Demographic parameters of the invasive species *Bactrocera invadens* (Diptera:Tephritidae) in guinean area of Côte d'ivoire

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Abstract

The invasive species, *Bactrocera invadens* Drew Tsuruta & White, is a major pest of mangoes (*Mangifera indica* L.) and sweet oranges (*Citrus sinensis* Osbeck) in Côte d'Ivoire.. A better knowledge of its biology can improve the control of this new destructive pest. The study was carried out under semi-natural conditions of orchard and in laboratory at the insectarium of Cocody University (28.2 ± 2.7 ° C and 81 ± 3% RH). Thirty newly emerged couples of *B. invadens* were gathered. Five healthy fruits were presented to each couple every 24:00 until the death of the females. The duration of the biological cycle was of 20.93 ± 0.96 days on mango and of 23.83 ± 1.21 days on sweet orange. It comprises 3 larval stages, all infesting for the two hosts and a pupa stage followed by the adult stage. The number of eggs laid by a female was higher on mango (269.13 ± 41.20 eggs) than on sweet orange (58.97 ± 6.35 eggs). The rates of emergence on the two substrates were respectively 74.17 ± 2.71%, and 35 ± 3.08%. The lifespan of adult males was 82.96 ± 5.82 days on mango and 64.3 ± 2.9 days on sweet orange while life span of females was 75.96 ± 4.49 days on the first substrate and 54.7 ± 2.99 days on the second. Mango was the most suitable fruit species for oviposition and larval development of *B. invadens*. These results will allow a better determination of periods and means of intervention in the control of *B. invadens*.

Introduction

The fruit crops are an important source of income but also jobs opportunities for farmers in the world. In Côte d'Ivoire, mango production is at least 100000 tons / year over an area of about 20000 hectares. Mango is the third cash crop after cotton (*Gossypium hirsutum* L.) and cashew (*Anacardium occidentale* L.) (Anonymous, 2007). Unfortunately, the fruit crops are attacked and depreciated by many pests mainly in the tropics. Globally, fruit flies, with about 250 species of economic importance, cause considerable damage (White and Elson-Harris, 1992). Tephritidae species subservient to mangoes and citrus are responsible for the implementation of strict international quarantine systems that limit the export of these commercially important fruits (Landolt and Quilici,

1996). Tephritidae are therefore, a major obstacle to the movement of tropical fruits in the world (Anonymous, 1994). These flies attack over 50 species including citrus fruit, mangoes (Oukil and *al.*, 2002) and various fruit species throughout sub-Saharan Africa (Mwatawala and *al.*, 2009; Rwomushna and *al.,* 2008; Umeh and *al.*, 2008; Vayssières and *al.*, 2008). The main species of fruit flies infesting mangoes in Benin are *B. invadens* (Drew, Tsuruta and White), *Ceratitis cosyra* (Walker), *C. silvestrii* Bezzi and *C. quinaria* (Bezzi) (Vayssières and *al.*, 2005). The main species of fruit flies infesting citrus are *B. invadens* and *Ceratitis capitata* (Wiedemann) in West Africa but primarily *B. invadens* in Benin in Guinea area (Vayssières et *al.* 2010a). These five species are polyvoltin likely to have damage to much of the year (Hala *and al.* 2008; Vayssières et 2009) on tropical fruit species cultivated but wild plants the latter being Tank (Vayssières *and al.*, 2010b). In Africa, only a fraction of the 1.9 million tons of mangoes produced is exported each year. The arrival of *B. invadens* in West Africa and especially in Côte d'Ivoire increased the importance of the damage caused by the Tephritidae of mangoes and oranges, as well as at regional level (Anonymous, 2007). In Côte d'Ivoire, *B. invadens* was listed for the first time in 2005 (Hala et *al.*, 2006). This species colonized all areas of mango production (N'Dépo et *al.*, 2009; N'Guessan, 2009). This work highlighted the importance of damage due to this species which biology is still poorly understood because it was recently described (Drew et *al*., 2005). Knowledge of main demographic parameters of this pest is a precondition to the establishment of effective control methods. Some preliminary biological studies were conducted on this species in East Africa (Ekesi *and al.*, 2006) but it seemed important to complete in West Africa with respect to its two main cultivated hosts, the mango and orange trees. Thus, this work, which is part of the fight against *B. invadens*, to improve the production and marketing in Côte d'Ivoire, aims to determine the number of laid eggs.

Material and methods

Study area

The study was conducted at the experimental station and production Marc Delorme (Lat. 5° 20 ' N, Length. 4° 01 ' W) and the insectarium of the University of Cocody (Lat. 5° 23 ' N, length. 4° 11 ' W), in Abidjan in the south Côte d'Ivoire. The climate is subequatorial with a mean annual rainfall of 2625 mm. Mean conditions of temperature and relative humidity at station Marc Delorme were 25.7 ± 3.3 °C and 78 ± 2 % HR; those of the University of Cocody insectarium were 28.2 ± 2.7 °C and 81 ± 3 % HR.

Material

The vegetable material were fruits of mango tree (*Mangifera indica* L.), variety mango and of orange tree (*Citrus sinensis* Osbeck), variety ambersweet. The fruits were harvested from orchards in Abidjan and were used as substrates of laying and larval development. The animal material was the fly species, *B. Invadens,* which emerged from infested mangoes and oranges and harvested in our experimental plots.

Methods

Breeding of fruits flies

The fruits, bitten by female flies collected from fruit trees, were brought to the laboratory. They were incubated in trays containing sterilized and humidified sand. Sterilization was carried out using an autoclave at a temperature of 121°C and a pressure of 1.5 bar. Five days later, the incubated fruits were immersed. The pupae were recovered by floating and the larvae by sieving. Larvae and pupae were then kept in plexiglas boxes (28 X 27 X 9.5 cm) containing sterilized and humidified sand, until adult emergence. The biological parameters studied were the number of eggs laid per female during its life, the duration of the incubation period of eggs, duration of the biological cycle, rate of emergence, sex ratio and adult longevity.

Determination of the number of eggs laid per female and the duration of eggs incubation period

Two batches of thirty pairs of *B. invadens*, newly emerged, were made up one for the tests in mangoes and the other for test on citrus. There were placed, each in a muslin sleeve containing 5 healthy fruits on the tree. Every 24 hours, the pitted fruits (mangoes and citrus), were removed from the sleeve and was moved to continue the experiments on other healthy fruits on the tree, until the death of the female. For the first batch, the eggs laid by the females under the skin of the fruit were counted, under binocular magnifying glass, removal of pulp, at the place of the deposit of eggs. The average number of eggs laid per female during its life (L) was calculated.

$$L = \frac{\sum eini}{\sum ni}$$

ei: number of laid eggs;   *ni*: number of the females

For the second batch, fruits harbouring eggs were placed in trays containing sterilized sand. These eggs were observed daily until hatching. The average incubation period (Pi), which is the time between egg lying (l) of the hatching (h) was noted.

$$Pi = \frac{\sum tivi}{\sum vi}$$

ti = h - l; vi = number of eggs

Determination of the duration of biological cycle, the rate of emergence, sex ratio and longevity of the adults
At the hatching of the second batch of females, the larvae stage 1 were isolated and deposited in healthy fruits having undergone a notch and then they were placed, each in a box with holes covered by muslin and containing sterilized sand. These fruits were monitored daily and the dates of successive exuviae were noted and the duration larval development (Dl), expressed in days, was given.

$$Dl = \frac{\sum xini}{\sum ni}$$

xi = Time between egg and larva stage 3 (TS3);
ni = number of larvae stage 3

Pupation (P), expressed in days was noted. It is the time between (TS3) the moment of obtaining the pupa (Tp).

$$P = \frac{\sum aibi}{\sum bi}$$

ai = Tp - TS3: time taken by the larva stage 3 to become a pupa; bi: number of pupae
The duration of pupal development (Dp), in days, was determined. It corresponds to the time between pupation (P) and the emergence of the adult (Ea).

$$Dp = \frac{\sum cidi}{\sum di}$$

ci = Ea - P: time taken for the pupa to become adult; di: number of adults

The average rate of emergence (Me) as a percentage was calculated.

$$Me = \frac{\sum eifi}{\sum fi}$$

$$ei = \frac{\text{number of adult}}{\text{number of egg laid}} \times 100$$

fi = number of female parent

The duration of the biological cycle (Dc.), expressed in days, the period egg-laying and adult stage, was determined.

$$Dc = Pi + Dl + P + Dp$$

The mean of the sex ratio (Sr), as a percentage, was calculated for the offspring of the 30 females.

$$Sr = \frac{\sum gihi}{\sum hi}$$

$$gi = \frac{\text{number of male emerging}}{\text{number of female emerging}}$$
hi = number of female parent

Adults were fed honey diluted in water to 5%, in muslin sleeves. The number of dead imagoes was increased each day until death of the last individual. The average longevity of adults (Fd), expressed in days, was determined.

$$Fd = \frac{\sum liki}{ki}$$

li: longevity; ki: number of insects

Statistical analysis
Data processing was carried out using the software Statistica version 6.0. Each test was repeated 30 times. The results were subjected to a variance analysis (ANOVA). Mean separations were done using the Newman-Keuls test and Mann-Withney U test at 5%.

Results

Number of eggs laid and eggs incubation period
During its life, a female laid eggs from 201 to 325, or an average of 269.13 ± 41.20 eggs in mango. In orange, the number of eggs ranged from 51 to 74 eggs with an average of 58.97 ± 6.35 eggs. Mann-Whitney U test reported a significant difference between the values obtained on both substrates (U = 3, P <0.001).
The period of incubation of the eggs ranged from 2 to 4 days, an average of 2.6 ± 0.55 days in the mango and 3.33 ± 0.75 days in the orange (F = 18.88, P <0.001) temperature and relative humidity equal (Table I).

Duration of biological cycle, emergence rate, sex ratio and longevity of adult flies

Larval development took place from 7 to 9 days in the mango and 8 to 10 days in the orange or the respective average duration of 7.97 ± 0.60 and 8.73 ± 0.68 days. Statistical analysis indicated a significant difference between the average duration of larval development in both remained breeding substrates (F = 20.592, P <0.001). The larvae stage 3, left the fruit, in the sand to turn into pupae.

The duration of the pupal stage ranged from 1 to 2 days in the mango and 1 to 3 days in the orange with respective averages of 1.37 ± 0.48 and 1.63 ± 0.55 days. Statistical analysis indicated a significant difference between the two periods of pupation (Newman-Keuls test at 5%: F = 491.4226, P <0.001). Pupation lasted from 8 to 10 days from the mango, or an average of 8.97 ± 0.66 days. From orange, pupation lasted from 9 to 11 days with an average of 10.27 ± 0.73 days. Statistical analysis revealed a significant difference (F = 50.99, P <0.001) (Table I).

The duration of *B. invadens* biological cycle ranged from 19 to 22 days in the mango with an average of 20.93 ± 0.96 days. In the orange, egg turn into adult from 22 to 27 days, or an average of 23.83 ± 1.21 days. Statistical analysis indicated a significant difference between the average durations of *B. invadens* on mango biological in the orange (F = 101.57, P <0.001).

The development of *B. invadens* included three (3) larval stages and a pupal stage. The rate of emergence of the fruit fly varied from 71 to 79% with an average of 74.17 ± 2.71% in the mango. This rate was ranged from 31 to 44% with an average of 35 ± 3.08% in the orange (Table II). Statistical analysis showed a significant difference between the rates of emergence (F = 2647.1, P <0.001).

The sex ratio was 0.74 ± 0.02 in the mango and 0.87 ± 0.03 in the orange (F = 382.7, P <0.001) (Table II). Statistical analysis indicated highly significant difference between these both values.

The average longevity of males was 82 ± 5.82 days in the mango and 64.3 ± 2.9 days in the orange; that of females was 75.96 ± 4.49 day in the mango and 54.7 ± 2.99 days in the orange. The Newman-Keuls test revealed a significant difference between the average longevity of males and females on the same substrate breeding. This difference was also observed between the average longevity of individuals of the same sex which development took place in each of these fruits (F = 137.11, P <0.001) (Figure 1).

Discussion

The demographic parameters of *B. invadens* were significantly different between two hosts grown and commercial interest such as the mango and the sweet orange. The average number of eggs laid by a female of *B. invadens* is 5 times higher than in the orange. This suggests that the sweet orange is not a very attractive host fruit for females of *B. invadens*. For *C. capitata*, the low number of eggs laid in the orange is due to the presence of toxic oils in the rind of the fruit (Salvatore and *al*., 2004; Papachristos and *al.*, 2008). Thus, the skin of citrus fruits appears to be a limiting factor for development of immature stages. This argument is similar to that of Papachristos and *al.* (2008) who worked on *C. capitata*. The number of eggs laid by *B. invadens* in the mango (269.13 ± 41.20 eggs) was different of that obtained in the same substrate (794.6 eggs) by Ekesi and *al.* (2006). This difference is related to experimental conditions. Indeed, this work focused only on the first generation of *B. invadens*. Adults of *B. invadens* can adapt to development conditions until the fourth generation (Ekesi and *al.*, 2007).

The average duration of eggs incubation was shorter in the mango than in the orange at temperature and RH equal. This was true for the average duration of larval and pupal development. The differences are linked to the structure, the chemical composition of the skin and pulp of host fruits. Indeed, the skin of citrus fruit is quite thick and contains toxic substances; their pulp has a high acidity (Papachritos and *al.,* 2008). As for the mango's skin is thin and non toxic. Its pulp has a low acidity. The difference between development times obtained in the mango and the orange could be explained by the richness of the mango pulp in nutrients conducive to larval development. The duration of *B. invadens* biological cycle was longer in the orange than in the mango. The difference may reflect a slowing of larval development probably due to the acidity of the orange pulp (Papachristos and *al.*, 2008). This argument joined that of Papadopoulos and Katsoyannos (2002). These authors reported that, on three apple varieties, the duration of larval development was longer when the acidity was very high. In the mango, the results showed that the duration of *B. invadens* biological cycle was shorter (20.93 ± 0.98 days) than that obtained by Ekesi and *al.* (2006) (25 days). This difference would be linked to the substrate breeding but also the fact that the HR ranged from 50% in Kenya to 80%, in Côte d'Ivoire. Indeed, experiments were conducted on natural substrates (mangoes and oranges) so that these authors used an artificial substrate made from wheat and carrot (Ekesi *and al.*, 2006).

The rate of adult emergence of *B. invadens* was two times higher in the mango than in the orange. The differences in thickness and chemical composition of the skin of two fruits could explain this variation. Indeed, the thick skin of the orange is a physical barrier that prevents the female to lay eggs in the pulp. Mortality is also related to the presence of

toxic substances in the orange zest. The orange skin contains chemicals close to oil (Duyck and *al*., 2004). Another factor, the partitioning of the orange pulp could also cause death of *B. invadens* larvae. Indeed, they should provide more effort to reach the quarters of orange and move around. These observations joined those of Papachristos and *al.* (2008) whose work focused on *C. capitata.* The sex ratio was higher in the orange than in the mango. This is due to the fact that the mango is more conducive to the reproduction of *B. invadens*. The results showed that the longevity of *B. invadens* adult is gendered. Indeed, for *B. invadens*, males lived longer than females, regardless of the substrate for the development of immature stages. Similar results were reported by Ekesi and *al*. (2006). This trend was also observed in other species of fruit flies such as *C. capitata* (Papadopoulos and *al.*, 2002; Carey and *al.*, 2005). The short longevity of the females could be attributed to the energy released during spawning. Indeed, female insects use their energy reserves to spawn eggs. This argument was also reported by Zannou (2000) studying the biological parameters of adults from a strain of *Callosobruchus maculatus* Fab in Benin. The reproductive activity (mating and egg laying) contributes to a strong decrease in the longevity of females. This is what Williams (1996) described as "reproduction cost", a concept that links the reproductive effort to the other functions of the insect. Longevities were shorter on orange than mango, both in males than in females. The nature of the host would influence the longevity of *B. invadens*. This result was also observed by N'Guessan (2009).

Conclusion

The study of demographic parameters showed that *B. invadens* is a holometabolous insect. It's the biological cycle which includes three instars and a pupal stage. The duration of the biological cycle was 20.93 ± 0.96 days in the mango and 23.83 ± 1.21 days in the orange. Males lived longer than females on two supports breeding. A female of *B. invadens* laid an average of 269.13 ± 41.20 eggs in the mango and 58.97 ± 6.35 eggs in the orange semi-field with a higher rate of emergence of the first fruit. The sex ratio in the orange was higher than in the mango. The biological cycle was shorter in the mango than in the orange. Mango is the most suitable substrate for the development of *B. invadens*. The study determined the average number of eggs lay per female and identified the infesting stages. The infestation is related to the ability of the female egg laying and the development of three larval stages, the number of eggs laid and the biological cycle could provide important information about the period of infestation. Thus, knowledge of the biological cycle of *B. invadens*

would implement the appropriate methods and establish a control calendar.

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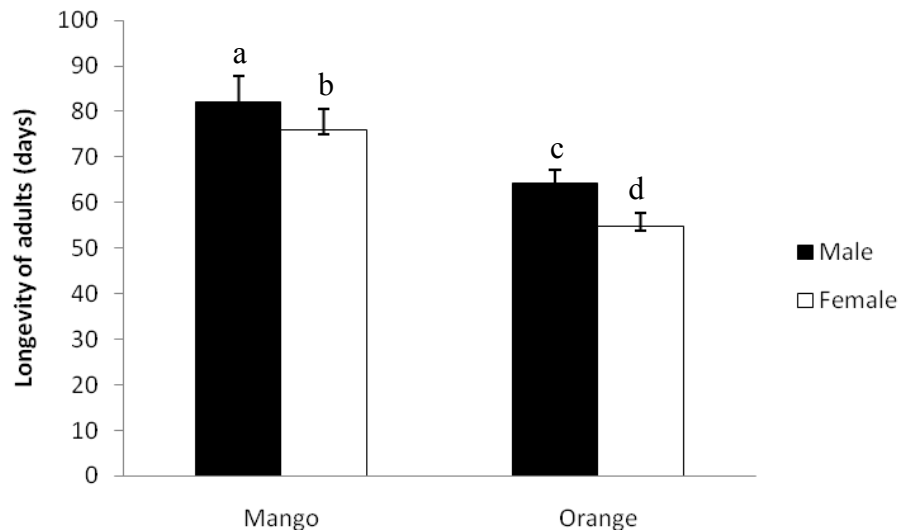
Figure 1: Longevity of the adults of *B. invadens* according to the substrate of laying (mango or orange)

Test Newman-Keuls au seuil de 5%

F = 137.11   df = 1   P < 0.001

Table 1: Mean durations of development of the immature stages of *B invadens* on two fruit-bearing species

Substrate of breeding	Mean duration (days)				
	Incubation of eggs	Larval development	Pupation	Pupal development	Biological cycle
(*Mangifera indica*) Mango	2.60 ± 0.55^a	7.97 ± 0.60^a	1.37 ± 0.48^a	8.97 ± 0.66^a	20.93 ± 0.96^a
(*Citrus sinensis*) Sweet Orange	$3.33 \pm 0,75^b$	8.73 ± 0.68^b	$1.63 \pm 0,55^a$	10.27 ± 0.73^b	23.83 ± 1.21^b

n = 30

Test Newman – Keuls au seuil de 5%

Duration of eggs incubation	F = 18.088	df = 1	P < 0.001
Duration of larval development	F = 20.592	df = 1	P < 0.001
Duration of Pupation	F = 491.4226	df = 1	P < 0.001
Duration of pupal development	F = 50.99	df = 1	P < 0.001
Duration of biological cycle	F = 101.57	df = 1	P < 0.001

Table 2: Mean rate of emergence and mean of sex-ratio adults of *B. invadens*

Substrate of breeding	Mean rate of emergence (%)	Mean of sex-ration
(*Mangifera indica*) Mango	$74{,}17 \pm 2{,}71^{b}$	$0{,}74^{a} \pm 0{,}025$
(*Citrus sinensis*) Sweet orange	$35{,}00 \pm 3{,}08^{a}$	$0{,}87^{b} \pm 0{,}026$

n = 30

Test de Newman–Keuls au seuil de 5%

Rate of emergence	F = 2647.15	df = 1	P < 0.001
Mean of sex-ratio	F = 382.7	df = 1	P < 0.001

Aims and Scope

The Journal of Asian Scientific Research is a monthly, peer-reviewed international research journal which deals with empirical as well as theoretical issues. The editors welcome papers in all the major issues including:

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Arrow, K. (1970) "The Organization of Economic Activity: Issues Pertinent to the Choice of Market Versus Non-market Allocations" in Public Expenditure and Policy Analysis by R.H. Havenman and J. Margolis, Eds., Markham: Chicago, pp. 67-81

Benabou, Roland (1994) "Education, Income Distribution, and Growth: The Local Connection". NBER working paper number 4798

Berglas, E. (1976) "Distribution of tastes and skills and the provision of local public goods". Journal of Public Economics Vol. 6, No.2, pp.409-423.

Edgeworth, F.Y. (1881) Mathematical Psychics, Kegan Paul: London.

Mas-Colell, A and J. Silvestre (1991) "A Note on Cost-Share Equilibrium and Owner- Consumers" Journal of Economic Theory Vol.54, No.1,pp. 204-14.

Appendix: At the end of the paper

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