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Population Dynamic of *Hilda Cameroonensis* Tamesse & Dongmo (Tettigometridae) Pest of *Vernonia amygdalina* Delile in Yaoundé-Cameroon

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ABSTRACT

Vernonia amygdalina Delile commonly called bitter leaf is a perennial shrub that belongs to the family of Asteraceae and grows throughout tropical Africa. It is probably the most used medicinal plant in the genus *Vernonia*. Insect pests caused serious damages on the leaves. In Cameroon, a new species of *Hilda* genus (Tettigometridae) was reported and described recently, *Hilda cameroonensis* Tamesse & Dongmo, for the first time on *V. amygdalina*. This insect caused leaves to shrivel. The population dynamic of that pest species was conducted in a natural farm, in the Yaounde Region, from November 2014 to October 2016. During the study, eggs, larvae, and adults were surveyed and counted once a week. Seven different generations of *H. cameroonensis* were recorded during the first years and six during the second years. The pest population was correlated with climatic parameters, mostly with relative humidity, temperature, wind speed, and rainfall. The main factors influencing the numerical fluctuation of the pest, and the outbreak periods of each of the developmental stages of the pest in the Yaounde region will be take into consideration by the integrated pest management.

Keywords: Tettigometridae, *Hilda cameroonensis*, *Vernonia amygdalina*, Pest control, Yaounde, Cameroon.

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INTRODUCTION

Planthoppers are herbivorous, sap-feeding homopterans that included several world-class agricultural pests. They constituted a cosmopolitan group of insects, feeding mainly on the phloem tissues of the woody or herbaceous plants, but sometimes on fungi, mosses, horsetails, or ferns [1]. They are highly polyphagous, feeding on numerous taxa of unrelated plants or sometimes strictly monophagous, feeding on only a single host plant species. Some studies have focused on the impacts of particular planthoppers on specific crop plants, especially rice, sugarcane, maize, oats, wheat, barley, and coconut palm [2]. Then, they are recorded as pests of economic plants.

Twenty families of planthoppers are recognized and collectively included in the superfamily of Fulgoroidea [2]. The Fulgoroidea are represented in all regions of the globe, from desert regions to Alaska beyond the Arctic Circle. However, they are found mainly in the tropics. This biogeographic diversity is very uneven across families, reflecting the historical constraints of each group. Thus, the Tettigometridae are absent from the Nearctic, Neotropical, and Australian regions. Tettigometridae is the most interesting and important family of the Fulgoroidea superfamily, although it is one of the smallest groups within this superfamily. The family of Tettigometridae is a pathogen vector for some plants familylike Fabaceae, Anacardiaceae, Rutaceae, Solanaceae, Poaceae, Annonaceae, and Asteraceae [2]. The plants of the Asteraceae family are known as preferential host plants of Tettigometrids of the genus *Hilda* [3, 4]. No record has been published concerning the impact of abiotic and biotic factors on the population dynamic of Tettigometrid species on *Vernonia amygdalina* (Asteraceae).

Vernonia amygdalina Del (Asteraceae) is a multipurpose and rapid regenerating soft wooded shrub of 2 to 10 m high with petiolate leaves of around 6 mm in diameter. Different ethnic groups around the world have called this plant by different names [5]. The plant belongs to the order Asterales, Asteraceae family, *Vernonia* genus, and the species *Vernonia amygdalina* [6]. *V. amygdalina* occurs in most countries of tropical Africa, South African, from Guinea East to Somalia and South to North Eastern, and Yemen. It is commonly grown as a vegetable in Benin, Nigeria, Gabon, RDC, and Cameroon [7]. In cultivation, *V. amygdalina* is mostly pruned to a shrub or hedge. It also can grow into a tree; but for commercial production, farmers prefer younger plants

The chemical composition of *V. amygdalina* makes it ideal for nutritional and medicinal uses [8]. The leaves are used for human consumption; these leaves are washed before cooking to get rid of the bitter taste [9]. There are some necessary active drug molecules and other substances, which are for maintaining good health and safe physiological functions of the body without any toxicity [10]. *V. amygdalina* has medicinal virtue according to Ebangue *et al.* [11]. It is probably the most used medicinal plant in the genus *Vernonia* [12]. Thomas *et al*. [13] showed that this plant has many therapeutic effects and can be considered as an antibacterial, antidiuretic, antipaludeen, antifungal, anticancer. Then, *V. amygdalina* can be classified as healthy food. The dried leaves of *Vernonia amygdalina* were also found to have insecticidal potency against the larvae of *Callosobruchus maculates* and *Sitophilus zeamais*; these insects caused heavy losses of stored cowpea and maize respectively [14]. These results seem to confirm that *V. amygdalina* could be considered an important plant in traditional medicine with subsequent pesticidal effects.

A few years ago, we collected for the first time in Cameroon a new species of Tettigometridae family described as *Hilda cameroonensis* Tamesse & Dongmo [4]. This new species was found feeding on plant tissue of *Vernonia amygdalina* and were responsible for their leaves shriveled [4]. There were no control measures for fighting against this pest of *V. amygdalina* in Cameroon. The insect pests' control is based on a thorough understanding of their population dynamic [15]. According to Milaire [16], studies on the population dynamic help to keep pest populations below the threshold of economic damage by insecticides. It allows finding strategies to optimize the use of chemicals in Integrated Pest Management [17] and provides useful information for taking preventive measures against the resurgence of infestations [18].

This work aimed to study the population dynamic of *Hilda cameroonensis* on its host plant *Vernonia amygdalina*. Abiotics and biotics factors that regulate the natural populations of the pests are investigated.

MATERIALS AND METHODS

Site and period of study

The study was conducted for two consecutive years from November 2014 to October 2016 in an experimental plantation in the locality of Nkolfoulou, Yaounde, Cameroon

Sampling

Host plants were planted by farmers. The total number of plants regularly surveyed was thirty (30). Ten plants were chosen randomly for Tettigometrids surveys and collection. These plants received no pesticide treatment before and during the study period. For the sampling, the visual inspection was adopted. Weekly inspections were out for 24 months, from November 2014 to October 2016. During each year, leaves of *Vernonia amygdalina*, the most commonly infested one, were examined. Tettigometrids adults, nymphs of stages III to V were counted directly. Five to ten infested leaves were collected and eggs and other nymphal stages were counted underhand magnifying lens. Meteorological data were obtained from the Meteorological Center of Yaounde.

Data analysis

SPSS statistical program was used to compare mean with non-parametric Mann Withney statistic tests. This program was also used to calculate the Spearman correlations between abiotic factors with the population dynamic of *Hilda cameroonensis*.

RESULTS AND DISCUSSION

The number of adults of *H. cameroonensis* counted on *V. amygdalina* was 826 adults with 260 males and 566 females. The sex ratio, in favor of females, was 2.17. The number of eggs recorded was 13186. The number of larvae of early-stage (larvae of 1st and 2nd stages) was 16367 and the number of larvae of advanced stages (larvae of 3rd, 4th, and 5th stages) was 3595.*Laying preference*

Adults of *H. cameroonensis* laid their eggs on leaves and stems of the host plant; more eggs were laid preferentially on stems and the Mann Whitney non-parametric U test shows a highly significant difference (P <0.0001) between the average number of eggs laid on stems and those laid on leaves.

Numerical variation of eggs

The number of eggs counted on each plant varied from one week to another during the experimental period. The numerical variation of the number of eggs of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, January 2015, February 2015, April 2015, June 2015, August 2015, and October 2015. During the second year of study, five peaks of outbreaks were recorded and the main peaks were obtained during April 2016, June 2016, July 2016, September 2016, and October 2016 **(Figure 1)**.

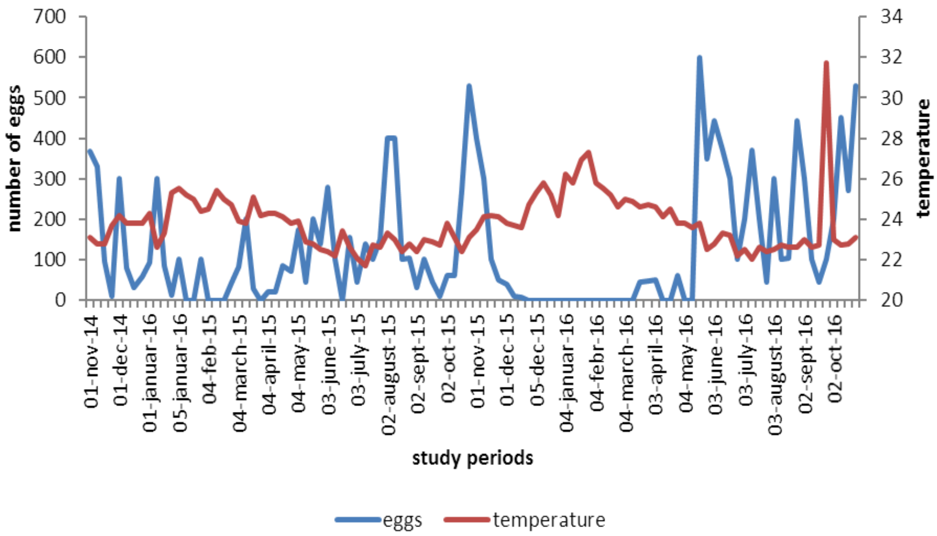

Figure 1. Numerical variation of eggs of *Hilda cameroonensis* depending on the temperature variation in a farmers plantation in Nkolfoulou (Yaoundé) from November 2014 to 0ctober 2016.

Numerical variation of larvae

Numerical variation of larvae of 1st stage

The number of 1st larval stage counted on each plant varied from one week to another during the experimental period. The numerical variation of the 1st larval stage of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, January 2015, April 2015, June 2015, August 2015, and October 2015. During the second year of study, four peaks of outbreaks of the pest were recorded and the main peaks were obtained during May 2016, July 2016, August 2016, and September 2016 **(Figure 2)**.

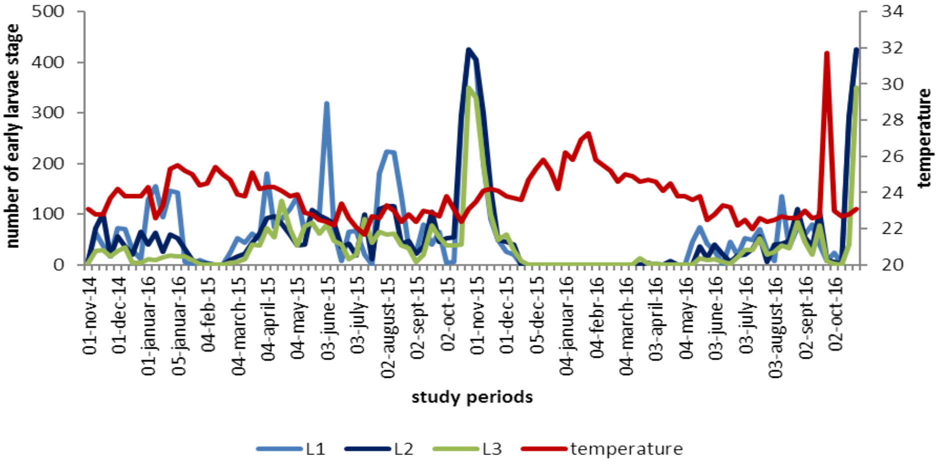

Figure 2. Numerical variation of stage I, II, III larvae of Hilda cameroonensis depending on the temperature variation in a farmers plantation in Nkolfoulou (Yaoundé) from November 2014 to 0ctober 2016.

Numerical variation of stage II larvae

The number of 2nd larval stage counted on each plant varied from one week to another during the experimental period. The numerical variation of stage II larvae of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, January 2015, February 2015, April 2015, June 2015, and October 2015. During the second year of study, four peaks of outbreaks were recorded and the main peaks were obtained during June 2016, August 2016, and October 2016 **(Figure 2)**.

Numerical variation of stage III larvae

The number of 3rd larval stage counted on each plant varied from one week to another during the experimental period. The numerical variation of stage III larvae of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, May 2015, June 2015, July 2015, September 2015, and October 2015. During the second year of study, three peaks of outbreaks were recorded and the main peaks were obtained during April 2016, June 2016, August 2016, and September 2016 **(Figure 2)**.

Numerical variation of stage IV larvae

The number of 4th larval stage counted on each plant varied from one week to another during the experimental period. The numerical variation of stage IV larvae of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, January 2015, April 2015, June 2015, August 2015, and October 2015. During the second year of study, four peaks of outbreaks were recorded and the main peaks were obtained during March 2016, June 2016, August 2016, and October 2016 **(Figure 3)**.

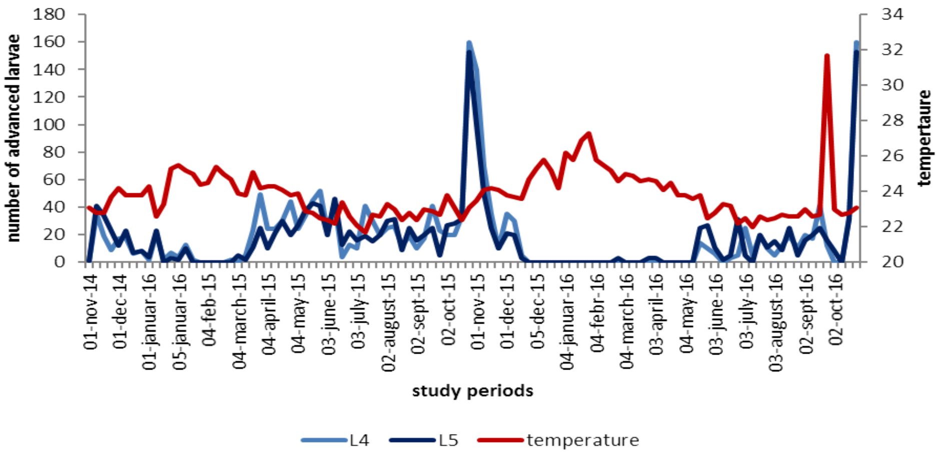

Figure 3. Numerical variation of stage IV and V larvae of Hilda cameroonensis depending on the temperature variation in a farmers plantation in Nkolfoulou (Yaoundé) from November 2014 to 0ctober 2016.

Numerical variation of stage V larvae

The number of 5th larval stage counted on each plant varied from one week to another during the experimental period. The numerical variation of stage V larvae of *H. cameroonensis* showed six peaks during the first year of study, during November 2014, January 2015, April 2015, June 2015, August 2015, and October 2015. During the second year of study, four peaks of outbreaks were recorded and the main peaks were obtained during March 2016, June 2016, August 2016, and October 2016 **(Figure 3)**.

Numerical variation of adults

Numerical variation of males

The number of males counted on each plant varied from one week to another during the experimental period. The numerical variation of males of *H. cameroonensis* showed seven peaks during the first year of study, during November 2014, January 2015, February 2015, March 2015, April 2015, August 2015, and October 2015. During the second year of study, six peaks of outbreaks were recorded and the main peaks were obtained during December 2015, May 2016, June 2016, July 2016, September 2016, and October 2016 **(Figure 4)**.

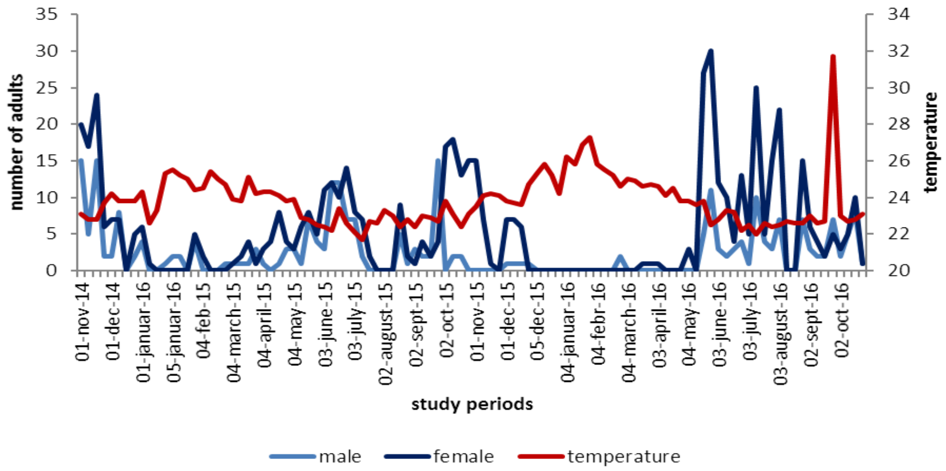

Figure 4. Numerical variation of adults (male and female) of *Hilda cameroonensis* depending on the temperature variation in a farmers plantation in Nkolfoulou (Yaoundé) from November 2014 to 0ctober 2016.

Numerical variation of females

The number of females counted on each plant varied from one week to another during the experimental period. The numerical variation of females of *H. cameroonensis* showed seven peaks during the first year of study, during November 2014, January 2015, February 2015, April 2015, June 2015, September 2015, and October 2015. During the second year of study, six peaks of outbreaks were recorded and the main peaks were obtained during December 2015, March 2016, June 2016, August 2016, September 2016, and October 2016 **(Figure 4)**.

The different peaks could express the number of generations of these Tettigometrids in our study region. The number of generations varied from one year to another during the two years of study. Then, seven generations during the first year and six during the second year of studies.

Numerical Variation of climatic factors

The ombro-thermic diagram **(Figure 5)** of the Yaoundé locality showed that there are two dry seasons and two rainy seasons in our study region. The major dry season starts from November until February and the small dry season from July to August. The major rainy season starts in September and ends in October and the small rainy season takes place from March to June. The comparison of the average of the main climatic parameters in our region gives no significant differences (P >0.05) from one year to another and the climate was relatively stable from November 2014 to October 2016 during our survey.

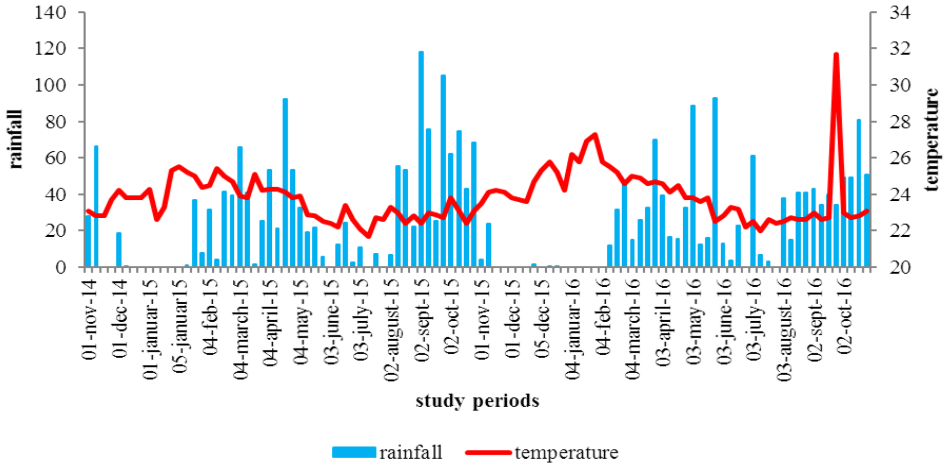

Figure 5. Rainfall and mean temperature of the study region from November 2014 to 0ctober 2016.

Effects of climatic factors on the numerical fluctuation of the population of H. cameroonensis

There is a positive and highly significant correlation between all developmental stages of larval (P≤0.0001) during the first year (October 2014-November 2015) and all climatic parameters **(Table 1).** During the second year (October 2015-November 2016), we recorded a positive and highly significant correlation between all developmental stages from eggs to adults **(Table 2)**.

Among the climatic factors, some were regularly recorded as important factors regulating the population dynamic of this pest among which are temperature and, relative humidity, wind speed, and rainfall.

Table 1. Spearman correlation between populations of *H. cameroonesis* sp.n. and some biotic parameters of the study site from 2014-2015

14-15	Eggs	L1	L2	L3	L4	L5	adults	Aver. T°	Rainfall	Winds	Rel.hum
Eggs	1.000										
L1	0,546**	1.000									
L2	0,476**	0,653**	1.000								
L3	0,293*	0,500**	0,749**	1.000							
L4	0,355*	0,437**	0,768**	0,851**	1.000						
L5	0,440**	0,378**	0,706**	0,675**	0,826**	1.000					
Adults	0,433**	0,028	0,226	0,292*	0,288*	0,433**	1.000				
Average T°	-0,710**	-0,247	-0,386**	-0,421**	-0,467**	-0,534**	-0,439**	1.000			
Rainfall	0,402**	-0,053	-0,028	0,148	0,168	0,151	0,173	-0,042	1.000		
Winds		-0,114	-0,106	0,037	-0,147	-0,196	-0,235	-0,041	0,308*	1.000	
Rel. hum	0,167	0,121	0,334*	0, 524**	0,494**	0,616**	0,543**	-0,710*	0,402**	0,167	1.000

*not significant; **significant; ***highly significant; L1, L2, L3, L4, L5: development stages

Effect of temperature

Data from the two years study showed that there is a highly significant negative correlation, between all developmental stages of *H. cameroonensis* (from eggs to adults) and temperature (P <0,05). This result showed that when the temperature is higher, the population of the pest decreased in our region **(Tables 1 and 2).**

Effect of relative humidity

During the two years of study, the data revealed that there was a positive correlation, between all developmental stages of *H. cameroonensis* and relative humidity. These correlations are highly significant for all developmental stages of the pest during the second year, while during the first year only larval of 3rd, 4th, 5th stages, and adults showed highly significant correlations. It should be noted, however, that egg and stage 1 larvae do not show a significant correlation with relative humidity **(Tables 1 and 2)**.

Effect of winds speed

During the second year of study, there was a negative highly significant correlation between wind speed and different developmental stages of *H. cameroonensis*; during this period, the increase of wind speed could reduce the number of the insect collected on host plants **(Tables 1 and 2)**.

Effect of rainfall

During the first year of study, the numerical variation of eggs showed a positive and highly significant correlation with rainfall (P <0, 05). During the second year of study, we recorded also a positive and highly significant correlation for the number of eggs counted, 1st instar larvae, and adults with the quantity of rainfall in our region **(Tables 1 and 2)**.

Table 2. Spearman correlation between populations of *H. cameroonensis* sp.n. and some biotic parameters of the study site from 2015-2016

15-16	Eggs	L1	L2	L3	L4	L5	adults	Aver.T°	Rainfall	Winds	Rel. hum
Eggs	1.000										
L1	0,752***	1.000									
L2	0,757***	0,868***	1.000								
L3	0,706***	0,852***	0,924***	1.000							
L4	0,616** *	0,847***	0,895***	0,917***	1.000						
L5	0,679***	0,809***	0,866***	0,866***	0,889***	1.000					
Adults	0,764***	0,554***	0,598** *	0,598***	0,568***	0,628***	1.000				
Aver.T°	-0,701** *	-0,650***	-0,625***	-0,619***	-0,558***	-0,568***	-0,675***	1.000			
Rainfall	0,421***	0,284* *	0,242*	0,182 *	0,153*	0,233*	0,286**	-0,354***	1.000		
Winds	-0,414***	-0,477***	-0,435***	-0,371***	-0,441***	-0,455***	-0,376***	0,300**	-0,184 *	1.000	
Rel.Hum.	0,661***	0,606***	0,522***	0, 472***	0,413***	0,453***	0,522***	-0,766***	0,631***	-0,234*	1.000

*not significant; **significant; ***highly significant; L1, L2, L3, L4, L5: development stages

The study of the population dynamic of *H. cameroonensis* on *V. amygdalina* was carried out over two years, from November 2014 to October 2016 in the Yaounde locality, center Region of Cameroon. *Hilda cameroonensis* Tamesse & Dongmo, the pest of Vernonia amygdalina (Asteraceae), was described as a new species, belonging to the family Tettigometridae [4]. Like other species of the family Tettigometridae, *H. cameroonensis* feed laid eggs and developed on its host plant *Vernonia amygdalina*. Indeed, the plants of the Asteraceae family are known as preferential host plants of Tettigometrids of the genus *Hilda*. Aléné *et al*. [3] reported the presence of *Hilda* spp on *V. amygdalina*. But the species collected by Aléné *et al.* [3] on *V. amygdalina* from the Yaounde region was wrongly identified as *H. patruelis*, the pest of groundnut in South Africa. In the Center Region of Cameroon, the species involved was described by Tamesse & Dongmo [4] as a new species, *H. cameroonensis.* All developmental stages from egg to adult were surveyed during the two years of study; *V. amygdalina* being the preferred host plant of *H. cameroonensis*, this plant would therefore constitute a source of food, shelter or protection, and compulsory support for mating and spawning [19, 20]. No significant difference was recorded between the numbers of eggs laid from one year to another; this result could reflect certain homogeneity of annual egg-laying by the female of *H. cameroonensis* on its host plant.

The study of the population dynamic of *H. cameroonensis* allowed us to record seven different generations of *H. cameroonensis* in the Yaoundé region during the first year of study (2014-2015) and six generations during the second year of study (2015-2016). The number of generations of this insect differed from one year to another. The same statement was made by Tamesse & Messi [21] when studying the factors influencing the population dynamic of the African citrus psyllid, *Trioza erytreae;* for this insect in the Yaounde region, seven generations were recorded during the first year and three generations during the second year of study. Similar observations were made by several authors working on the bionomics of insects in our region: Elisabeth *et al*. [22] studying *Phytolyma fusca*, psyllids of *Milicia excelsa* in Cameroon, Joly *et al*. [23] for *Diclidophlebia eastopi* and *D. harrisoni*, psyllids pest of *Triplochiton scleroxylon*, Soufo & Tamesse [24] for *Blastopsylla occidentalis*, psyllids pest of *Eucalyptus* spp. Thes population dynamic of this pest could be influenced by some biotics or abiotics factors that may directly or indirectly affect the biology of the insect. The number of generations thus defined reflects that *H. cameroonensis* could be polyvoltine, unlike other insect species developing on *V. amygdalina*, such as *Sphaerocoris annulus,* which is a monovoltine species according to Mbondji & Pluot-Sigwalt [25].

The comparison of each developmental stage of *H. cameroonensis* from one season to another season of the same year showed a significant difference, there was a highly significant difference during the second year of study. Alternation of seasons has an influence on the development of an individual and consequently on the population size [26]. This influence can be direct. This means that the climatic conditions prevailing during a given season would directly affect the development of the insect at all developmental stages. Thus, season influence also the phenology of the host plant; most of the plants renew their leaves during the rainy season and these new leaves are eggs laying preferential sites and are more suitable for nymphal development of *H. cameroonensis*. The same statement was made by Tamesse & Messi [21] when studying the impact of the phenology of *Citrus* spp on the population dynamics of *Trioza erytreae*, citrus psyllid. Similarly, Miller, *et al*. [27] and Cirino & Miller [28] showed that the phenology of *Opuntia mesacantha* (cactus) affected the size, sexual dimorphism, and reproduction of *Narnia femorata* (Coreidae). Thus, the small number of eggs observed during the higher dry and smaller dry season would be due to the impact of the plant phenology.

The correlation between weekly recorded temperatures and the numbers of individuals of different developmental stages of *H. cameroonensis* is negative and significant; this result explained that the development of *Hilda cameroonensis* could be influenced by the increase in temperature. Thus, during the major dry season, a period when the temperature is usually very high in our region, the number of insect pests counted was very low. From January to March of each year the population level of *H. cameroonensis* is low. The same statement was made by several authors working on a decrease in the survival during the eggs hatching to the larval stages, subjected to a constantly increasing temperature. This statement is supporting by the work of Tamesse & Messi [21] on citrus psylla, Nurhayati *&* Koesmaryono [29] on brown rice borers.

The correlation between relative humidity and the numbers of individuals of different developmental stages of *H. cameroonensis* are positive and significant; this result could explain that the population of the insect pest could increase when the relative humidity is higher. The same statement was made by several authors as Tamesse & Messi [21], Benhadi-Marin, *et al.* [30], when studying the effect of low relative humidity in the reduction of the developmental stages of citrus psylla, *Trioza erytreae* respectively in Cameroon and South Africa.

The rainfall showed positive correlations with the different developmental stages during two years of study, reflecting the importance of rainfall on the flushing rhythm of the host plant and consequently the better development of the population of *H. cameroonensis.* This statement is following the result obtained by several authors working on the bionomics of insect pests of plants in the tropic [22, 23].

Adults of *Hilda cameoonensis* are negatively correlated with weekly variations in wind speed.

This result reflects the possible negative impact of the increase of wind speed on the adult population of the insect. Indeed, when the number of adults decreased, the wind speed increased. According to Faivre d'Arcier *et al.* [31], populations of pear psyllids reach high densities in several regions when there was an inhibition of their flight as the wind speed was very high. The wind speed constituted, among other factors, the main passive transport of adults of *H. cameroonensis* from one plant *V. amygdalina* to another.

CONCLUSION

Hilda cameroonensis feed and developed on its hot plant, *V. amygdalina* where they achieved the entire life cycle. The number of adults of *H. cameroonensis* counted on *V. amygdalina* was 826 adults with 260 males and 566 females. The number of eggs recorded was 13186. The number of larvae of early-stage (larvae of 1st and 2nd st stages) was 16367 and the number of larvae of advanced stages (larvae of 3rd, 4th, and 5th stages) was 3595. The number of individuals of each developmental stage of *H cameroonensis* counted varied from one week to another; seven different generations of *H. cameroonensis* have been recorded during the first year of study (2014-2015) and six different generations during the second year (2015-2016). The number of generations of this insect varied from one year to another. Among the biotic and abiotic factors regulating the population dynamic of this pest, the result encountered showed that temperature, relative humidity, rainfall, and wind speed are the main factors regulating the numerical variation of this pest in our region. The integrated pest management will take into consideration these factors and the outbreak periods of *H. cameroonensis* in the Yaounde region should be considered for the control measures to be taken. The next challenge will be important to identify natural enemies of *H. cameroonensis* for biological control against this Tettigometrid pest.

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CONFLICT OF INTEREST: none

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ETHICS STATEMENT: This study was carried out following the rules established in research ethics.

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