

# GROWTH, HISTO-CYTOLOGY AND BIOCHEMISTRY OF BANANA PLANTAIN (*MUSA AAB CV HORN 1*) BUD SUCKERS

L. TURQUIN<sup>1</sup>., S. AKE<sup>1</sup>., L. GRILLET<sup>2</sup>, P. K. ANGUI<sup>3</sup>, P. A. ANNO<sup>1</sup>

<sup>1</sup>Laboratoire de Physiologie végétale, Université de Cocody-Abidjan,  
22 B.P. 582 Abidjan 22, Côte d'Ivoire.

<sup>2</sup>Laboratoire Population et Environnement, UFR de sciences naturelles,  
Université d'Aix-Marseille 1, 3 place Victor Hugo, 13331 Marseille cedex 3, France.

<sup>3</sup>Laboratoire de Pédologie, UFR des Sciences et Gestion de l'Environnement,  
Université d'Abobo-Adjamé, 02 B.P. 801 Abidjan 02, Côte d'Ivoire.

## ABSTRACT

The banana plantain (*Musa AAB cv Horn 1*) is one of the most important staple crop in the world, particularly in Côte d'Ivoire where it plays a major role in the economic system. In traditional plantations, farmers use sizeable suckers, also called bayonets, which, generally, are reduced to a single plant per mother stand at the expense of the numerous buds or bud suckers. This farming practice brings about several disadvantages of which the lack of potential propagation units such as bud suckers. Faced with this crucial slip-seed shortage, a more emphasis was put on bud suckers. The objective of this work is to improve banana plantain culture by pointing out the agronomic importance of bud suckers in farming situation. Suckers were introduced to a hydroponic media, at 35 °C for 16 days and their morphological and biochemical parameters were monitored with time. A nutritive solution was prepared by a 1200-fold dilution of a base solution. This solution served as a medium for the growth of suckers. Results showed that suckers were vigorous and roots healthy. Such conditions of stimulation of bud suckers growth, which were naturally inhibited by the mother plant, allowed for the awakening, subsequent rapid growth and the development of lanceolate leaves in a 15-day period. Anatomic studies revealed a typical scaly parenchyma characterized by the absence of a palissadic tissue. Biochemical analyses showed that phenols (essentially tannins) and carbohydrates were more prominent than proteins.

**Keywords :** Banana plantain, bud suckers, hydroponic culture, carbohydrates, phenols, Côte d'Ivoire.

## RESUME

### CROISSANCE, HISTO-CYTOLOGIE ET BIOCHIMIE DES REJETS ÉCAILLES CHEZ LE BANANIER PLANTAIN (*MUSA AAB, CV CORNE 1*)

*Le bananier plantain (*Musa AAB, cv Corne 1*) constitue une des principales cultures vivrières dans le monde, notamment en Côte d'Ivoire, où son importance est capitale dans l'économie. Dans les plantations traditionnelles, les paysans utilisent de grands rejets encore appelés baïonnettes, généralement réduits à l'unité par pied-mère, au détriment de nombreux bourgeons ou rejets écaillés. Cette pratique culturale engendre plusieurs inconvénients dont l'insuffisance des unités potentielles de propagation que constituent les rejets. Face au problème crucial de disponibilité de ces semenciboutures, les rejets écaillés constituent une solution. L'objectif de ce travail est l'amélioration de la culture du bananier, par la mise en évidence de l'intérêt agronomique des rejets écaillés pour les replantations paysannes. Les rejets ont été mis en culture hydroponique, à 35 °C durant 16 j. Leurs paramètres morphologiques et biochimiques ont été déterminés, ainsi que leur évolution dans le temps. Une dilution*

appropriée d'une solution nutritive de base, d'un facteur de 1200, a permis la croissance de rejets vigoureux dont toutes les racines sont saines. Ces conditions de stimulation de l'éveil des rejets écailles, dont la croissance est naturellement inhibée par le pied-mère en plantation, ont permis leur croissance rapide avec un développement de feuilles lancéolées en quelques jours (15 j). L'étude anatomique a montré un parenchyme écailleux typique caractérisé par l'absence de tissu palissadique. L'analyse biochimique a permis de mettre en évidence l'abondance des phénols (essentiellement des tanins) et des glucides, à l'opposé des protéines.

**Mots clés :** Bananier plantain, rejets écailles, culture hydroponique, glucides, phénols, Côte d'Ivoire.

## INTRODUCTION

The banana plantain is one of the most important staple crop in the world, with a production of nearly 87.4 Mt per year. In Côte d'Ivoire, it plays a crucial role in the economic system. The cultivars concerned with this study belongs to the AAB triploid group of the *Horn* type. It is the cv Corne 1 banana, which produces both 'bayonets' and bud-suckers. The latter have, until now, been neglected by farmers for the benefit of bayonet suckers. This farming practice presents the following disadvantages :

- bud-cuttings are often voluminous and cumbersome, making hauling difficult ;

- flowering is erratic (nonuniform). It varies from few weeks to several months causing harvest not only to be spread over time but also difficult to monitor ;

- a high monitoring and time costs from plant establishment to end of harvest ;

- finally, a somewhat long planting - harvest cycle which exceeds, by far, 12 months.

Faced with this propagation problems related to plantain cultivation, different alternatives were considered, notably the development of several plantain cultivars under hydroponic culture conditions (Swennen *et al.*, 1986), the rapid establishment of the banana *in vivo* (Cordeiro and Dos Santos, 1991) with a

long delay in production. Finally, the obtention of a somatic embryo from *in vivo* culture by Bierberach and Escalant (1996). Thus, *in vivo* culture, which could be a good planting alternative for reasons downstream of vitroplants production techniques, is out of the reach of most farmers due to the following reasons :

- an important investment is needed ;

- vitroplants production infrastructure is far from site of utilisation ;

- purchasing costs to farmers is prohibitive.

This study is aimed at experimenting a rapid growth stimulation and development of banana plantain *Musa* AAB cv Corne 1 bud suckers. The ultimate objective being to improve the banana production through :

- An important gain in time (several months) in the establishment of a banana plantation, eventually a reduction in the growth cycle ;

- a revealing of the agronomic importance of this type of bud suckers (type *b*) ;

Expected results from this study will, on one hand deal with the development of early dormancy breaking conditions and of type *b* shoot growth, and on the other hand, reveal the agronomic importance of bud-suckers replantations by farmers.

## MATERIAL AND METHODS

### PLANT MATERIAL

Plant material, used for the different trials, were made of type *b* suckers, of banana plantain (*Musa* AAB cv Horn 1, a hybrid obtained from a cross between *Musa Acuminata* and *Musa Balbisiana*. Type *b* suckers or simply *b*-suckers originated from deep underground and appeared to the surface of the ground always around the mother stand, apparently were never directly associated to the mother bulb (Turquin, 1989). Moreover, the suckers evolved to adult plants through different development stages. At stage 3, *b*<sub>3</sub>-suckers had buds bigger than those from the preceeding stage. Their rhizomes were curve-shaped and voluminous. The aerial part was well developed. The bulb was provided with long roots capable of reaching 20 cm. Because of the important number, and frequency, and low volume, only *b*<sub>3</sub>-suckers were considered in these trials (figure 1 : A-B).

Hydroponics cultures of bud-suckers were conducted at the Laboratory of Plant Physiology at the University of Cocody. »

### METHODS

#### SHOOT GROWTH STIMULATION TRIALS UNDER CONTROLLED CULTURE CONDITIONS

Shoots originating from banana plantations at the IDEFOR-DFA Station in Azaguié, and from the collection plots of the Laboratory of Plant Physiology at the University of Abobo- Adjamé (Abidjan).

#### Banana plantain bud-suckers sample collection

Bud-suckers sampling from parent stands was conducted in the morning.

Buds freed from dirt and roots were subject to a careful pralinage, that is, dipped into a binding agent solution at 10 % (nemacure). After drying, samples were prepared for calibration trials which consisted in making sets of suckers of similar masses and dimensions, or of the same order of magnitude. Suckers were first identified, before weighing and dimensions were recorded.

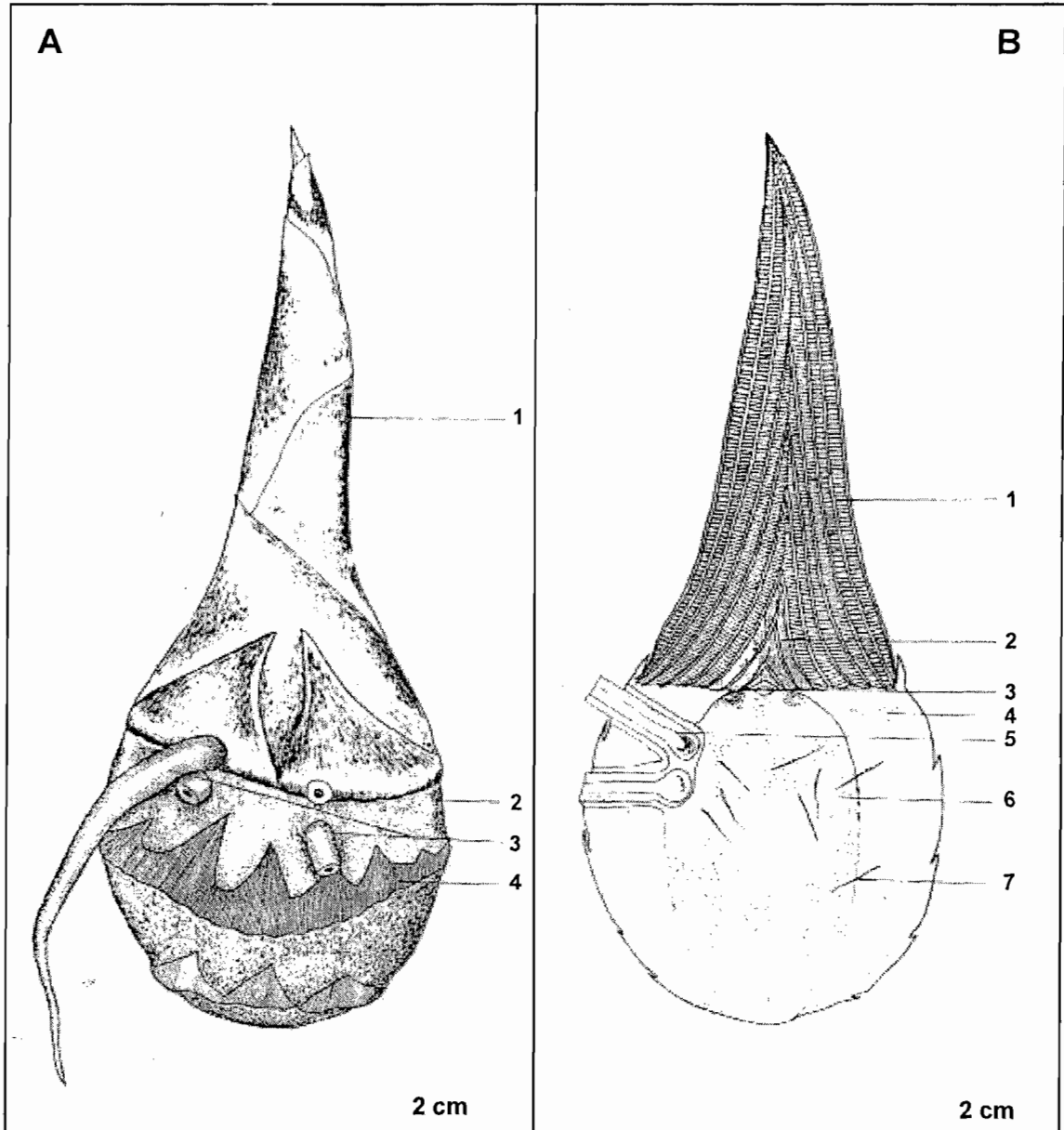
### Experimental set up

Calibrated shoots were introduced to culture media by following experimental design described in the following manner : cultures were conducted in 40 liter-glass containers. Temperature was set at 35 °C and culture medium aeration was achieved using a thermo-plunger (Polystat II from Bioblock). Evaporation was reduced to a minimum with the help of a plexiglas tank. Photoperiod was set at 13 hrs of light (figure 2 : A-B). Culture medium was made up with a nutritional solution supplement.

### Determination of adequate nutrition levels

In preliminary trials, a culture without a base nutritional value was found to limit shoot growth (necrosis at the end of the first week following culture, using distilled water alone as the nutritive medium, plant were also stressed with running water).

A nutritional mineral solution supplement (complexal from SOFACO Groupe AGREVO), normally used as a foliar spray, was employed. It was adjusted it to our experimental conditions, hence the necessity to determine the concentration that will constitute a base nutritive solution for shoots under culture. The nutritional supplement was added to the culture medium right at start. Total volume in culture tank was 14 l. Nutritive solution level inside the tank was adjusted as needed.



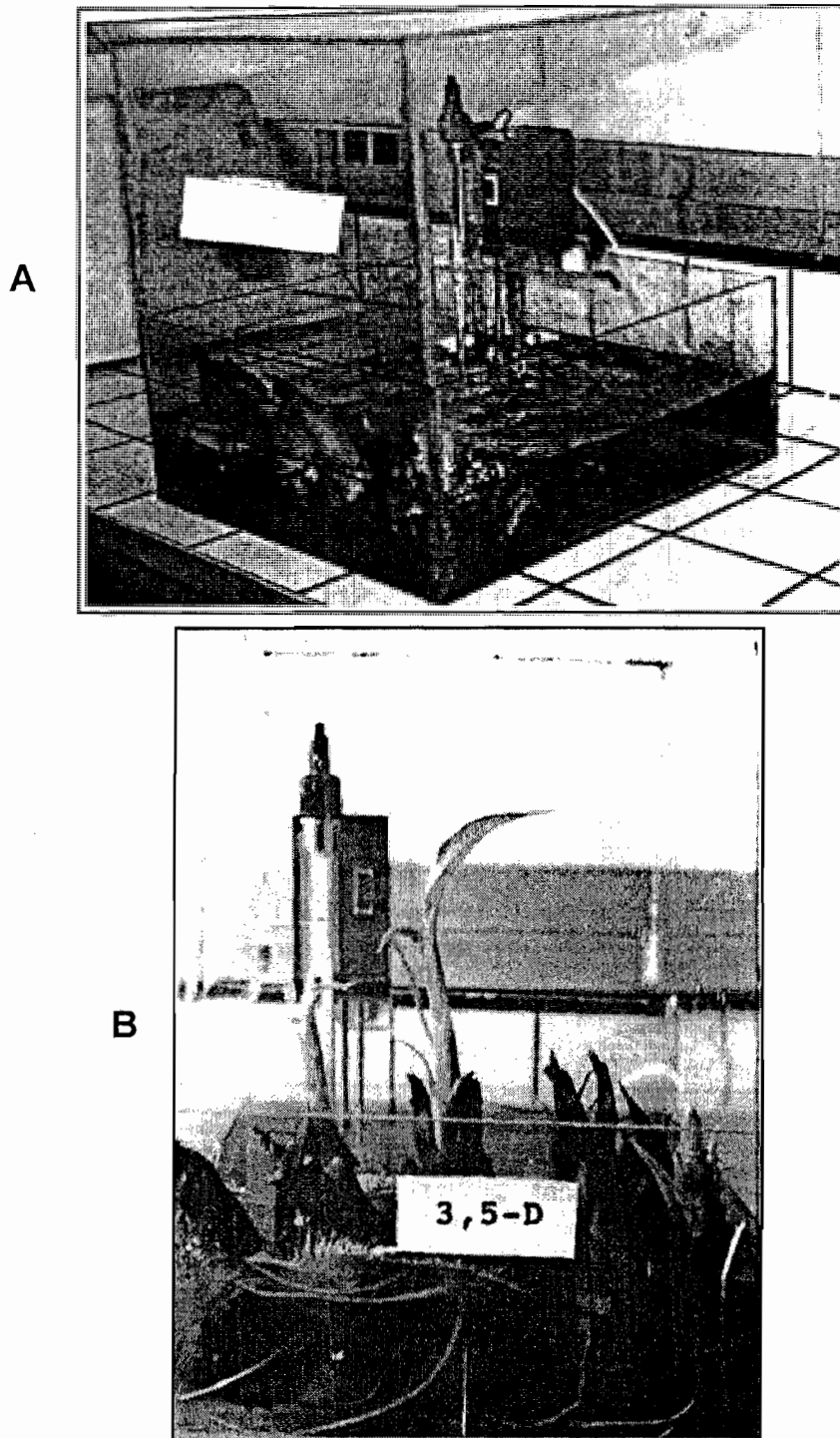
**Figure 1 :** A bud sucker (type  $b_3$ ) of banana plantain cv Horn 1  
*Rejet de type  $b_3$  du bananier plantain cultivar Come 1.*

A- Overall external view of a bud sucker

1. scale 2. bulb 3. roots 4. Cankered scale cicatrice

B- A longitudinal section of a bud sucker

1. Old or external scales	2. Two internal young scales
3. apical meristem area	4. peripheral bulb area
5. roots	6. central bulb



**Figure 2 :** Experimental device for the hydroponic culture of the banana plantain cv Horn 1 bud suckers.

*Dispositif expérimental de la culture de rejets de bananier plantain cultivar Come 1 en milieu hydroponique.*

A- Suckers in culture at 0 days

B- 16 aged cultured suckers

### Measurements of suckers growth parameters

For kinetic measurements, each sucker was used once at 0, 4, 8, 12, and 16 days. The suckers were freeze-dried then analysed. For each determination, three suckers were used because of a limited number per mother stand.

Each trial was conducted at least twice. Bud sucker weight and height were recorded every three days. Qualitative observations, such as scale opening time, the general state of the sucker and the number of lanceolate leaves, were also made. Roots were counted during the entire experiment ; the level of growth that corresponded to the presence or the absence of root ramifications was also recorded.

### Biochemistry of the banana bud suckers

A study of the biochemical properties of the bud suckers was conducted in order to quantitatively evaluate markers with respect to histochemical characteristics. All analyses were carried out using freeze-dried plant materials.

### Carbohydrates

After grinding and filtering through a fine mesh, a 2 g plant material was introduced into a box which was then refluxed with a 50 ml 80° boiling ethanol solution for 30 min. Alcohol extracts were evaporated to dryness at 37 °C. The enzymatic dosage method (Bergmeyer, 1978) was used. Results were expressed in  $\mu$ moles of glucose or fructose or  $\mu$ g sucrose (or equivalent glucose) per ml of solution.

### Proteins

Proteins were extracted for 2 min. using a 2 g- plant material in the HEPES buffer (25 mM DTT, 5 mM and Triton X-100, 0,1 %, pH 7). Then the supernatant

solution was precipitated in presence of ammonium sulfate at 80 % saturation. Protein debris were again treated with a minimum amount of the buffer without a non oxidizing agent. Bradford (1976) carried out soluble protein content determinations. Protein content was expressed in  $\mu$ g/ml of dry matter using a calibration curve.

### Phenols

Soluble phenols were extracted using the method by Bastide (1978), modified and adapted to our plant experimental conditions. Phenolic compounds were determined using the procedure described by Marigo (1973). Results were expressed in meq. chlorogenic acid (méq. ac.chl. per gram of dry material).

### Cytological techniques

Parts observed during histological studies were the scales, the meristematic zone, central bulb, peripheral bulb and junction point between the two parts of the bulb. Explants were prepared then subject to a double coloration : first, by the Schiff Reactant, then by the Naphtol Blue-black. A histolocalisation of the peroxydases enzymes by 3,3- diaminobenzidine (DAB) was carried out on other samples. Samples observed using a scanning microscope were treated as follow : fixation of explants with glutaraldehyde-paraformaldehyde, then conservation in alcohol at 70°. Before observations were made, samples were first dehydrated with three alcohol washings at 100°, then passed through a CO<sub>2</sub> atmosphere according to the Critical Point Method and finally metalized with a palladium ion. Observations were made and photographs taken with the use of a model GEOL 59H 35 microscope.

### Parameter measurements

For growth stimulation, of type *b* suckers subjected to a hydroponic culture,

morphological parameters were measured over a period of 16 days. Three suckers were used for each measurement during culture. Each experiment was repeated at least once. The average from three measurements was used per experimental point.

For biochemical parameters, in another experiment, three suckers were used once for each determination. For every biochemical compound determined, average DO data was computed. However, as a comparison, a *t*-test (STATWORK Program) was used with respect to morphological parameters as variables.

## RESULTS

### REQUIREMENTS FOR MINIMUM GROWTH AND THE VIGOR OF BUD SUCKERS UNDER CULTURE

Several dilutions of the nutritional supplement solution were tested. A lower dilution of the mother solution (1/120) showed toxicities resulting in necrosis

of important tissues leading to rotting : only less than 20 % of shoots survived. A salt content corresponding to dilutions between 120 to 600-folds showed mild toxicities characterized by an absence of scale opening, poor root development and vegetative growth in hydroponics culture. For the dilutions investigated, only a nutritive mineral « complexal » solution diluted 1200 fold maintained minimum suckers growth in a hydroponic culture medium.

### Morphological parameters of the suckers

Table 1 presents the evolution of some morphological parameters of the suckers subjected to hydroponic culture conditions. Weight gain was 42 g over a period of 16 days. The increase in weight was not evident before two days of culture. Sucker height increased by 4 cm, or a growth rate of 0.3 cm/day. Root onset began at the 4<sup>th</sup> day, with an average length of 10 cm, or a growth rate of 0.6 cm/day in the same period. Lanceolate leaves appeared at 12 days following a progressive development of the scales.

**Table 1** : Some morphological characteristics of type b suckers of the banana plantain cv Horn 1 under hydroponic culture medium.

*Evolution de quelques paramètres morphologiques de rejet b de bananier plantain cv Corne 1 en culture hydroponique.*

Morphological parameters	Time of culture (days)					
	0	4	8	12	16	
Suckers	Height (cm)	19±2	19±3	20±3	20±4	23±2
	Mass (g)	236±10	254±32	248±40	262±35	278±45
Roots	Number	0	1±0	2±1	4±1	4±1
	Total length (cm)	-	-	-	-	11±3
Lanceolated leaves	0	0	0	1	1	

### **Cytological and histochemical structure of the suckers**

Growing bud suckers were self-rolled with a void structure (figure 3 : A-B). Starting from the back, toward the front, the following could be distinguished :

- an epidermis covered with a thin cuticule, and stomata (figure 4) ;

- a parenchyma around which were arranged sclerenchyma islands ;

- a lacunal parenchyma housing a ray of woody tissue. The structure of the lacunal parenchyma reminds of that of a ladder (figure 5 : A). On the adaxial side only, complete partitions alternated here and there with incomplete partitions (figure 5 : B) ; whereas, on the abaxial side, partitions were visibly ordered (figure 5 : C) ;

- a parenchyma with less important voids/spaces, poor in sclerenchyma tissue on the abaxial side ;

A palisadic parenchyma was not apparent in the structure of the suckers.

The bulb was made of a non chlorophyllous tissue. It is a parenchyma containing numerous egg-shaped amyloplasts. It is also made of two distinct parts separated by the Mangin Line, rich in woody rays tissue (figure 6 : A-B). The central bulb (medular zone) is made of parenchyma cells of increasing size, more and more differentiated from the apex to the base. The peripheral bulb (cortical zone) contains the central bulb. It is made of large size parenchyma cells and limited to the outside by a layer of epidermic cells.

Tracheids that were observed were scarred (figure 7). In the different parts of the bulb some cells contained

calcium oxalate crystals of raphid shapes (figure 7 : B).

At the root level, a longitudinal section shows that the meristematic tissue was surrounded by a root cap made of an amyloplast-rich tissue (statoliths). The outer cells of the root cap is peeling off. A cross section through the rear part (young stage) shows that the root was divided into two zones of unequal importance (figure 8 : A). One can distinguish from the outside to the inner part, the cortex and the central cylinder. The well-developed tissue had two parts : it was limited to the outer part by piliferous tissue/bed cells. Under this bed of cells, four constituted the external part of the cortex with non lined-up large cells. The internal part of the cortex was limited to the inner part by a frame-like endodermis.

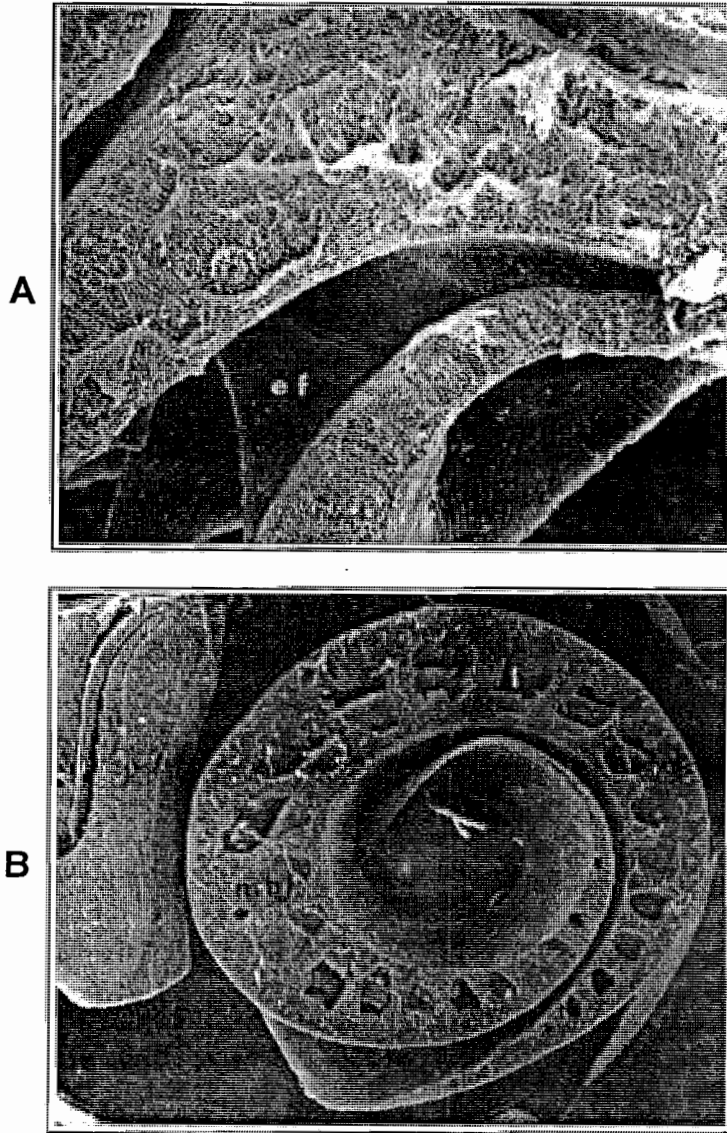
A transversal section through the nearest part of the root (advanced stage) shows the structural changes of the cortex (figure 8 : B). Certain cells evolved with voids between them ; this caused the cortex to be divided into two parts :

- a peripheral area, where the peripheral layer had nearly disappeared, made of a parenchyma ;

- an intermediate area made of lacuna, and parenchyma ;

- an internal area made of a parenchyma with line-up cells. Pericycle cells seemed to alternate with those from the endodermis. The central cylinder was limited by a layer of cells the pericycle. The rays of xylem and primary phloem, had a very distinct internal disposition on the outer part of the stele (figure 8 : C). Secondary ramifications of the root originated from the pericycle.

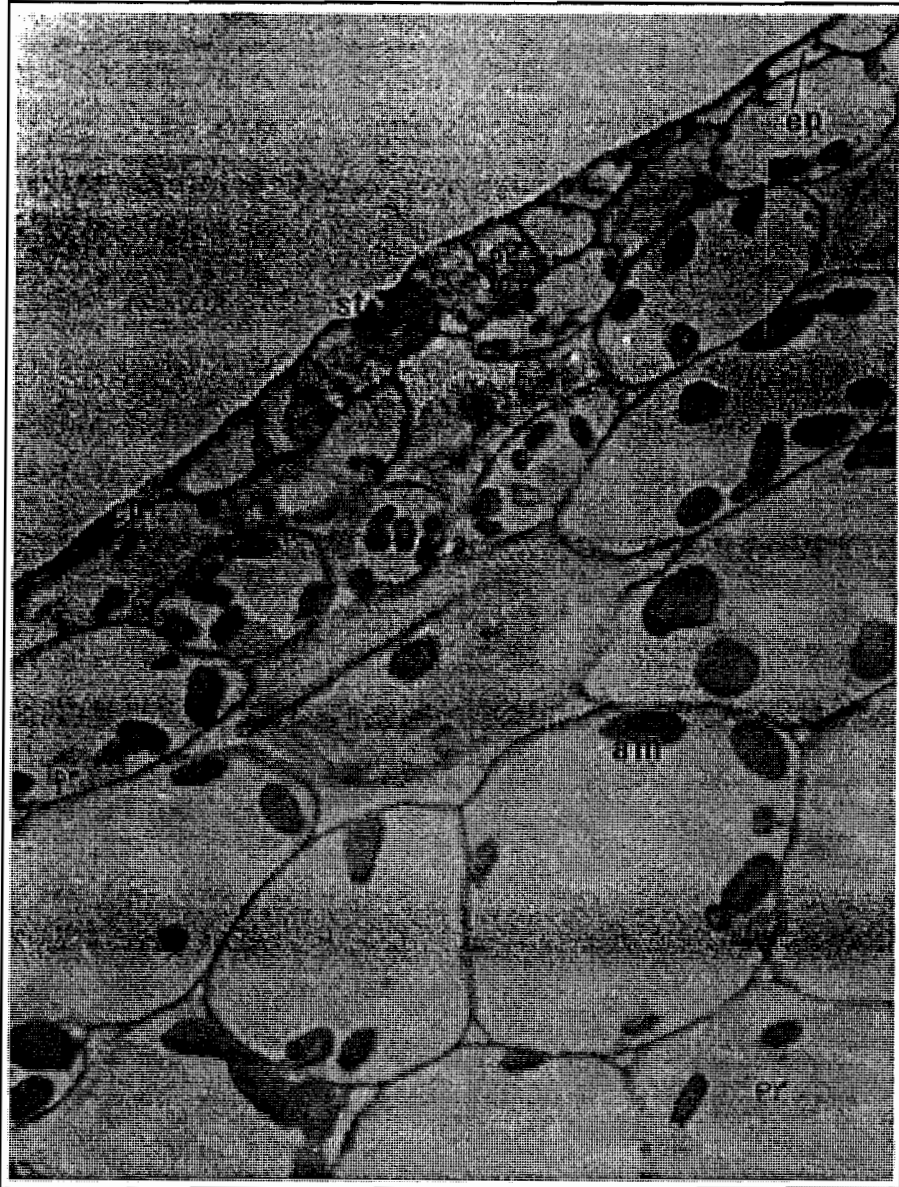




**Figure 3** : A cross section of inner leaf scales of the banana plantain cv Horn 1 under a scanning electron microscope.

*Coupe transversale d'écaillés internes de rejet du bananier plantain cv Come 1 en microscopie électronique à balayage.*

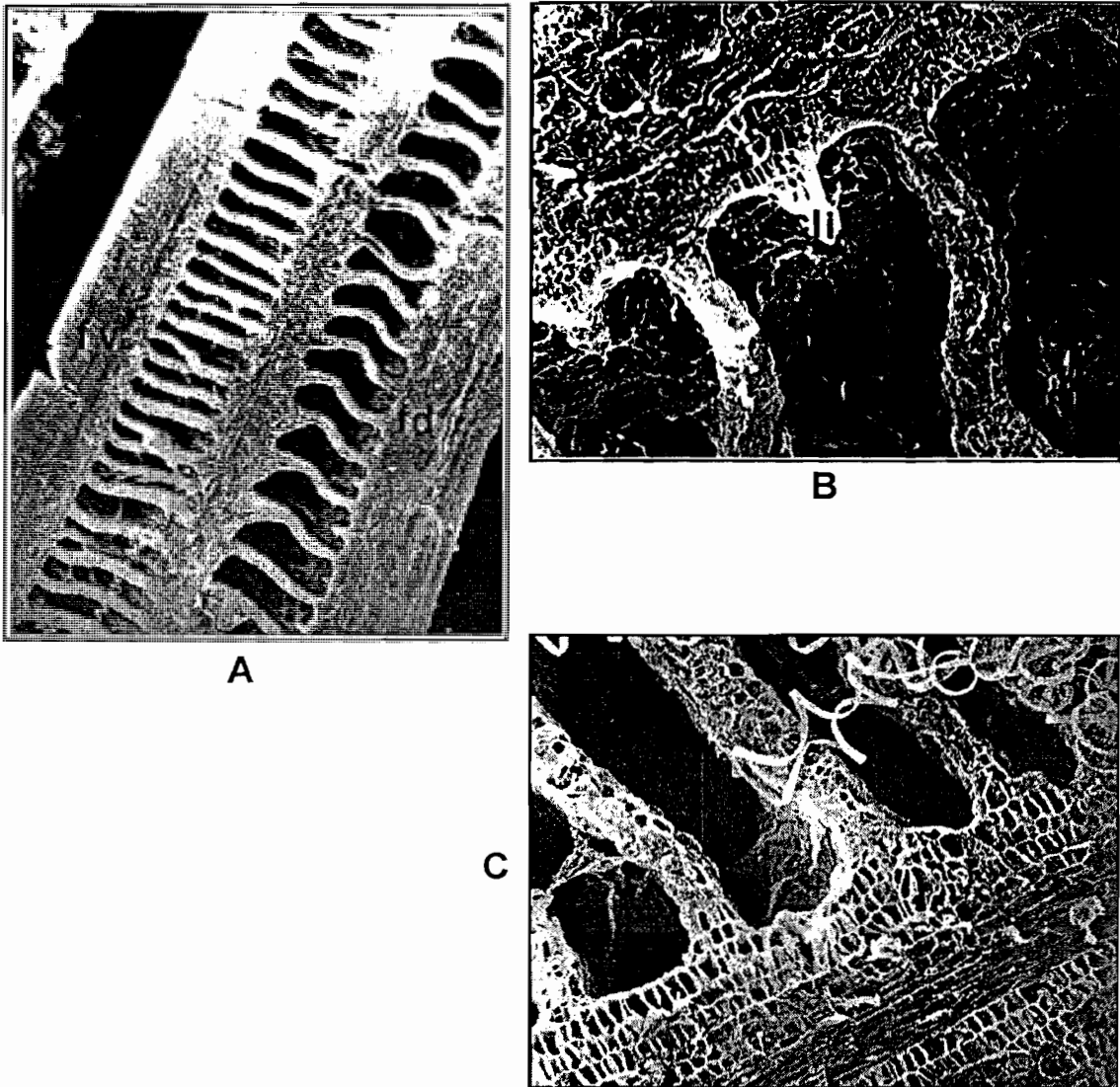
- A- Over view of a rough leaf scale (G x 30). Base of the rough leaf scale (ef) before differentiation of lacunas.
- B- Overview of a young enrolled leaf scale (G x 30).



**Figure 4 :** Detail of a cross-sectional view of the edge of a scale (G x 400) of a banana plantain cv Horn 1 bud-sucker.

*Détail du bord d'un rejet d'écaillés de bananier plantain cv Corne 1, en coupe transversale (G X 400).*

The layer of epidermis cells (ep) contains from time to time stomates (st) made of reniform amyloplasts rich cells (am), a cuticle, (cu) and proteins in the nucleus (pn).



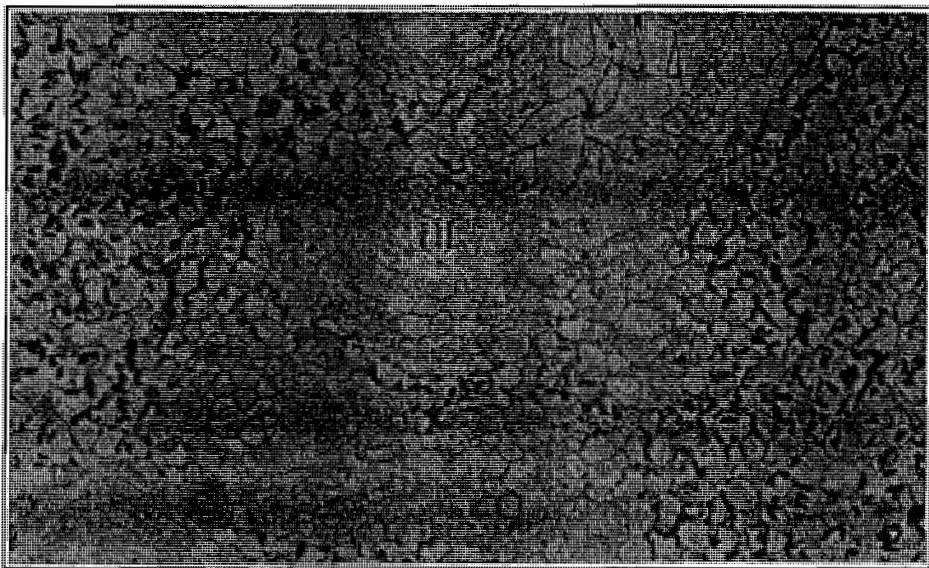
**Figure 5 :** A longitudinal section of an outer scale a banana plantain cv Horn 1 bud sucker under a scanning electron microscope.

*Coupe longitudinale d'écaille externe de rejets de bananier plantain cv Come 1 en microscopie électronique à balayage.*

- A- Overview of a longitudinal section of an outer scale (G x 18) ladder-like scale structure with dividing walls (cli) on back side (fd) at times on the ventral side (fv).
- B- Detail of a longitudinal section of a portion of a scale (G x 100). Note the presence at times of incomplete partitions (cli) on the ventral side (fv).
- C- Detail of a longitudinal cross-section of a portion of an outer scale (G x 100). Note the presence of complete partitions (cli).



A



B

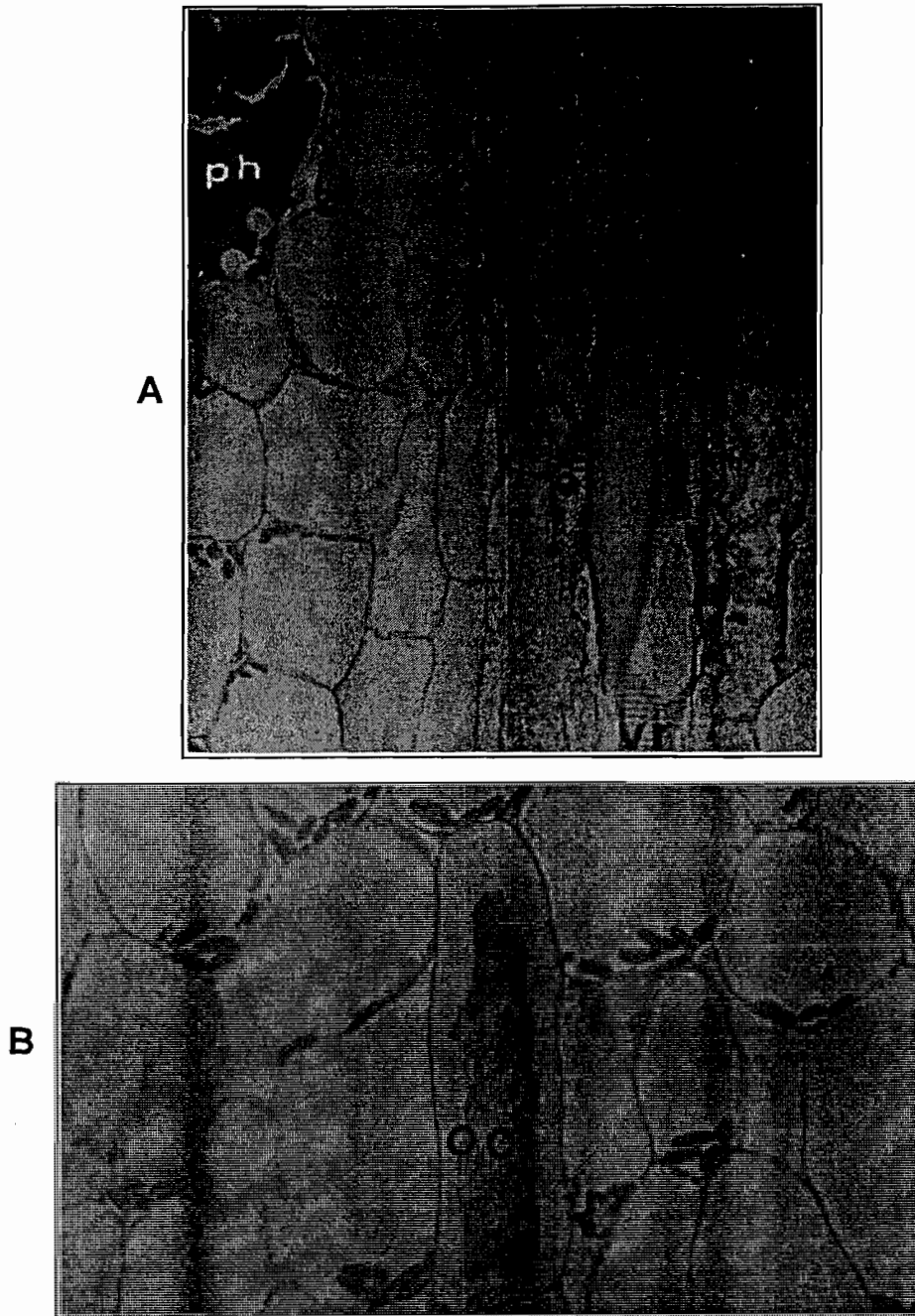
**Figure 6** : The structure of the bulb of a banana plantain cv Horn 1 bud sucker (G x 100).

*Structure du bulbe de rejet du bananier plantain cv Come 1 (G X 100).*

A. Longitudinal section

B. Cross section

Tissue observation reveals the presence of two distinct zones : the central bulb (bc) and the peripheral bulb separated by the Mangin Line (M.). It is mainly made of woody lignous type rays and contains very little starch and almost no phenolic compounds.



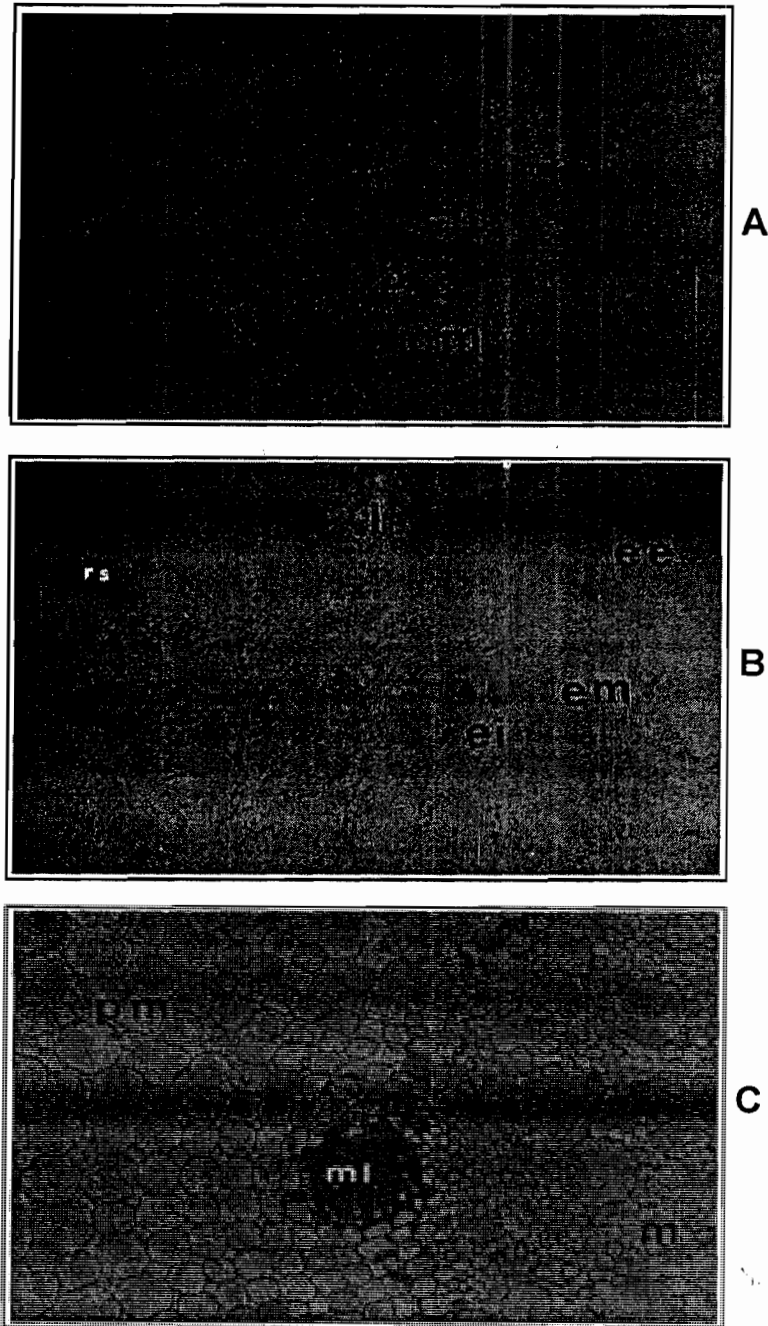
**Figure 7** : A longitudinal section of a bulb of a banana plantain cv Horn 1 sucker.

*Bulbe de rejet de bananier plantain cv Come 1 en coupe longitudinale.*

A- A portion of a bud sucker bulb showing conductive sored vessels of scare type (vr) and natches of phenols (ph) (G x 160).

B- Detail view of a portion of a bulb of a sucker. Cellular inclusions, and raphid shape oxalate cristals (oc).





**Figure 8** : A cross-section view of roots from a banana plantain cv Horn 1 bud sucker.

*Racines de rejet de bananier plantain cv Come 1 en coupe transversale.*

- A- A general view of a root (G x 25). Parenchyma with voids (pm) moella (m).
- B- A general view of a proximal area of a root (advanced stage) (G x 25). Very lacunous structure of the cortex. Progression of meristematic tissue of two secondary roots is throughout the cortex (rs).
- C- Detail distal area of a root (G X 160) showing initial stage differentiation of a secondary root (sr) from pericycle (p) ; endoderm (en) central cylinder (cc) bulk (é) secondary root (rs).

## Histochemical observations

A very abundant starch was differently distributed according to tissue type. As a matter of fact, the backside of the foliar scales was richer than the ventral part. Inside the bulb, the central zone was much richer than the outer part (figure 9 : A, B). The Magin line and the central cylinder contained less starch. A polar localisation of this starch was evident inside all the cells of the organs and a parietal localization of the peroxidase enzymes (figure 10). No protein reserves were revealed by the method used. Protein content was low compared to phenols and starch in all tissue.

All tissues observed were rich in phenolic groups inside cells localized along the vessels or scattered in the different tissues (figure 11 : A). tanins were the most prominents, while the flavonoids and cafeic acids were less represented. At the Mangin line level, no phenolic compounds were apparent. Certain cells contain both phenols and starch (figure 11 : B).

## Characteristics of the biochemical parameters of the bud suckers under hydroponics culture conditions

### *Total phenol levels*

Within the bulb, total phenol contents decrease during the early days (4 days) of culture. Beyond that, total phenols temporary accumulated with an optimum of 260 meq.ac.chl/g MS at 12 days of culture (figure 12 : A). At the scales level, a drop in total phenol content was also

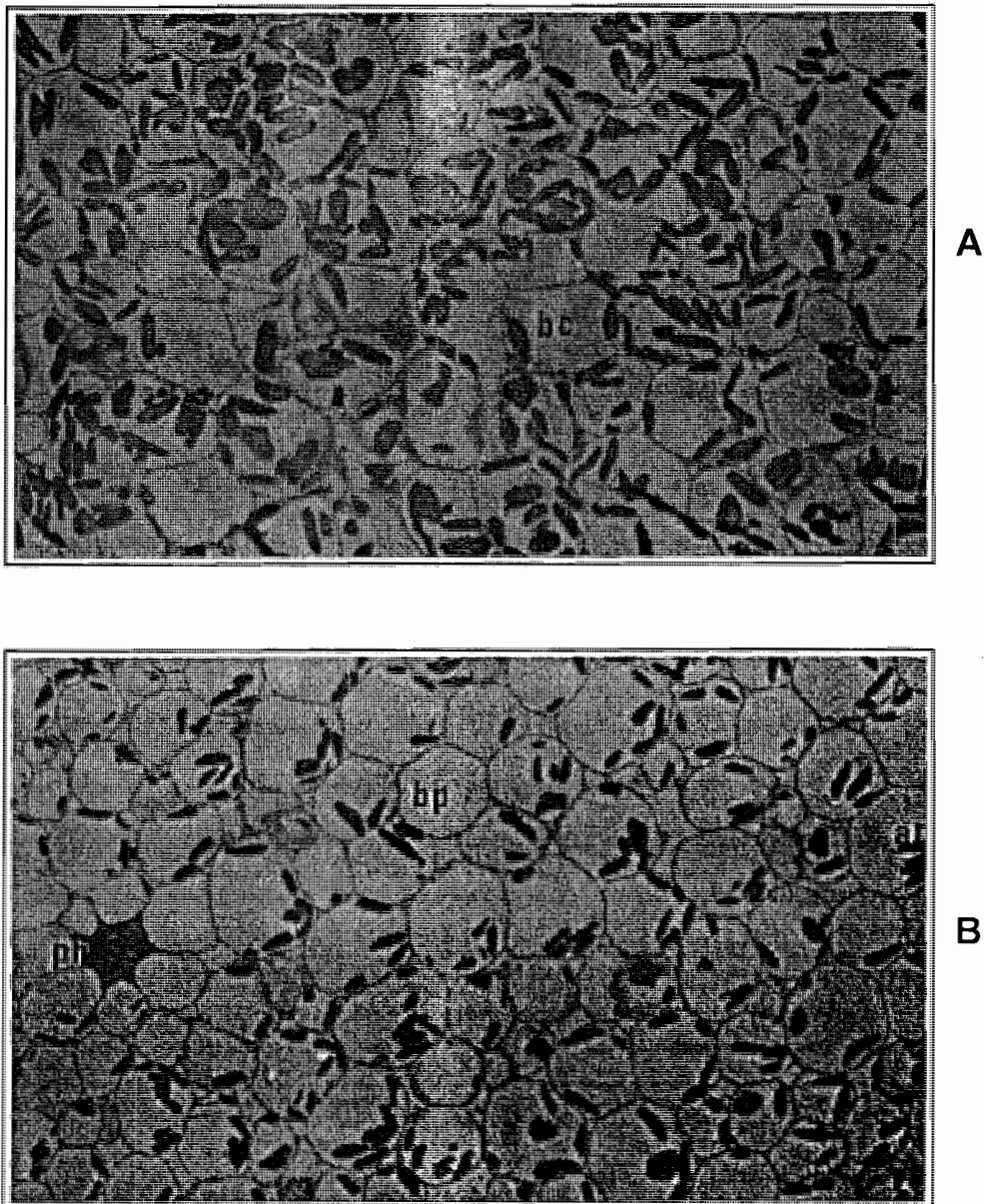
apparent, as in the bulb. However, it continues until the 8<sup>th</sup> day of culture (figure 13 : A). There was no accumulation of phenolic compounds as was the case in the bulb. The amplitude of the variations was very small as compared to that of the bulb. Total phenol contents were higher (three times more) within the bulb (150 meq.ac.chl/g MS) than in the scales (50 meq.ac.chl/g MS) of the suckers under culture.

### *Total protein levels*

Protein levels were lower in the shoots during culture regardless of treatment. Protein levels were higher within the bulb (290 µg/ml) than in the scales (140 µg/ml) of the shoots under culture (figure 12 B).

### *Reducing sugars and sucrose*

Glucose content was three times higher in the bulb (0.06 µmole/ml) than in the foliar parts (0.02 µmole/ml) ; this was not the case for foliar parts whose levels were lower. Glucose levels of the young foliar parts always accumulate rapidly in the bulb as well as in the leaf/scale parts (0.11 µmole/ml) with an optimum of 0.17 µmole/ml) at 8 days, followed by a decrease up to 12 days, then a second accumulation until the end of culture (figure 13 A). Fructose levels were nearly constant during culture period. In the leaf/scale part, fructose levels tend however to decrease gradually, while they tend to increase within the bulb (figure 13 B). Sucrose content of the bulb were always higher than those of the leaf/scale part of the banana shoots (figure 13 : C). Sucrose level was similar to that of glucose.

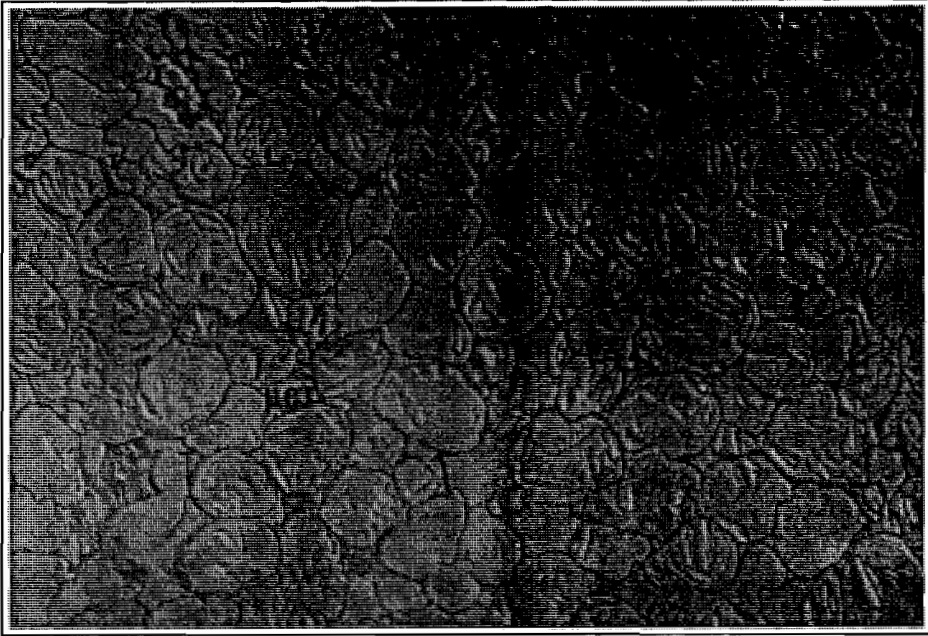


**Figure 9** : A microscopic view showing the presence of starch in the two parts of the bulb of a banana plantain (cv Horn 1) bud sucker (G x 160).

*Observation microscopique mettant en évidence l'amidon dans les deux parties du bulbe de rejet de bananier plantain cv Come 1 (G X 160).*

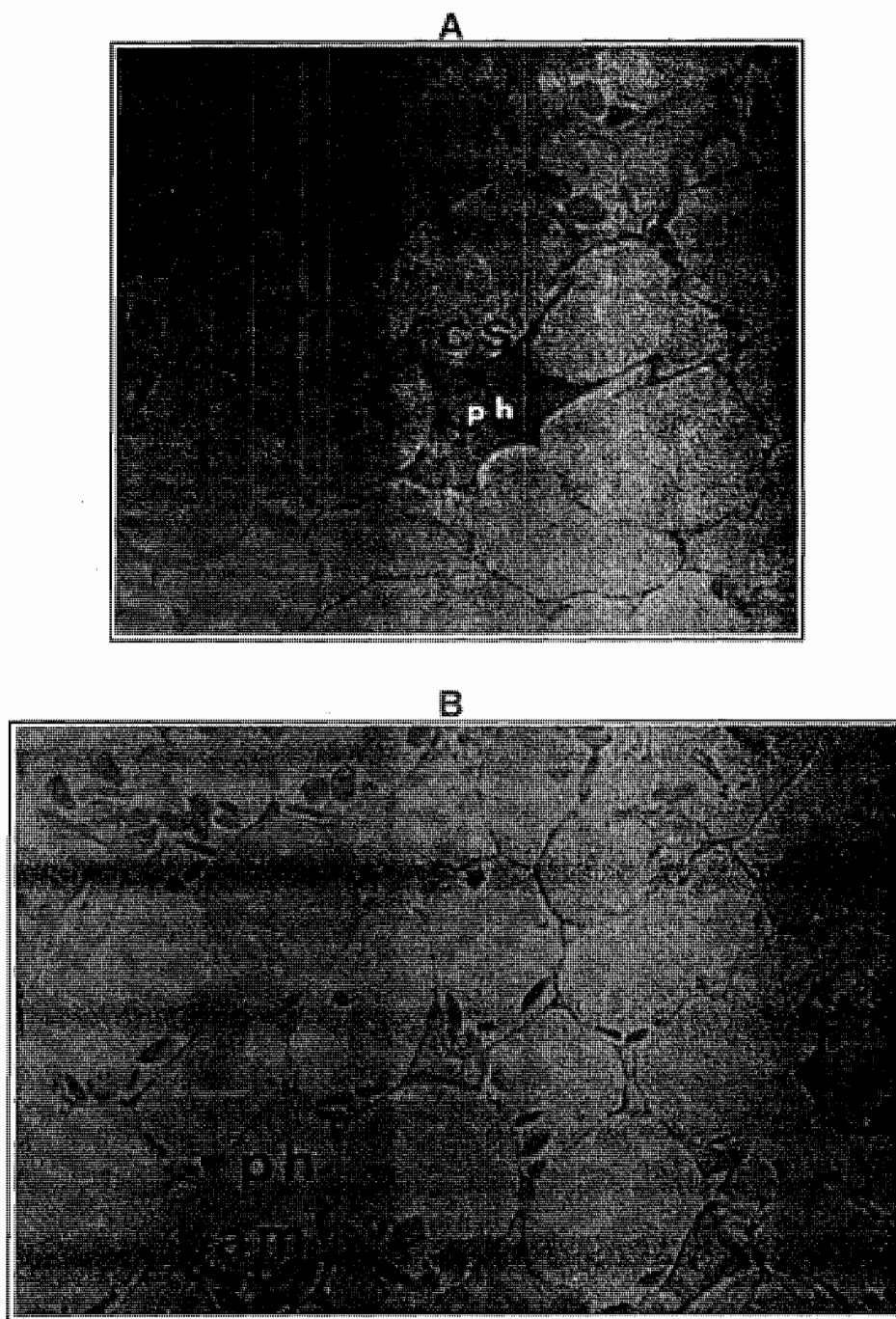
- A. Parenchyma cells of the central bulb (bc) showing a high concentration of starch (am).
- B. Peripheral bulb (bp) containing less starch (am).





**Figure 10 :** A microscopic view (G x 160) of a parietal peroxidase in bulb cells of a banana plantain bud sucker by Diaminobenzidine (DAB).  
Note the parietal localization of the peroxidase (pei).

*Mise en évidence des peroxydases pariétales dans les cellules du bulbe de rejet de bananier plantain cv Come 1 par le diaminobenzidine (DAB) (G X 160).  
Remarquer la localisation pariétale de la peroxydase (pei).*

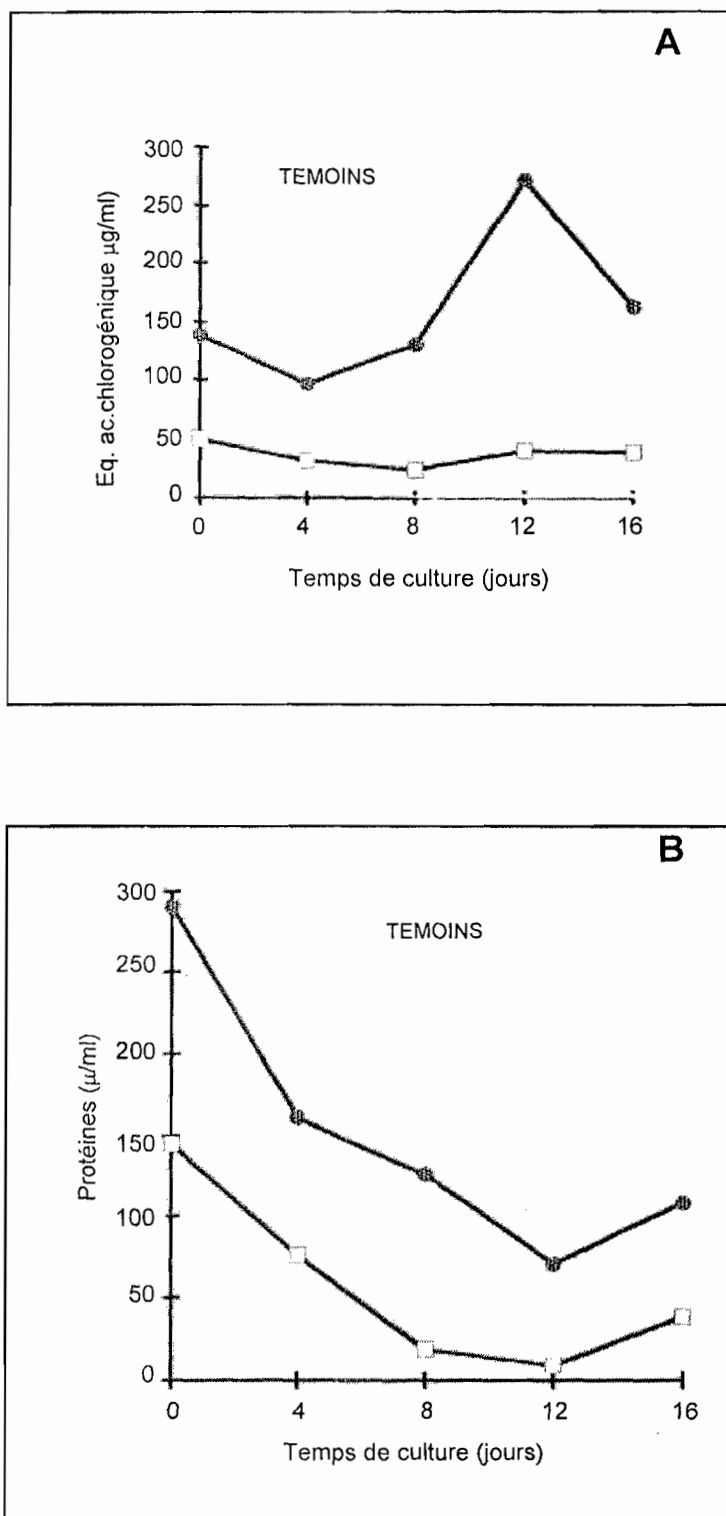


**Figure 11** : Total phenols in the bulb of of banana plantain cv Horn 1 suckers.

*Phénols totaux dans le bulbe de rejet écailles de bananier plantain cv Come 1.*

A. Overall view of a cross section of a bulb (G x 160) showing a high phenols content (pH).  
Secretion cells (cs).

B. Overall view of a cross section of bulb cells (G x 160) containing both phenol (pH) and  
starch (am).

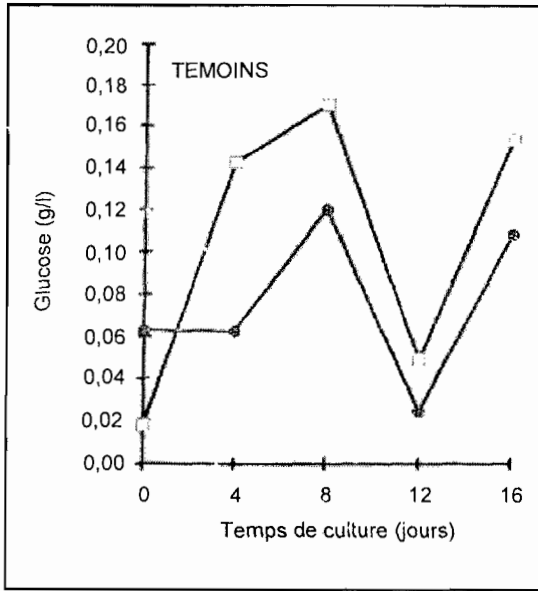


**Figure 12** : Phenol and protein contents with time of a banana plantain (cv Horn 1) bud-sucker.

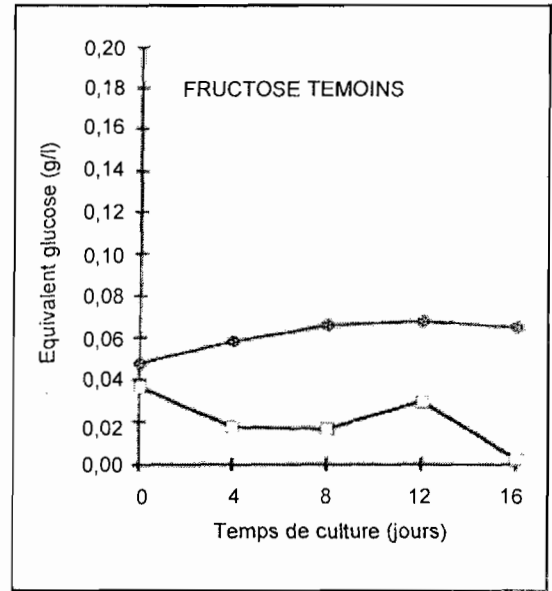
*Evolution dans le temps des teneurs en phénols et protéines de rejets de bananiers plantain cv Corne 1.*

A. Total phenols.  
(Bulb)

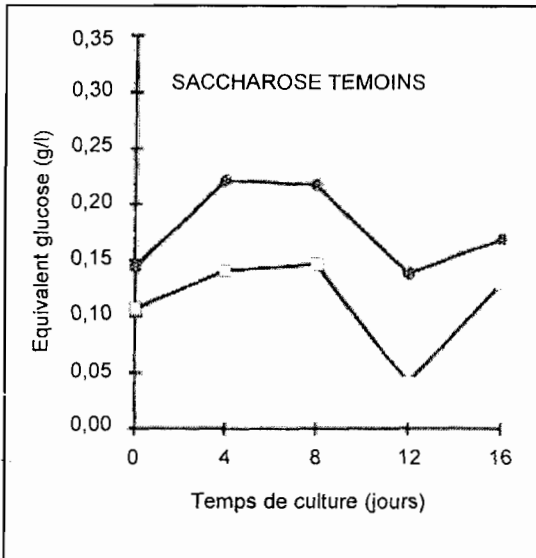
B. Soluble proteins  
(Scales)



A



B



C

**Figure 13** : Reducing and non-reducing sugar contents with time of banana plantain (cv Horn 1) suckers in hydroponic culture medium.

*Evolution dans le temps des teneurs en sucres réducteurs et sucres non réducteurs des rejets de bananiers plantain cv Come 1, en culture hydroponique.*

- A. Glucose content
- B. Fructose content
- C. Sucrose content.

## DISCUSSION

Growth stimulation of type b bud suckers was possible under hydroponic culture conditions. A 1200-fold dilution of the base nutritive solution permitted minimum sucker growth. Suckers were vigorous, with an important early root development and without necrosis. In early experiments, conducted in containers under confined and non agitated medium, the delay in field transfer of young was found to be at least 30 days. Thus under stimulation conditions, in hydroponic culture, this period could be reduced to two weeks. In relation to morphological parameters, studies of the plantain growth characteristics show that suckers underment a strong inhibiting influence from the mother stand. This inhibition, which persists even after weaning *in situ*, can go up to a complete halt of sucker growth during regime development (Robinson and Anderson, 1991). The earliness in the opening of the bud suckers constitutes a determining step in the growth process, which in some respect, readily resulted in an important weight gain, and also, by the development of lanceolate leaves after only few days of culture. In relation to farming practices, Swennen *et al.* (1986) experimented a similar system using 11 banana cultivars *Musa* (AA, AAA, and AAB), of which the Horn banana plantain. Environmental conditions of culture (roots under darkness, longer period of culture, with the medium renewed daily) are different from ours. This system allowed for better conditions for root growth (95 % of healthy roots) with the different banana cultivars and to differentiate two root types (nourishing and pioneer).

Our set up adapted from that of Swennen *et al.* (1986) allows for healthy root development. It came as a solution to the 5 % loss in root number observed by these authors, by insuring to the plant better growth conditions which resulted in a better behaviour towards transplantation in the field, in a two week-period. Results

from our hydroponics culture confirmed the usefulness of such system for the study of the banana plantain growth.

Anatomical studies show that the young foliar parenchyma was typical and characterized by an absence of a palissadic tissue. The different parts of the bud suckers were made of a parenchyma containing very few xylem tissue. Only the young foliar part and old roots were provided with aeriferous lacuna. As far as the cytological structure of suckers were concerned, our results on cv Horn 1 banana plantain were in accordance with descriptions made elsewhere on several banana cultivars (French Grande naine) at more advanced growth stages, even at adults stages in some cases (Kwa, 1993). Do the two distinct zones identified in the bulb played different physiological or metabolic roles ? The difference in starch the accumulation in the central and in the peripheric bulb may help to explain this hypothesis. With regard to the histo-chemical study of the suckers, the abundance of starch in the different tissues, the oval shape of the amyloplasts, as well as the poorness of the tissues in protein, agree with observations made by Gnakry and Kamenan (1994) on several banana plantain cultivars (*Musa sp.*) and the low protein content of the tissues observed with desert bananas (Chandler, 1995). At a biochemical standpoint, our study showed that metabolite levels of the bulb were higher than those of the leaf scale part ; this could be associated with the nature of the bulb. As a matter of fact, the bulb of the banana rhizome is a reserve tissue. High glucid contents in the banana plantain can be linked to important quantities found by Saya (1991) with Cavendish (*Musa AAA*) vitroplant, another juvenile unit of propagation.

## CONCLUSION

Hydroponic culture techniques, applied to our study conditions using a

nutritive solution as supplement, allowed to :

- reduce the delay time in bud dormancy breaking, thus allowing for an early plantation as compared to an awakening in nonstimulating conditions ;

- increase bud survival and insure a minimum growth rate of the suckers.

- analyze the morphological parameters to reveal an important quick weight gain, a development of lanceolate leaves within few days and a circular root development ; roots are in sets of at least 2-3 roots ;

- obtain young vigorous plants ready for transfer to the farm for replantations in a short period of time.

Anatomic studies revealed that the young foliar parenchyma was unique and characterized by an absence of a palisadic tissue. The different parts of the bud suckers were made of a parenchyma containing very few primary woody tissue. Calcium oxalate crystals accumulated as raphids in the two bulb zones. Only young foliar tissues and old roots were provided with aerenchymatous spaces.

Histochemical studies allowed for a qualitative assessment of biochemical parameters such as starch, proteins, and phenols. Phenols (mainly tanins) and

starch were abundant as opposed to protein, which were poorly represented.

Biochemical analysis show an accumulation of total phenolic compounds capable of reaching a maximum value in the 12<sup>th</sup> day, for proteins and sugar, with the exception of a drop in fructose content that can reach its minimum value the 12<sup>th</sup> day. The evolution of phenols, proteins and sugars within the bulb, with the exception of fructose, were parallel.

According to the hydroponic culture system, it may be envisioned to simplify bud sucker preparation by modifying the dormancy breaking step in the laboratory. As a matter of fact, weaned suckers may first be subjected to relatively high temperatures (<40 °C), before being transplanted in the field, where they could be irrigated or sprayed with the base medium in order to stimulate growth. These conditions open the possibility of putting at the disposal of the farmer, a more practical know-how in banana farming.

## ACKNOWLEDGEMENT

We wish to express our recognition to Pr. N. Michaux-Ferriere from the «Laboratoire de Cytologie» CIRAD-BIOTROP, Montpellier and also to Dr. M.A. D'almeida of the «GERME d'Abidjan».

## REFERENCES

- BASTIDE (S.), 1987. Evolution des composés phénoliques des fèves de Cacao durant leur développement au cours de la maturation du fruit de *Theobroma cacao* L. Thèse U. S. T. L. Montpellier 147 p.
- BIERBERACH FORERO (C.Y.) et (J.V.) ESCALANT. 1996. *Embriogenesis somatica* en cuatro cultivares de *Musa* sp. in CATIE. Unidad de biotecnología. Informe bianual 1994-1995, n.p.
- BRADFORD (M.M.), 1976. A rapid and sensitive method for the quantitation of microgram quantities of proteins utilizing the principle of protein dye binding. *An. Biochem.* 72, p. 248 - 254.
- CHANDLER (S.), 1995. The nutritional value of bananas. "Bananas and plantains, Ed. GOWEN, Chapman and Hall London p. 468- 480".
- CORDEIRO (Z. J. M.) , (W.) DOS SANTOS SOARES FILHO. 1991. Propagação da bananeira por fracionamento do rizoma. *Embrapa / CNPMF - Banana em foca* 45, p. 1-2

- GNAKRI (D.) et (A.) KAMENAN. 1994. Caractéristiques physicochimiques de l'amidon de plantain (*Musa* AAB). *Agronomie Africaine* vol.6 n° 1, 1994 p. 20-25.
- KWA (M.), 1993. Architecture, morphogenèse et anatomie de quelques cultivars de bananiers. *Thèse de Doctorat. Université de Montpellier II*. p. 22-224; 244-250.
- MARIGO (G.), 1973. Sur une méthode de fractionnement et d'estimation des composés phénoliques chez les végétaux. *Analysis* 2, n° 2 fev. 1973, p. 106-108.
- ROBINSON (J. C.) and (T.) ANDERSON, 1991. Growth of banana ratoon suckers in relation to mother plant. *Citrus Subtrop Fruits Res. Inst Information Bulletin Sept. 1991* n° 229 p.
- SAYA (R.A.), 1991. Amélioration de la croissance et du développement de pousses feuillées de bananier (*Musa acuminata* AAA) : Passage du stade hétérotrophe *in vitro* au stade autotrophe *ex vitro*. *Thèse de doctorat de l'Université d'Aix-Marseille*.
- SWENNEN (R.), (E) DE LANGHE (J.) JANSSEN (D.) et DECOENE. 1986. Study of root development of some *Musa* cultivars in hydroponics. *Fruits* 41, 9, p. 515-524
- TURQUIN (L.), 1989. Etude des potentialités rhizogènes du bananier plantain, cultivar Corne 1 du sous-groupe parthénocarpique, AAB de *Musa*, L. (*Musa acuminata* x *Musa balbisiana*) : Influence de quelques substances de croissance. *D.E.A d'Ecologie Tropicale (Option : physiologie végétale). Université Nationale de Côte d'Ivoire*.