MORPHOMETRIC CHARACTERIZATION OF THE MANGO TREE’S MEALY COCHINEAL: RASTROCOCCUS INVADENS ON CITRUS TREE IN SENEGAL

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http://doi.org/10.35410/IJAEB.2019.331349

ABSTRACT

Morphometric data is a useful complement of information that allows precise identification of the different parts of Rastrococcus invadens in Senegal. This study was carried out in four farms (Diatock and Oussouye) all located in natural Casamance and (Santhie and Khay) in the region of Thies. The work took place in February 2016 where citrus production is important. In each farm, we chose the lemon tree and the orange tree which are host plants of R. invadens.

From each plant we collected 10 specimens. This enabled us to get 40 specimens from Casamance and 40 other specimens from Thies. Specimens were coded with regard to both the area and the type of plant they were collected from. The average measurements of the different parts of the insect showed a difference between the populations of the two agro-ecological zones with homogeneity of the populations of Thies and a heterogeneity of the populations of Casamance with a large zone of introgression. The results of this study should allow easier recognition of the different parts of R. invadens.

Keywords: Morphometric data, Rastrococcus invadens, Introgression

1. INTRODUCTION

Fruit production in Senegal, mainly concentrated on mango, citrus fruits and bananas in respective proportions of 67, 23 and 5%, was destined for domestic consumption (Parfonry, 1989).

Since 1981 (the year of the first exports to the French market), it has become the third largest agricultural production in southern Senegal, after cotton and cashew (COLEACP, 1995).

Although the volume exported is small relative to production, Senegal has strongly increased trade abroad and especially to Europe during the last 10 years, reaching average quantities between 5,000 to 6,000 tons per year, despite the fall of 50% in 2010 (about 3,000 tones) due to the quality of the fruits as well as the attacks of insect pests which compromised the marketing campaign (Strebelle, 2013).
However, fruit production is threatened by phytosanitary problems, the most important of which are fruit flies belonging to the family Tephritidae and the mordant scale mango *Rastrococcus invadens* (Williams, 1986) (Homoptera: Pseudococcidae) (N’guetta, 1995). This latter pest was accidentally introduced in Africa in the early 1980s from the Southeast Asia where it originates (Williams, 1986). Mealybug was first observed in Togo and Ghana before spreading to most West African countries, causing damage to mango and other fruit trees (Agounké *et al.*, 1988).

In Côte d’Ivoire, its appearance was reported in 1989 (Neuenschwander *et al.*, 1994). Very polyphagous, *R. invadens* quickly became one of the main enemies of mango and several other fruit trees including citrus and various ornamental and shade plants. *R. invadens* is a species of bisexual and ovoviviparous cochineal that lives in colonies on the leaves of host plants (ANSES, 2015). On plants, mealybugs can be found on leaf petioles, on fruits and peduncles (ANSES, 2015).

Various studies have been undertaken with a view to setting up an integrated pest management program against this pest. Morphometric data is a useful complement of information that allows precise identification of different stages of insect development (Berkani, 2003).

The present work aims to take stock of the morphological and morphometric characteristics of the pest of fruit plants in space and time since its appearance in Senegal.

### 2. MATERIAL AND METHODS

#### 2.1 Sampling

Two populations of *R. invadens* (Williams, 1986) [2] were compared during this analysis: one population from Natural Casamance region and another from Thies region. In each region we chose two farms in two different localities. In Casamance, we chose one farm in Diatock locality and another one in the locality of Oussouye. In Thies region, we sampled in Santhie and Khay localities.

In each farm, we chose the lemon tree and orange tree which represents the host plant of *R. invadens*.

From each plant we collected 10 specimens. This enabled us to get 40 specimens from Casamance and 40 other specimens from Thies. Specimens were coded with regard to both the area and the type of plant they were collected from. Data are clustered in Table 1 below. In general, morphometric analysis of *R. invadens* larvae is usually destructive. Actually, it requires prior death of the specimens and their fixation in alcohol (Osafune *et al.*, 2005) [12].

#### 2.2 Morphometric study

We chose 10 measurable variables with a reasonable degree of accuracy. These mainly include body length, body width, length and width of head, length of rear, median, and front legs. To these are added lengths of abdomen, thorax, and average diameter of sternum (Figure 1).
Table 1: Choice of values

| Body       | Head            | Thorax          | Abdomen     |
|------------|-----------------|-----------------|-------------|
| Lc : Body length | LT : Head length | Th : Thorax length | La : length of abdomen |
| lc : body width  | lt : Head width  | Lp1 : length of the first pair of legs |
|             |                 | Lp2 : length of the second pair of legs | Ls1 : length of first sternum |
|             |                 | Lp3 : length of the third pair of legs |

The relevant parts were mounted on a binocular stereoscope equipped with a camera connected to a computer. Observations were made on L3 (3rd larval stage) specimens which correspond to pre-pupa for males. Specimens are then cleaned in alcohol 70 before proceeding to measurements, each relevant part being carefully separated from the next one. Each specimen of a given sample is associated to a code, using the capital letter of the gender name followed by the first letter of the specific epithet of the corresponding plant, the first letter of the locality of origin and finally the first letter of the region of origin (Table 2).

Table 2: Summary table of the sampling
2.3 Statistical analyses

2.3.1 Raw measurements

A discriminant factor analysis (DFA) of populations with raw measurements of variables in regard to the sampled agro-ecological zones was carried out with the help of the software R version 3.2.3 (Bloomfield, 2014) [13]. This analysis enables to set off the contribution of each variable with respect to their bald measurement, in order to see the most discriminating ones, and to bring out morphometric groups regarding the agro-ecological zones, too.

2.3.2 Converted measurements

- **Size effect**

According to Santos (2015) [14] size effect appears by a circle of correlation that groups all the variables in a single plane for a given axis. This concerns a very undesirable effect which metrical studies try to overcome. The principle of elimination is then to bring all specimens to the same size so as to observe on the PCA only differences in shape.

- **Data conversion**

Eliminating the size effect that affects almost all biometric studies was done using the following approach by Santos (2015) [14]:

- Log-transformation of data: initial data table consists of the variables X1, X2, ..., Xp, subsequently a new data table consisting of the variables log (X1), log (X2), ..., log (Xp) has been created.
- For each specimen, the average over all Log-transformed variables was calculated. This average score is a good idea of the "size" for this specimen.
- Finally, for each specimen, the average size obtained with Log-transforms was deducted from each of these raw measurements.

Size effect is thus eliminated and only the difference in shape will be observed on the PCA. Decreasing the weight of this factor (Size) results in a decrease of the global discrimination between populations and the reduction of the distance between centres of gravity of populations. This transformation was performed in Excel version 2011.

2.3.2.1 Discriminant Factor Analysis (DFA)
A discriminant factor analysis (DAF) of the populations with the transformed data of the variables according to the localities and host plants sampled was carried out with the software R version 3.2.3 (Bloomfield, 2014) [13]. This analysis aims to bring out the contribution of each variable after elimination of the size effect in order to see the differences in form between population groups revealed by the discriminating power of each of the variables.

2.3.2.2 Correspondence Factor Analysis (CFA)
Factorial Correspondence Analysis (CFA) is performed to visualize relationships between specimens of different populations from converted data and test for possible metric similarities between these populations. It is a multi-varied analysis method that considers converted measurements of all populations as variables (Hoda and Marsan, 2012) [15]. For this purpose, a graphical representation is produced from the transformed data that are adapted using the Genetix version 4.05.2 program (Belkhir et al., 2004) [16] to estimate the distribution of morphological diversity at all levels (individuals, populations and total population).

I.3.2.3 Confusion matrix for cross-validation results
The confusion matrix summarizes the reclassifications of specimens to infer both rates of good and bad ranking. This makes it possible to determine the "correct%" which is the ratio of the number of well-ranked specimens to the total number of specimens. Cross-validation is done using transformed data according to agro-ecological zones and plant species sampled with the XLSTAT software version 2016.03.30882 (Addinsoft, 2018) [17].

I.3.2.4 Hierarchical Ascending Classification (CAH)
It represents a method of trees construction that is often delicate and difficult to generalize if the learning data are poorly representative of reality. Automatic classification methods that do not require learning are of great interest when data are completely unknown. They thus make it possible to release subsequent classes that are not obvious a priori. Therefore, the CAH consists of grouping specimens regarding their resemblance or difference. The ascending hierarchical classification is carried out in Excel version 2011.

3. RESULTS
3.1 Raw data
3.1.1 Contribution of the variables with raw measurements in terms of the localities of origin of specimens collected from lemon tree
The discriminant factor analysis obtained from specimens collected from lemon tree shows that the first two factorial axes (dimension) best explain the morphometric variability with 83.14% of inertia power. Following the factorial axis 1 (dimension 1), we find that variables such as Lp2 (F1=12.8), Lp1 (F1=12.3), Lp3 (F1=12.2), Lt (F1=11.6), Lc (F1=11.2), lc (F1=10.9), LT (F1=9.6) and Ls1 (F1=7.10) have largely participated in the construction of the first factorial axis with 70.70% of the power of inertia. Only the variable Th (F1=0.003) contributes slightly to the construction of the first axis. The factorial axis 2 (dimension 2), with a low power of inertia (12.44) is constructed largely by the variables Th (F2=80) and Lc (F2=11.9). Other variables such as Lt (F2=3.29), La (F2=2.82), LT (F2=1.36), Lp3 (F2=0.34), Lp1 (F2=0.09), Ls1 (F2=0.009), Lp2 (F2=0.08) and lc (F2=0.003) contribute little to the construction of this axis. On the first factorial axis, all the variables are positively correlated. Obviously, the size effect affects our ACP. (figure 1) A globally positive correlation for the variables, along the factorial axis of dimension 1, seems to suggest an influence of the data by the "size effect".

Figure 1: Contribution of lemon tree populations’ variables

3.1.2 Discrimination of populations according to raw measurements according to the localities of individuals from the orange tree

The discriminant factor analysis obtained from specimens collected from mango tree shows that the first two factorial axes (dimension) best explain the morphometric variability with 87.33% of inertia power. Following the factorial axis 1 (dimension 1), we find that variables such as LT (F1=11.9), Lt (F1=11.7), Lp1 (F1=11.6), Lp2 (F1=11.3), lc (F1=10.8), Lc (F1=10.2), Lp3 (F1=10.1), La (F1=9.16) and Ls1 (F1=8.7) have largely participated in the construction of the first factorial axis with 74.46% of the power of inertia. Only the variable Th (F1=4.41) contributes slightly to the construction of the first axis. The factorial axis 2 (dimension 2), with a low power of inertia (12.87) is constructed largely by the variables Th (F2=45.2), Ls1 (F2=21.8), lc (F2=10.8) and Lc (F2=9.89). Other variables such as Lp3 (F2=4.13), Lp2 (F2=2.89), Lp1 (F2=2.07), La (F2=1.27), Lt (F2=0.93) and LT (F2=0.91) contribute little to the construction of this axis. On the first factorial axis, all the variables are positively correlated. Obviously, the size effect affects our ACP. (Figure 2) A globally positive correlation for the variables, along the factorial axis of dimension 1, seems to suggest an influence of the data by the "size effect".
3.1.4 Discrimination of populations according to raw measurements according to the localities of individuals from the lemon tree

With the lemon tree, following the two factorial axes with a power of inertia of 83.10%, the AFD (Discriminant Factor Analysis) reveals two groups. A group consisting of Santhie and Khay populations and another group composed by that of Diatock and Oussouye with a zone of significant introgression between the two populations. Khay specimens have some resemblance to those from Diatock.

3.1.4 Discrimination of populations according to raw measurements according to the localities of individuals from the orange tree

Following the two factorial axes with a power of inertia of 87.40%, the AFD (Discriminant Factor Analysis) reveals two groups. A group consisting of the populations of Santhie and Khay and another group composed by that of Diatock and Oussouye with a zone of significant introgression between the two populations. Khay specimens have some resemblance to those from Diatock.
Figure 4: Representation in the main AFD plan of the populations of *Rastrococcus invadens* on the orange tree

3.1.5 Variables allowing the discrimination of lemon tree populations

In regard to the Tukey test, among the 10 variables studied, only the length of the body (Lc), length of the head (Lt), length of the first pair of legs (Lp1), intermediate leg length (Lp2) and the length of the third pair of legs (Lp3) make it possible to differentiate Diatock and Khay populations. The same result is obtained between Diatock and Santhie. Between Oussouye and khay, practically all variables are identical except body length (Lc) and thoracic length (Lth). This is the same result that was observed between Oussouye and Santhie.

Table 3: Morphometric study of specimens (*Rastrococcus invadens*) collected from lemon tree

| localities | Diatock     | Khay        | Oussouye    | Santhie     |
|------------|-------------|-------------|-------------|-------------|
| Lc         | 2.69±0.68a  | 2.81±0.42a  | 2.66±0.54a  | 3.08±0.18a  |
| Lc         | 1.52±0.54a  | 1.63±0.23a  | 1.56±0.42a  | 1.63±0.14a  |
| Lt         | 0.57±0.35a  | 0.75±0.09a  | 0.63±0.22a  | 0.61±0.17a  |
| Lt         | 0.75±0.40a  | 1.09±0.14b  | 0.84±0.15ab | 0.97±0.16ab |
| La         | 1.16±0.55a  | 1.63±0.36bc | 1.46±0.21ab | 1.98±0.22c  |
| Lth        | 0.94±0.23b  | 0.42±0.16a  | 0.57±0.19a  | 0.48±0.20a  |
| Lp1        | 0.53±0.35a  | 0.76±0.12a  | 0.71±0.16a  | 0.60±0.17a  |
| Lp2        | 0.61±0.35a  | 0.85±0.14a  | 0.81±0.08a  | 0.64±0.19a  |
| Lp3        | 0.66±0.35a  | 0.91±0.13a  | 0.89±0.04a  | 0.68±0.20a  |
| Los1       | 0.31±0.17a  | 0.26±0.05a  | 0.29±0.11a  | 0.26±0.03a  |
3.1.6 Variables allowing the discrimination of orange tree populations

According to the Tukey test, among the 10 variables studied, except the length of the thorax (Th), all the other variables make it possible to differentiate Diatock and Khay populations. Between Diatock and Santhie, except the body width (lt) and the length of the thorax (Th), all the variables make it possible to discriminate these two populations. Between Oussouye and khay, the differentiation noted is due to variables such as body length (Lc), body width (lc), head length (Lt), head width (lt) and length of the first sternum (Ls1). Between Oussouye and Santhie, variables such as body length (Lc), body width (lc), head length (Lt), length of the abdomen (La), length of the thorax (Th) and length of the first sternum (Ls1) discriminate their populations.

Table 4: Morphometric study of specimens (*Rastrococcus invadens*) collected from orange tree

| variables | Diatock | Khay | Oussouye | Santhie |
|-----------|---------|------|----------|---------|
| Lc        | 3.95±0.6b | 2.86±0.43a | 4.12±0.33b | 2.67±0.15a |
| lc        | 2.47±0.57b | 1.75±0.31a | 2.28±0.35b | 1.54±0.14a |
| Lp1       | 1.09±0.18b | 0.64±0.20a | 1.05±0.86b | 0.56±0.07a |
| Lp2       | 1.72±0.41b | 1.09±0.21a | 2.01±0.13b | 1.02±0.08b |
| Lp3       | 2.26±0.22c | 1.91±0.40ab | 2.06±0.25bc | 1.64±0.13a |
| Lp4       | 0.61±0.46a | 0.38±0.23a | 1.01±0.04b | 0.46±0.14a |
| Lp5       | 1.57±0.29b | 0.89±0.14a | 1.08±0.07a | 0.91±0.06a |
| Lp6       | 1.68±0.24b | 1.06±0.26a | 1.14±0.12a | 0.98±0.09a |
| Lp7       | 1.74±0.24b | 1.17±0.33a | 1.20±0.16a | 1.04±0.09a |
| Ls1       | 0.37±0.12bc | 0.30±0.04ab | 0.41±0.04c | 0.27±0.03a |

3.2 Converted data

3.2.1 Contribution of the variables with converted measurements in terms of the localities of origin of specimens collected from lemon tree

Unlike raw data, the factor analysis with the transformed data of individuals from the lemon tree shows a reduction of the inertia percentage of 14.68% for the first dimension (factorial axis 1) following a decrease in the discriminating power of most of the variables taxpayers namely: Lp2 (F1=15.2), Lp1 (F1=13.9), Lp3 (F1=13.9), Lp4 (F1=13.8), lt (F1=11.3), Lc (F1=11.1), lattice (F1=10.7) and LT (F1=6.28).

Only the variables Th (F1 = 2.41) and Ls1 (F1 = 1.33) contribute weakly to the construction of this first axis.

The second factorial axis with a very noticeable decrease (2.04%), shows a situation almost identical, compared to the results with the raw data, with an increase in the discriminating power of almost all the variables and a significant contribution of some variables such as Th (F2 =
50.9), Ls1 (F2 = 18.6), Lc (F2 = 11.3) and LT (F2 = 6.26) The best quality of representation is always obtained with the plane formed by axis 1 and 2 with a total inertia percentage of 70.5%.

Figure 5: Contribution of lemon tree population variables

3.2.2 Contribution of the variables with converted measurements in terms of the localities of origin of specimens collected from orange tree
In contrast to raw data, the factor analysis with the transformed data of individuals from the orange tree shows a reduction of the inertia percentage of 14.68% for the first dimension (factorial axis 1) following a decrease in the discriminating power of most taxpayer variables namely: Th (F1 = 13.7), F1 (F1 = 12.8), Lp1 (F1 = 12.3), Lp2 (F1 = 11.7), LT (F1 = 10.5), La (F1 = 9.69), Lp3 (F1 = 8.92), lc (F1 = 8.32), Lc (F1 = 8.32). Only the variable Ls1 (F1 = 3.61) contributes slightly to the construction of the first axis.

The second factorial axis with a very noticeable decrease (2.04%), shows a situation almost identical, compared to the results with the raw data, with an increase in the discriminating power of almost all the variables and a significant contribution of some variables such as Lc (F2 = 23.1), lc (F2 = 18.6), Lp3 (F2 = 15.5), Ls1 (F2 = 12.1), Lp2 (F2 = 11.1) and Lp1 (F2 = 6.29). The best quality of representation is always obtained with the plane formed by axis 1 and 2 with a total inertia percentage of 76.2%.

Figure 6: Contribution of orange
3.2.3 Discrimination of the populations, according to the converted measurements, in terms of the localities of origin of specimens collected from lemon tree

With the lemon tree, following the two factorial axes with a power of inertia of 83.10%, the AFD (Discriminant Factor Analysis) reveals two groups’ one constituted by the populations of Santhie, Khay and another composed of individuals from Diatock and Oussouye. However, we note a certain similarity between the populations of different agro-ecological zones.

![Figure 7: Representation in the main AFD plan of Rastrococcus invadens populations on the lemon tree](image)

3.2.4 Discrimination of the populations, according to the converted measurements, in terms of the localities of origin of specimens collected from orange tree

On the orange tree, along the two factorial axes with a power of inertia of 87.40%, the AFD (Discriminant Factor Analysis) reveals two groups. A group consisting of the populations of Santhie and Khay and another group composed by that of Diatock and Oussouye with a zone of significant introgression between the two populations. Khay individuals have some resemblance to Diatock individuals.

![Figure: Representation in the main AFD plan of Rastrococcus invadens populations on the orange tree](image)
3.2.5 Variables allowing the discrimination of lemon tree populations
According to the Tukey test, among the 10 variables studied, only the length of the body (Lc), head length (Lt), length first pair of legs (Lp1), length intermediate leg (Lp2) and the length of the third pair of legs (Lp3) make it possible to differentiate the Diatock and Khay populations. The same result is obtained between Diatock and Santhie. Between Oussouye and Khay, practically all variables are identical except body length (Lc) and thoracic length (Lth). This is the same result that was observed between Oussouye and Santhie.

Table 5: Variables allowing the discrimination of lemon populations (converted data)

| localities | Diatock | Khay | Oussouye | Santhie |
|------------|---------|------|----------|---------|
| Lc         | 2.82±0.50<sup>a</sup> | 2.86±0.36<sup>a</sup> | 2.73±0.46<sup>a</sup> | 3.16±0.14<sup>a</sup> |
| lc         | 1.66±0.36<sup>a</sup> | 1.66±0.18<sup>a</sup> | 1.63±0.34<sup>a</sup> | 1.71±0.10<sup>a</sup> |
| Lt         | 0.71±0.18<sup>a</sup> | 0.80±0.07<sup>a</sup> | 0.69±0.14<sup>a</sup> | 0.69±0.13<sup>a</sup> |
| lt         | 0.89±0.23<sup>a</sup> | 1.14±0.08<sup>b</sup> | 0.91±0.09<sup>a</sup> | 1.05±0.13<sup>b</sup> |
| La         | 1.30±0.38<sup>a</sup> | 1.68±0.31<sup>b</sup> | 1.53±0.14<sup>ab</sup> | 2.06±0.18<sup>c</sup> |
| Lth        | 1.08±0.41<sup>b</sup> | 0.47±0.15<sup>a</sup> | 0.64±0.14<sup>a</sup> | 0.56±0.24<sup>a</sup> |
| Lp1        | 0.67±0.17<sup>a</sup> | 0.81±0.06<sup>a</sup> | 0.78±0.13<sup>a</sup> | 0.68±0.12<sup>a</sup> |
| Lp2        | 0.74±0.17<sup>ab</sup> | 0.91±0.09<sup>c</sup> | 0.88±0.06<sup>bc</sup> | 0.72±0.14<sup>a</sup> |
| Lp3        | 0.80±0.17<sup>a</sup> | 0.96±0.08<sup>b</sup> | 0.96±0.07<sup>b</sup> | 0.76±0.16<sup>a</sup> |
| Ls1        | 0.45±0.06<sup>c</sup> | 0.30±0.02<sup>a</sup> | 0.36±0.04<sup>b</sup> | 0.34±0.02<sup>ab</sup> |

3.2.6 Variables allowing the discrimination of orange tree populations
According to the Tukey test, among the 10 variables studied, except the length of the thorax (Th), all the other variables make it possible to differentiate the Diatock and Khay populations. Between Diatock and Santhie, except for the width of the body (lc) and the length of the thorax (Th), all the variables make it possible to discriminate between these two populations. Between Oussouye and Khay, the differentiation noted is due to variables such as body length (Lc), body width (lc), head length (Lt), head width (lt) and length of first sternum (Ls1). Between Oussouye and Santhie, variables such as body length (Lc), body width (lc), head length (Lt), length of the abdomen (La), thoracic length (Th) and length of the first sternum (Ls1) discriminate their populations.
Table 6: Variables allowing the discrimination of orange populations (converted data)

| Localities | Dilotok | Khay | Ouissouye | Santhie |
|------------|---------|------|-----------|---------|
| Lc         | 3.36±0.53<sup>b</sup> | 2.64±0.16<sup>a</sup> | 3.15±0.25<sup>b</sup> | 2.63±0.19<sup>a</sup> |
| lc         | 1.83±0.20<sup>a</sup> | 1.67±0.11<sup>a</sup> | 1.80±0.24<sup>a</sup> | 1.67±0.13<sup>a</sup> |
| Lt         | 0.82±0.13<sup>b</sup> | 0.58±0.05<sup>a</sup> | 0.69±0.18<sup>ab</sup> | 0.61±0.04<sup>a</sup> |
| lt         | 1.14±0.18<sup>b</sup> | 1.07±0.05<sup>ab</sup> | 0.99±0.11<sup>a</sup> | 1.08±0.07<sup>ab</sup> |
| La         | 1.88±0.33<sup>a</sup> | 1.65±0.15<sup>a</sup> | 1.69±0.25<sup>a</sup> | 1.69±0.18<sup>a</sup> |
| Lth        | 0.56±0.28<sup>b</sup> | 0.48±0.11<sup>a</sup> | 0.81±0.39<sup>ab</sup> | 0.42±0.10<sup>a</sup> |
| Lp1        | 1.11±0.32<sup>b</sup> | 0.87±0.08<sup>a</sup> | 0.74±0.15<sup>a</sup> | 0.88±0.08<sup>a</sup> |
| Lp2        | 1.20±0.29<sup>b</sup> | 0.97±0.08<sup>a</sup> | 0.92±0.17<sup>a</sup> | 0.99±0.09<sup>ab</sup> |
| Lp3        | 1.27±0.27<sup>b</sup> | 1.04±0.09<sup>ab</sup> | 1.02±0.22<sup>a</sup> | 1.07±0.10<sup>ab</sup> |
| Ls1        | 0.29±0.12<sup>a</sup> | 0.35±0.01<sup>a</sup> | 0.30±0.05<sup>a</sup> | 0.32±0.01<sup>a</sup> |

3.2.7 Matrice de confusion pour les résultats de la validation croisée des populations

The confusion matrix summarizes reclassifications of specimens to infer the rates of good and bad ranking. This makes it possible to determine the "correct%" which is the ratio of the number of well-ranked specimens to the total number of specimens. Thus, specimens from different populations are well ranked in their original populations.

Table 7: confusing mastery of cross-validation of specimens from lemon tree

|         | Casamance | Thies | Total | % correct |
|---------|-----------|-------|-------|-----------|
| Casamance | 20        | 0     | 20    | 100.00%   |
| Thies   | 0         | 20    | 20    | 100.00%   |
| Total   | 20        | 20    | 40    | 100.00%   |

Table 8 : confusing mastery of cross-validation of specimens from orange tree

|         | Casamance | Thies | Total | % correct |
|---------|-----------|-------|-------|-----------|
| Casamance | 20        | 0     | 20    | 100.00%   |
| Thies   | 0         | 20    | 20    | 100.00%   |
| Total   | 20        | 20    | 40    | 100.00%   |
3.3 Hierarchical Ascending Classification (HAC)

3.3.1 Hierarchical ascending classification on the orange tree

The hierarchical ascending classification brings out several morphometric groups based on similarities, from the variables. On the orange tree, the ascending hierarchical classification allows to have 4 groups with group 1 where we find only the populations of Diatock, the group 2 in which we meet all the populations (Oussouye, Diatock, Khay and Santhie), in the group 3 we find only the populations of Santhie and in group 4 we find all the populations except that of Khay. These results show that the population of Diatock is very heterogeneous because it is almost present in all groups. The populations of Oussouye and Santhie are not very heterogeneous because of the 4 groups, they are present on both. Only the population of Khay is homogeneous because it is present only in group 2.

Figure 9: Ascending classification of specimens from the orange tree

Gr1: group 1 Gr2: group 2 Gr3: group 3 Gr4: group 4

3.3.2 Hierarchical ascending classification on the lemon tree

The hierarchical ascending classification brings out several morphometric groups based on similarities, from the variables. On the lemon tree we can distinguish 4 groups with the group 1 which consists mainly of Diatock and Oussouye populations, the group 2 includes all the populations (Diatock, Oussouye, Santhie and Khay). In groups 3 and 4 we find respectively of Oussouye and Diatock populations. These results show that Khay and Santhie populations are homogeneous while those of Diatock and Oussouye are heterogeneous because they are found in all groups.
Figure 10: Ascending classification of specimens from the lemon tree

Gr1: group 1   Gr2: group 2   Gr3: group 3   Gr4: group 4

3.4 Correspondence factor analysis (CFA)

3.4.1 Correspondence factor analysis (CFA) on the lemon tree

The discriminant factorial analysis reveals that the first five factorial axes explain all the morphometric variability of this cochineal. However, the plan formed by the first three axes best explains the discriminative situation of agro-ecological zones with an inertia 100%. The first factorial axis with an inertia of 37.2% discriminates the group formed in majority by the Oussouye individuals. The second factorial axis with an inertia of the order of 34.6% discriminates the other groups of the one containing almost all the individuals of Diatock. The third factorial axis with an inertia of 28.19%, allows the discrimination of the group which contains the majority of the individuals of Khay and Santhie, of the group formed in majority by the individuals of the area of Thies.

Figure 11: Simulation of the morphometric variability of specimens of *R. invadens* on lemon tree following the first three axes of the AFC

3.4.2 Correspondence factor analysis (CFA) on the orange tree

The discriminant factorial analysis reveals that the first five factorial axes explain all the morphometric variability of this cochineal. However, the plan formed by the first three axes best explains the discriminative situation of agro-ecological zones with 100% inertia. The first factorial axis with an inertia of 34.41% discriminates the group formed in majority by the Oussouye individuals. The second factorial axis with an inertia of the order of 34.01%, discriminates the other groups of the one containing only the individuals of Santhie. The third factorial axis with an inertia of 31.58%, allows the discrimination of the group which contains all the individuals of Diatock, and a large part of the individuals of Khay with introgressions of some individuals of the other ecotypes.
Figure 12: Simulation of the morphometric variability of specimens of *R. invadens* on orange tree following the first three axes of the AFC

4. DISCUSSION

The raw data with the lemon tree reveal two groups made up on the one hand by the populations of Santhie and Khay and on the other by those of Diatock and Oussouye. Considering the measurement of each variable obtained after transformation, we can see that the individuals harvested from lemon trees have a certain dissimilarity with, on the one hand, individuals from the Niayes zone that form a group and, on the other hand, individuals from Casamance.

On the orange tree, they also reveal two groups. A group consisting of the populations of Santhie and Khay and another composed by that of Diatock and Oussouye with a zone of significant introgression between the two populations. Considering the measure of each variable obtained after transformation, we find that Khay individuals have a certain resemblance to Diatock individuals. But also we can say that the individuals of Diatock have the largest dimensions for all the variables: length of the body (3.82mm), width of the body (2.32mm), length of the head (0.95mm), width head (1.57mm), length of the abdomen (2.11mm), length first pair of leg (1.43), length second pair of leg (1.54mm), length third pair of leg (1, 60mm) while on the lemon tree the largest dimensions of the length of the first segment (0.35mm) are observed in the individuals of Khay and Santhie.

The discriminant factorial analysis of the lemon tree reveals the group formed mainly by the Oussouye individuals and discriminates the other groups from the one containing almost all the Diatock individuals. It also allows the discrimination of the group which contains the majority of the individuals of Khay and Santhie, the group formed mainly by individuals from the region of Thies.

On the orange tree, it makes it possible to discriminate the group formed in majority by the Oussouye individuals, that containing only the individuals of Santhie and a third group which
contains all the individuals of Diatock, and a large part of the individuals of Khay with introgressions of some individuals from other ecotypes. From these results we can say that some individuals belonging to a previously defined area have more similarities with other individuals from neighboring agro-ecological zones: this is the case of Santhie and Khay individuals who are all in the same Ecological agro zone but also with less close area individuals like Diatock.

This shows that the distance corresponds to a discriminative criterion because it intervenes in the variation of the morphology of the species, as well as the climatic conditions (Gbagni et al., 2015). Apart from this semblance of homogeneity, Khay is revealed as the most homogeneous population while Diatock is the most heterogeneous. This could lead us to consider the Niayes area as the focus of the infestation (Han et al., 2007).

The results with the hierarchical ascending classification show that:

On the lemon tree we can distinguish 4 groups with the group 1 which consists essentially of the populations of Diatock and Oussouye, group 2 in which we find all the populations (Diatock, Oussouye, Santhie and Khay). In groups 3 and 4 we find respectively populations of Oussouye and Diatock. As the populations of Diatock and Oussouye are almost found in all groups, we can consider that the individuals from Casamance (Diatock and Oussouye) are more heterogeneous than those from the Niayes (Khay and Santhie).

With the orange tree, the ascending hierarchical classification also allows to have 4 groups with group 1 where we find only the populations of Diatock, the group 2 in which we meet all the populations (Oussouye, Diatock, Khay and Santhie), in group 3 we find only the Santhie populations and in group 4 we find all the populations except that of Khay. These results confirm the heterogeneity of the Diatock population whose individuals appear in all groups. The populations of Oussouye and Santhie are not very heterogeneous because of the 4 groups they are present only on both. Only the population of Khay is homogeneous because it is present only in group 2.

However, greater similarity between Khay and Oussouye individuals is noted, but also between Diatock individuals and others who seem even closer. According to these two agro-ecological zones, we can consider that the individuals are more homogeneous in Thies than in Casamance. The morphological homogeneity of intragroup individuals in the Niayes biotype is explained by the fact that the plants in this zone constitute the primary specul of R. invadens (FALL et al., 2017).

5. CONCLUSION
The study of the morphometric characterization of Rastrococcus invadens populations made it possible to acquire additional information on the biology of this phytophage. It revealed morphometric groups more under less distinct especially between the zone of niayes and low Casamance. All of its results should be taken into account when developing IPM techniques against this pest in citrus orchards in Senegal.

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