

Population dynamics of the mango mealybug, *Rastrococcus invadens* Williams (Homoptera: Pseudococcidae) in northern Côte d'Ivoire

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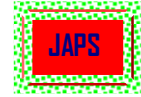
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1 SUMMARY

To collect data for a biological control strategy against mango mealybug (*Rastrococcus invadens*), the infestation of three varieties of mango (*Kent*, *Keitt* and *Early Gold*) were studied. The periods of *R. invadens* fluctuations and those of its two main natural enemies, the parasitoids *Gyranusoidea tebygi* and *Anagyrus mangicola*, the impact of parasitoids/hyperparasitoids complex and the influence of abiotic factors on mealybug/parasitoids complex, were also studied. Dynamics of pest primary and secondary parasitoids was measured from September 1999 to September 2000 in northern Côte d'Ivoire, the main producing region of mangoes and the most affected by negative effects of *R. invadens*. Live (adults and larvae), dead and mummified mealybugs, were counted directly in the field on 20 leaves per tree. Primary and secondary parasitoids were identified and counted at their emergence from full mummies, after 2 weeks of incubation in laboratory at room temperature. *Keitt* was more infested by the pest, followed by *Kent* then *Early Gold*. *R. invadens* population is more abundant in rainy season than in dry season. Under natural conditions in northern Côte d'Ivoire, mealybug is able to develop five generations from 60 to 70 days per year. Abiotic factors that appear to affect populations of pest are mainly rainfall and temperature variations and to a lesser extent humidity. The level of live and dead insects is positively correlated with rainfall in contrast to temperature variations. With only 17.95 % of parasites developed by *G. tebygi* and *A. mangicola* on mealybug, their impact seems weak on the pest. *Gyranusoidea tebygi* and *Anagyrus mangicola* are attacked in their turn by several hyperparasitoids including *Marietta Léopardina* and *Chartocerus hyalipennis* as the most important when considering their level of hyperparasitism (about 65.18 %). Mass production of *G. tebygi* and *A. mangicola* and their release is considered to control *R. invadens*. The presence of hyperparasitoids responsible of high level mortality of parasitoids, however, represents a constraint. The best periods suited to mass production of *G. tebygi* and *A. mangicola* are from February to September and for first releasing, from March to April.



1 INTRODUCTION

With nearly 9 000 tons of mangoes exported annually since 1997, Côte d'Ivoire has become the second supplier of mangoes to the European market (OCAB, 2001). Development of mango crop, especially in the northern region, brings more and more substantial income to producers and other participants of the channel (trackers, exporters, carriers.) (ANADER, 2001). Mango production in the country faces phytosanitary problems, whose most important are fruit flies belonging to the family of Tephritidae and mealybug *Rastrococcus invadens* Williams (Homoptera: Pseudococcidae) (N'Guetta, 1995; Hala et al., 2004; Hala and Coulibaly, 2007). Accidentally introduced into Africa in the early 1980's from South-East Asia where it originates (Williams, 1986), *R. invadens* has been observed for the first time in Togo and Ghana before spreading into most countries of West Africa where it causes damage to mango trees and other fruit trees (Agounke et al., 1988; Ivbijaro et al., 1992). Due to its polyphagia and high propagation favored by human activity, *R. invadens* after its appearance in Côte d'Ivoire in 1989 (Neuenschwander et al., 1994), reached very quickly the northern country (especially around Korhogo), the main production area of mangoes for export (Hala et al., 2004). According to Neuenschwander et al., (1994) and Hala et al. (2004), the average loss of potential for mango production can reach 53% in this region. Production can fall to zero when the producers are obliged to fell infested trees. Similarly, when heavily affected, ornamentals and shade plants can no longer perform these first functions of ornament and shelter and are severely pruned because of honeydew and

sooty mold present on the leaves (Figures 1 and 2), stems and soil (Agounke et al., 1988; Hala et al., 2004). Biological control of *R. invadens* by the introduction of its parasitoids *Gyranusoidea tebygi* and *Anagyrus mangicola* (Hymenoptera: Encyrtidae) has proven effective in Togo (Agricola et al., 1989) and Benin (Neuenschwander et al., 1994; Bokonon-Ganta and Neuenschwander, 1995). Dissemination of mealybug in Côte d'Ivoire had been largely facilitated by the transport of infected plant material. Parasitoids and hyperparasitoids have logically accompanied the pest during its travels (Hala et al., 2004). According to Hala et al. (2004), although its two main parasitoids (*G. tebygi* and *A. mangicola*) are present in this country, the level of their populations currently remains too low to ensure control of the pest. In Côte d'Ivoire, rational chemical control and biological control were selected as components of possible methods of an integrated pest management of mealybug populations (Hala and Coulibaly, 2007). The biological control component will be characterized by massive release of *G. tebygi* and *A. mangicola*, natural enemies of the pest. Before the establishment of such a program, study of dynamics of populations of pest-parasitoids-hyperparasitoids complex is essential.

This work aims to:

- compare the sensitivity of three varieties of mango tree with mealybug infestations,
- determine the periods of fluctuations of pest, *Gyranusoidea tebygi* and *Anagyrus mangicola* populations,
- determine abiotic factors that influence these different populations,
- measure the impact of primary and secondary parasitoids on mango mealybug.



Figure 1: Leaves and fruits of mango *Keitt* infested by mealybug.



Figure 2: Mango leaves covered with sooty mold following honeydew secreted by mealybug of mango tree.

Form Bold

2 MATERIALS AND METHODS

2.1 Climatic characteristics of the area of study: In general the northern region of Côte d'Ivoire has six months of rainy season (May-October) with peaks from July to September. During this period, average daily temperatures fluctuate from 24 to 26 °C with minimum at 19 °C and maximum at 28 °C. Relative hygrometry (RH) varies from 60 to 80 %. There are six months of dry season (November-April) during which the average

daily temperatures range from 23-30 °C. March and April are the hottest months. The dry season shows the greatest daily difference of temperature: minimum from December to February can drop down to 14 or 12 °C and maximum up to 32 °C. These three months presents also the peaks of drought and the lowest hygrometry (40 to 60 %) (Figures 3 and 4).

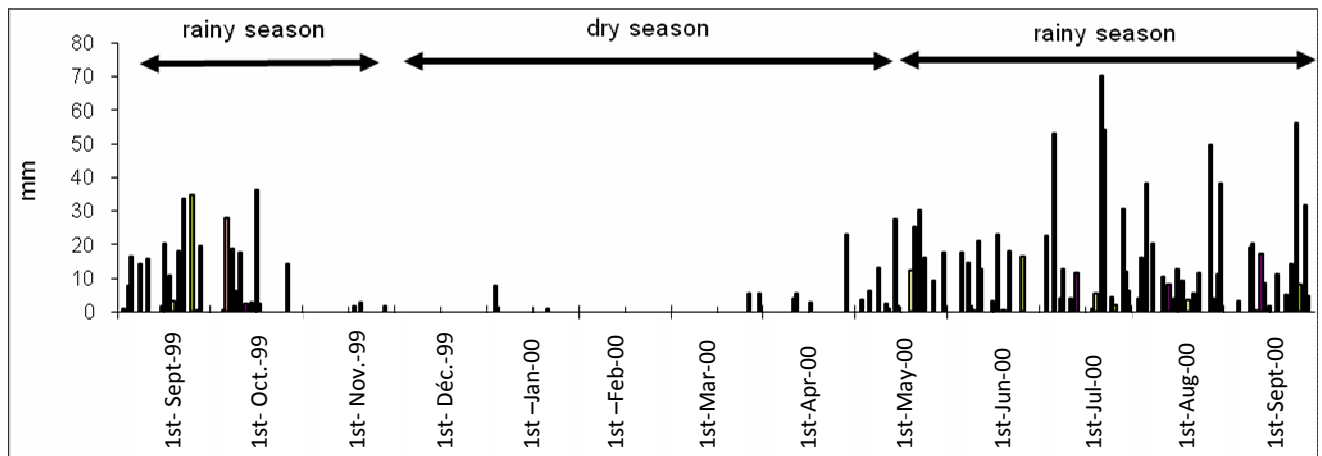


Figure 3: Daily rainfall from September 1999 to September 2000

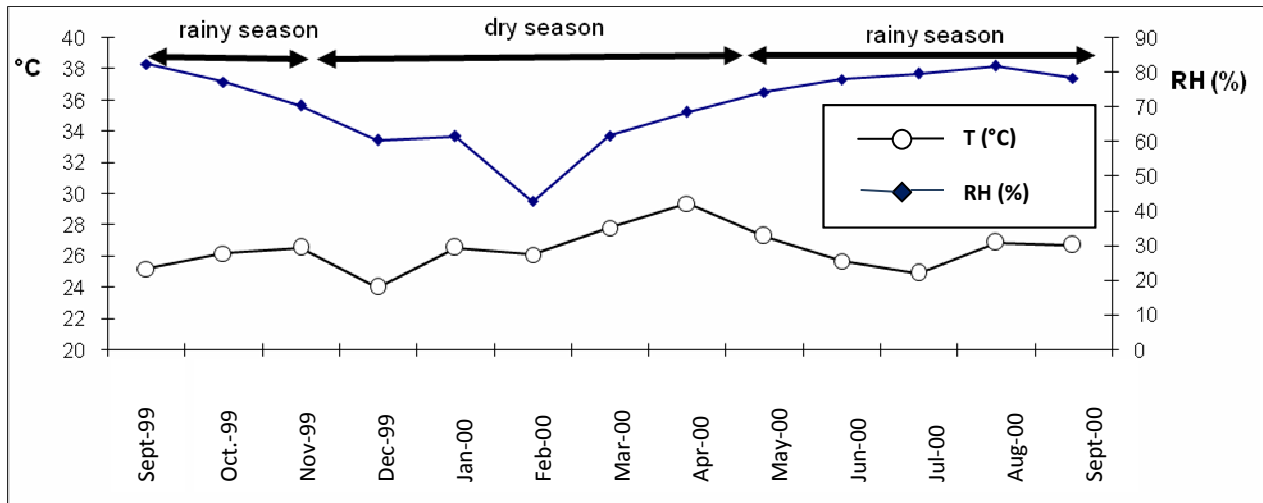


Figure 4: Averages monthly temperatures and relative humidity from September 1999 to September 2000

2.2 Plant material: Plant material used was mango (*Mangifera indica*) from three grafted varieties of American origin (Akassey and Daubrey, 1992):

- *Keitt*, fruit of large caliber (average 500 g), variety of semi-late to late season, very broad tree port and long and large leaves (23 cm x 7.5 cm). Its shows a very good force tree and regular fructifications, - *Kent*, medium to large size fruit (average 400 g), variety of mid-season, vigorous and spreading tree with leaves of medium-sized (18.5 cm x 4.5 cm) and sharp edges,

- *Early Gold*, medium size fruit (200 to 300 g), variety of early season with short and wavy leaves (16 cm x 4 cm).

2.3 Experimental design: Study of population dynamics of mango mealybug (adults and larvae) was set in the field from September 1999 to September 2000. The plot of 2.42 ha broad, density of 158 plants/ha in gauge 7 m x 9 m, consisted of 24 rows of 16 trees. The 24 lines were divided into four blocks (or replicates) of six lines each (2 lines per variety). The basic plot consisted of two consecutive lines of the same variety. A tree was chosen at random in each line. Sampling method

derived from Schneider *et al.* (1987). On each tree, leaves were sampled outside the canopy at an average height of 2 meters above the ground. Each tree was divided into four quadrants: North, South, East and West. In each quadrant, five leaves were selected at random for a total of 20 leaves/tree. The leaves were placed by quadrant in polythene bags and brought to the laboratory for recounting.

2.4 Variables observed: Mango mealybug was counted directly in the field using a hand lens, mentioning its different scales: live (adults and larvae), dead, mummified. Primary and secondary parasitoids were determined from full pest mummified collected on the leaves and placed in capsules for 2 weeks at room temperature. After their emergence, they were counted and identified with a dissecting microscope.

2.5. Data analysis: Data were classified on an Excel spreadsheet to determine the graphics and statistical analysis performed using SPSS (Statistical Package of Social Science) version 7.5. After the analysis of variance, Duncan's test was used at 5% to separate means when a significant difference between them.

3 RESULTS

3.1. Comparison of annual densities of mealybug on three varieties of mango: Statistically significant differences of densities of pest were found over the varieties of mango (Table

1). *Keitt* presented the higher density (90.10/20 leaves) followed by *Kent* (73.23/20 leaves) then *Early Gold* (55.43/20 leaves).

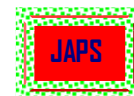


Table 1: Comparison of annual average density of mealybug on three varieties of mango

Varieties	Density (mealybugs/20 leaves)
<i>Early Gold</i>	55.43 a
<i>Kent</i>	73.23 ab
<i>Keitt</i>	90.10 b
$F_{\text{varieties}} = 3.563 (S)$ Prob. = 0.047	
S: Significant at 5 %	

3.2 Seasonal variations in populations of mango mealybug: The evolution of infestation levels showed the same trend for the three varieties of mango (Figure 5). Two periods of fluctuation were observed:

- a latency period, from October 1999 to mid-March 2000, characterized by a low and nearly constant level of the pest, especially during harmattan (November 1999-February 2000) with

densities of populations ranged between 0.20 and 0.85 insect/leaf,

- a grading period, from mid-March 2000 to August 2000 marked by a significant variation in the density of the pest. Its population varied from 2.79 to 24.75 insects/leaf. Peaks of abundance of pest occur in the rainy season (June-August). The grading period is followed by a rapid decline in densities of adult and larval populations in September 2000.

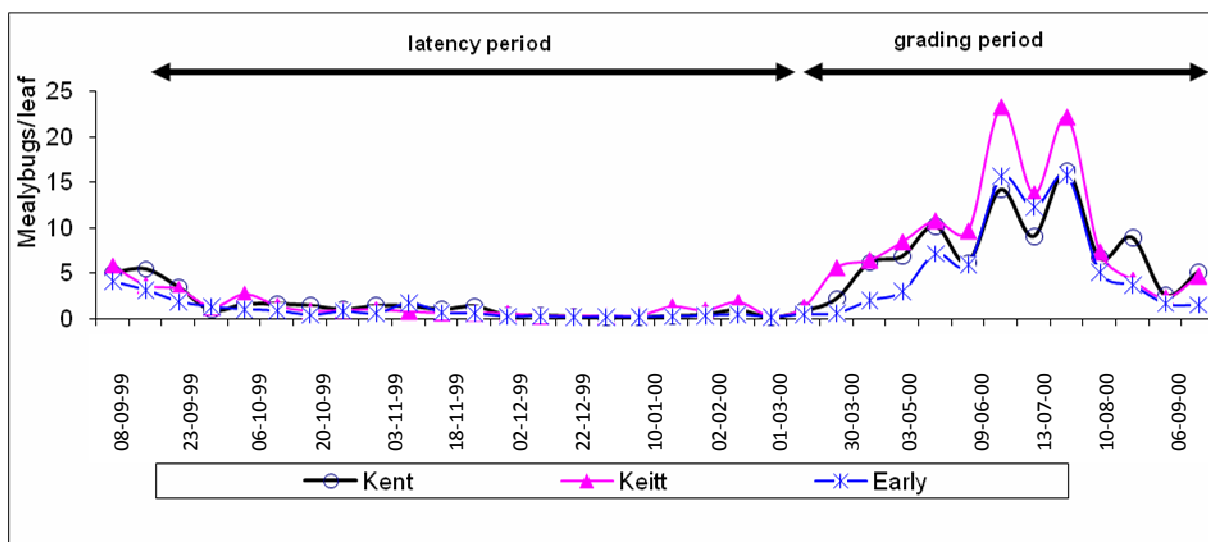


Figure 5: Evolution of the density of live mealybug on mangoes *Kent*, *Keitt* and *Early Gold*

Accounting of live mango mealybug indicates five peaks of larvae output recorded at the following dates: September 23rd 1999, December 1999 2nd, February 2000 2nd, April 2000 12th and June 2000 22nd (Figure 6). These peaks correspond to

generations. Interval between two successive peaks of abundance represents a generation time. It lasts 60 to 70 days.

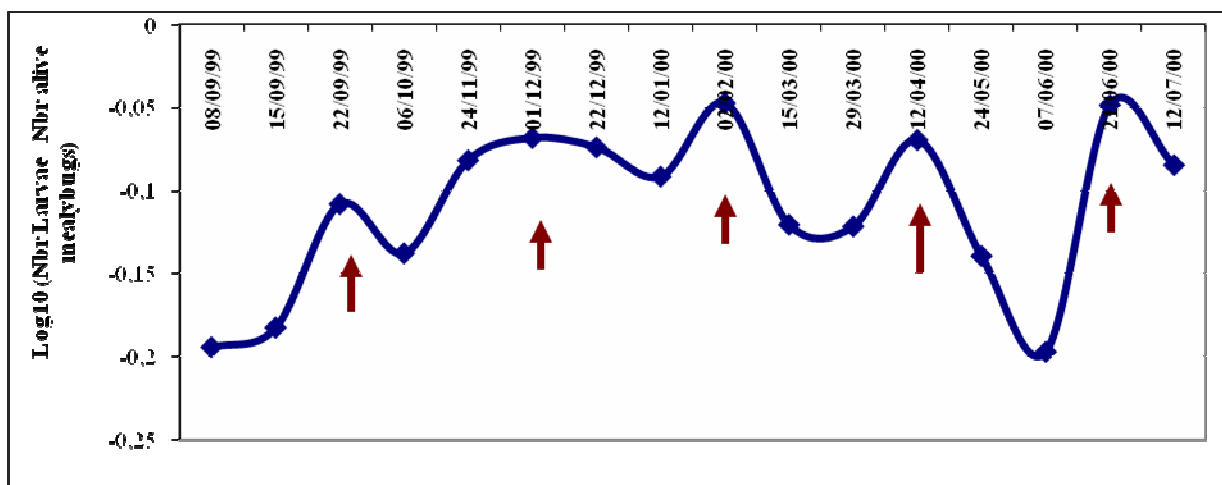
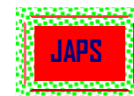


Figure 6: Generation time of mango mealybug in Korhogo area

3.3 Impact of abiotic factors: With respectively significant correlation coefficients 0.583 and 0.693, the levels of live and dead mealybug are positively correlated with rainfall (Table 2). In the contrary, they show negative correlations with temperature ranges, respectively -0.578 and -0.697. The populations of insects, with corresponding

correlation coefficients of 0.117 and -0.393, do not link with average monthly temperatures. The rates of dead mealybug present a highly significant correlation coefficient 0.739 with relative humidity. In the opposite, with an insignificant correlation 0.117, live insects presented a weak dependence with this factor.

Table 2: Linear regression data of the levels of mealybug and non biotic factors

	Log (Number of live mealybug +1)		Log (Number of dead mealybug +1)	
	Equations	Correlation coefficients	Equations	Correlation coefficients
Rainfall (mm)	$y = 0.129x + 2,514$	R = 0.583 (S)	$y = 0.102x + 2,109$	R = 0.693 (HS)
Relative humidity (%)	$y = 0.028x + 0,981$	R = 0.434 (NS)	$y = 0.032x + 0,217$	R = 0.739 (HS)
Average temperatures (°C)	$y = 0.064x + 1,279$	R = 0.117 (NS)	$y = -0.136x + 6,053$	R = -0.393 (NS)
Temperature ranges (°C)	$y = -0.126x + 4,548$	R = -0.578 (S)	$y = -0.101x + 3,731$	R = -0.697 (HS)

S: Significant at 5 %, HS: Highly significant at 5 %, NS: Not significant at 5 %

3.4 Parasitic complex

3.4.1 Importance of different species of primary and secondary parasitoids: Four species were identified among the parasitoids and hyperparasitoids that emerged from mealybug mummified (figure 7a). These are two Encyrtidae (*Gyranusoidea tebygi* Noyes and *Anagyrus mangicola*), one Aphelinidae (*Marietta Léopardin*) and one Signiphoridae (*Chartocerus hyalipennis*). The rate of emergence of parasitoids (34.82 %) is significantly lower than hyperparasitoids (65.18 %). When

considering all the parasitoids and hyperparasitoids, the dominant species is *Marietta* sp, followed in descending order of rate of emergence by *Gyranusoidea tebygi*, *Chartocerus hyalipennis* (Hayat) and *Anagyrus mangicola* (Figure 7a). *Gyranusoidea tebygi* is the dominant species in parasitoids (71.10 % of their population) (Figure 7b) whereas with 65.07 % attendance, *Marietta Léopardin* predominates in hyperparasitoids (Figure 7c). The unidentified species are respectively 2.47 % and 2.15 % of the

population in parasitoids and hyperparasitoids (Figures 7b and 7c).

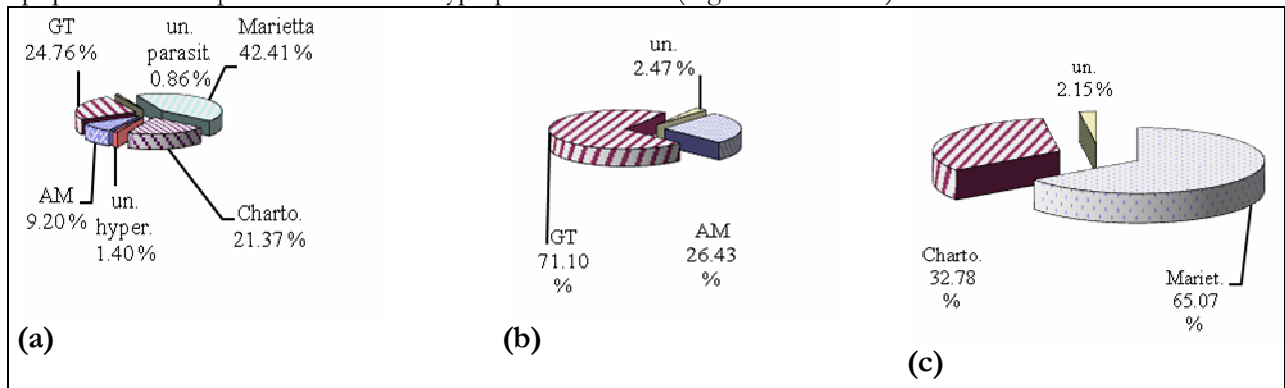


Figure 7: Proportion of different species of parasitoids and hyperparasitoids

(a): Proportion of different species of primary and secondary parasitoids, (b): Proportion of primary parasitoids, (c): Proportion of secondary parasitoids. GT = *G. tebygi*, AM = *A. mangicola*, un. = unidentified parasitoids or hyperparasitoids

3.4.2 Parasitic activity and predominance of different species over time:

Evolution of parasitism rates distinguished two periods of parasitoids (Figure 8). A period of high parasitic activity from September 1999 to March 2000 characterized by a decrease in the density of mummified mealybug and a period of low parasitoids activity from May 2000 to September 2000, marked by a high density of mummies.

The figure 9 illustrates the periods of dominance of primary and secondary parasitoids. There are 4 periods of abundance: the first dominated by hyperparasitoids, begins in September 1999 and ends in November 1999. This phase shows two

peaks of emergence about 41.4 % of the population in September and November 1999. The second begins in mid-November 1999 and ends in February 2000. It is characterized by an alternation rate of emergence of parasitoids and hyperparasitoids, during which neither can dominate. The third goes from March to July 2000. It is marked by high rates of emergence of primary parasitoids, with a peak of 63.93 % recorded in April 2000. The fourth starts in July 2000 and expires in September 2000. It is dominated by hyperparasitoids. A peak of 54.61% is observed in July 2000.

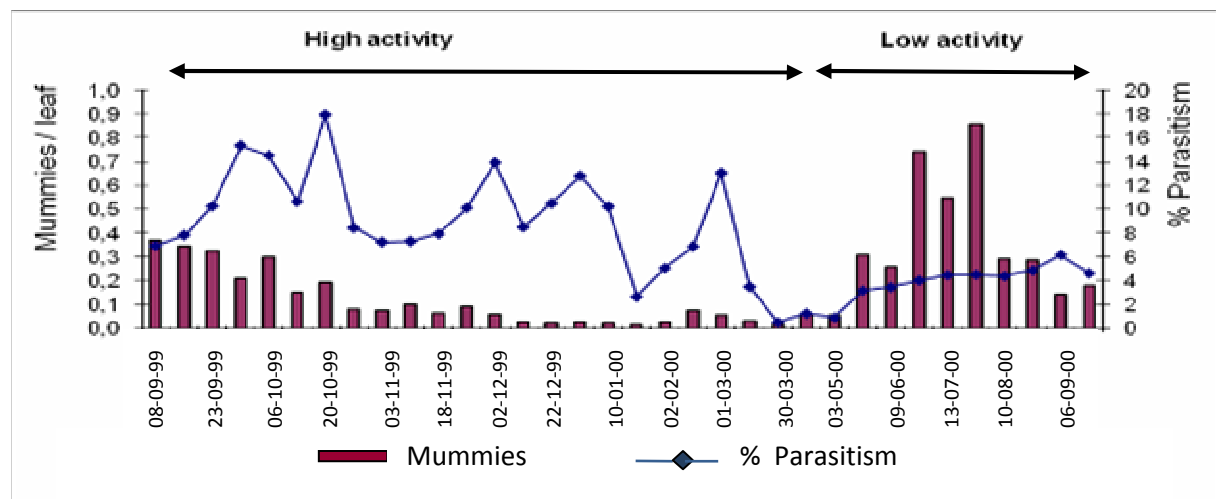


Figure 8: Evolution of the density of mummified mealybug and the rate of parasitism

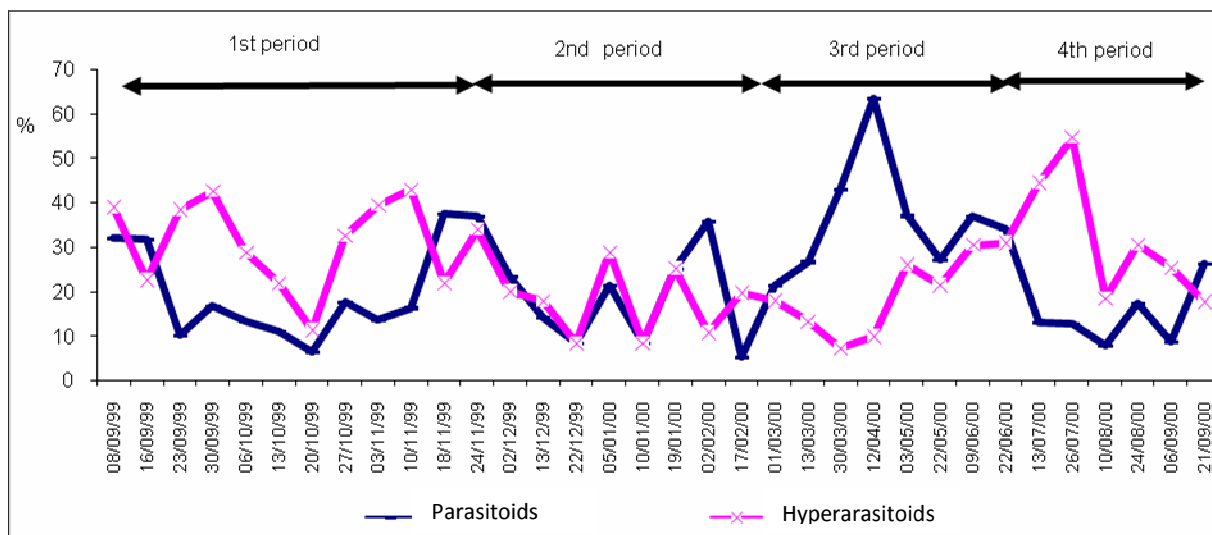
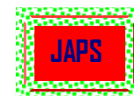


Figure 9: Evolution of primary and secondary parasitoids

3.4.3 Relationships of mango mealybug-parasitoids and parasitoids-hyperparasitoids: Relationships of mango mealybug-parasitoids and parasitoids-hyperparasitoids, show strong correlations between live and mummified

mealybugs, between live mealybug and parasitoids and between mummified mealybugs and hyperparasitoids. Correlation coefficients are positive and statistically significant (Table 3).

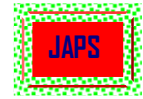
Tableau 3 : Correlation matrix

	Mealybug	Mummies	<i>G. tebygi</i>	<i>A. mangicola</i>	<i>M. leopardina</i>	<i>C. hyalipennis</i>
Mealybug	1					
Mummies	0.770	1				
<i>G. tebygi</i>	0.815	0.837	1			
<i>A. mangicola</i>	0.742	0.772	0.786	1		
<i>M. leopardina</i>	0.722	0.961	0.788	0.729	1	
<i>C. hyalipennis</i>	0.720	0.849	0.802	0.643	0.828	1

4. DISCUSSION

4.1. Sensitivity of three varieties of mangoes to mealybug infestations: Although the varieties of mango benefit from the same macro-climate, the graduation of pest profiles varies from one to the other. This study results confirm those of Boavida *et al.* (1992). *Keitt* variety presented the highest annual density of insects while *Early Gold* indicated the lowest infestation. One explanation could be the differences in leaf area. Leaves of *Keitt* are wider than those of the other two varieties with logically more surfaces exposed to mealybug (Akassey and Daubrey, 1992). These differences in density of infestation can also be explained by the renewing of leaves during tree phenology. Issued new leaves

provide new sites for colonization by mealybug (Boavida and Neunschwander, 1995a). According to Matokot *et al.* (1992), changes in the number of the pests seem to be more related to the host plant than climatic factors. Physiology of mango trees, including the renewal rate of leaves, may partly explain the differences in the populations of mealybug. Indeed, mango trees have a growth rate varying from one tree to another and a rate of vegetative growth and production of branches which depend on the variety (Laroussilhe, 1979). Such physiological characteristics can lead to variability in the level of mealybug populations from a tree to another.



4.2. Graduation profile of the populations of *R. invadens* in the region of Korhogo:

Fluctuations in populations of *R. invadens* showed a low level of infestation from September 1999 to February 2000 and a high density of population from March to August 2000. These periods of abundance of mealybug in the northern Côte d'Ivoire have been identified by N'Guetta (1999). Five generations of 60 to 70 days were obtained in this region. In Congo, Matokot *et al.* (1992) identified five generations from 70 to 80 days (Willink and Moore, 1988; Moore, 1992). In this study, abiotic factors influencing population dynamics of mango mealybug are rainfall, temperature variations and to a lesser extent relative hygrometry. In the northern region of Côte d'Ivoire characterized by a monomodal climate, peaks of abundance of mango mealybug coincide with those of rainfall. This situation is not unique to all ecological zones affected by the pest. Indeed, in southern Benin of bimodal climate, the density of mealybug populations is low during rainy seasons and very high during dry seasons (Boavida and Neunschwander, 1995b).

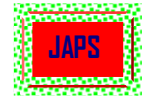
This suggests that rainfalls appear to be an abiotic factor that can affect negatively or positively seasonal variations of the pest. The positive effect of rainfall on the population of pest results of vegetative growth, particularly the issue of new leaves that lead to positive physicochemical changes in the nutritional resources of the tree. The negative effect of rainfall is probably due either to the mechanical action of rain that could dislodge the young larvae out of the leaves, or with the spread of disease in populations of insects favored during the wet season (Akalach *et al.*, 1992). Causal agents of disease are likely entomopathogenic fungi. According to Moore (1992), pathogenic fungi whose presences were confirmed on mealybug are *Neozygites fumosa*, *Hirsutella cryptosclerotium*, *Cephalosporium* sp, *Aspergillus parasiticus*, etc. In this study, it is likely that the negative effect of precipitation (either mechanically or disease) was small because there is an increase in population of *R. invadens* during heavy rains.

4.3. Impact of parasitoids on the graduation profile of the populations of *R. invadens* :

Population levels of mummified mealybug is substantially synchronized with those of live

mealybug. Growth period of live insect begins in March while that of mummies starts in May, meaning 2 month lag between these two phases, corresponding also to destructive actions of the pest by the parasitoids. These results corroborate those of N'Guetta and Kéhé (1998). *Gyransoïdea tebygi* and *Anagyrus mangicola* are the two major primary parasitoids while *Marietta Léopardina* and *Chartocerus hyalipennis* are the major secondary parasitoids of mealybug. *C. hyalipennis* is a gregarious hyperparasitoid (Goergen and Neunschwander, 1994).

During the eight years since their introduction in northern Côte d'Ivoire, a level of 17.95 % of parasitism was developed by *G. tebygi* and *A. mangicola* and hasn't significantly affected the populations' dynamics of mealybug. This situation is not observed where *G. tebygi* and *A. mangicola* have been introduced accidentally. In Democratic Republic of Congo, *G. tebygi* developed from 50 to 90 % parasitism four months after its introduction and thus caused a decrease in the populations of mealybug (Matokot *et al.*, 1992). Despite this low rate of parasitism, establishment of *G. tebygi* and *A. mangicola* seems to be confirmed in the north of Côte d'Ivoire. Indeed, these parasitoids are able to remain on low population densities of their host (less than 2 mealybugs/20 leaves) as it is the case in Togo (Agricola *et al.*, 1989). *G. tebygi* and *A. mangicola* have biological characteristics that may explain their effectiveness as biological control agents (Moore, 1992; Bokonon-Ganta *et al.*, 1995 and 1996; Agricola *et al.*, 1989). But the rapid adaptation of a hyperparasitoids complex in Korhogo area, responsible of nearly 65.18 % mortality of parasitoids appears to be an obstacle to establishing a balance between the populations of mango mealybug and parasitoids (Hala *et al.*, 2004) even though the level of hyperparasitism do not limit significantly the effectiveness of parasitoids (Matokot *et al.* (1992); Moore (2004). Our field studies indicate that the response of hyperparasitoids is highly dependent on the density of parasitoids. This corroborates the results of Hammond and Neunschwander (1992) who reported high percentages of hyperparasitoids when parasitoid *Epidinocarsis lopeszi* (Hymenoptera: Encyrtidae) is abundant on cassava mealybug *Phenacoccus manihoti* (Homoptera: Pseudococcidae),



and low when the populations of *P. manihoti* and *E.*

5 . CONCLUSION

The study of population dynamics of *Rastrococcus invadens* has, firstly, allowed clarification of the biology of this pest under natural conditions, and secondly, to know its seasonal variations. The profile of graduation of mealybug populations varies greatly from one mango tree to another of the same variety and from one variety to another. *Keitt* variety seems to be more conducive to infestations by mealybug than *Early Gold* and *Kent*. The pest is able to develop five generations from 60 to 70 days per year under natural conditions in northern Côte d'Ivoire. Abiotic factors that affect significantly the levels of infestation of mango mealybug are mainly rainfall and temperature variations and to a lesser extent relative humidity. The presence of exotic parasitoids *Gyranusoidea tebygi* and *Anagyrus mangicola* did not significantly affect the dynamics of adult and larval populations of mealybug. Maintaining of these parasitoids on low

lopezi are small.

populations of the pest indicates their acclimatization to the area of study. However, the rapid adaptation of a hyperparasitoids complex could hamper the establishment of a balance between the populations of mealybug and those of parasitoids. To fight against mealybug by massive releases of *G. tebygi* and *A. mangicola*, multiplication of parasitoids on their host can be carried out from February to September, a period that corresponds to a natural increase of the different populations. Also the evolution of the densities of the pest, parasitoids and hyperparasitoids shows it would be wise to make the first releases of parasitoids in March-April, during which all populations are low. For successful biological control against mealybug, the impact of hyperparasitoids and all natural enemies, including predators (ladybugs, thrips), should be taken into account.

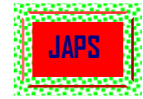
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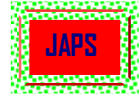
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