Journal of
Entomology

ISSN 1812-5670

Academic
Journals Inc.

www.academicjournals.com

OPEN ACCESS Journal of Entomology

ISSN 1812-5670
DOI: 10.3923/je.2021.47.54

Research Article

Diversity of Mealybugs Vectors of *Cacao Swollen Shoot* in Nawa Region (Southwest, Cote d'Ivoire)

[1]N'guettia Akoua Miezan Claudine, [2]Oro Zokou Franck and [1]Ouali N'goran San-Whouly Mauricette

[1]Laboratory of Zoology and Animal Biology, UFR Biosciences, Felix Houphouët Boigny University, Centre of Excellence on Climate Change, Biodiversity and Sustainable Agriculture, 22 BP Abidjan 22, Cote d'Ivoire
[2]Department of Plant Biology, Training and Research Unit (UFR) of Biological Sciences, Pelefero Gon Coulibaly University of Korhogo, BP 1328 Korhogo, Cote d'Ivoire

Abstract

Background and Objective: The *Swollen shoot* virus is transmitted by several species of mealybugs. The objective of this work was to assess the status of mealybugs vectors of the *Swollen shoot* disease and specifically to determine the diversity of these mealybugs in the southwest of Cote d'Ivoire. **Materials and Methods:** The inventory was conducted at three sites Nawa area. The surveys were conducted on fixed plots of one hectare (1 ha) subdivided into 5 blocks (20×20 m). Each block consisted of 26 Cacao trees on which observations were made. Sampling consisted of checking in each block the presence or absence of mealybugs on the different organs of the Cacao tree at a height of 2 m. The mealybugs collected were brought to the laboratory for identification. **Results:** Eleven (11) species of mealybugs belonging to the family *Pseudococcidae* were inventoried. Among the eleven (11) species identified, *Pseudococcus viburni* and *Pseudococcus* sp. were the two species not yet recognized as *Swollen shoot* vectors in our study area. *Formicococcus njalensis*, the dominant species at the three sites, represented about 80% of the mealybugs population followed by *Planococcus citri* species (about 15%). The other species were in the minority with scale abundance ranging from 0-1%. Shannon's diversity index values varied by site H' = 0.22 for the Kipiri site, H' = 0.55 for the Petit Bondoukou site and H' = 0.77 for the Koda site. **Conclusion:** The study revealed several species of mealybugs of which *Formicococcus njalensis* and *Planococcus citri* were the most dominant.

Key words: *Swollen shoot*, south-west, mealybugs, canopy, biodiversity, *Pseudococcidae*, Shannon's diversity index

Citation: Claudine, N.A.M., O.Z. Franck and O.N.S.W. Mauricette, 2021. Diversity of mealybugs vectors of *Cacao Swollen Shoot* in nawa region (Southwest, cote d'ivoire). J. Entomol., 18: 47-54.

Corresponding Author: N'guettia Akoua Miezan Claudine, Laboratory of Zoology and Animal Biology, UFR Biosciences, Felix Houphouët Boigny University, Centre of Excellence on Climate Change, Biodiversity and Sustainable Agriculture, 22 BP Abidjan 22, Cote d'Ivoire

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Cacao, known as the food of the gods, is a plant of the Malvaceae family and the genus *Theobroma*. This crop originated in tropical America and was introduced to West Africa by colonists in the 20th century. Since then, West Africa has held the monopoly of world production with over 70% of the supply[1]. This Cacao production is now an important issue in the growth and economic development of several West African countries such as Côte d'Ivoire[2]. This Ivorian Cacao production contributes to more than 20% of the national GDP and represents nearly 70% of the country's export earnings[3].

However, Cacao cultivation in West Africa, mainly in Côte d'Ivoire, is faced with various sanitary problems. Indeed, the majority of Ivorian Cacao farms are more than 30 years old[4]. In addition to this constraint, there is the pressure exerted by insect pests and diseases, including *Swollen shoot* disease. This disease is viral in origin and the pathogen is *Cacao Swollen Shoot* Virus (CSSV). The virus is naturally transmitted in a semi-persistent manner by several species of mealybugs belonging to Hemiptera and the family of Pseudococcidae. CSSV is considered the most important viral disease limiting Cacao production in West Africa[5]. This disease causes defoliation of Cacao trees, destruction of the canopy, desiccation of branches and death of trees[6].

Although some work by Obodji *et al.*[7] and N'Guessan *et al.*[8] on CSSV vectors has been conducted in Côte d'Ivoire, the most intensive studies have been conducted in Ghana, Nigeria and Togo. In Côte d'Ivoire, there is a lack of data on monitoring the Cacao mealybug population. Moreover, a good control strategy against a disease vector starts with a good knowledge of these insects in their living environment. This work, therefore, aims to take stock of the mealybugs that are vectors of the *Swollen shoot* disease and specifically to determine the diversity of these mealybugs in the southwest of Côte d'Ivoire, the new Cacao production area of the country.

MATERIAL AND METHODS

Study area: The study was conducted in the Department of Soubré, the capital of the Nawa area in south-western Côte d'Ivoire. This area was chosen because it is the main production area currently threatened by *Swollen shoot* disease. The climate is equatorial, characterized by high rainfall, high atmospheric humidity, two rainy seasons and two dry seasons that alternate. The vegetation is of dense, tropical and humid forest type. The soils encountered are of the ferralitic type.

Experimental setup: The study was conducted from January, 2018-December, 2019 in peasant plantations aged 15-50 years. These plantations or sites are located in three localities of Soubré. These are Koda (6°38'W and 5°56N), Kipiri (6°29'W and 6°8'N) and Petit Bondoukou (6°38'W and 5°56N). At each site, three plots were selected. The size of the different plots selected varied from 3-4 ha. However, to facilitate the implementation of the system, an area of 1 ha was delimited in the centre of each selected plot. That hectare plot was divided into 5 blocks of 20×20 m (Fig. 1). Within each block, 26 Cacao trees were randomly selected and numbered with red paint.

Inventory and collection of mealybugs: For the mealybug inventory, observations were made on all the Cacao trees numbered in the blocks. It consisted of checking the Cacao trees in each block to count the mealybugs present on the different parts such as the pods, cherelles, flowers and the trunk at a height of 2 m. The scale insects are counted in situ up to 100 individuals. Beyond this number, the mealybugs were collected with flexible forceps and put in pillboxes containing 70 alcohol. The name of the site, the plot, the block, the number of the tree and the date of observation were written on each pillbox. The samples were brought to the laboratory for enumeration. The sum of the mealybugs per organ was used to find the total number of mealybugs per tree. Infested organs were marked with ribbons to track the population over time and space. Observations were made every two weeks.

Identification of mealybugs: The mealybugs collected from Cacao trees were identified in the laboratory using

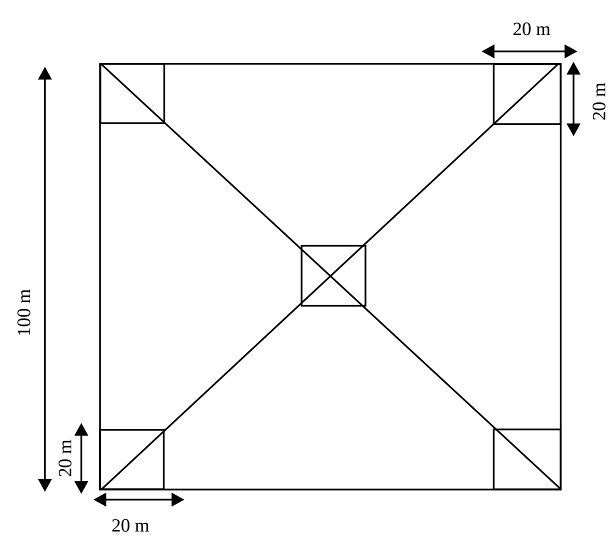

Fig. 1: Experimental setup

identification[9-11] and other collectors' works. Species identification was confirmed by entomologists at the National Center for Agronomic Research (CNRA) in Divo.

Data processing: The data collected were processed by ecological indices such as species richness (S), Relative Abundance (RA), Frequency of Occurrence (Fo), Shannon Weaver diversity index (H'), Equitability (E) and similarity of scale species according to different experimental sites:

- Species richness (S) is one of the fundamental parameters characteristic of the biodiversity of a stand[12]. It represents the total number of species in the stand considered in a given ecosystem:

$$S = \sum \text{species with } S\text{: specific richness}$$

- Relative Abundance (RA) is a concept that allows us to evaluate a species, a category, a class or an order ($\eta\iota$) to all the animal populations of all species combined (N) present in a faunal inventory[13]:

$$RA\ (\%) = \frac{\eta\iota * 100}{N}$$

where, RA (%) is the relative abundance of scale insects, $\eta\iota$ is the number of individuals of species i considered and N is the total number of individuals of all species.

- Frequency of occurrence of a species is the ratio, expressed as a percentage, of the number of samples where this species is noted to the total number of samples taken[14]:

$$Fo\ (\%) = \frac{N * 100}{P}$$

where, Fo is the frequency of occurrence of the species, N is the total number of samples containing the species under consideration and P is the total number of samples taken. According to Dajoz[14] we distinguish rare species if Fo<5%, incidental species ($F \leq 25\%$), incidental species (25%<F<50%), constant species ($F \geq 50\%$), regular species if $50\% \leq Fo < 75\%$ and omnipresent species if Fo = 100%.

- Shannon index (H'): It is necessary to combine a relative abundance of species and total richness to obtain a mathematical expression of the general index of the diversity of Shannon-Weaver (H'). It is calculated by the following Eq:

$$H' = -\sum \varphi\iota \ \ln \varphi\iota$$

$$\varphi\iota = \frac{\eta\iota}{N}$$

where, H' is the Shannon index and i is the one specie.

- Equitability (E) is the ratio between the actual diversity of the community and its theoretical maximum diversity[12]. Equitability varies between 0 and 1. It tends towards 0 when almost all the numbers correspond to a single species of the stand and tend towards 1 when each species is represented by a similar number of individualy[12]:

$$E = \frac{H'}{\text{In } S}$$

where, E is the equitability, S is the specific wealth and H' is the Shannon index.

- Sorensen's Similarity Index (β) which is a measure of beta diversity ranging from (0 = no similarity) to (1 = perfect similarity)[15] was used to compare the biodiversity of mealybugs at the different sites in this study. It was calculated according to the Eq:

$$\beta = \frac{2C}{S_1 + S_2}$$

where, β is the sorensen's index of Similarity, C is the number of species common to both localities, S_1 is the specific richness of site a and S_2 is the specific richness of site b.

RESULTS

Species richness: A total of 11 species of mealybugs belonging to the family *Pseudococcidae* were inventoried on the three study sites. They are *Formicococcus njalensis*, *Planococcus citri*, *Ferrisia virgata*, *Pseudococcus longispinus*, *Dysmicoccus brevipes*, *Phenacoccus hargreavesi*, *Planococcus kenyae*, *Maconellicoccus hirsutus*, *Pseudococcus* sp., *Pseudococcus viburni*, *Pseudococcus*

Fig. 2(a-i): Mealybug species inventoried on Cacao

(a) *Formicococcuss njalensis*, (b) *Planococcus citri*, (c) *Pseudococcus viburni*, (d) *Pseudococcus* sp., (e) *Pseudococcus jackbeardsleyi*, (f) *Planococcus kenyae*, (g) *Ferrisia virgata*, (h) *Pseudococcus longispinus* and (i) *Maconellicoccus hirsutus*

jackbeardsleyi Fig. 2(a-i) of the 11 species recorded, 9 species were commonly encountered at the three study sites during the two years of observations. At each site*, F. njalensis* and *P. citri* were the most commonly encountered species in the orchards.

Relative abundance of species by site and observation year: Species abundance varied on the three sites and during the two years of observation. Of all the scale insects surveyed, *F. njalensis* and *P. citri* were the most abundant at all three sites and in both years of observation.

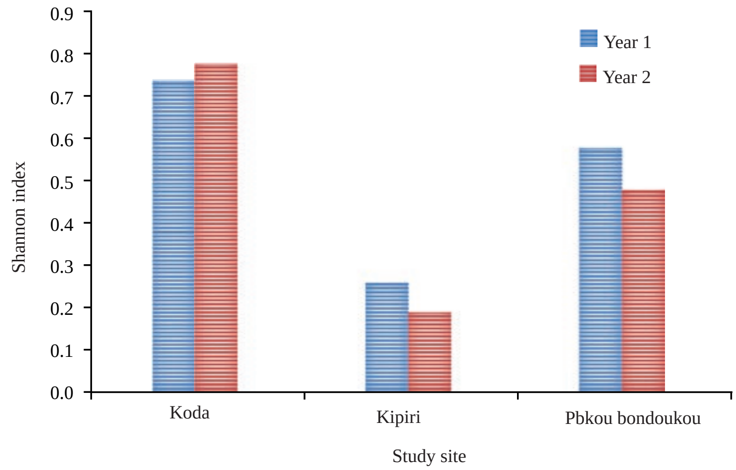

Fig. 3: Shannon diversity index by site and year of observation
Year 1: January-December, 2018, Year 2: January-December, 2019

Table 1: Relative abundance of species according to sites and years of observation

	Year 1 (January-December, 2018)			Year 2 (January-December, 2019)		
Species	Koda	Kipiri	Pbkou	Koda	Kipiri	Pbkou
Formicococcus njalensis	72.73	94.87	82.44	69.06	85.32	86.37
Planococcus citri	23.57	4.44	15.15	28.13	2.37	11.56
Ferrisia virgata	1.62	0.29	0.77	0.77	0.44	1.12
Pseudococcus longispinus	1.07	0.19	0.98	0.99	11.35	0.24
Dysmicoccus brevipes	0.0	0.02	0.13	0.0	0.0	0.0
Phenacoccus hargreavesi	0.013	0.015	0.11	0.099	0.04	0.03
Planococcus kenya	0.17	0.007	0.12	0.19	0.26	0.09
Maconellicoccus hirsutus	0.59	0.04	0.12	0.63	0.04	0.27
Pseudococcus sp.	0.0	0.0	0.0	0.0	0.0	0.27
Pseudococcus viburni	0.06	0.01	0.02	0.07	0.03	0.0
Pseudococcus jackbeardsleyi	0.13	0.08	0.11	0.03	0.12	0.0

Pbkou: Petit bondoukou

During the first year of observation (January-December, 2018), the species *F. njalensis* was dominant at the three sites with a proportion of 72.73% of the mealybugs population at Koda, 94.87 at Kipiri and 82.44% at Petit Bondoukou. The abundance of *Planococcus citri* was 23.57% in Koda, 15.15% in Petit Bondoukou and 4.44% in Kipiri. In the second year of observation (January-December 2019), the species *F. njalensis* represented 69.06% of the scale population in Koda, 85.32% in Kipiri and 86.37% in Petit Bondoukou. The abundance of *Planococcus citri was* 28.13, 2.37 and 11.56% in Koda, Kipiri and Petit Bondoukou, respectively (Table 1).

The other species were in the minority during the two years of observation at the three study sites, with mealybug abundance ranging from 0-1%. In addition to the scale species already known on Cacao, two species were identified. These are *Pseudococcus viburni* and *Pseudococcus* sp.

Mealybugs diversity by site and frequency of occurrence: Analysis of the Shannon-Weaver diversity index shows a variation in diversity from one site to another and as a function of time. The Koda index was the highest followed by the Petit Bondoukou (Pbkou) site, the lowest index was noted at Kipiri (Fig. 3).

The frequency of occurrence applied to the collected species was used to identify mealybug species categories. Thus, out of eleven mealybug species collected, we have 2 rare species, 2 accidental species, 2 accessory species, 2 species are classified as constant, 1 regular species and 2 omnipresent species (Table 2).

Equitability and Similarity of mealybugs across sites: The equitability index values obtained at the three study sites ranged from 0.09-0.30. Equitability was the lowest at Kipiri with an index of 0.09. In Koda and Petit Bondoukou, the values of the equitability index were 0.33 and 0.22, respectively

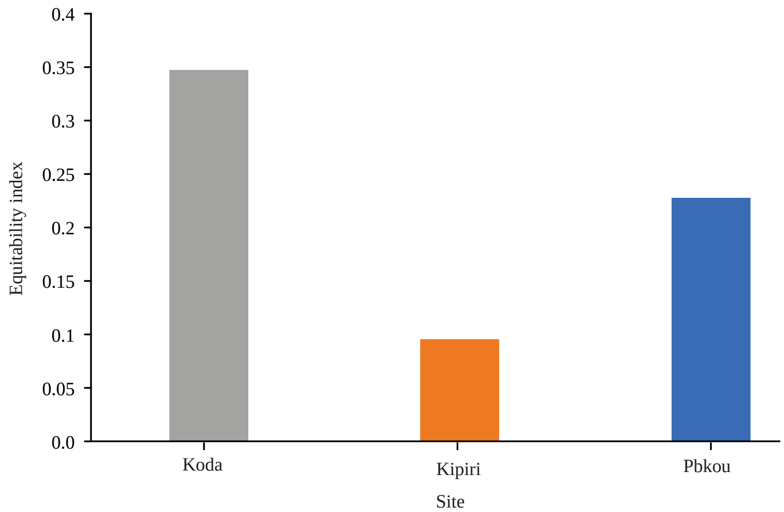

Fig. 4: Equitability of mealybugs across sites

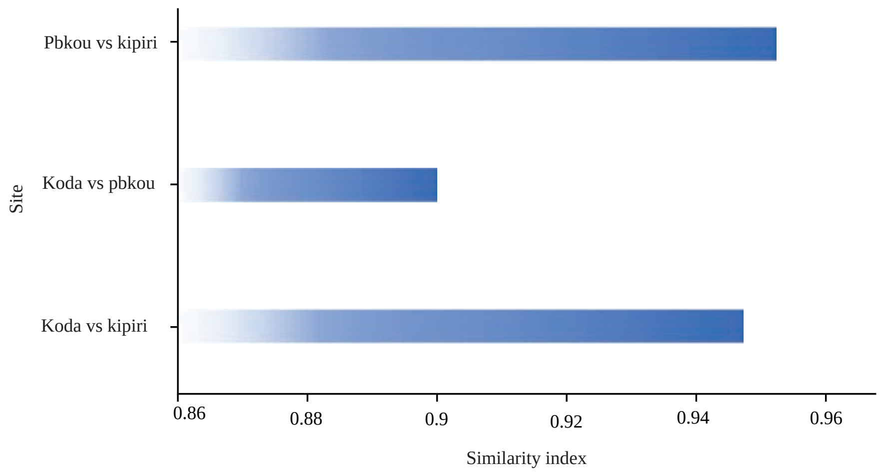

Fig. 5: Similarity of mealybugs across sites

Table 2: Frequency of occurrence of different scale species

Species	Pi	Fo (%)	Categories
Dysmicoccus brevipes	2	4	Rare
Ferrisia virgata	39	81	Constant
Maconellicoccus hirsutus	15	31	Accessory
Planococcus citri	48	100	Omnipresent
Phenacoccus hargreavesi	26	54	Regular
Pseudococcus jackbeardsleyi	10	21	Accidental
Planococcus kenyae	18	38	Accessory
Pseudococcus longispinus	41	85	Constant
Formicococcus njalensis	48	100	Omnipresent
Pseudococcus sp.	1	2	Rare
Pseudococcus viburni	7	15	Accidental

Fo: Frequency of occurrence of the species and Pi: Total number of samples taken

(Fig. 4). In terms of similarity, the mealybugs species found at the different study sites were very similar, with values tending towards 1. The sites of Koda/Kipiri and Petit Bondoukou/Kipiri have a similarity of 0.94 and 0.95, respectively, while that of Koda/Petit Bondoukou is 0.9 (Fig. 5).

DISCUSSION

The study inventoried eleven (11) species of mealybugs belonging to the family *Pseudococcidae* on the three experimental sites. These results are close to those of

Obodji *et al.*[7] and by N'Guessan *et al.*[8] who, respectively had identified 8 and 9 species of mealybugs in Cacao orchards in Côte d'Ivoire. The other nine (09) species already identified are recognized as vectors of *Swollen shoot* disease. The present study has therefore made it possible to update the list of mealybugs species infested with the Cacao crop in Côte d'Ivoire. In addition to the species of mealybugs previously recorded on Cacao in Côte d'Ivoire, two new species were identified. These are *Pseudococcus viburni* and *Pseudococcus* sp., which despite their low recorded numbers deserve special attention because their implications in the transmission or not of the *Swollen shoot* disease remain to be determined.

In terms of abundance, *Formicococcus njalensis* and *Planococcus citri* were the most abundant species at all three sites and in both years of observation. *Formicococcus njalensis* was the dominant species at the three sites with a proportion of about 80% of the scale population followed by *Planococcus citri* with about 15% of the population. Franco *et al.*[16] showed that *F. njalensis* accounted for more than 80% of the mealybug population recorded during his study in Togo. These results go along with those of Obodji *et al.*[7] and N'Guessan *et al.*[8] who showed that *F. njalensis* species was the most abundant, followed by *P. citri* species. The abundance of the *F. njalensis* population compared to the other species can be explained by the favourable conditions for its development. Indeed, according to Franco *et al.*[16], *F. njalensis* develops in abundance in well-covered Cacao fields. A dense shade formed by the forest cover or the shade of the Cacao canopy favours its pullulation. When this shading decreases or disappears, the numbers of *F. njalensis* also decrease in contrast to the *P. citri species* whose population-level increases to be dominant in the plot. In addition, the age of the orchards could be a factor in the distribution of *F. njalensis* species as reported by N'Guessan *et al.*[8]. Indeed, *F. njalensis* is rampant in mature Cacao farms and its full activity is limited by the dry season.

Other species were in the minority during the two years of observation at the three study sites with scale abundance ranging from 0-1%. This is probably due to the ecology of each species. Indeed, some species of scale insects such as *F. njalensis* and *P. citri* are strongly associated with ants, which provide them with assistance and protection. Under these conditions, ant-assisted species are favoured and will increase at the expense of unassisted species[17]. Campos *et al.*[18] compared free-living insects with those associated with ants and observed that the proportions were higher in scale insects receiving protection from ants as opposed to other species without protection.

The diversity of mealybugs at the three sites was determined by calculating the Shannon diversity index. According to Net *et al.*[19] a community will be more diverse the higher the H' index. In general, the values of the Shannon diversity index varied from one site to another and as a function of time. This could be explained by the difference in temperature and other environmental factors. Both abiotic and biotic factors remain important in the variation of mealybugs abundance.

Equitability was the lowest one in Kipiri with an index of 0.09. In Koda and Petit Bondoukou, the values of the equitability index were 0.33 and 0.22, respectively. These values reflect an unequal distribution of individuals of the species on the different study sites. This confirms an imbalance in numbers among the species present at the study sites. The mealybug species found on the different study sites show very high similarities. These great similarities recorded show that from one site to another the mealybugs species encountered are approximately the same in the study area.

CONCLUSION

The study aimed to assess the status of mealybugs vectors of the *Swollen shoot* disease and to make an analysis of the diversity in the southwest of Cote d'Ivoire, the new Cacao production area. Eleven species of mealybugs were identified. The species *Ferrisia virgata*, *Pseudococcus longispinus* were constant, whereas *Planococcus citri* and *Formicococcus njalensis* were omnipresent. *Pseudococcus viburni* and *Pseudococcus* sp. are reported for the first time on Cacao in Côte d'Ivoire. Future specific studies on these species in the transmission of *Swollen shoot* disease seem necessary. There is a dominant relationship between the species that could be explained by the favourable conditions for their development from one site to another.

SIGNIFICANCE STATEMENT

This study discovers two new species which has allowed us to update the exciting data on mealybugs of Cacao. Thus, this discovery will allow reorienting the knowledge on the transmission of the *Swollen shoot* disease.

ACKNOWLEDGMENT

The authors would like to thank the African Centre of Excellence on Climate Change, Biodiversity and Sustainable Agriculture (CEA-CCBAD) and Bayer Crop Science for funding this work (grant number {2400000 FCFA}). We would also like to thank the World Agroforestry Centre (ICRAF) for providing the study framework and Cacao farmers for their availability during this study. Finally, we would like to thank the technicians for their support in the execution of the various field works.

REFERENCES

1. Wessel, M. and P.M.F. Quist-Wessel, 2015. Cocoa production in West Africa, a review and analysis of recent developments. NJAS Wageningen J. Life Sci., 74-75: 1-7.
2. Duguma, B., J. Gockowski and J. Bakala, 2001. Smallholder cacao (*Theobroma cacao* Linn.) cultivation in agroforestry systems of West and Central Africa: Challenges and opportunities. Agrofor. Syst., 51: 177-188.
3. Coulibaly, S.K. and C. Erbao, 2019. An empirical analysis of the determinants of cocoa production in Cote d'Ivoire. Econ. Struct., 10.1186/s40008-019-0135-5.
4. Assiri A.A., G.R. Yoro, O. Deheuvels, B.I. Kebe, Z.J. Keli, A. Adiko and A. Assa, 2009. The agronomic characteristics of the cacao (*Theobroma cocoa* L.) orchards in Côte d'Ivoire. Elewa Biosci., 2: 55-66.
5. Muller, E., 2016. *Cacao Swollen Shoot* Virus (CSSV): History, Biology, and Genome. In: Cacao Diseases, Bailey, B. and L. Meinhardt (Eds.)., Springer International Publishing, New York, ISBN-13: 978-3-319-24787-8, pp: 337-358.
6. Oro, F.Z. H.D. Lallie, K.G. Brou, J.K. Koigny and H.A. Diallo, 2020. Impact of plot maintenance and level of Cacao tree leaf cover on spread of *Swollen shoot* disease in Côte d'Ivoire: Case of Petit-Bondoukou site. Int. J. Environ. Agric. Biotechnol., 5: 517-524.
7. Obodji, A., W.P. N`guessan, K.F. N`guessan, P. Seri-Badama, L.R.N. Aboua, I. Kébé and R. Aka, 2015. Inventory of the mealybug species associated to the cocoa tree (*Theobroma cacao* L.) in four producing areas infected with the swolen shoot disease in côte d'ivoire. J. Entomol. Zool. Stud., 3: 312-316.
8. N`Guessan, P.W., A. Yapi, F.K. N`Guessan, N.N. Kouamé and C.N. Gouamené *et al*., 2019. Inventory and abundance of mealybug species in immature and mature cocoa farms in Côte d`ivoire. J. Appl. Entomol., 143: 1065-1071.
9. S. Abd-Rabou, H. Shalaby, J.F. Germain, N. Ris, P. Kreiter and T. Malausa, 2012. Identification of mealybug pest species (Hemiptera: Pseudococcidae) in Egypt and France, using a DNA barcoding approach. Bull. Entomol. Res. 102: 515-523.
10. Miller, D.R., A. Rung and G. Parikh, 2014. Scale Insects, edition 2, a tool for the identification of potential pest scales at USA ports-of-entry (Hemiptera, Sternorrhyncha, Coccoidea). zookey, 431: 61-78.
11. Correa, M.C.G., J.F. Germain, T. Malausa and T. Zaviezo, 2012. Molecular and morphological characterization of mealybugs (Hemiptera: Pseudococcidae) from Chilean vineyards. Bull. Entomol. Res. 102: 524-530.
12. Das, p., S. Kar, U. Das, M. Bimola, D. Kar and G. Aditya, 2020. Day time variations of zooplankton species composition: Observations from the wetlands of Assam, India. Acta Limnologica Brasiliensia, 10.1590/S2179-975X1418.
13. Faurie, C., C. Ferra, P. Médori, J. Dévaux and J.L. Hemptinne, 2011. Ecology-Scientific and practical approach. 6th Edn., Technique and Doc, Pages: 488.
14. Travlos, I.S., N. Cheimona, I. Roussis and D.J. Bilalis, 2018. Weed-species abundance and diversity indices in relation to tillage systems and fertilization. Front. Environ. Sci., 10.3389/fenvs.2018.00011.
15. Christopher, A.O., 2020. Comparative analyses of diversity and similarity indices of West bank forest and block A forest of the international institute of tropical agriculture (IITA) Ibadan, Oyo State, Nigeria. Int. J. For. Res., 2020: 1-8.
16. Franco, J.C., A. Zada and Z. Mendel, 2009. Novel Approaches for the Management of Mealybug Pests. In: Biorational Control of Arthropod Pests, Ishaaya I. and A. Horowitz (Eds.)., Springer, Netherlands, pp: 233-278.
17. Ackonor, J.B., 2002. Current levels of incidence of parasitism and predation in *Planococcus citri* risso (Homoptera: Pseudococcidae) in ghanaian cocoa (*Theobroma cacao* L.) farms. Int. J. Trop. Insect Sci., 22: 105-112.
18. Campos, R.I. and G.P. Camacho, 2014. Ant-plant interactions: The importance of extrafloral nectaries versus hemipteran honeydew on plant defense against herbivores. Arthropod-Plant Interact., 8: 507-512.
19. Prudence, D.J., Z.T.S. Hubert, T.K.R. Polycarpe, D.H. Charly, N.T.J. Guy, F.M. Samuel and N. Thomas, 2015. Physico-chemistry characterization and zooplankton specific diversity of two fishponds in Yaoundé (Cameroon, Central Africa). J. Biodivers. Environ. Sci., 6: 16-30.