Establishment and Spread of *Gyranusoidea tebygi* Noyes and *Anagyrus mangicola* Noyes (Hymenoptera: Encyrtidae), Two Biological Control Agents Released against the Mango Mealybug *Rastrococcus invadens* Williams (Homoptera: Pseudococcidae) in Africa

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Two specific endophagous parasitoids, Gyranusoidea tebygi and Anagyrus mangicola, of Indian origin, were mass-reared at the International Institute of Tropical Agriculture in Cotonou and released against the mango mealybug, Rastrococcus invadens, in collaboration with national biological control programmes. G. tebygi was released in the following countries: Benin, Gabon, Ghana, Nigeria, Sierra Leone and Zaire. In Togo, it had been released earlier and studied during another project. This parasitoid is now established in all areas infested by the mango mealybug. In addition, it established itself without previous release in Congo and Côte d'Ivoire. A. mangicola has been released in Benin, Gabon and Sierra Leone since 1991, and by mid-1993 was recovered from a few sites. It seems locally established in southern Benin.

Keywords: Rastrococcus invadens, Gyranusoidea tebygi, Anagyrus mangicola, mango, mango mealybug, Africa, biological control

INTRODUCTION

Around 1981–82, a new mealybug was accidently introduced into West Africa and quickly spread through Togo, Benin and Ghana. It was identified as a new species, *Rastrococcus invadens* Williams (Homoptera: Pseudococcidae), of Indian origin (Williams, 1986). Although reported on 45 species of crop plants, ornamentals and wild plants (Agounké *et al.*, 1988), its main attack was on mango (*Mangifera indica*, Anacardiaceae) and, to a lesser extent, on citrus (*Citrus* spp., Rutaceae), frangipani (*Plumeria* spp., Apocynaceae) and *Ficus* spp. (Moraceae). Owing to the heavy accumulation of honeydew and the resulting sooty mould, growth, flowering and fruiting of attacked trees were arrested. This often led farmers to cut them down.

R. invadens and its natural enemies were investigated in India (Narasimham & Chako, 1988), and two promising parasitoids, *Gyranusoidea tebygi* Noyes and *Anagyrus mangicola* Noyes

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(Hymenoptera: Encyrtidae), were sent to quarantine by the International Institute of Biological Control (IIBC) in Silwood Park for further study (Noyes, 1988, 1990; Willink & Moore, 1988; Cross & Moore, 1992). A biological control project, financed by the Food and Agriculture Organization (FAO), was started at the Plant Protection Services in Cacaveli, Togo, and Porto-Novo, Benin. Both services were supported by the Gesellschaft für Technische Zusammenarbeit (GTZ).

At an FAO-sponsored conference in Lomé, Togo, in 1987, a regional project on R. invadens was set up with the agreement of the Inter-African Phytosanitary Council of the Organization of African Unity (OAU). Close collaboration between the International Institute of Tropical Agriculture (IITA) and the IIBC in training, introduction of natural enemies and research was recommended. By then, a special project on biological control of the mango mealybug, separately funded by Switzerland, had already been added to IITA's biological control programme. The aim of the project was to introduce exotic parasitoids to supplement indigenous predators that were not capable of controlling R. invadens (Agounké et al., 1988). In addition, the use of a pathogen was investigated (Fernandez-Garcia & Moore, 1988; Akalach et al., 1992), but this has not reached the stage of trials in the field as yet.

G. tebygi was released in Togo in late 1987. First results indicated that it was successful in controlling R. invadens (Agricola et al., 1989). However, recent observations of 'hot spots' of infestation despite the presence of G. tebygi (A. Bokonon-Ganta & P. Neuenschwander, IITA, Berlin, unpublished results) prompted investigations in Benin and other countries, which led to release of the second parasitoid, A. mangicola. From laboratory experiments, it had been concluded that, under certain circumstances, this parasitoid was able to compete successfully with G. tebygi (Moore & Cross, 1992). A simulation model, however, gave ambiguous predictions about the outcome of competition between the two species (Godfray & Waage, 1991).

Rearing and release of these parasitoids followed the model established in the successful campaign against the cassava mealybug (Herren & Neuenschwander, 1991). It involved close collaboration with national biological control programmes already supported by IITA. The present paper documents the release, establishment and spread of the exotic biological control agents up to October 1993. Further details on spread and the results of impact studies will be presented later by scientists in the various countries concerned.

MATERIALS AND METHODS

Origin of Biological Control Agents

G. tebygi, which had been quarantined in Britain, was received from the GTZ project in Cacaveli, Togo, in January 1988. *A. mangicola* was obtained directly from the IIBC quarantine in Silwood Park in September 1990. Both parasitoids were checked by the quarantine service in Benin before being brought to the IITA insectary.

Rearing

For rearing of *R. invadens*, different host plants including oleander (*Nerium oleander*, Apocynaceae), seedlings of mango and *Ficus polita* were tested. *F. polita* was retained because:

- (1) it is easily propagated from cuttings;
- (2) its broad leaves are not shed in the insectary (as was often the case with the other species);
- (3) its high carrying capacity.

Potted 30-cm tall plants were used for infestation.

Gravid *R. invadens* females were placed on the leaves with a fine brush. The infested plants were transferred to wooden cages $(44 \times 45 \times 58 \text{ cm})$ with fine screen sides and glass tops, and kept in the insectary at $28 \pm 2^{\circ}$ C and 60-80% relative humidity, under a 12:12 h light/dark regime. Since adult *R. invadens* are not susceptible to parasitism by *G. tebygi* and hyper-parasitoids, contamination of the culture was prevented.

After 2 weeks, adult G. tebygi of both sexes were introduced into the cages and kept for about

10 weeks. Starting 4 weeks after infestation, adult wasps were collected with an aspirator for release or for further rearing. Setting up three new cages every 2 weeks led to a stable production of 2000–3000 G. tebygi per week.

Rearing of *A. mangicola* initially proved more difficult. When a broad spectrum of host instars was offered, better results were achieved. Thus, about 600 *A. mangicola* were produced weekly from two cages, which were set up every 2 weeks.

Packaging, Transport and Release

Adults were collected from the rearing cages by aspirating them in 20-cm³ plastic vials, containing a strip of filter paper with a drop of honey diluted with water. The vials were closed with a gauze cover. If necessary, they were stored overnight at $15 \pm 1^{\circ}$ C and transported in styrofoam cool boxes.

Delivery to countries other than Benin was only made upon receipt of an official request with a valid import permit, and all releases were made in collaboration with national programme scientists.

G. tebygi was also delivered for further rearing to the Plant Protection Services in Porto-Novo, Benin; the National Institute of Horticulture (NIHORT) in Ibadan, Nigeria; and the National Biological Control Programme at the University of Gabon in Libreville, Gabon. The latter two laboratories also received *A. mangicola* for culture.

Under the prevalent conditions of transport, G. tebygi proved to be as hardy as Epindinocarsis lopezi (De Santis) (Hymenoptera, Encyrtidae) released against the cassava mealybug (Herren et al., 1987). A. mangicola, by contrast, survived only 1-2 days. However, most releases could be done within one day of packaging. Mortalities during transport in Benin were generally low (<10%). They were considerably higher when transport lasted longer. Owing to logistic difficulties, two shipments to Ghana and two to Nigeria suffered 100% mortality; they are not considered further.

Releases were made throughout the year on heavily infested mango trees. A. mangicola was released exclusively into small pockets of infestation, which remained after G. tebygi had already become established in the same area.

Monitoring

A protocol was set up to follow the population dynamics of *R. invadens* and its parasitoids and predators in Benin. In addition, a survey protocol was established to assess the impact of the biological control project. Both protocols, which are not further presented here, required searching for parasitized mealybugs, the so-called mummies. The mummies were kept in gelatine capsules inside a vial, closed with a gauze cover, until emergence of the wasps. In the present paper, only presence or absence of parasitoids is recorded. Establishment of an exotic species is declared if adults have been recovered at least 1 year after the release in a given site (De Bach & Bartlett, 1964). This means that the parasitoid populations have passed through a year's cycle of adverse conditions, like rains, dry winds ('harmattan'), etc. For the present parasitoids, 1 year represents about 17 generations.

In Benin, country-wide surveys were made in February–March 1989, August 1990, and during February–March 1990 and 1991. An additional survey was made in the north in 1993. Mango trees were selected without bias at intervals of 10 km along all major roads in rural areas and of 5 km in urban areas. In order to cover the entire country, stops were also made along dirt roads where necessary. At each stop, four adjacent mango trees, and any surrounding fruit trees known to be susceptible to *R. invadens* attack, were inspected. Sampling procedures followed the results of a study on spatial distribution of mango mealybug in mango trees (Boavida *et al.*, 1992). Further results of these surveys will be presented elsewhere.

RESULTS AND DISCUSSION

All shipments and releases of *G. tebygi* and *A. mangicola* from the IITA insectary in Cotonou are detailed in Table 1. Release areas are indicated in Figure 1, together with the known spread of *G. tebygi*. The situation in the different countries is presented below, more or less in the order in which these countries were infested by *R. invadens*, and in which they started their biological control projects.

Togo

The first country to release G. tebygi in late 1987, Togo, was also the first to measure the impact of the parasitoid on its host populations (Agricola et al., 1989). G. tebygi spread over the entire area infested by R. invadens at a speed of about 100 km per year. Indigenous hyper-parasitoids attacking G. tebygi proved to be the same ones as those recorded from E. lopezi (Agricola & Fischer, 1991). On the basis of a sharp decline in mealybug population densities on the few sampling trees, which were evenly distributed over the two infestation zones, the project was declared a success by the executing agencies. Further studies became impossible because of the political situation in the country.

Benin

Surveys were done in collaboration with the Plant Protection Services in Porto-Novo. After its detection in 1986, *R. invadens* quickly spread in the southern half of the country. Spread in the north was, and is, limited to some big towns and their surroundings.

Releases of *G. tebygi* started in May 1988. Surveys indicated that *G. tebygi* established itself in all release sites and spread slowly at first, then at the rate of about 100 km per year. It was recovered not only in the large mealybug populations sometimes observed in towns, but also on isolated mango trees in farmers' fields. On the isolated trees, mealybug populations were often so low that a few mealybugs only were found on a tree despite a search of about 15 min.

During the first survey, *R. invadens* and *G. tebygi* were found on many different tree species. As mango mealybug populations declined, the pest became uncommon on all trees except mango. By the time the last survey was executed in 1991, mango mealybug was found only in a few 'hot spots' in the towns. In the 'hot spots', mealybug populations were high despite the presence of *G. tebygi*. Generally, only a few, heavily infested, trees could be found.

A. mangicola was released repeatedly in such 'hot spots'. In the Guinea savanna zone in Djougou and Ouaké, mealybug populations crashed after the releases, to the extent that no mealybugs could be found and, later, no establishment of A. mangicola could be demonstrated. The observed crash was probably caused by G. tebygi. In the south, however, A. mangicola was regularly recovered around Cotonou. The situation in the 'hot spots' is now being studied in more detail.

Ghana

The Quarantine and Plant Protection Services in Pokoase were supplied with *G. tebygi* and assisted in their releases over all infested zones. *G. tebygi* was initially released in the commercial mango plantations, FARMEX, in Greater Accra, and CAMSI, in the Central Region. Quick establishment and spread of the exotic parasitoid were noted. By the end of 1988, *R. invadens* had virtually disappeared. By 1993, mango trees on farms and in villages showed no signs of attack or sooty mould. Periodic but severe infestations of the mango mealybug, accompanied by heavy sooty mould, persisted, however, locally in Accra, Kumasi, Sunyani, Tamale and Wa.

A. mangicola was supplied in 1991. Survival in transport, except for the last shipment, was bad. No recoveries of the parasitoid have yet been made (A. R. Cudjoe, K. K. Antwi and J. A. Timbilla, Ministry of Agriculture, Accra, Ghana, unpublished results).

Species	Date of release	Number packaged	Mortality (%)	Country of destination	Locality of release
Gvranusoi	dea tebygi	·····	· · · · · · · · · · · · · · · · · · ·		
-)	17-03-88	550	15	Ghana	Accra
	20-04-88	750			Accra, Camsi
	25-05-88	1800	0	Benin	Abomey Calavi
	01-06-88	650	ŏ		Abomey Calavi
	10-06-88	1000	ŏ		Abomey Calavi
	21-08-88	1000	25	Ghana	Koforidua
	22-08-88	1000	95	Ciruina	Kumasi
	28-08-88	2750	48		Sunyani
	12-10-88	1200	-40	Benin	Porto-Novo
	28-10-88	3000	23	Ghana	Central Region
	22-11-88	800	20	Benin	Parakou
	23-11-88	800		Denni	Natitingou
	20-12-88	1100			Porto Novo
	27-12-88	4000			Covè, Savalou
	11-02-89	4000	10	Ghana	Aburi, Tema, Accra
			10	Onana	
	11-04-89	· 3400		Zaire	Ejura ^a
	09-06-89	1100			Kinshasa
	20-06-89	1130	16	Benin Zaire	Parakou
	03-08-89	320	10		Kinshasa
	03-08-89	120		Nigeria	Lagos
	10-08-89	280			Ijebu-Ode, Abeokuta Shagamu
	24-08-89	400	21	C 1	Ibadan
	30-08-89	450	31	Ghana	Accra
	19-09-89	1300		Nigeria	Ibadan
	28-09-89	1100		Ghana	Accra
	17-10-89	2450		Nigeria	Ibadan
	03-11-89	200			Ibadan
	03-11-89	1500		Zaire	Kinshasa
	15-12-89	1200	12	Nigeria	Ibadan
	09-01-90	3050		Gabon	Libreville
	29-03-90	1500	57	Nigeria	River, Imo, Anambra, Bendel State
	02-04-90	1400		Ghana	Tamale
	27-04-90	2600	55		Tolon-Tamale ^a
	22-08-90	2900		Zaire	Kinshasa
	07-09-90	2600	40	Nigeria	Eastern states
	21-12-90	1300	34	Ghana	Industrial area Accra
	14-03-91°	500	29	Nigeria	Ibadan, NIHORT
	09-10-91	900	67	Ghana	Accra
	11-12-91	1280		Sierra Leone	Lunsar
	11-02-92	1100	86	Ghana	Wa
	30-05-92°	650		Nigeria	Ibadan, NIHORT
	19-04-93	800	20	Benin	Natitingou
nagvrus n	nangicola				
	21-06-91	1700	10	Benin	Ouaké
	21-06-91	1650	15		Djougou
	08-10-91	1100	58	Ghana	Accra
	23-10-91	500	65	Gabon	Libreville
	11-12-91	900	50	Sierra Leone	Lunsar
	30-05-92	450		Nigeria	Ibadan, NIHORT
	24-06-92	700	82	Sierra Leone	Lunsar
	11-09-92	300	Õ	Benin	Godomey
	23-09-92	200	ŏ	point .	Abomey Calavi
	24-09-92	300	ŏ		Godomey
	06-10-92	400	0		Abomey Calavi
	16-10-92	550	5		Ouidah
	18-11-92	450	0		Ouidan
	20-11-92	300	0		
	03-12-92	300	0		Cotonou
					Cotonou
	21-12-92	150	0	Chart	Cotonou
	10-04-93	650	10	Ghana	Accra
	16-04-93	1500	15	Benin	Djougou
	19-04-93	350	80		Natitingou
	21-05-93	1650	20	CI .	Kouandé
	02-07-93	850	2	Ghana	Accra, Achimota
	13-10-93	650	0	Benin	Ouidah

TABLE 1. Shipments and releases of R. invadens parasitoids from the IITA insectary in Cotonou, Benin, from 1988 to 1993

^{*a*} Aerial release. ^{*b*} For rearing.

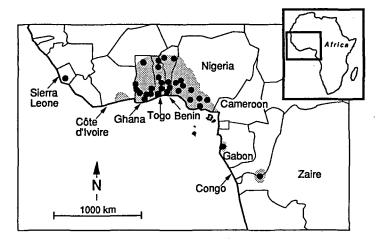


FIGURE 1. Distribution in mid-1993 (shaded) of *R. invadens* and its parasitoid *G. tebygi*, in Africa. Dots = release sites.

Nigeria

The quarantine authorities were informed by IITA in 1987 of the presence of *R. invadens* near Lagos and were later assisted in a survey of the southern half of the country to determine the spread of the mealybug. By mid-1989, *R. invadens* was widely distributed in Lagos, Ogun and Oyo States (Ivbijaro *et al.*, 1992). By early 1990, it had reached Ondo, Bendel, Anambra, Imo, Rivers and Akwa Ibom States (F. V. Anno-Nyako, National Horticultural Research Institute, Ibadan, Nigeria, personal communication).

One release of *G. tebygi* was made with material imported from Togo. Subsequently, the Nigerian biological control committee, in collaboration with NIHORT, made numerous releases of *G. tebygi* obtained from IITA in all three southwestern states and, later, in the southeastern states. *G. tebygi* was recovered at all release sites, from where it spread to new areas (F. V. Anno-Nyako and N. Echendu, National Root Crops Research Institute, Umudike, Nigeria, personal communication).

Côte d'Ivoire

R. invadens was found in a small strip along the coast in 1988. Though no releases had ever been made, *G. tebygi* was also recovered (K. Amany and K. Kouamé, Ministère de l'Agriculture, Abidjan, Côte d'Ivoire, personal communication).

Gabon

R. invadens was first observed in 1988, and its spread was studied in detail (Boussienguet & Herren, 1992). For biological control, both exotic parasitoids were sent from the IITA insectary; they were partly released and partly used for rearing in the laboratory. *G. tebygi* was established in all release sites in the Libreville area, and a large reduction in the number of host plants attacked was observed after this establishment. *A. mangicola* was recovered; but it is not clear whether long-term establishment will occur (J. Boussienguet, Université du Gabon, Libreville, Gabon, personal communication).

Zaire

Following an attack by *R. invadens* in Kinshasa around 1987, *G. tebygi* was sent for release in 1989. It was later found established (Loma Tongomo, Université Nationale du Zaire, Kinshasa, Zaire, and H. D. Nsiama She INERA/PRONAM, Kinshasa, Zaire, personal communication). Due

to the political situation in the country, further data about the spread of the mealybug and its parasitoid are not available.

Congo

R. invadens was found for the first time in Central Africa in the Congo in 1986 (Moussa & Matile-Ferrero, 1988). Its population dynamics have been studied since October 1988. In May 1989, a few weeks before the releases of *G. tebygi* in Kinshasa across the Zaire river, this parasitoid appeared in collections. It later greatly reduced population levels of *R. invadens* (Matokot *et al.*, 1992).

Sierra Leone

In 1991, a locally very restricted *R. invadens* infestation appeared in Lunsar, in the vicinity of a popular hospital in the interior, and not, as would be expected in the busy port town of Freetown. Both parasitoids were released, but separately in different places. *G. tebygi* became abundant and spread; *A. mangicola* was recovered only from a few specimens (B. James, Fourah Bay College, Freetown, Sierra Leone, personal communication).

Cameroon

R. invadens was recently found in Douala and surroundings. No mummies of *G. tebygi* were seen (M. Tchuanyo, Institut de Recherche Agronomique, Buea, Cameroon, personal communication).

Other Countries

Guinea Conakri and Angola were suspected to have *R. invadens*. On subsequent inspection by an entomologist, however, these reports turned out to be of other Homoptera.

CONCLUSION

It seems that, on several occasions, *R. invadens* was accidentally introduced in West and Central Africa, mainly in large market centres, sometimes great distances apart. In some instances, like in the Congo and Côte d'Ivoire, already parasitized mealybugs might have been introduced and no releases of *G. tebygi* were needed. (Dispersal of adult *G. tebygi* by flight and wind seems less likely because of large distances to be crossed where hosts were absent.) It must be assumed that these introductions occurred on introduced fruits brought to the markets, irrespective of quarantine regulations.

To our knowledge, G. tebygi is established over the entire area of infestation of R. invadens in Africa, though it might be absent very locally. A. mangicola, by contrast, has been recovered only in small 'hot spots' of R. invadens infestation. In view of regular recaptures up to October 1993, establishment in Benin seems permanent.

Distribution of *G. tebygi* over such a large area, and obtained in such a short time, was achieved thanks to close collaboration between national and international scientific institutions as outlined at the FAO-sponsored conference in Lomé in 1987. Official recognition of the project culminated in two ceremonial releases, which were made in the presence of government ministers in Ibadan, Nigeria in December 1989 and Libreville, Gabon in January 1990.

Population dynamics studies in Togo (Agricola *et al.*, 1989), Congo (Matokot *et al.*, 1992) and Benin (C. Boavida, IITA, Berlin, unpublished results) suggest that *G. tebygi* is responsible for successful biological control. Repeated detailed surveys in Benin (A. Bokonon-Ganta, IITA, Berlin, unpublished results) confirm that effective control occurs over large areas. Due to *G. tebygi*, mango mealybug infestations have been mostly restricted to mango in local 'hot spots', into which *A. mangicola* was later released. The competition among the two parasitoids in these 'hot spots' and their influence on the level of *R. invadens* control merit further studies.

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ESTABLISHMENT OF PARASITOIDS OF MANGO MEALYBUG

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