Weevil resistant sweetpotato through biotechnology: the weevil is still winning…

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Unfortunately, weevils continue every year to damage about a quarter of the sweetpotato harvest in countries like Uganda and alter the quality and safety of storage roots. A long effort to express Cry proteins that would kill weevil larvae in the infected storage root has not resulted in obtaining fully resistant sweetpotato varieties. A new strategy based on the production of small RNA to weaken specific genes of the weevils to block their development has been developed but lacks the final proof of concept in the sweetpotato crop. It is clear that a single solution is not at hand from the on-going biotech and breeding efforts. Options for the way forward are discussed.

Fig. 1 The Evil Weevil, Cylas puncticollis (credit M. Ghislain)

Weevils threaten food security of the poorest

A farmer survey conducted in Uganda revealed that weevils are responsible for 28% of crop losses every year. Losses can be up to 90% during dry periods, which can be quite devastating (Fig. 1). Weevils can affect not only food security, but also sweetpotato production, marketability, healthiness, and sustainability, especially in areas experiencing longer dry periods. With weather extreme predictions for Sub-Saharan Africa (SSA) foreseeing an expanding dry season, the threat and impact of weevils may increase further. Adapting conventional integrated pest management practices among smallholder farmers is difficult due to the challenges controlling field sanitation in small-scale production systems. Extensive efforts to develop weevil-resistant sweetpotato through conventional breeding have not yet succeeded but have led to the identification of a repellent compound in moderately resistant sweetpotato variety New Kawogo grown in Uganda. As a result, there is currently little farmers can do when weevils infest their fields, other than quickly harvest and salvage what is left of their crop.

What do we want to achieve?

The aim of this project is to develop weevil-resistant sweetpotato varieties through breeding and biotechnology. Bacillus thuringiensis (Bt) is a soil bacterium that is well-known for its insecticidal activity. Synthetic Bt genes that produce the proteins active against the two weevil species attacking the sweetpotato were introduced into the plant but failed to confer complete pest resistance.

Fig. 2 The three African PhD students (Star) with the Extended Weevil Resistance Biotech team

Because the storage root has low protein content, a non-protein-based approach (RNA interference - RNAi) is targeting essential genes of the weevils and can be combined in the future with the insecticidal proteins. This Bt – RNAi combined gene technology has already been used successfully to increase resistance to rootworm in maize. Other proteins and metabolites with higher anti-weevil activity including repellent compounds are under development. Combining the most effective tools will result in protection of the storage roots from devastating weevils. In addition, health benefits may be expected because farmers will not consume partially damaged roots containing toxic compounds as they do currently under severe food shortage.

How are we working with partners?

Research on the identification of insecticidal proteins from Bt (Cry proteins) has taken place in the US at the Auburn University and at the National Crops Resources Research Institute (NaCRRI) in Uganda. Genetic transformation of sweetpotato was first developed at the CIP biotechnology lab in Peru, later at Makerere University and NaCRRI in Uganda, then at BecA and Kenyatta University in Kenya, and lately at the Donald Danforth Plant Science Center in the USA. A confined field trial has been conducted at the University of Puerto Rico Mayaguez. The University of Valencia elucidated the mode of action of the Cry proteins and Ghent University builds mainly biosafety capacities and is developing the RNAi strategy. AgBiome, (USA) is collecting soil and sweetpotato samples to sequence microbes to develop anti-weevil biological(s). North Caroline State University in partnership with the University of Greenwich (UK) and the National Agricultural Research Organisation (Uganda) are characterizing hydroxycinnamic acids as repellent and associated SSR markers in genetics and breeding materials. Building on the successes of the Attract-and-Kill technology to control potato tuber moth, CIP entomology team is now evaluating the same to control sweetpotato weevils. The project is targeting Uganda and, if successful, other SSA countries.

What have we achieved so far?

During these eight years of intense research, human capacities were developed with 4 PhD (3 women, 1 man) of which 3 were African (Fig. 2).

We have introduced synthetic cry genes that produce Cry proteins with activity against sweetpotato weevils into various sweetpotato varieties, including some grown in SSA. Three cry gene combinations and three promoters have been tested in a total of 132 transgenic events. Bio-assays using storage roots infested by oviposition from female adults resulted in few transgenic events with apparent differences with the non-transgenic materials but none of them turned out after a second and third bioassay to be fully resistant to weevils. Early attempts to detect and quantify Cry proteins in storage roots yielded results indicating low level of accumulation of the Cry proteins. Thus, we conclude that the combination of low level of expression and level of activity of the Cry proteins have not been high enough to confer full resistance. The non-protein based strategy, the anti-weevil RNAi strategy, was developed after the identification of 3 target genes that have given good mortality results for both weevil species in both soaking and artificial diets: proteasome 20 kD subunit, ribosomal protein S13e and snf7 genes. Five hairpin gene constructs were designed based on Prot20kd and snf7 from Cylas puncticollis (Cp) and C. brunneus (Cb) in single and double combinations. Transgenic events are being produced and should be screened for high expressers of small RNA and then storage roots produced and tested at the BecA facilities for activity against weevils.

What are the next steps?

The conclusion of the Bt strategy without success clearly points at the need to identify new anti-weevil proteins with higher activity than the Cry proteins tested. In addition, a non-protein based strategy such as the anti-weevil RNAi targeting essential genes and identification of metabolites with direct or repellent activities against weevils must be pursued. Hence, new efforts to control the weevil pests must focus on the most promising strategies: anti-weevil gene RNAi, microbial anti-weevil proteins and metabolites including repellent compounds such as hydroxycinnamic acids. Weevils remain the single most important threat on sweetpotato food availability to the poor in many SSA countries. We should not let them win!

CONTACT
Marc Ghislain (CIP)
m.ghislain@cgiar.org