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HOST-PARASITE INTERACTIONS THROUGHOUT THE *STRIGA* LIFE CYCLE, AND THEIR CONTRIBUTIONS TO *STRIGA* RESISTANCE

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ABSTRACT

Field screening for resistance to *Striga* has been slow and difficult, with only modest success. Successful *Striga* parasitism is dependent upon a series of chemical signals produced by its host. Interruption of one or more of these signals results in failure to establish parasitism. We have embarked upon a programme of characterising these signals, developing simple laboratory assays for them; identifying genotypes which are resistant because they produce abnormal levels of signals; elucidating the mode of inheritance to genotypes with high yield potential and broad adaptation. Focusing on the initial stage, the signal required for *Striga* seed germination, our collaborative research work in sorghum has resulted in the development of elite sorghum genotypes which combines *Striga* resistance with yield potential and good quality characteristics. These germplasm are currently under wide international testing in several African countries. We hope to follow this pattern for each host-dependent step in the development of *Striga* plants, then pyramid the resulting resistance genes into a single genotype which should exhibit durable, broad-based resistance.

Key Words*: Host-parasite interactions, *Striga

RÉSUMÉ

La recherche de facterurs de résistance contre le *Striga* est lente et difficile et donne peu de résultats. Le succès d'une infection par le *Striga* dépénd de toute une série de stimulus produits par son hôte. La suppression d'un ou plusieurs de ces stimulus cause un échec du parasitisme. Nous avons établi un programme de recherche pour caractériser ces stimulus, développer un simple test de laboratoire, identifier les genotypes qui sont résistants parce qu'ils en produisent un niveau anormal, examiner l'hérédité des caractéristiques qui sont responsable de la résistance, et transférer cette résistance dans des génotypes qui ont un haut rendement et une large adaptation. Au stade initial, nous nous sommes concentrés particulierement sur les stimulus qui sont nécessaires pour la germination des graines de *Striga*. Notre groupe de recherche a developpé des genotypes élites de sorgho qui combinent la résistance contre le *iga* avec un haut rendement potentiel et une bonne qualité nutritive. Ces génotypes sont maintenant testés internationalement dans des différents pays d'Afrique. Nous poursuivons notre recherche de cette manière afin de développer des résistance au *Striga* pour chaque stade qui est hôte-dépendant. Après, tous les gènes résistants seront recombinés dans un seul génotype qui devrait avoir une résistance large et durable.

Mots Clés*: Entr'action hôte-parasite, *Striga

INTRODUCTION

Striga may have become one of the greatest biological constraints to food production in the drier parts of Africa, probably a more serious agricultural problem than insects, birds, or plant diseases. These parasitic weeds also pose a serious problem in India and other parts of the world where sorghum (*Sorghum bicolor* (L) Moench), pearl millet (*Pennisetum glaucum* (L) R.Br), maize (*Zea mays* L.), rice (*Oryza sativa L.*), cowpeas (*Vigna unguiculata* (L) Walp) and sugarcane (*Saccharum spp.)*, are widely cultivated. Yield losses from damage by *Striga*, on crops grown under heavy infestation, are significant. Estimates vary from 10–70% depending on crop cultivar and degree of *Striga* infestation (Lagoke *et al.*, 1991; Doggett, 1984). In many places in Africa, the *Striga* problem has reached epidemic proportions, presenting a desperate situation to small subsistence agriculture in these areas.

Many cultural, chemical and biological methods have been suggested as control measures for *Striga*. Yet none has been found to be highly effective or economically feasible when evaluated under subsistence agriculture prevalent in semi-arid Africa. Combining several approaches may be necessary to reliably curtail the *Striga* epidemic. Host plant resistance is central to any effective *Striga* control strategy, and is perhaps the most practical and economically feasible means for lessening the damage caused by *Striga* infestation. Progress in breeding for *Striga* resistance in African crops has been rather limited, for reasons varying from complexity of the trait (Ejeta *et al.*, 1991) to lack of research support as well as the lack of a functional and rational approach to selection strategy.

At Purdue University, we have established an interdisciplinary *Striga* research programme devoted to developing new approaches based on a growing understanding of the basic biology of the intricate relationships between *Striga* and its hosts (Ejeta *et al.*, 1993). The purpose of this report is to briefly describe our approach and to incite reactions and create greater awareness to the challenges and exciting opportunities for alleviating the *Striga* problem.

RESEARCH RATIONALE

We have made the characterisation of mechanisms of resistance the focal point of our effort to develop resistant crop genotypes. The central theme of our effort is around the basic biology of the elaborate host-parasite interactions leading to the life cycle of *Striga* or successful parasitism (Fig. 1). Mechanisms of resistance are defined on the basis of host-dependent developmental processes, and the essential signals exchanged between *Striga* and its host. Our working hypothesis is that the complex trait of resistance can be dissected into simpler components based on increased understanding of these specific signals involved at each stage of the life cycle.

Because it is an obligate parasite, interactions between *Striga* and its host plant play a crucial role in the survival of the parasite. Hence, disrupting these interactions offers unique opportunities for controlling *Striga* by multiple interventions through its life cycle (Fig. 1). Our overall plan is to identify crop genotypes in which the host-parasite interaction does not develop normally, at each of the different stages of parasitism.

Implicit to our working hypothesis and our overall strategy are the following simple but valid assumptions derived from cumulative knowledge of host-parasite biology and its interaction with the environment:

1. *Striga* resistance is a complex trait controlled by a number of genes and field selection for resistance is often confounded by environmental influences.

2. *Striga* is an obligate parasite of tropical grasses and legumes and through co-evolution with these crops has developed intricate inter-relationships involving exchange of vital signals.

3. Many of these signals are of a chemical nature and unusually low production of these signals by the host could lead to resistance.

4. Characterisation of these signals could lead to development of appropriate laboratory essays for screening genotypes.

5. Genetic variation in crop genotypes, for each of these signals essential for successful parasitism, exists in nature or can be artificially created.

6. Host-plant resistance to *striga* as a result of

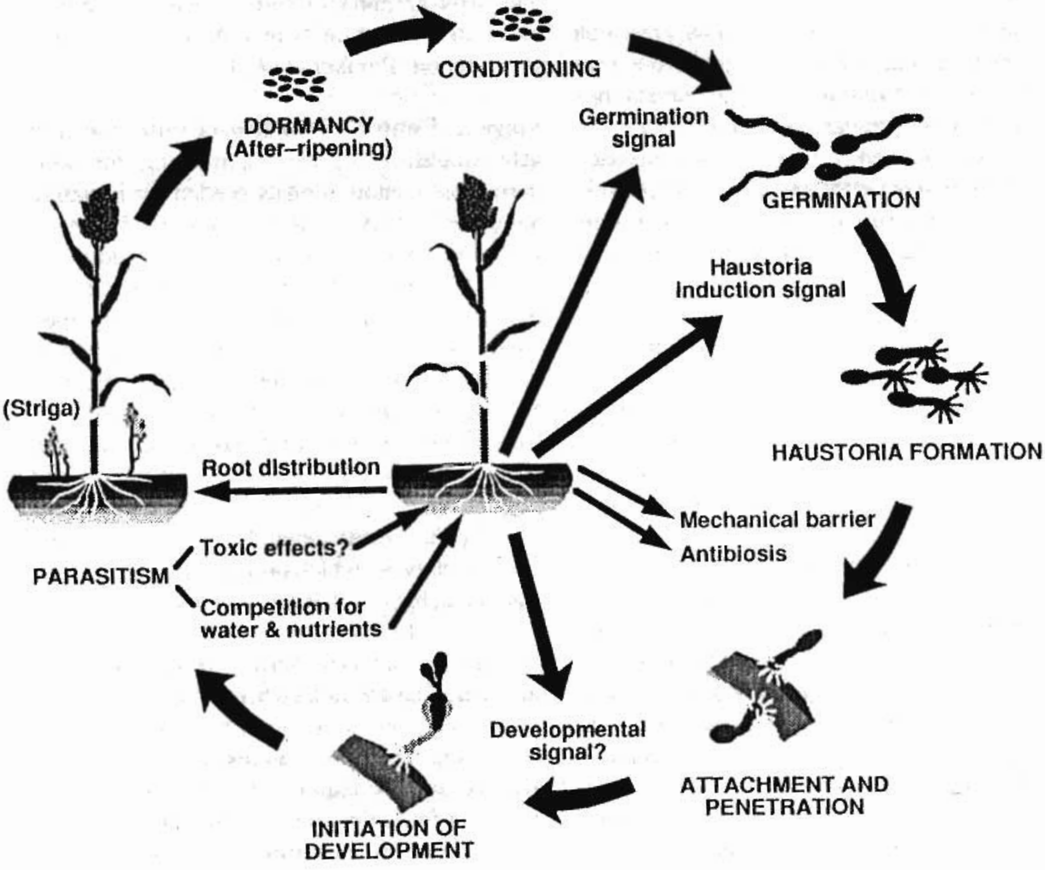

Figure 1. Life cycle of *Striga spp.* and potential mechanisms for host-plant resistance

disruption in one critical signal exchange is likely to be simply inherited.

7. Resistance to *Striga* may also be due to chemical defense mechanisms, morphological barriers or other mechanisms conferring tolerance.

8. Crop germplasm can be catalogued on the basis of their mechanisms of *Striga* resistance based on the signal disrupted or defense mechanisms identified.

9. With availability of appropriate assays and genetic differentials, conventional plant breeding technologies can be used to combine more than one mechanism of resistance into one genetic background.

10. Resistance to *Striga*, resulting from a combination of mechanisms thus developed, would be more durable and stable across ecological zones and *Striga* strains than single gene resistance sources currently available.

RESULTS

Using this approach, our interdisciplinary research team has made considerable progress, We have documented much of this in several publications including our new research bulletin (Ejeta *et al.*, 1993) where detailed coverage of our approach and the results of our efforts are given. We provide below an abridged summary of some of our results to help illustrate the functionality of the approach we have proposed.

Stage 1. Control of *Striga* germination. Low production of host plant root exudate compounds that are essential for *Striga* germination has been the best understood mechanism of resistance to *Striga* (Lynn and Change, 1990). Most of our recent-past effort has also focused on this mechanism of resistance.

Using our unique approach described above, we have fully exploited low production of germination stimulants in selecting for *Striga* resistance in sorghum. We have identified and characterised several host root-produced chemical signals of two different types required for *Striga* seeds to germinate (Netzly *et al.*, 1986; Slate *et al.*, 1993). We developed simple methods to screen host genotypes for the production of germination signals (Hess *et al.*, 1992) and have identified sorghum genotypes that are resistant because they produce limited amounts of the signals (Hess *et al.*, 1992; Weerasuriya *et al.*, 1993). We have determined the mode of inheritance for low germination stimulant production (Vogler, R. K., Ejeta, G. and Butler L. G., unpublished), and have incorporated this resistance-conferring trait into improved sorghums for use in *Striga*-infested areas (Ejeta *et al.*, 1993). International testing of these newly derived *Striga* resistant sorghums for broad adaptation and utilisation is currently being undertaken.

We have also began the molecular characterisation (Melake Berhan, 1993) of the genes conferring *Striga* resistance, using RFLP (Restricted Fragment Length Polymorphism) and RAPD (Randomly Amplified Polymorphic DNA) technologies. Our work confirmed earlier reports (Jackson and Parker, 1991; Logan and Steward, 1991) that the ultimate signal for *Striga* germination is ethylene, and identified various chemical compounds, including the cotton defoliant thidiazuron in mixture with herbicides such as 2,4 D, which stimulate ethylene production and *Striga* germination and which may be used to clean up infested fields by induction of suicidal germination (Babiker *et al.*, 1993).

Stage 2. Control of haustoria initiation and attachment. While the requirements for host-derived exogenous signals needed for initiating *Striga* germination has been known for many years, evidences for the existence of an additional signal from the host root to produce specialized rootlets for attachment of host roots have emerged only in the last few years. A number of compounds have been shown to function as haustoria initiators in *Striga* but the active signals from host roots have not yet been identified. We are developing a quantitative assay for production of this second host-derived developmental signal. Preliminary work by Fasil Reda (unpublished) has identified maize genotypes which produce low amounts of this signal, but they have not yet been field tested. Host genotypes which encourage *Striga* germination but lack other haustoria initiation signal may have the added benefit of depleting the *Striga* inoculum in addition to producing crop *Striga*-free. Our observations, to date, indicate that the signals required for germination and haustoria formation are inherited and produced independently by host roots, thus upholding our working hypothesis.

Stage 3. Control of attachment and penetration. As early as the 1930s, host plant resistance mechanism based on mechanical barriers which impede invasion of cortical cells by haustoria, thickened inner walls of the endodermal cells, and hardened vascular cylinders of host roots have been described (Sanders, 1933). Matti *et al.* (1984) have since determined that certain *Striga* resistant sorghums obstruct haustoria attachment to their roots because of liquefied pericycle cells, and endodermal cells that thicken with silica deposits. Associated with these physical influences, chemical defense mechanisms which regulate host development and metabolism (antibiosis) may be present at this stage.

Oliver *et al.* (1991) evaluated resistant and susceptible sorghum genotypes cytochemically and suggested some evidence for some cellulolytic enzymes from haustoria accompanied by secondary thickening of roots of resistant genotypes but not susceptible varieties. While relatively little is known about this mechanism of resistance, the probable role for chemical regulation of attachment and penetration provides opportunity for developing a quicker assay than slow histochemical evaluations which do not allow screening of crop germplasm in large numbers.

Stage 4. Control of *Striga* differentiation. Post-attachment events involving penetration of haustoria to the host root vascular tissue and subsequent normal development of the parasite also provide opportunities for disrupting the life cycle, and thus allow avenue for resistance to *Striga* parasitism. We have recently shown that additional host-derived signals are required for *Striga* seedlings, once attached to host roots, to develop normally into shoots (Cai *et al.*, 1993). We are developing an assay for these signals using *Striga* cells grown in tissue culture. Cultures of *Striga* cells are also being used to screen extracts of host plants for factors which inhibit elongation and/or proliferation of *Striga* cells, possibly preventing invasion of host root by the parasite.

Stage 5. Control of host growth and development. Host damage due to *Striga* results in stunting, chlorosis and wilting, symptoms that appear even before emergence of the *Striga* plants. Such symptoms have suggested a possible involvement of toxins produced by *Striga* that are transported to the host plant metabolic machinery. These observations prompted Efron *et al.* (1988) to inject crude extract of *Striga* into the stem of maize seedlings resulting in necrotic symptoms at a distance from the injection point. We have also found that crude extracts of *Striga* leaves and stems can induce loss of chlorophyll and wilting of susceptible sorghum plants. These observations, at best, provide anecdotal evidence that *Striga* may produce toxic compounds which have negative effect in the development of host plants. We also find that host genotypes differ in their ability to tolerate *Striga* toxins. We are currently using the *Striga* toxin as a screen for identifying clones of *in vitro* cultured sorghum cells with enhanced resistance to the toxin. Such an approach may result in host plants with enhanced tolerance which when used in combination with genes for resistance (as in SRN39, our *Striga* resistant sorghum cultivar) may provide for a good source of crop cultivar.

Root growth habit of host plants (Cherif Ari *et al.*, 1990) and premature haustoria formation (Reopel and Timko, 1992) have also been implicated as avoidance mechanisms which can be lumped to Stage 5 as they tend to contribute to factors affecting the normal development of host plants. It is interesting to note that the same sorghum genotype, P-967083, has been found to have limited root length density (Cherif Ari *et al.*, 1990) to describe its mechanisms of resistance to *Striga*.

SUMMARY

We have made the exploitation of the intricate relationship between *Striga* and its host plants the focal point of our effort in the development of crop varieties with resistance to *Striga*. We have developed an interdisciplinary research agenda based on the premise that with increased knowledge of the nature of host-parasite signals exchanged, appropriate and rapid assays can be developed, germplasm can be separated, and gains from selection can be speeded up.

Our initial focus has been on the control of *Striga* germination via essential host-produced chemical signals. We have identified and characterised these signals, used this knowledge to develop appropriate assays, and, with the use of this rapid assay identified and/or confirmed sources of *Striga* resistance, but more significantly transferred resistance genes into agronomically superior sorghum lines. We have also used this assay in genetic studies including establishing mode of inheritance, as well as undertaking molecular marker studies for genetic mapping and market assisted selection. We hope to apply this approach to subsequent stages of signal exchange between *Striga* and its host with similar success.

Our goal is to expand our *Striga* research programme utilising modern biotechnology based on genetics, physiology, biochemistry, and molecular biology for the development of crop cultivars with durable resistance to *Striga*.

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