RESEARCH ARTICLE

INSECT DIVERSITY OF COCOA (THEOBROMA CACAO L.) BEANS STOCKS FROM HAUT-SASSANDRA REGION AND THEIR IMPACTS (DALOA, CÔTE D’IVOIRE).

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Manuscript Info

Abstract

Cocoa storage is a potential opportunity for insect infestation. The objective of this study was to identify insects and assess their damage to stored cocoa beans from the Haut-Sassandra region. To do this, 3 samples of 3 Kg of cocoa beans were collected per locality. These samples were stored for 30 days in jute bags. After this month of storage, each sample was sieved every 15 days in a muslin observation cage for 120 days. The living or dead adults collected in each lot were observed with a binocular magnifying glass, counted and identified using manuals. The livings are returned to the batches from which they were removed. For the estimation of the damage, three samples of 1 kg cocoa beans without holes were made and followed for 120 days. Counting after sieving revealed the presence of 876 individuals divided into 2 orders, 10 families and 12 species. The most abundant species was Ahasverus advena (24.89%) followed by Tribolium castaneum with 18.38% presence. The estimated damage caused by insects to cocoa beans was 26.88% at the 120th day of storage.

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Introduction:

The cocoa tree (Theobroma cacao L.), is a perennial plant grown for the beans of its fruits. Its fruit beans are used in human nutrition, animal nutrition, drug formulation and cosmetics (Barrel et al., 2006). It is a major economic resource for many countries. World production in growing areas, such as Latin America, South Asia and Africa, is estimated at 4.06 million tonnes or the equivalent of US$3 billion (Lass, 2006). Since its independence, Côte d’Ivoire has made agriculture, particularly the coffee and cocoa pair, the mainstay of its economy (Sangaré et al., 2009). Cocoa, the main cash crop, contributes to the country’s development, accounting for 20% of the Gross Domestic Product (GDP). About 4 to 5 million people live from this production. Most of the cocoa is exported in beans (70%) and semi-finished products (30%) (ICCO, 2018). Cocoa production is mainly ensured by the Haut-Sassandra region (30.6%), Bas-Sassandra (13.8%), Moyen Comoé (7.66%) and Moyen Cavally (7.15%). Eight other regions in the south of the country whose production varies from 3 to 7%, contribute to fill the rest of the national production (Sangaré et al., 2009). Ivorian production represents 44% of the world harvest with an annual

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average of 1,700,000 tonnes (Kébé et al., 2006; ICCO, 2018). Despite this remarkable performance, the sustainability of cocoa farming in Côte d'Ivoire is compromised by several biotic constraints, including diseases, pest, rodent and parasitic plant attacks (Kébé et al., 2006; Kouamé et al., 2016). Concerning diseases, Phytophthora and Loranthus attacks lead to significant production losses. Virus disease such as swollen shoot destroys the cocoa tree after 3 to 5 years (Chingandu et al., 2017). Stored cocoa beans are often subject to losses of a variable and complex nature, most of which are attributed to insects, micro-organisms and rodents (Kekeunou et al., 2006; Tah et al., 2016). Indeed, cocoa can be infested initially in the field, during post-harvest treatments and storage by several insect species and other pests. If these infestations are not initially treated with effective fumigation before shipment (Doumbia and Kouassi, 2009), these species will survive during transport to traders, processors and manufacturers. Thus, it is therefore necessary to know the entomofauna of cocoa bean stocks in the Haut-Sassandra region. This knowledge will allow effective pest control. This study is part of the improvement of cocoa storage conditions. The objective of this work is to update the list of insects associated with cocoa bean stocks and to assess their damage to the beans during storage.

**Study site**
The cocoa beans samples from the 2017-2018 seasons were collected from farmers in 6 localities (Bazra, Bédiala, Boguedia, Gadouan, Zaïbo and Zoukougbeu) in the Haut-Sassandra region, Center West of Côte d'Ivoire (Fig 1). These localities were chosen for their high production. The samples were sent to Jean Lorougnon Guédé University in Daloa for study. The city of Daloa is located at 6°53 North latitude and 6°27 West longitude from Côte d'Ivoire. During the study period, the temperature of the storage room varied between 28.4 and 30.5°C and the relative humidity between 67.53 and 82.42%.

**Fig 1:** Presentation of the localities of the collected samples

**Materials and Methods:**

**Inventory and insect dynamics of cocoa bean stocks**

Inventory of stock insects were carried out on the lots of field cocoa beans which had not undergone any chemical treatment. These beans were collected from 6 localities in the Haut- Sassandra region. In each locality, a 2 kg lot was taken from the stocks of cocoa of 5 producers. The beans were removed at different locations from the cocoa bags (two ends and in the center) according to regulation N° 401/2006 (CE) (Commission of the European Communities, 2006). These 5 elementary samples were grouped together to form an overall sample of 10 kg (Regulation No. 401/2006). From this overall homogenized sample, 3 samples of 3 kg of cocoa beans were made per locality. These batches were stored for one month to allow the hatching of any insect eggs. After one month of storage, each sample was sieved every 15 days in a muslin observation cage for 120 days. Live or dead adults insects harvested in each lot were observed under binocular magnifying glass and counted. The living are returned to the lots from which they
were removed. Counting is done directly by species after sorting. The number of adults per species has been noted. Species identification was done using identification key (Weidner and Rack, 1984; Delobel and Tran, 1993).

Evaluation of damage during storage time
The estimate of damage to cocoa beans is done by evaluating the proportion of beans carrying pest attack holes in a sample. It is made according to the counting and weighing method (MCP). Thus, three samples of 1 kg of cocoa beans not bearing holes were made. The number of beans corresponding to 1 kg has been accounted. Each month, after the sieving session, each sample was divided into two fractions: the first consisting of perforated beans and the second consisting of beans without holes. The damage of each fraction was estimated as a percentage after counting. The evaluation was done monthly, for four months. The estimate of damages was done by the following formula (Boxall, 1998):

\[
\text{Damage} (\%) = \frac{B}{A} \times 100
\]

With: A : total number of cocoa beans; B : number of cocoa beans with holes

Estimate the relative weight loss of cocoa beans
Relative loss is the degree of transformation of a bean following a pest's infestation by comparing it to an intact bean. It is evaluated every month for four months. The Counting and Weighing (MCP) method is used to determine the relative weight loss of cocoa beans. It ignores the determination of the beans moisture content; then it compares the standard internal value of the intact cocoa beans with that of the damaged cocoa beans. The sample removed from the stock after sieving is divided into two batches: on the one hand the intact cocoa beans and on the other hand the infested cocoa beans. The beans of each batch are weighed and counted. The relative weight loss is then evaluated according to the formula of Adams and Schulten (1978):

\[
P(\%) = \frac{(ExB) - (CxD)}{ExA} \times 100
\]

A : total number of cocoa beans; B : number of damaged beans
C : number of intact beans; D : weight of the damaged beans
E : weight of intact beans; P : relative weight loss
After each survey, the counted cocoa bean samples are returned to the bags of the corresponding batches.

Data analysis:-
Data processing was performed using the software Statistica 7.1 version. The variance analysis followed by the Newman-Keuls test at the 5% threshold allowed to compare the average number of insects of the cocoa beans of each batch, the number of cocoa bean bearing or no holes, the damage and the relative weight loss of cocoa beans. The XLSTAT 7.5 software was used to perform factorial correspondence analysis (AFC) between insect species and localities in the region to highlight the characteristic species of the region. The relative abundance of the species was determined by the formula of Zame and Gautier (1989):

\[
Ar (\%) = \frac{Ni}{N} \times 100
\]

Ni: number of species’ individuals considered; N: total number of all species’ individuals combined; Ar (%): relative abundance.

Results:-
Insects identified in the six localities
The identification of the collected specimens revealed on all 6 localities two orders (Coleoptera and Lepidoptera) with 10 families and 12 species. Of these 12 species, only one *Ephesia cautella* belonging to the order Lepidoptera and the family Pyralidae was collected.
Boguedia’s locality

On the 30th day of storage, the insects collected are composed of 5 species: Ahasverus advena (Silvanidae), Ephestia cautella (Pyralidae), Tribolium castaenum (Tenebrionidae), Rhizopertha dominica (Bostrichidae) and Lasioderma serricorne (Anobiidae). On the 45th day of monitoring, two species Korynetes sp (Cleridae) and Oryzaephilus surinamensis (Silvanidae) appear in the stocks with average numbers of 0.66±0.57 individuals. The species A. advena, L. serricorne, Korynetes sp, O. surinamensis and R. dominica disappeared from the stocks as from the 90th day. As for the species T. castaenum, it has been present throughout the duration of the inventory control. E. cautella was the most abundant species with a population of 44 individuals, representing 40.37% of the total number of insect’s harvested (109 individuals) (Table 1). The variance analysis followed by the Newman-Keuls test at 5% threshold revealed a significant difference (P-value < 0.001) between averages numbers of species collected.

### Table 1: Average number of insects counted in the cocoa beans in Boguedia’s locality during 120 days of storage

| Insect species | Storage time (Days) |
|---------------|---------------------|
|               | 30                  | 45                  | 60                  | 75                  | 90                  | 105                 | 120                 |
| A. advena     | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>e</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| E. cautella   | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 5.66±1.15<sup>b</sup>| 0.66±0.57<sup>c</sup>| 0±0<sup>g</sup>    | 11.66±2.51<sup>a</sup>| 6.66±1.15<sup>b</sup>|
| L. serricorne | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| Korynetes sp  | 0±0<sup>g</sup>    | 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>c</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| O. surinamensis| 0±0<sup>g</sup>    | 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>c</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| R. dominica   | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| T. castaenum  | 1.66±1.15<sup>g</sup>| 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>c</sup>| 3.66±2.08<sup>d</sup>| 1.66±1.52<sup>a</sup>| 2.66±0.57<sup>a</sup>| 1.66±1.15<sup>g</sup>|

F = 57.42; df = 48; P-value = 0.00001. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

Bédiala’s locality

On the 30th day of storage, the insects collected in the bean stocks of this locality are mainly dominated by the order Coleoptera represented by 5 species: L. serricorne, R. dominica, T. castaenum, O. surinamensis and Stegobium paniceum (Anobiidae) (Table 2). The S. paniceum and O. surinamensis species appear in the stocks at 45 days with mean numbers of 0.66 ± 0.57 and 1.66 ± 1.15, respectively. Species E. cautella and T. castaenum were present throughout the stock control period. The most abundant species is E. cautella, with a population of 57 individuals, representing 36.54% of the total number of insects harvested (156 individuals). The species T. castaenum comes in second place with 33 individuals, whether 21.15%. The variance analysis followed by the Newmans-Keuls test at 5% threshold revealed a significant difference (P-value < 0.001) between averages numbers of species collected.

### Table 2: Average number of insects identified in the cocoa beans in Bédiala’s locality for 120 days of storage

| Insect species     | Storage time (Days) |
|--------------------|---------------------|
|                    | 30                  | 45                  | 60                  | 75                  | 90                  | 105                 | 120                 |
| A. advena          | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>e</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| E. cautella        | 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>d</sup>| 2.66±0.57<sup>c</sup>| 3.33±0.57<sup>d</sup>| 5.66±1.15<sup>b</sup>| 7.66±1.52<sup>b</sup>| 11.66±2.51<sup>a</sup>|
| L. serricorne      | 0±0<sup>g</sup>    | 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>c</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| S. paniceum        | 0±0<sup>g</sup>    | 1.66±1.15<sup>c</sup>| 3.33±0.57<sup>d</sup>| 1.33±0.57<sup>c</sup>| 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>c</sup>| 0.66±0.57<sup>c</sup>|
| O. surinamensis    | 0±0<sup>g</sup>    | 0.66±0.57<sup>d</sup>| 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    | 0±0<sup>g</sup>    |
| R. dominica        | 1.66±1.15<sup>g</sup>| 0.66±0.57<sup>d</sup>| 0.66±0.57<sup>c</sup>| 3.66±2.08<sup>d</sup>| 1.66±1.15<sup>c</sup>| 2±0<sup>c</sup>    | 2.66±0.57<sup>c</sup>|

F = 62.24; df = 48; P-value = 0.00001. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

Bazra’s locality

From the 30th to the 120th day, the insects recorded in the stocks of cocoa beans are represented by 6 species: A. advena, Araecerus fasciculatus (Anthribidae), Cryptolestes ferrugineus (Cucujidae), O. surinamensis, Sitophilus zeamais (Curculionidae) and T. castaenum. At the end of the first 30 days, of the 6 species, 5 were present in stocks with mean numbers ranging from 2 ± 1 to 13.66 ± 2.08 individuals. The species A. fasciculatus appears on the 60th
day and then disappears 75th day of storage. The most abundant species is *A. advena*, whose number of individuals is 63, whether 36.62% of the total number of insects harvested (172 individuals). *O. surinamensis* comes second with 47 individuals, whether 27.32% (Table 3). The variance analysis followed by Newmans-Keuls test at 5% threshold showed a significant difference (P-value < 0.001) between averages numbers of species collected during storage.

| Insect species   | Storage time (Days) |
|------------------|---------------------|
|                  | 30      | 45      | 60      | 75      | 90      | 105     | 120     |
| *A. advena*      | 14±2.64 | 18.33±1.52 | 22.33±3.21 | 12.66±4.61 | 6.66±1.15 | 5.66±0.57 | 5±1     |
| *A. fasciculatus*| 2±0     | 3.66±1.15 | 4±1     | 2.66±0.57 | 1.33±0.57 | 1±1     | 0±0     |
| *C. ferrugineus* | 5.33±1.52 | 7.33±2.52 | 8.33±2.88 | 5.66±1.15 | 3.66±1.15 | 2.33±0.57 | 2±1     |
| *E. cautella*    | 0±0     | 2.33±0.57 | 4.33±0.57 | 2.66±0.57 | 1±1     | 0±0     | 0±0     |
| *L. serricorne*  | 2±0     | 2.66±0.57 | 1.66±0.57 | 0.66±0.57 | 0±0     | 0±0     | 0±0     |
| *O. surinamensis*| 5.33±1.52 | 7.66±2.52 | 9.66±2.88 | 5.33±1.52 | 2.66±0.57 | 2.33±0.57 | 1.33±0.57 |
| *S. zeamais*     | 0±0     | 3.33±0.57 | 4±1     | 1.33±0.57 | 0.66±0.57 | 0±0     | 0±0     |
| *T. castaneum*   | 2±1     | 3.33±1.52 | 2.66±0.57 | 1.66±0.57 | 1.33±0.57 | 1.33±0.57 |

F = 42.12; df = 41; P-value = 0.00001. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

**Gadouan’s locality**

On the 30th day of storage, the recorded insects were represented by 5 species (*A. advena, C. hemipterus, L. serricorne, O. surinamensis* and *T. castaneum*) with mean numbers varying from 2 ± 0 to 14 ± 2, 64 individuals. Species *A. fasciculatus, E. cautella* and *S. zeamais* appeared on the 45th day of stock monitoring. The most abundant species was *A. advena* with 81 individuals, whether 32.66% of the total (248 individuals). It is followed by *C. hemipterus* with 33 individuals, whether 13.31% (Table 4). The variance analysis followed by Newmans-Keuls test at 5% threshold showed a significant difference (P-value < 0.001) between averages numbers of species collected during storage.

| Insect species   | Storage time (Days) |
|------------------|---------------------|
|                  | 30      | 45      | 60      | 75      | 90      | 105     | 120     |
| *A. advena*      | 14±2.64 | 18.33±1.52 | 22.33±3.21 | 12.66±4.61 | 6.66±1.15 | 5.66±0.57 | 5±1     |
| *A. fasciculatus*| 0±0     | 3.66±1.15 | 4±1     | 2.66±0.57 | 1.33±0.57 | 1±1     | 0±0     |
| *C. ferrugineus* | 5.33±1.52 | 7.33±2.52 | 8.33±2.88 | 5.66±1.15 | 3.66±1.15 | 2.33±0.57 | 2±1     |
| *E. cautella*    | 0±0     | 2.33±0.57 | 4.33±0.57 | 2.66±0.57 | 1±1     | 0±0     | 0±0     |
| *L. serricorne*  | 2±0     | 2.66±0.57 | 1.66±0.57 | 0.66±0.57 | 0±0     | 0±0     | 0±0     |
| *O. surinamensis*| 5.33±1.52 | 7.66±2.52 | 9.66±2.88 | 5.33±1.52 | 2.66±0.57 | 2.33±0.57 | 1.33±0.57 |
| *S. zeamais*     | 0±0     | 3.33±0.57 | 4±1     | 1.33±0.57 | 0.66±0.57 | 0±0     | 0±0     |
| *T. castaneum*   | 2±1     | 3.33±1.52 | 2.66±0.57 | 1.66±0.57 | 1.33±0.57 | 1.33±0.57 |

F = 73.21 df = 55 ; P-value = 0.00001. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

**Zaïbo’s locality**

From the 30th day to the 120th day, seven species of insects were recorded in this locality. They are represented by *A. advena, A. fasciculatus, E. cautella, L. serricorne, O. surinamensis, S. zeamais* and *T. castaneum*. On the 30th day of storage, 5 species (*A. advena, L. serricorne, O. surinamensis, S. zeamais* and *T. castaneum*) were present at the control with populations ranging from 0.66 ± 0.57 to 3.66 ± 1.15 individuals. *A. advena* was the most abundant species with a total of 56 individuals, representing 35.44% of the total number of insects harvested (158 individuals) (Table 5). The variance analysis followed by the Newmans-Keuls test at 5% threshold revealed a significant difference (P-value < 0.001) between the average numbers of species collected.
Table 5: Average number of insects identified on cocoa beans in the locality of Zaïbo for 120 days of storage

| Insect species | Storage time (Days) |
|---------------|---------------------|
|               | 30                  |
| A. advena     | 3.66±1.15<sup>a</sup> |
|               | 45                  |
| A. fasciculatus| 2.66±0.57<sup>b</sup> |
| E. cautella   | 0±0<sup>c</sup>     |
| L. serricorne | 0.66±0.57<sup>c</sup> |
| O. surinamensis| 1.33±0.57<sup>c</sup> |
| S. zeamais    | 3.66±1.15<sup>c</sup> |
| T. castaneum  | 2±0<sup>c</sup>     |
|               | 60                  |
| A. advena     | 11.33±1.52<sup>a</sup> |
| A. fasciculatus| 1.33±0.57<sup>de</sup> |
| E. cautella   | 1.33±0.57<sup>de</sup> |
| L. serricorne | 1.66±1.57<sup>d</sup> |
| O. surinamensis| 2.66±0.57<sup>de</sup> |
| S. zeamais    | 2.66±0.57<sup>de</sup> |
| T. castaneum  | 2.66±0.57<sup>de</sup> |
|               | 75                  |
| A. advena     | 8.66±2.08<sup>b</sup> |
| A. fasciculatus| 2.66±0.57<sup>cd</sup> |
| E. cautella   | 2.66±0.57<sup>de</sup> |
| L. serricorne | 0±0<sup>c</sup>     |
| O. surinamensis| 6.66±1.15<sup>bc</sup> |
| S. zeamais    | 0.66±0.57<sup>bc</sup> |
| T. castaneum  | 1.66±1.52<sup>c</sup> |
|               | 90                  |
| A. advena     | 4±1<sup>c</sup>     |
| A. fasciculatus| 1.66±0.57<sup>de</sup> |
| E. cautella   | 0±0<sup>c</sup>     |
| L. serricorne | 0±0<sup>c</sup>     |
| O. surinamensis| 6.66±0.57<sup>c</sup> |
| S. zeamais    | 0.66±0.57<sup>bc</sup> |
| T. castaneum  | 2±0<sup>c</sup>     |
|               | 105                 |
| A. advena     | 3.66±0.57<sup>ad</sup> |
| A. fasciculatus| 1.66±0.57<sup>d</sup> |
| E. cautella   | 1±0<sup>c</sup>     |
| L. serricorne | 0±0<sup>c</sup>     |
| O. surinamensis| 6.66±0.57<sup>bc</sup> |
| S. zeamais    | 0±0<sup>c</sup>     |
| T. castaneum  | 2±0<sup>c</sup>     |
|               | 120                 |
| A. advena     | 2.33±0.57<sup>d</sup> |
| A. fasciculatus| 3.66±0.57<sup>ad</sup> |
| E. cautella   | 2.66±0.57<sup>de</sup> |
| L. serricorne | 1.66±1.52<sup>c</sup> |
| O. surinamensis| 3.66±1.52<sup>ad</sup> |
| S. zeamais    | 1.66±0.57<sup>c</sup> |
| T. castaneum  | 0.66±0.57<sup>c</sup> |

F = 82.25 df = 48; P-value = 0.34. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

Zoukougbou’s locality

From the 30th to the 60th day of storage, only one species of the order Coleoptera, T. castaneum was present in stocks with an average size of 3.33 ± 0.57 individuals (60th day). S. zeamais and E. cautella appear in the stocks at 75 and 90 days respectively. T. castaneum was the most abundant species with a total of 21 individuals, representing 63.64% of the total number of insects harvested (33 individuals) (Table 6). The variance analysis followed by Newmans-Keuls test at 5% threshold showed a significant difference (P-value < 0.05) between averages numbers of species collected during storage.

Table 6: Average number of registered insects of cocoa beans in the Zoukougbou’s locality during 120 days of storage

| Insect species | Storage time (Days) |
|---------------|---------------------|
|               | 30                  |
| E. cautella   | 0±0<sup>c</sup>     |
| S. zeamais    | 0±0<sup>c</sup>     |
| T. castaneum  | 0±0<sup>c</sup>     |
|               | 45                  |
| E. cautella   | 0±0<sup>d</sup>     |
| S. zeamais    | 0±0<sup>d</sup>     |
| T. castaneum  | 0±0<sup>d</sup>     |
|               | 60                  |
| E. cautella   | 0±0<sup>d</sup>     |
| S. zeamais    | 0±0<sup>d</sup>     |
| T. castaneum  | 1.66±1.15<sup>b</sup> |
|               | 75                  |
| E. cautella   | 0±0<sup>d</sup>     |
| S. zeamais    | 3.33±0.57<sup>a</sup> |
| T. castaneum  | 2.66±0.57<sup>ab</sup> |
|               | 90                  |
| E. cautella   | 0±0<sup>d</sup>     |
| S. zeamais    | 2.66±0.57<sup>ab</sup> |
| T. castaneum  | 1.66±1.15<sup>b</sup> |
|               | 105                 |
| E. cautella   | 0±0<sup>d</sup>     |
| S. zeamais    | 1.66±1.15<sup>b</sup> |
| T. castaneum  | 0.66±0.57<sup>c</sup> |
|               | 120                 |
| E. cautella   | 1.66±1.15<sup>b</sup> |
| S. zeamais    | 0±0<sup>c</sup>     |
| T. castaneum  | 0.66±0.57<sup>c</sup> |

F = 132.94; df = 20; P-value = 0.000001. The averages numbers which behave the same letters are not significantly different according to the Newman-Keuls test at 5% threshold.

Relative abundance of species in the Haut-Sassandra region

The insects encountered throughout the sampling in the stocks of cocoa beans in the Haut-Sassandra region were 868 individuals represented by 12 species (Fig 2). The most abundant species was A. advena with 218 individuals or 24.89%, followed by T. castaneum (18.38%), E. cautella (16.89%) and O. surinamensis (15.41%). The least abundant species was S. paniceum with a population of 3 individuals. Factorial correspondence analysis revealed that 4 species, A. advena, T. castaneum, E. cautella and O. surinamensis are characteristic of the Haut- Sassandra region (Fig 3). The species A. advena was the most abundant in the localities of Gadouan, Bazra and Zaïbo. As for the species T. castaneum and E. cautella, they had very high numbers in the stocks of cocoa beans of the localities of Bediala and Boguedia. Concerning the species O. surinamensis it was very abundant in the locality of Bazra (Fig 3).
Fig 3: Factorial analysis of the correspondences representing the characteristics in the Cartesian plane of the insects’ species in the Haut-Sassandra

Estimation of damage and relative weight loss of the cocoa beans during storage time
Insect damage to cocoa beans results in holes and galleries. After a month of monitoring an average total of 937.33 ± 5.68 cocoa beans, only 14.67 ± 2.52 of them carry at least one hole. At the 120th day of storage, the average number of beans carrying at least one hole is 252.00 ± 8.19. The amount of damage is 12.20 ± 0.88% at the end of the second month. It increases very significantly to reach at the third and fourth month of storage the values of 18.60 ± 1.06 and 26.88 ± 0.81% respectively (Table 7). The relative weight loss by the cocoa beans is very low (0.63 ± 0.04%) at the end of the first month of storage. It grows insignificantly to reach 3.73 ± 0.17% at the end of the second month of storage. Beyond this period, the loss becomes more significant with 6.84 ± 0.10% at the end of the third and 8.32 ± 0.21% at the end of the fourth month (Table 7).

Table 7: Relative weight loss caused by insect pests on 1kg of cocoa beans during storage time

| Storage time (Days) | Total number of cocoa beans | Number of cocoa beans carrying hole | Number of cocoa beans without hole | Damage (%) | Relative weight losses of cocoa beans (%) |
|---------------------|-----------------------------|------------------------------------|-----------------------------------|------------|------------------------------------------|
| 0                   | 937.33±5.68                 | 0^e                                 | 937.33±5.68                       | 0^e        | 0^e                                      |
| 30                  | 937.33±5.68                 | 14.67±2.52^d                        | 922.67±4.24^b                     | 01.57±0.28^d | 0.63±0.04^d                             |
| 60                  | 937.33±5.68                 | 114.33±7.57^c                       | 823.00±3.54^e                     | 12.20±0.88^e | 3.73±0.17^c                             |
| 90                  | 937.33±5.68                 | 174.33±9.07^b                       | 763.00±2.83^d                     | 18.60±1.06^b | 6.84±0.10^b                             |
| 120                 | 937.33±5.68                 | 252.00±8.19^a                       | 685.33±9.19^e                     | 26.88±0.81^a | 8.32±0.21^a                             |

Number winnings with at least one hole: F=879.59 ; df = 4; P-value = 0.00001
Number of grains not bearing holes: F=1066.50 ; df = 4; P-value = 0.00001
Damage: F=735.51 ; df = 4; P-value = 0.00001
Relatives losses grain weight: F= 2262.21 ; df = 4; P-value = 0.00001

Note: The average numbers followed by the same letter in a column are not significantly different according to the Newman Keuls test at the 5% threshold.
Discussion:-
The inventory of insects of cocoa bean stocks in the Haut-Sassandra region recorded a total of 12 insect species (A. advena, A. Fasciculatus, C. hemipterus, C. ferrugineus, E. cautella, L. serricorne, O. surinamensis, S. zeamais, T. castaneum, R. dominica, S. paniceum and Korynetes sp). Of these 12 species, 11 are from the Lavabre (1961) listed species on cocoa beans in different parts of the tropics. The results obtained differ from those of Tah et al. (2011); Sadia-Kacou et al. (2015), which recorded 10 species of insects in the stocks of cocoa beans respectively in the Tonkpi region and the Abidjan Autonomous Port. Although our work took place in Côte d’Ivoire like those of Tah et al. (2011) and Sadia-Kacou et al. (2015) only 4 species are common to the three repertoires (A. advena, A. Fasciculatus, C. ferrugineus and T. castaneum). The difference would be related to the extent of the experimental areas, to the storage conditions including temperature, relative humidity and the way in which the beans are dried by farmers. The range of relative humidity between 67.53% and 82.42%, values encountered under experimental conditions, could explain the importance of the number of insects. The value of 80% relative humidity is qualified as optimal. The results are in agreement with those of Mossu (1990); Asiedu (1991). According to these authors, relative humidity values above 80% affect the state of the cocoa bean and, therefore, promote its infestation and mold development (Tah et al., 2016). Temperature values observed during storage ranging from 28.4 to 30.5 °C are favorable for the development of insects such as E. cautella (Aldawood et al., 2013). The presence of Korynetes sp, a species first encountered in cocoa bean stocks, could be the cause of cross infestation during drying by storage of dried cassava by farmers. Korynetes sp, a species that grows in cassava chips during drying, only consumes plant products that have undergone rancidity and infestation by other insects (Delobel, 1992). Four species (A. advena, T. castaneum, E. cautella and O. surinamensis) were present during the 120 days of storage. This presence of these 4 species could be explained by their biological parameters. Females of these species can lay between 50 to 300 eggs (Delobel and Tran, 1993; Goergen et al., 2005; Aldawood et al., 2013). A. Advena was the most abundant species with 218 individuals, whether 24.89%. This abundance may also be due to the longevity of the insect which is 2 to 10 months following the abiotic conditions and duration of the development cycle which lasts approximately one month (Delobel and Tran, 1993). Of a total of 937.33 healthy cocoa beans seen at the beginning of storage, 252 had at least one hole after 120 days of storage. This is linked to the gradual reinfestation of cocoa beans. This reinfestation could be the consequence of long-term storage. Indeed, a long-term conservation seems to favor the setting up of conditions (high humidity, mold) allowing both the development of insects already existing in the stock, as well as the appearance of new species.

Conclusion:-
This study made it possible to carry out an inventory of cocoa bean insect species in the Haut-Sassandra region. The batch monitoring of cocoa beans indicated that all lots from the fields are infested with insects. The identification of the collected specimens revealed 12 species, 10 families and 2 orders that are Coleoptera and Lepidoptera. The species encountered during all the sampling in the cocoa bean stocks were: A. advena, A. fasciculatus, C. hemipterus, C. ferrugineus, E. cautella, L. serricorne, O. surinamensis, S. zeamais, T. castaneum, R. dominica, S. paniceum and Korynetes sp. Among these species, Korynetes sp is a species encountered for the first time in the stocks of cocoa beans. A study of the biocology of this species on cocoa beans will determine its status as a pest or insect occasionally encountered in cocoa stocks. The most abundant species was A. advena (24.89%). Factorial analysis of the correspondences indicated that four species A. advena, T. castaneum, E. cautella, and O. surinamensis are characteristic of the region. Insect damage to cocoa beans was 26.88% at the 120th day of storage. This inventory will help to better target pests in order to develop an appropriate response.

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