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Population dynamics of *Phytolyma fusca* Alibert (Hemiptera: Homotomidae), psyllids pest of *Milicia excelsa* (Welw) (Rosales: Moraceae) in Cameroon

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Abstract

Milicia excelsa (Welw) C.C. Berg is among the most valuable important timber species in Cameroon. National programs of establishing plantation of *M. excelsa* have been constrained by the gall forming psyllid, *Phytolyma fusca*. This paper focused on the population dynamics of *P. fusca*, was conducted in an experimental *M. excelsa* plantation in Yaounde. Psyllid galls were counted in situ, collected and dissected under laboratory conditions. Adults and larval stages were counted. The results showed, in 2009, three periods of psyllids outbreaks: February - April, August - October and November - December. Seven generations of the pest are noted. In 2010, three periods of psyllids outbreaks are noted: January - February, May - September and November - December. Six generations of the pest existed. The number of generations varies from one year to another and depends on the phenology of the host plant which depends on the climatic factors.

Keywords: *Milicia excelsa*, *Phytolyma fusca*, psyllid, population dynamics, Cameroon

1. Introduction

Jumping plant lice or psyllids are sap-sucking insects of the plants. They belong to the order Hemiptera and are mainly associated with perennial plants^[1, 2, 3]. However, some species are associated with Monocotyledons^[4] and other species are recorded on conifers^[5]. The known psyllid species are mainly those of the temperate and subtropical regions of the world. Several species of economically important agricultural crop plants and commercial timbers are under the permanent attack of the devastating indigenous psyllids pests. Therefore, psyllids are responsible for huge losses in orchards and plantations and also during regeneration of commercial timber species. Among the commercial timbers under the attacks of psyllids in Cameroon is *M. excelsa*.

M. excelsa is a forest tree presents naturally in Sub-Saharan tropical Africa. *M. excelsa* is known as Iroko or African Teak as commercial common name. It is also known under various names according each country of origin: "Abang bang" from Cameroon, "Odum" from Ghana and Cost Ivory, "Rokko" from Nigeria, "Kambala" or "Lusanga" from Congo Democratic Republic^[6]. In Cameroon, this plant is considered as one of the main timber export. It is particularly preferred for the construction of outdoor, furniture especially for flooring and paneling. The timber is strong, moderately hard and very durable with interlocked and sometimes irregular grain. It seasons well in air and does not warp or shrink afterwards^[7, 8]. According to authors^[7, 8, 9], Iroko is highly resistant to termites and fungal attack.

M. excelsa hosted insects responsible for damages on leaves and young seedlings. Among these insects a psyllid species, *Phytolyma fusca* is the main pest of this important forest plant. Taxonomic revision of psyllids made recently^[10] confirms that *Phytolyma* is member of Homotomidae Heslop-Harrison family, Macrohomotominae While and Hodkinson subfamily and Phytolymini While and Hodkinson tribe. Several authors have held discussions based on *Phytolyma*/Moraceae relationship. *Phytolyma lata* Walker seems to be restricted to the host *M. regia* which occurs naturally between Gambia and Volta region of Ghana. *Phytolyma lata* var *P. fusca* Alibert should be regarded as a distinct species attacking *M. excelsa* which occurs in a broad belt across Africa from Liberia to northern Mozambique^[11]. Where the two species of *Milicia* naturally overlap in Liberia, Ivory Coast and Ghana, *P. lata* and *P. fusca* seem to remain host specific. Hollis^[12] described four species within the *Phytolyma* genus and Tamesse *et al*^[13] described *Phytolyma tchuentei*, a new psyllid species of *Morus mesozygia*

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(Moraceae), from Cameroon. Host species of *Phytolyma* are normally species of Moraceae family. Attempts to grow *M. excelsa* in plantations have generally failed due to attack by gall-forming *P. fusca* in Cameroon as it is also the same case with *P. lata* and *P. tuberculata* infestations of *M. regia* [14]. Exploitation of *M. excelsa* is mainly done from natural forest; however, regeneration has proven to be inadequate to match with the rate of their susceptibility to *P. fusca* attack [9]. The attack leads to the formation of galls on the shoots, which is followed by dieback of foliage down to the woody tissue. Consequently, these damages disrupt physiological processes causing growth reduction and kills seedlings [15]. Many studies focus on natural regeneration of *M. excelsa* [16] and the evaluation of a resistant line of *M. excelsa* [17]. In Ghana and Nigeria, several chemicals have been evaluated for control of *Phytolyma* species, but there were ineffective because of cryptic nature of the pest.

P. fusca was recorded in Cameroon for the first time by [12], this author identified specimens collected on *M. excelsa* in the Njombe region. No others investigations were published on the biology of *P. fusca* in Cameroon. The control of the insect pests is based through understanding of their population dynamics [18]. According to some author [19], studies on population dynamics help to keep pest populations below the threshold of economic damage by using insecticides. It allows finding strategies to optimize the use of chemicals in Integrated Pest Management [20] and provides useful information for taking preventive measures against the resurgence of infestations [21]. The aim of this work was to study the population dynamics of *P. fusca* psyllid of *M. excelsa* from Cameroon. Abiotic factors that regulated natural populations of the pests are investigated.

2. Materials and methods

2.1 Site and study period

The study was conducted for two consecutive years from February 2009 to January 2011 in an experimental plantation at Nkolbisson, Yaounde VIIth Sub-division. The altitude and geographical coordinates are : 740m, 03°52'290" N and 11°25'420" E. Host plants were planted three years ago before and they are about 2 m height. The total number of investigated plants was ten. These plants received no pesticide treatment before and during the study period.

2.2 Sampling method

Sampling was done once a week from February 2009 to January 2011 (during 24 months).

Branches of each of the ten plants were inspected during our survey. One branch of three leaves or more leaves per plant were examined and galls formed on infested leaves were counted. During psyllids outbreaks, up to seven infested leaves with galls forms were collected. Since adults and larvae leave inside the galls, we opened the closed gall with the razor blade and larvae and adults of the psyllid are captured using a mouth aspirator and conserved in 70% alcohol. Some of the adults were captured directly on the plant using a sweep net of 0.5 mm mesh sizes. In the laboratory, psyllids were examined using a Leica Stereomicroscope. The number of larvae of different instars, males and females and galls recorded on each plant were counted.

Meteorological data were obtained from the Institute of Agricultural Research for Development (IRAD), Nkolbisson. The selected parameters are temperature, relative humidity, rainfall, wind speed and insolation.

2.3 Data analysis

The SPSS statistical program (19.0) was used to compare mean of gall with non-parametric Wilcoxon test and changes in abundances of different development stages of psyllids of *P. fusca* with Mann-Whitney test ($P < 0.05$). This program was also used to perform Spearman's correlations between abiotic factors that may regulate the population dynamics of *P. fusca* in the Yaounde region (Cameroon).

3. Results

Adult psyllid females of *P. fusca* laid their eggs on young leaves of the buds. Eggs are inserted along the veins of the young leaves. After hatching, the first instar larvae dig a hole in the plant tissue and its development inside the hole produced a closed gall. The successive molting process of larvae took place inside the galls. Very adjacent galls communicate through galleries. Female continue laying eggs on the top of the galls and various larval instars and adults were recorded inside the same gall. During the psyllids outbreaks, young buds and leaves were totally used to form galls of this pest (Fig 1a). At the end of the larval development, the closed gall open (Fig 1b) and adults escape. The infestation bud of *M. excelsa* dried after the last molt of the psyllid (Fig 1c). Sometimes for unknown reasons, gall may dry and never release adult psyllid. Inside the galls lived together nymphs (Fig 1d) of various stages and adults (Fig 1e).

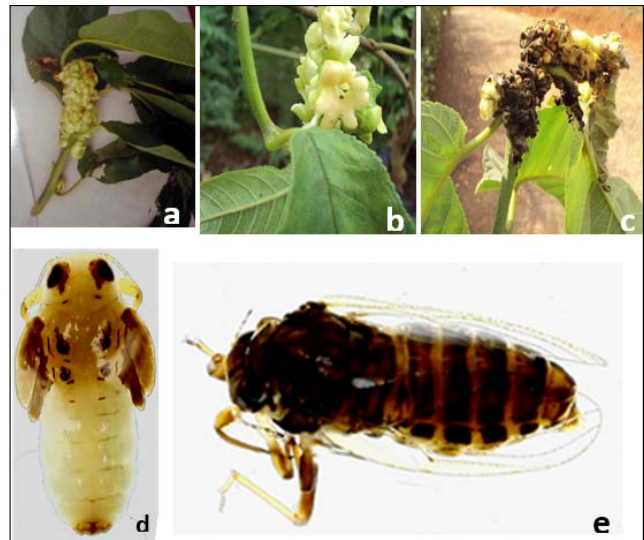


Fig 1: *P. fusca* and damages causes on *M. excelsa*: **a:** young bud of *M. excelsa* with galls formed by *P. fusca*; **b:** galls open after the last molt of the nymphs; **c:** young bud of *M. excelsa* dried after infestation by *P. fusca*; **d:** nymph; **e:** adult.

P. fusca is responsible for the loss of young seedlings in nursery and constituted the main limiting factor for the regeneration of *M. excelsa*. The damages were expressed by the variation of the number of galls formed on young leaves. In 2009, the galls were numerous in February, August, September and October (Fig 2). In 2010, the galls formed were abundant in February, March, June, July, August, November and December (Fig 2).

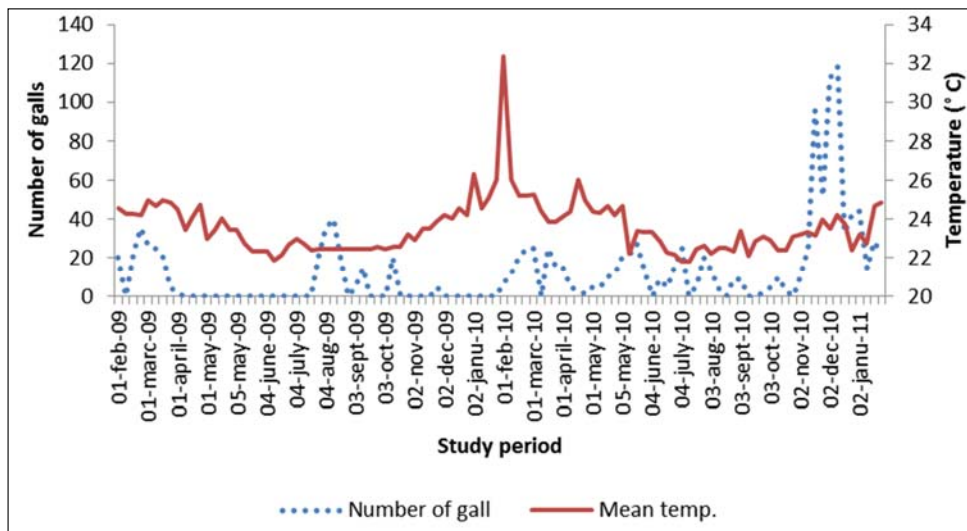


Fig 2: Numerical variations of the number of galls of *P. fusca* on leaves of *M. excelsa* in an experimental plantation depending on temperature in Yaounde from February 2009 to January 2011.

The comparison of the mean numbers of galls formed during the two years of study shows a very highly significant difference ($Z = -4.79, P < 0.001$). This suggests that the fluctuation in the number of galls on host plant was not identical during the two years of study. The Spearman correlation test between temperature and the number of galls produced by *P. fusca* is positive but not significant ($r = 0.127; P = 0.19$). The same trend was observed considering the influence of rainfall; but the Spearman correlation is negative and not significant ($r = -0.063; P = 0.52$).

3.1 Numerical variations of larvae of early stages

In 2009, four peaks were recorded for the first, second and

third instar larvae of *P. fusca* respectively in March, August, October and December. The most important one was observed in March (Fig 3).

In 2010, three peaks were recorded for the first instar larvae respectively in March, June and December. For the second instar larvae, three peaks were recorded respectively in February, May and December. For the third-instar larvae three peaks were recorded in March, May and December. The most important one was obtained in March. (Fig 3)

Among the early larvae stages, the highest peak was recorded in March 2009. No early larval stage was spotted on young leaves between early May and late July 2009.

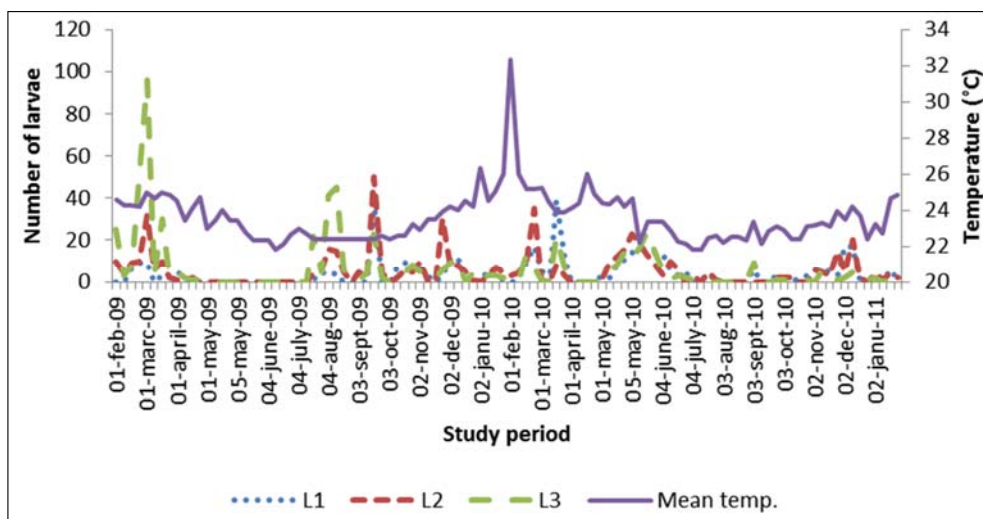


Fig 3: Numerical variations of early larval stages of *P. fusca* on *M. excelsa* in an experimental plantation depending on temperature in Yaounde from February 2009 to January 2011.

The comparison of the variation in the abundance of first instar larvae in the two years of study shows a significant difference ($U = 1050.0, P < 0.05$). For the second larval stage, the difference is not significant ($U = 1328.5; P = 0.88$); also the difference was not significant for the third larval stage ($U = 1159, P = 0.20$). In general, there were no significant differences from one year to another for nymphs of 1st, 2nd and 3rd larval stages of *P. fusca*.

3.2 Numerical variations of larvae of advanced stages

In 2009, we recorded five peaks for the 4th larval instar respectively in February, April, August, October and December with the highest one was recorded in October. The numerical variations of the 5th larval instar also showed five peaks respectively in March, August, October, November and December with the most important one in August (Fig 4). The most important peak was recorded in September 2009 for the fourth larval stage.

In 2010, the numerical variations showed five peaks for the fourth larval instar recorded respectively in February, April, May, September and December while for the fifth larval

stage, four peaks was recorded respectively in February, April, September and December (Fig 4). The highest peak in 2010 was recorded for the fifth larval stage in February.

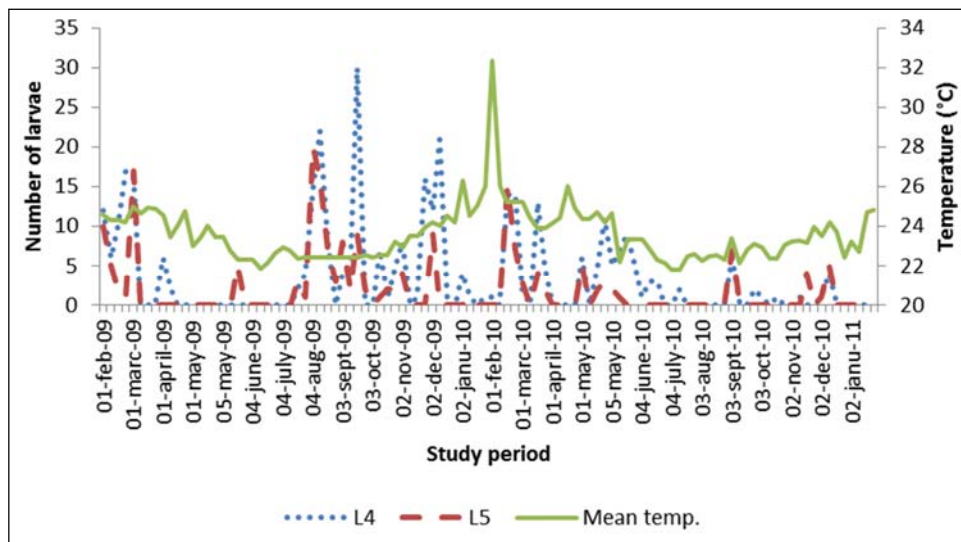


Fig 4: Numerical variations of advanced larval stages of *P. fusca* on *M. excelsa* in an experimental plantation depending on temperature in Yaounde from February 2009 to January 2011.

The comparison of the variation in the abundance of the advanced larval stages during the two years of study, shows that there was a no significant difference between for the fourth larval instar ($U = 1202.0, P = 0.29$) and for the fifth larval instar ($U = 1130.50, P = 0.09$). In general, results showed that advanced larvae stages show no significant difference ($P > 0.05$) from one year to another.

3.3 Numerical variations of adults

In 2009, we observed seven peaks, the 3 most important peaks

were obtained respectively in March, October, and December. Comparing the average number of males and females collected on *M. excelsa*, we noticed that, the numbers of females were more important in March, while the numbers of males were more important in October and December (Fig 5). In 2010, six peaks were recorded and the most important one was noted in February, May, and December. The number of females was higher in February and May, while the number of males was higher in December (Fig 5).

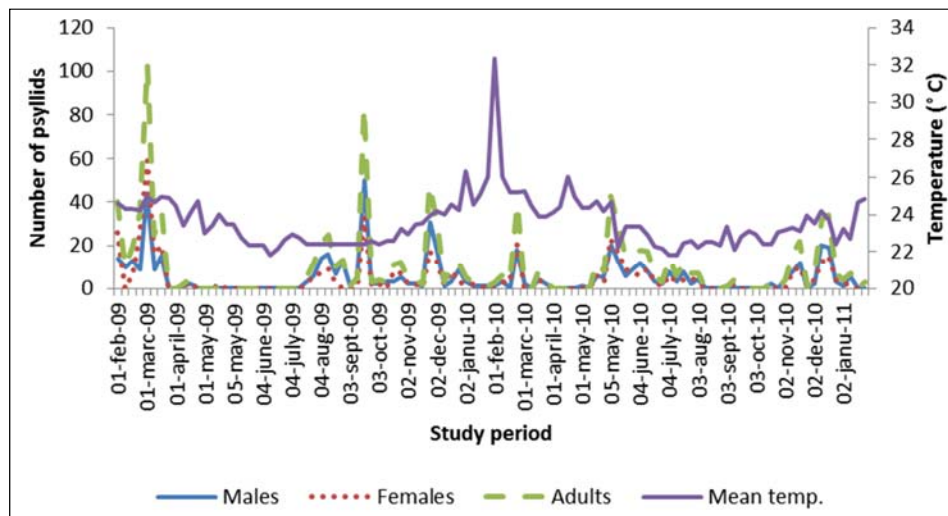


Fig 5: Numerical variations of adults of *P. fusca* on *M. excelsa* in an experimental plantation depending on temperature in Yaounde from February 2009 to January 2011.

The comparison of the variation in the abundance for males and females during the two years of study, showed that there were no significant difference from one year to another for both males ($U = 1291.0, P = 0.68$) and females ($U = 1282.5, P = 0.64$). This suggests that the numerical variation of males and females do not significantly vary from one year to another.

3.4 Impact of abiotic factors on the numerical variation of P. fusca in Yaounde

The ombrothermic diagram of Yaounde (Fig 6) shows 4 seasons, two rainy seasons and two dry seasons. Rainy season: August–November and March - July. Dry season: November - March and July - August. During the two years of study (2009 and 2010), we recorded in February, 7 days which the temperature was more than 32 °C.

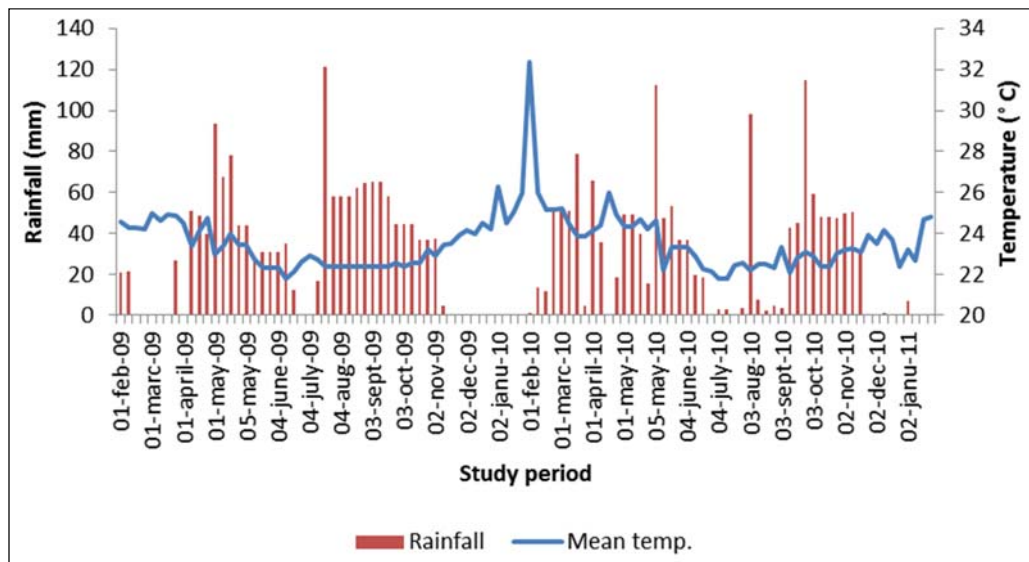


Fig 6: Ombrothermic diagram of Yaounde region from February 2009 to January 2011.

3.5 Effects of temperature on the numerical variations of *P. fusca*

The Spearman correlation between the numerical variation of individuals of various developmental stages of *P. fusca* and temperature was: $r = 0.234$; $P < 0.01$ for the first larval instar; $r = 0.273$; $P < 0.001$ for the second larval instar; $r = 0.206$; $P < 0.05$ for the third larval instar; $r = 0.137$; $P = 0.164$ for the fourth larval instar; $r = 0.040$; $P = 0.684$ for the fifth larval instar; $r = 0.034$; $P = 0.736$ for the males and $r = 0.116$; $P = 0.241$ for the females. Considering these values of Spearman correlation test (Table 1), we noticed that the correlation coefficient is positive for all developmental stages but significant only for the first, second, and third instars larvae. Then the larval of the earlier stages are more vulnerable to the variation of the temperature in our region. This result expresses the fact that when the ambient temperature was higher, the numbers of psyllids counted were low.

3.6 Effects of relative humidity on the numerical variations of *P. fusca*

The Spearman correlation between the numerical variation of individuals of various developmental stages of *P. fusca* and the relative humidity was: $r = -0.123$; $P = 0.215$ for the first larval instar; $r = -0.105$; $P = 0.287$ for the second larval instar; $r = -0.089$; $P = 0.367$ for the third larval instar; $r = -0.002$; $P = 0.988$ for the fourth larval instar; $r = 0.051$; $P = 0.610$ for the fifth larval instar; $r = 0.021$; $P = 0.835$ for the males and $r = -0.067$; $P = 0.502$ for the females. According to Spearman correlation test (Table 1), we noted that the correlation coefficient was negative or positive but always not significant ($P > 0.05$).

3.7 Effects of rainfall on the numerical variations of *P. fusca*

The Spearman correlations between the numerical variation of individuals of various developmental stages of *P. fusca* and rainfall was: $r = -0.055$; $P = 0.58$ for the first larval instar; $r = -0.030$; $P = 0.762$ for the second larval instar; $r = 0.041$; $P = 0.678$ for the third larval instar; $r = 0.113$; $P = 0.253$ for the fourth larval instar; $r = 0.222$; $P < 0.05$ for the fifth larval

instar; $r = -0.043$; $P = 0.663$ for males and $r = -0.138$; $P = 0.162$ for females. According to Spearman correlation test (Table 1), we noted that the correlation coefficient was negative and not significant with 1st and 2nd larval instar, and adults; it is positive and not significant for 3rd and 4th larval instars. The correlation is positive and significant for 5th larval instar. Then, the numerical variation of 5th larval instar of *P. fusca* was correlated with the quantity of rainfall. Rain fall should be favorable to the development of this larval instar in our region.

3.8 Effects of wind speed on the numerical variations of *P. fusca*.

The wind, because of its speed is being proved to have negative influence on all the developmental stages of psyllid as well as the adults. This result suggests that the wind, because of its mechanical action, could help to clean the leaf's surface and transport individuals. The Spearman correlation between the numerical variation of adults of *P. fusca* and wind speed was: $r = -0.148$; $P = 0.134$ for males and $r = -0.051$; $P = 0.607$ for females (Table 1). According to the Spearman correlation test, we noted that the correlation coefficient was negative and not significant for adults of *P. fusca*.

3.9 Effects of insolation on the numerical variations of *P. fusca*

The Spearman correlations between the numerical variation of individuals of various developmental stages of *P. fusca* and insolation was: $r = 0.515$; $P < 0.001$ for the first larval instar $r = 0.706$; $P < 0.001$ for the second larval instar; $r = 0.617$; $P < 0.001$ for the third larval instar; $r = 0.532$; $P < 0.001$ for the fourth larval instar; $r = 0.459$ $P < 0.001$ for the fifth larval instar; $r = 0.967$; $P < 0.001$ for the males and $r = 0.920$; $P < 0.001$ for females. According to these values of Spearman correlation test (Table 1), we noted that the correlation coefficient was positive and highly significant with all the larval instars and adults. Then, the numerical variation of individuals of various developmental stages of *P. fusca* depends on the variation of insolation in our region.

Table 1: Spearman correlations test between developmental stages of *P. fusca* and some climatic parameters in Yaounde from February 2009 to January 2011.

Parameters		Developmental stages							
		L1	L2	L3	L4	L5	Males	Females	Adults
Temperature	r	0.234	0.273	0.206	0.137	0.040	0.034	0.116	0.075
	P	0.017*	0.005**	0.036*	0.164	0.684 NS	0.736 NS	0.241 NS	0.448 NS
Relative humidity	r	-0.123	-0.105	-0.089	-0.002	0.051	0.021	-0.067	-0.021
	P	0.215 NS	0.287 NS	0.367NS	0.988 NS	0.610 NS	0.835 NS	0.502 NS	0.830 NS
Rainfall	r	-0.055	-0.030	0.041	0.113	0.222	-0.043	-0.138	-0.084
	P	0.580 NS	0.762 NS	0.678 NS	0.253 NS	0.024*	0.663 NS	0.162 NS	0.396 NS
Wind speed	r	0.126	-0.042	-0.180	-0.150	-0.226*	-0.148	-0.051	-0.117
	P	0.203 NS	0.675 NS	0.068 NS	0.129 NS	0.021 NS	0.134NS	0.607 NS	0.237 NS
insolation	r	0.515	0.706	0.617	0.532	0.459	0.967	0.920	0.036
	P	0.000***	0.000***	0.000***	0.000***	0.000***	0.000***	0.000***	0.714 NS

NS= no significant ; * =significant ; ** =highly significant ; *** = very highly significant

4. Discussion

The study of the numerical variations of *P. fusca* enabled us to identify in 2009 seven peaks which could be consider as seven different generations of this psyllid living on the young leaves of *M. excelsa* in the Yaounde region. In 2010, the number of peaks of numerical variation of individuals of various developmental stages recorded was six which could represent six different generations of this psyllid in our region. All peaks obtained in 2010 were lower than those observed in 2009; the most important peak was observed in June 2010. The number of generations of *P. fusca* varied from year to another, seven in 2009 and six in 2010. [22] reported that *P. lata* is multivoltine and ten generations or more can be recorded in a year; this author affirmed that the numbers of generation varied from one year to another. The same statement was made by various authors studying the population dynamic of psyllids in the Yaounde region: for citrus psyllid, *Trioza erytrae* [23]; for *Diclidophlebia eastopi* and *D. harrisoni*, psyllids of *Triplochiton scleroxylon* [24]; for *Blastopsylla occidentalis*, psyllid of *Eucalyptus* [25]; for *Pseudophacopteron* spp, psyllids of *Dacryodes edulis* [26]. The numbers of generations obtained during this study (7 and 6) is less that the number recorded by other (10 or more) for the sister species, *P. lata* [22].

This study showed that the damages on young buds were greater in November and December; during this period, the numbers of galls formed was very high. This period corresponds to the main flushing period and regeneration of leaves on *M. excelsa* plant in Yaounde region. The availability of young leaves is important for eggs laying sites for female. The higher number of galls on host plant leaves reflects the receptivity of the plant during this phase of plant growth towards the psyllids. These observations are consistent with this author [22], where he noticed a significant correlation between the numbers of galls formed by *P. lata* and the proportion of young leaves of *M. regia*. However, we have not observed particularly severe damage resulting to the death of the host plant. Host plant, of 3 to 5 years old, cannot succumb to the attacks of psyllid but it remains vulnerable and should be monitored during periods of renewed of leaves. This statement confirms previous observations on Citrus psyllid in Cameroon [23], in South Africa [27] and *Eucalyptus* psyllid [25] in Cameroon. Climatic factors such as temperature, rain fall, and insolation influenced notably the numerical variation of individual of various developmental stages of *P. fusca*. Spearman correlation showed us a significant correlation between some larval stages and temperature. Temperature affects negatively the abundance of *P. fusca* in our region. This result is the contrary of the findings of author who indicated that there was no significant difference

between the numerical variation of individuals of various developmental stages of *D. harrisoni* and *D. eastopi*, psyllids of *T. scleroxylon* and the temperature [24]. In contrast, this result confirmed the work of others who noted that all developmental stages of *B. occidentalis*, psyllid of *Eucalyptus* spp. are significantly correlated with temperature [25]. The phenology of the host plant could be the main factor influencing the population dynamic of *P. fusca* on *M. excelsa*. The Integrated Pest Management will take into consideration the main flushing periods of the plant and the psyllid outbreak periods in the Yaounde region. A good knowledge of the biology of *P. fusca* is important to ensure the effectiveness of chemical control and protection of seedlings and saplings. Similarly, the search for natural enemies of the psyllid is a major challenge for the limitation of natural populations of these pests.

5. Conclusion

P. fusca is a major pest of *M. excelsa* in Cameroon. Females of *P. fusca* laid their eggs on the buds, seedlings and young leaves. After hatching, first instar larvae dig a hole in the plant tissue and development inside the hole produced a closed gall. The larvae complete its life cycle inside the gall. Afterward, the gall burst to release adults. The numerical fluctuations of individuals of various developmental stages showed seven distinct generations of *P. fusca* in 2009 and six generations in 2010. Abiotic factors such as temperature, rain fall and insolation have an impact on numerical variation of the pest in the Yaounde region. The host plant phenology seem to be the main factor influencing the population dynamic of this psyllid on *M. excelsa*, The integrated pest management will take into consideration the main flushing periods of the plant and the psyllids outbreak periods in the Yaounde region for the control strategies.

6. Acknowledgements

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