioherbicide technology based on field data/surveys and interviews, current market information, and links with other *Striga* control strategies will be undertaken. After

validation, dissemination and commercialization will be promoted through private sector partnerships and integrated with other control options such as resistant varieties, IR varieties combined with seed treatment with imazapyr, crop rotation with legumes, and soil fertility management practices, to achieve sustainable management of *Striga*.

Partners

IITA (Dr F. Beed, Dr A. Elzein & Dr A. Menkir), Institute for Agricultural Research – Ahmadu Bello University (Dr A. Zarafi), Nigeria; University of Hohenheim (Prof G. Cadisch, Dr F. Rasche & Prof J. Kroschel), Germany; The Real-IPM Company Ltd (Dr H. Wainwright), Kenya; University of Stellenbosch (Prof A. Viljoen), South Africa; and McGill University (Prof A. Watson), Canada.

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