IPM CRSP Trip Report for L. Strathie & A. McConnachie to Ethiopia, 2009

Country(s) Visited: Ethiopia

Dates of Travel: 17-23 December 2009

Travelers Names and Affiliations: Ms Lorraine Strathie and Dr Andrew McConnachie, Agricultural Research Council – Plant Protection Research Institute, Private Bag X6006, Hilton 3245, South Africa

Purpose of Trip: To participate at the planning & training meeting of the USAID IPM CRSP Parthenium project in Ethiopia from 19-22 December 2009. Specific objectives of the trip were to:

(i) provide training on parthenium distribution surveys
(ii) provide training on biological control research and field implementation techniques, and the development of monitoring and evaluation techniques, with particular reference to parthenium biocontrol
(iii) to visit the weed biocontrol facility and view research at EIAR PPRC, Ambo
(iv) to visit potential field sites for mass-rearing of parthenium biocontrol agents

Sites Visited: (*locations within countries: institutions, cities, villages, or regions*)
Ethiopian Institute of Agricultural Research, Ambo
Addis Ababa
Addis Ababa to Sodore

Description of Activities/Observations:
All trip objectives were achieved.

Itinerary:
17 December: Travel from South Africa to Addis Ababa, Ethiopia
18 December: Visit Ethiopian Institute of Agricultural Research Plant Protection Research Centre weed biocontrol facility at Ambo. Tour of facilities and discussions with researchers and technicians on parthenium biocontrol research, and rearing and testing techniques for *Listronotus setosipennis*.
19 December: Field visit to Sodore area (assess potential mass-rearing sites)
20 December: Prepare presentations; travel from Addis Ababa to Ambo
21 December: Planning & training workshop of the USAID IPM CRSP parthenium project (Ambo Abebech Hotel)
22 December: Planning & training workshop of the USAID IPM CRSP parthenium project; visit to EIAR PPRC weed biocontrol facility, Ambo and training of research technicians. Travel from Ambo to Addis Ababa.
23 December: Travel from Addis Ababa to South Africa

Ms L. Strathie gave oral presentations on:
(i) Culturing and host-specificity testing of insect agents for parthenium
(ii) Overview of release and evaluation general principles
(iii) Growing parthenium in greenhouses

Dr A. McConnachie gave oral presentations on:
(i) Methodologies for collection of distribution data, mapping and predictive modeling
(ii) Biological control of weeds in South Africa and importation (quarantine) protocols
(iii) Mass-rearing of *Zygogramma bicolorata*
(iv) Evaluation of the impact of released agents

A second phase of the IPM CRSP parthenium project runs from 2010-2014. The focus of the project has shifted to Eastern Africa and the project is entitled 'Abating the Weed Parthenium (*Parthenium hysterophorus* L.) Damage in Eastern Africa using Integrated Cultural and Biological Control Measures'. Kenya and Tanzania are new participating countries in this phase.

EIAR has received permission to release the leaf-feeding beetle *Zygogramma bicolorata* (Coleoptera: Chrysomelidae) from their Ministry of Agriculture, following host-specificity testing and an evaluation process, but must follow the official process to receive permission from USAID before the biocontrol agent can be removed from quarantine or releases can begin. Particular emphasis, training and discussion during this workshop was given to the field implementation phase (insect and plant culturing techniques, mass-rearing, releases, evaluation) of the biocontrol component of this project, as well as training of new project participants (Tanzania, Kenya) in the methods for conducting parthenium distribution surveys. Planning for the forthcoming year's activities was undertaken.

The EIAR PPRC weed biocontrol facility was visited by L. Strathie and A. McConnachie; discussions were held with entomological researchers and technicians, and the parthenium biocontrol facilities and research were viewed and discussed. Discussions were held with the project coordinator regarding the involvement of PPRI in the 2010-2014 phase of the project.

There was some debate around the host-specificity of *Z. bicolorata* during the workshop, based on experience in India of spill-over feeding on sunflower in the field. This was resolved and it was concluded that this should not be problematic for Ethiopia as it has been demonstrated that *Z. bicolorata* was not a pest of sunflower in India. Sunflower is not a major crop in Ethiopia either.

The role of gender in the collection of project data was discussed in a presentation by Dr Y. Chiche, EIAR. Gender roles in intra- and inter-household activities should be analysed. Consideration should be paid to the impact of parthenium on productivity; knowledge and perception of women/men in different activities of weed management (who is pulling, burning, transporting weed); trend analysis (what is the understanding of parthenium invasion); pairwise ranking on the problem identification; how to enhance role of women in management of parthenium.

Dr L. Nigatu reported that surveys conducted by Haramaya University in eastern and western Haraghe demonstrated that plant species diversity decreased in grazing lands due to parthenium. Competitive plant studies were conducted using *Panicum*, *Cenchrum*, *Chloris*, *Bothriocloa* and *Cynodon*; *Chloris* and *Cynodon* were the best competitors against parthenium. These are currently being investigated in field trials. Studies conducted by Dr Nigatu have shown that parthenium infestations resulted in reduced carrying capacities, and the expansion of parthenium into crop fields (e.g. sorghum) increased the cost of cultivation and had a substantial impact on crop yield.

Dr K. Zewdie reported that in Central Ethiopia, 75% of farmers surveyed in that region considered parthenium a serious problem. The main methods used to control parthenium are hand-pulling, slashing and burning. K. Zewdie reported that there was a collection of parthenium by Haramaya University in 1968; there may be parthenium specimens from an earlier date in Addis Ababa University.

Dr J. Biskwa reported that parthenium has been found in 12 districts in Uganda – Busia, Namutuba, Bugiri, Mbale, Jinja, Mbarara, Ibanlla, Masaka, Kampala, Kabale, Kases. Infestations are present in the north-east area above Lake Victoria. Parthenium is being used for decoration in the florist industry. There is a large infestation of parthenium in no-man's land at the Busia border, just across the border from Kenya.

Dr E. Wabuyele reported that the East African herbarium in Nairobi is a regional herbarium, and that 10 specimens of *P. hysterophorus* are lodged there, 6 of which are from Kenya, and the others from Madagascar, Zimbabwe, western Indian Ocean region. Parthenium is reported to have appeared in Kenya around 1973. The first known collection was in 1975 by J. Cordingly, when resistance to herbicides in a coffee farm were reported. 6 other specimens were collected in the same general area (Kiambu district – coffee plantations; close to Nairobi) by farmers who took them to the herbarium for identification. Recent sight records of parthenium in western Kenya by East African herbarium staff suggest a wider distribution than currently recorded. Herbarium records indicate the presence of parthenium in Kenya for about 4 decades, but it has been under-recorded so its current distribution is unknown. Other than a 2009 record of parthenium around Arusha, K. Clark reported that there are currently no other records of parthenium for Tanzania; no surveys have been undertaken there yet.

Training Activities Conducted:

Program type (workshop, seminar, field day, short course, etc.)	Date	Audience	Number of Participants		Training Provider (US university, host country institution, etc.)	Training Objective
			Men	Women		
Visit	18 Dec 2009	Ethiopian Institute of Agricultural Research PPRC Ambo weed biocontrol staff	3	2	Organized by Virginia State University and Ethiopian Institute of Agricultural Research	Visit EIAR PPRC Ambo weed biocontrol quarantine facility and parthenium research activities; advise staff on parthenium biocontrol
Field visit	19 Dec 2009	IPM CRSP project participants	5	1	Organized by Virginia State University and Ethiopian Institute of Agricultural Research	Field visit to Sodore area to view potential field site for mass-rearing of parthenium biocontrol agents
Workshop	21-22 Dec 2009	IPM CRSP project participants	27	5	Workshop organized by Virginia State University and Ethiopian Institute of Agricultural Research	IPM CRSP Partners planning and training workshop

Suggestions, Recommendations, and/or Follow-up Items:

Recommendations from visit to EIAR PPRC Ambo weed biocontrol facility

A set of recommendations (appendix 1) were compiled by L. Strathie and A. McConnachie following a visit to the weed biocontrol quarantine facility and research programme at EIAR PPRC Ambo on 18 December. Recommendations for improved quarantine procedures, quarantine facility integrity, and *L. setosipennis* culturing techniques were compiled. Parthenium biocontrol research activities were found to be progressing well in general, with a few suggested modifications to protocols and techniques. Staff retention within the EIAR PPRC parthenium biocontrol project is vital for the success of the biocontrol component of the project, to maintain the knowledge base of expertise and transfer technology to new participants for expansion and field implementation of parthenium biocontrol in Ethiopia.

Recommendations from field visit to Sodore area:

On 19 December 2009 a field visit was conducted to the east of Addis Ababa as far as Sodore in the Rift Valley, to an area that was purported to be suitable for the establishment of a pilot mass-rearing centre for *Z. bicolorata*. The area there and en route was found to be unsuitable by PPRI researchers for the establishment of a parthenium biocontrol mass-rearing centre due to the arid conditions of the region. While parthenium was widespread along roadsides en route from Addis Ababa to Sodore, the plants were generally quite small, had numerous flowers but with limited foliage, and less suitable for a leaf-feeding agent such as *Zygogramma bicolorata*. Recommendations were made that the pilot mass-rearing centre for biocontrol agents be situated further east, near Dire Dawa, where parthenium infestations are reported to be dense and conditions less-arid (although potential sites still need to be checked). The site that is selected for the establishment of a mass-rearing centre should be in close

proximity to large parthenium infestations, where parthenium plants remain in lush, good condition for the growing season. It was recommended by PPRI researchers that a pilot mass-rearing centre be set up in the east of the country, teething problems sorted out and people trained there first, before the biocontrol programme is expanded further and other parthenium biocontrol mass-rearing centres are set up in other parts of Ethiopia. It was suggested that the pilot mass-rearing centre in the east could involve Haramaya University. Staff could be trained in parthenium biocontrol for several months at the EIAR PPRC Ambo centre before embarking on mass-rearing, so that there is good understanding of biocontrol principles and the agents to be mass-reared.

Greenhouse / shadehouse structures (see Fig. 1 a & b) used by the nursery industry were observed en route along the road between Addis Ababa and Sodore, and it is recommended that similar basic structures are erected for a parthenium biocontrol mass-rearing setup, using *Eucalyptus* (gum) poles and screening material (Fig.2; approximately 20% shading; for exclusion of predators and prevention of escape of *Zygogramma bicolorata* adults and larvae). Screening should be well sealed at the soil – screen interface to exclude predators and reduce escape of biocontrol agents (see Fig. 3) and have a lockable gate with additional screening (Fig. 4). For further recommendations for the suggested design for the mass-rearing facility refer to A. McConnachie's workshop presentation.

Figure 1a & b: Greenhouse structure (note that a biocontrol agent mass-rearing structure should be lower in height than the structure shown in photo, to enable easier collection of insects from ceiling) used by nursery industry en route from Addis Ababa to Sodore. It is recommended that a similar design is followed for parthenium biocontrol mass-rearing structures.

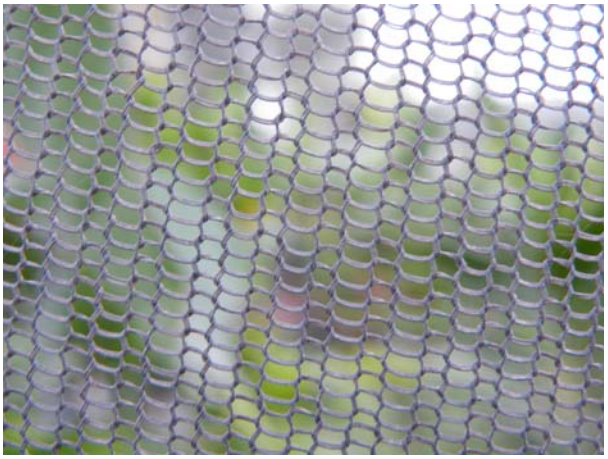

Fig. 2 Screening material (approx. 20% shade screening)

Fig. 3 Bottom of screening (rolled around pole) to be buried for a biocontrol agent mass-rearing structure, to prevent entry of insect predators from outside and exit of insects to be mass-reared inside the structure at the soil-screening interface.

Fig. 4 Lockable gate (will need fine screening on gate for biocontrol agent mass-rearing structures)

List of Contacts Made:

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Appendix 1:

Recommendations following visit to EIAR PPRC Ambo, Ethiopia weed biocontrol quarantine facility on 18/12/2009

Listronotus quarantine facility:

1. Install (well sealed against floor) wooden strips across doorways on 3 baffle entrance doors (outer, middle, inner) so that doors close against foam to form better seal
2. Close off all holes/gaps of walk-in-cage, especially at access zip near airconditioners
3. Fill up soil in pots to nearer top of pot
4. Ensure no gaps at soil-pot interface (adults will hide here) – fill up soil
5. After changeover of plants (after 2 weeks exposure to adults) hold the old plants in separate cages for another 2 weeks to allow larvae to develop. Only then dissect plants and collect larvae and pupae to be put into containers with soil.
6. Put no more than 100 larvae and pupae in each container of the size that we saw. Use lids for larval/ pupal development containers – glue gauze inserts onto lids for air. Do not place these pupal development containers in a cage with a plant – makes it more difficult to find eclosed adults. Rather check (sealed) containers daily for newly eclosed adults, collect and transfer to breeding cages.
7. Spray soil in larval/pupal development containers with 2% bleach solution 2 x / week (but do not make soil too wet).
8. Check larval/pupal containers every day or 2 days to collect newly eclosed adults. Transfer these to breeding cages.
9. Mark age (date of eclosion) of adults on labels of breeding cages
10. Group adults of a similar age (within 1-2 weeks of similar age) in cages – e.g. put adults that eclose within the same week in one cage (up to 100 per cage), then start another cage i.e. don't spread newly eclosed adults across all cages. Maintain within these groups as much as possible.
11. Changeover of plants – increase time spent changing over each cage.so that at least 95% of adults are collected / recovered.
12. Get larger, preferably white, trays to work on as trays that are being used are too small
13. Lighting is dim - get headlamps to give better lighting for easier sighting when collecting adults from cages
14. Keep all quarantine doors closed at all times – immediately repair doors that don't close
15. Foam position around doors for sealing is in incorrect place – re-position the foam within the door frame
16. Place new foam around doors of insect cages where foam has become compressed (or *Listronotus* will squeeze through gaps)
17. Ensure that there are no gaps in foam around cage doors etc. – add extra foam pieces to frames that have gaps in foam strips
18. Repair cages where necessary (holes in gauze, frame of cage, glue gauze back onto cage frame where it has come apart) – check and repair regularly. Inspect cages 1-2 times per week and repair immediately.
19. Move a dissecting microscope and light source to *Listronotus* quarantine glasshouse, and do not remove this from quarantine room. Microscope is needed for examination of plants for eggs in host range tests and prior to that when workers familiarise themselves with the insect's biology / behaviour.
20. Researchers and technicians should familiarise themselves with all practicalities of *Listronotus* biology long before starting host specificity testing. Using plants from the culture, flowers, stems, leaves and petioles should be inspected under a microscope. Check for eggs and position where they are deposited. Determine difference between frass only and frass-covered egg so can easily distinguish these apart. Dissect stems under microscope to observe larval tunnelling from egg into main stem. Get a "feel" for the insects in terms of egg-laying behaviour and

development (this behaviour changes on some test plants – eggs may sometimes be laid in different parts of the plant e.g. on petioles on some test plant species).

21. Get dedicated quarantine suits to be used for visitors
22. Get shoes that are only for quarantine use for researchers and technicians – these should remain with lab coats /suits etc. in the baffle – i.e. change from outside shoes and leave these in the baffle when working in quarantine. Do not wear quarantine shoes outside of quarantine.
23. Check gas level of freezers
24. Don't overload freezers with pots and other material.

Plants:

Problems:

(i) Insufficient plants of suitable stage (need tall, thick-stemmed plants with numerous flowers)

(ii) Shortage of flowers on plants (the more flowers the better for *Listronotus*, but also need tall, thick stems for optimal larval development).

We recommend:

1. Remove half of fibreglass sheeting above lath house / greenhouse to increase light for plants
2. Use fertiliser for flowering to increase number of flowers per plant (speak to rose growers re. best fertiliser to use to promote flowering)
3. Need lots more flowers on plants for *Listronotus* culturing – current plants are inadequate w.r.t. flower quantity
4. Increase number of stock plants (maintain at least 500 plants in different stages of development at all times – seedlings / plants with foliage / mature, flowering plants).
5. Unnecessary to field collect plants every few weeks – only need to field-collect plants a few times per year, if at all, if there is a critical shortage of plants. By collecting parthenium seedlings (from pots and by planting seeds in seedling trays) and potting about 50 seedlings or more per week every week throughout the year, a continuous supply of plants of correct stage for *Listronotus* will be available.
6. Use glasshouses outside of quarantine to propagate seedlings – better light and temperature (note that pest levels will likely increase if using glasshouse)
7. Use a tunnel or glasshouse in cool, dry season if these are available, for seedling propagation and more rapid plant growth.

Host-specificity testing of *L. setosipennis* (more detailed instructions for host-specificity testing to follow):

1. Use adults that are 3-4 weeks old after eclosion for host specificity tests (ensures that they are in good condition for egg-laying) – so must keep record of eclosion dates (ages) on cages.
2. Adults used in no-choice / choice tests can be used in culture afterwards for breeding purposes but must not be re-used in other tests. Mark cages with e.g. 'test adults' so you know they have been used in tests.
3. Run oviposition tests for minimum of 5 days, maybe longer because of shading of quarantine glasshouse but probably maximum of 7 days (larvae hatch from eggs after about 5 days)
4. All plants used for tests must be flowering, with equivalent floral material.
5. Use 5 mating pairs of adults per test plant (10 adults) – collect from cages and place in Petri dish, collect pairs while they are copulating – i.e. must see them mating – adults often mount others even of same sex so must actually observe them in copulation to ensure they are pairs. Best time to collect mating pairs is in early morning before 9am, and when they have been newly removed from plants and placed together in Petri dish. Place 5 pairs per Petri dish to be used per test plant. Practice collecting mating pairs of adults before any tests are initiated.

Other:

EIAR PPRC to send ARC-PPRI final updated host specificity test results for *Zygogramma bicolorata*.