

# Pineapple News

Newsletter of the Pineapple Working Group, International Society for Horticultural Science  
Issue No. 19, June, 2012

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## Pineapple Working Group News

### From the Editor

Dear Colleagues:

November of 2012 will be the 20<sup>th</sup> anniversary of the 1<sup>st</sup> International Pineapple Symposium (IPS) and there is good reason to celebrate this anniversary. The idea to organize such a symposium was first proposed by Dr. J.J. Lacoëuilhe of what was then CIRAD/IRFA at the XXII<sup>nd</sup> Horticultural Congress held in Davis, California in 1990 (Lacoëuilhe, J.J., 1990, *Pineapple*, *Chronica Horticulturae* 30, p. 59). As a justification for holding such a symposium, Dr. Lacoëuilhe stated that:

- Few people in the world are involved in pineapple but they do not communicate or exchange information.
- Research is often initiated by local circumstances and problems, which need quick solutions.
- Publications are rare and of irregular interest compared to other crops of the same economic importance.

Dr. Lacoëuilhe proposed to begin to solve this problem by publishing a list of people involved with pineapple research and production and invited people interested in this initiative to contact him.

Dr. Kenneth Rohrbach and I did not attend that IHC meeting and so missed that important announcement. However, we also had occasional conversations about the limited communication among the community of pineapple researchers and the difficulty that growers in developing countries have in getting access to information on modern pineapple cultural practices. As a result of these discussions we independently began to discuss the idea of hosting an international pineapple symposium in Hawaii. We sent out an inquiry of interest, also in 1990, and the response was positive. That first symposium was held November 2-6, 1992.

I have no recollection of why we held the symposium under the auspices of the International Society for Horticulture Science (ISHS), but give the credit for that recommendation to Pierre Martin Prevel, a senior researcher at CIRAD in France. Most importantly, it assured that our proceedings would be available in a well-recognized publication that has wide distribution. I am certain that it was Dr. Martin Prevel who at the end of the 1<sup>st</sup> IPS encouraged the establishment of a Pineapple Working Group (PWG) under Section Tropical and Subtropical Crops of the ISHS. I agreed to serve as chairman of the PWG, a position I held for 10 years, and stated my intent to establish a pineapple newsletter in an attempt to improve the exchange of information among pineapple growers and researchers.

The success of that initial effort is indicated by the fact that the 7<sup>th</sup> IPS was held in 2010, seven volumes of *Acta Horticulturae* document the contributions of the PWG to the pineapple literature, and the trend in pages in *Pineapple News* since the first issue in 1994 has been consistently upward (Fig. 1). Thanks to the strong support of the international pineapple community, we are a success! Readers of *Pineapple News* also know that planning for our 8<sup>th</sup> IPS has been underway in Australia since that country was selected as the site of the 8<sup>th</sup> symposium at the PWG meeting in Johor, Malaysia in 2010. Congratulations to all of us and I hope that many of us will meet again at the 8<sup>th</sup> IPS in Brisbane.

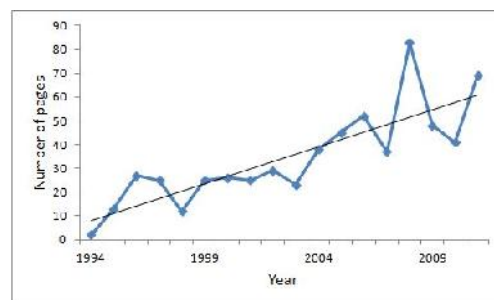


Fig 1. Pages in *Pineapple News* by year with trend line.

### Assuring continuation of Pineapple News

Age and family health issues have delayed the publication of this issue of the newsletter and caused me to realize that there is need to make provision for an orderly transition in the responsibilities of editorship of *Pineapple News*. Several years ago Brent Sipes, and soon thereafter Alain Soler, offered to assist with or take on the responsibility of editing *Pineapple News*. Recently Garth Sanewski made the same offer. I recently spoke to Brent about passing on some of the responsibility for production of the newsletter to him. He encouraged me to continue and said he would help as needed and be prepared to take over any time I decided to retire from the job. I expect Alain, Garth and probably others will also offer their assistance. So I am reassured that there are people ready and willing to see that the newsletter continues if I/when I decide the time has come for me to step aside.

### Are pineapple growers missing an opportunity?

I must confess that the following is written from the perspective of a long-time student of pineapple physiology and culture who has absolutely no background in marketing. That said I begin by relating a local story of a so-far minor success that could grow to become a significant money maker.

In 2010 I helped write the technical description for the plant patent application for 'Franklynn', a new pineapple hybrid selected from seedlings produced from a natural cross between a 'Smooth Cayenne' clone known as "Dry Sweet" and "Hilo White", which was most likely 'Monteliro' (G. Coppens d'Eeckenbrugge, personal communication). Dry Sweet was the seed parent. 'Franklynn' was developed slowly as the owners thought about how to capitalize on what they believed was a unique pineapple fruit with an unappealing appearance but an exceptionally sweet and flavorful flesh. The fruit is pale cream in color with a mild pina colada taste but the °Brix consistently ranged from 22 to 26. Acidity was more typical of the 'Smooth Cayenne' parent but the Brix:acid ratio was atypically high for a 'Smooth Cayenne' offspring. Recently it was found that 'Franklynn' fruits store well at normal refrigerator temperatures and are firm enough that they likely would ship well too.

Once the population had reached several hundred plants the owners took fresh-cut fruit to a local gourmet farmers market. Trays of cut fruit priced at \$5.00 a pound quickly sold out. As more fruits became available, whole fruits priced at \$5.00 per pound also sold well. Some shoppers purchased three and four fruits, a total sale of \$40 to \$60. In one instance, fresh cut fruit priced at \$10.00 per pound also sold well. The owners are currently looking for land on which to expand the production of their new hybrid and have enlisted the assistance of a retired pineapple farm manager.

As the supply of 'Franklynn' fruits increase, the challenge will be to find markets that are willing to pay the premium prices currently being obtained for the fruit. Could prices being paid be due to the fact that Hawaii is a small tourist market where visitors are willing to spend more for a unique taste or experience? However, when scanning the array of fresh fruit offered for sale in Hawaii and on U.S. mainland markets, I see odd looking fruits, the offspring of crosses among plums, peaches and cherries, which sell at what seem like exorbitant prices each year they come in season. Based on the numbers of cultivars of peaches, apples and other fruits in the supermarket with pricing differentials based on uniqueness, flavor, °Brix, etc., it would seem that there are opportunities for pineapple growers to compete in the exotic fruit trade if cultivars with exceptional flavor or other appealing characteristics are developed.

There are at least three real-world examples where small-scale growers produce non-standard (not 'MD-2' or 'Smooth Cayenne') pineapple cultivars for local or export fresh fruit markets. Growers in Australia typically grow two to three different cultivars (Sanewski, personal communication) and farmeres in Malaysia and Taiwan produce locally developed cultivars that are not grown elsewhere. The extent to which those cultivars are marketed by cultivar rather than company name is not known, but Taiwan growers successfully export Tainung 17 to Japan in direct competition with 'MD-2' imported from the Philippines. Brazil and the French in Reunion or Martinique, or perhaps both, have cultivars that presumably have distinctinve characteristics that would allow them to be marketed by cultivar name in competition with other fruits sold at premium prices. Perhaps the marketing of premium fresh fruits is a niche for the small farm rather than the large plantation, but the Hawaii experience related above and that of Taiwan would suggest that an opportunity exists to differentiate pineapple fruits by cultivar, perhaps at a premium price, which could increase total income or offset the impact of low yields on total return. A caveat is that growers considering importing cultivars from different regions might want to proceed with some caution because cultivar quality or performance may not be uniform across environments.

### Water relations of pineapple

In a soon-to-be published article in Experimental Agriculture (Carr, MKV. 2012. The water relations and irrigation requirements of pineapple (*Ananas comosus* var. *comosus*): A Review. Experimental Agriculture 38 (4): pages not yet assigned.), the author summarizes the existing research on the water relations and irrigation requirements of pineapple. The objective was to link fundamental studies on crop physiology to crop irrigation practices. The author reviews crop development, plant water relations, water requirements, and the effects of water on productivity and irrigation systems. The impact of the crassulacean acid metabolism (CAM) carbon assimilation pathway in pineapple on water-use efficiency is discussed. Also mentioned is the role of leaf water storage and aerial roots in absorbing water and the water economy of the plant. Though not a surprise to those

with knowledge of the subject, the author uses the word “surprising” more than once in the paper summary when commenting on published work on pineapple water relations and management. For example:

There are surprisingly few published reports of field measurements of crop water use and water productivity of pineapple. Two reports show evapotranspiration only occurring during the daytime. There is more uncertainty about the actual water use of pineapple, the value of crop coefficient (Kc) and relative rates of water loss (transpiration) and carbon gain (net photosynthesis), during the daytime and at night, under different water regimes. This is surprising given the amount of fundamental research reported on photosynthesis of CAM plants in general. .... There is a lack of reliable published data quantifying where irrigation of pineapple is likely to be worthwhile, how it is best practised and the benefits that can be obtained. This is remarkable considering the importance of pineapple as an internationally traded commodity.

Typically, the kind of research the author writes about is supported by public funds and such funds are not available in many areas where pineapple is grown. The long-term nature of the pineapple crop also makes such research prohibitively expensive for the average farmer. Until more research fills the knowledge gap, growers in many areas where plant growth is interrupted by water stress will have to make educated guesses about the water requirements of their crop.

The above author, an expert in soil/plant/water relations and irrigation agronomy with considerable experience in tropical agriculture, is also the author of the newly published book *Advances in irrigation agronomy: Plantation crops*. The book, which was published by Cambridge Univ. Press, includes the following chapters: Introduction, Banana, Cocoa, Coconut, Coffee, Oil Palm, Rubber, Sisal, Sugarcane, Tea and Synthesis.

## **Tropical Fruits Network**

In December of 2011 The International Tropical Fruits Network (TFNet) announced the return of its newsletter, Tropical Fruit Net ([www.itfnet.org](http://www.itfnet.org)), after a brief hiatus. It was announced that beginning in 2012 Tropical Fruit Net will be produced every other month to update readers with the latest news, projects, features, and events.

TFNet also welcomes contributions from individuals, especially from TFNet member countries, associate and ordinary members. Contributions can include articles on production, marketing and consumption of tropical fruits. Tropical Fruit Net is distributed to more than 1,500 recipients from more than 30 countries. The articles will also be predominantly featured in our website. Visit [www.itfnet.org](http://www.itfnet.org) for more information.

## **Proceedings of the 7th International Pineapple Symposium**

In case you missed the announcement, the proceedings of the 7<sup>th</sup> IPS were published in September of 2011. The abstracts of all papers presented at the 7<sup>th</sup> IPS are available as an addendum to Pineapple News No. 17 at <http://www.ishs-horticulture.org/workinggroups/pineapple/> and information about the proceedings, Acta Horticulturae volume 902, can be found at <http://www.actahort.org/books/902/index.htm>.

## News from Australia

### 8<sup>th</sup> International Pineapple Symposium

The 8<sup>th</sup> International Pineapple Symposium will be held from 17 to 22 August in Brisbane, Australia in 2014 as part of the International Horticultural Congress (IHC 2014). IHC 2014 will include many symposia covering a range of tropical and subtropical horticultural industries.

Information on the IHC2014, including accommodation and venue information, can be found at the website, <http://www.ihc2014.org/icmsa/index.html>. The **8th International Pineapple Symposium** is No. 32 under the **Tropical Fruits and Vegetables** sub-theme.

The pineapple symposium program will run from Monday 18 August to Friday 22 August (registration on 17 August or early on 18 August) and will include presentations, posters and a technical tour to view pineapple farms and research in South-East Queensland. The program, which will include invited speakers, will cover the latest developments in pineapple research and development from around the world. The format will be similar to those of previous pineapple symposia but will offer concurrent IHC sessions in a range of other symposia for those with broader horticultural interests.

Key dates for all symposia including the pineapple symposium can be found on the IHC2014 home page.

Key dates for presenters are;

Open Abstracts	1 April 2013
Close Abstracts	1 Sept 2013
Notification to Authors	17 Nov 2013
Presenter Registration	17 Dec 2013

Key dates for attendees are;

Attendee Registration	30 Sept, 2013
Early Bird Closes	17 Feb, 2014

For further queries regarding the 8<sup>th</sup> International Pineapple Symposium contact G. Sanewski ([garth.sanewski@daff.qld.gov.au](mailto:garth.sanewski@daff.qld.gov.au)).

On behalf of the 8<sup>th</sup> International Pineapple Symposium, ISHS, IHC 2014 and sponsors, the organising committee invites you to attend the 8<sup>th</sup> International Pineapple Symposium and IHC2014 in Brisbane in August, 2014.

Organising Committee  
8<sup>th</sup> International Pineapple Symposium

## News from Benin

### Introduction to Pineapple Industry in Benin

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Key words: Ananas comosus, Pineapple Cultivation, Benin, Hawaii, Farming System.

#### I. History of Pineapple in Benin

Benin (formerly Dahomey) is a coastal country in West Africa, with a population of eight million inhabitants and an area of 114.763 km<sup>2</sup>. The economy of the country depends essentially on agriculture and trade. Indeed, agriculture contributes 36.3% to the GDP, whereas industry represents 14.3% and trade and services 49.4%. The presence of pineapple in Benin goes back to the period of intensification of slavery in the 18<sup>th</sup> century under King Agadja, approximately corresponding to the introduction of the first suckers of pineapple in Hawaii. Moreover, pineapple became the emblem of King Agonglo, King of Abomey (1787-1797). He made his mark on history by his social reforms and the defence of the kingdom of Abomey, by saying: “*The lightning strikes palm oil tree, but pineapple escapes*”, hence his name Agonglo and the allusion to his capability to escape and avoid traps and difficulties during his reign. It is interesting that that fact was reproduced by the famous Martinique poet Aimé Césaire in the poem: “*The Tragedy of King Christophe*” in 1963. The first introduction of pineapple to Benin might have been by Portuguese who most influenced the culture in the Ouidah region, in the south of Benin. At the beginning, it was cultivated traditionally and in association with food crops for family consumption and subsistence. Gradually, pineapple cultivation gained in importance because statistics attest that the first exports of pineapple from Benin to neighbour countries occurred in 1963<sup>1</sup>. The pioneer was the *the Société Dahoméenne des Fruits et Légumes* (SODAF). It was a private company and after about two decades of State Farming System, controlled by SONAFEL (*Société Nationale des Fruits et Légumes*), the farms were bought back in 1990 by Fruitex-Benin. Mass production for local market and for export started in 1972. The production zones are situated in the Departments of Atlantique/Littoral, Ouémé/Plateau, Mono/Couffo and Zou/Collines (Fig. 1). The main producing communes of pineapple in Benin are: Abomey-Calavi (42%, top in 2006), Zê (31%) and Allada (17%). In general farms are of small size, from 1 to 2 ha. Some producers have farms of up to 150 ha.



Fig 1. Benin with pineapple production areas circled.

#### II. Beninese Pineapple industry

**1. Pineapple in Benin's Economy.** The Beninese pineapple industry has experienced significant growth from virtual nonexistence in the 1980's to 220,800 tonnes in 2010 when it ranked first in horticulture crops (Fig. 2) and eighth in terms of export value (Fig. 3). German Technical Cooperation Agency (GTZ) estimated the contribution of the pineapple sector to GDP at 13 billion FCFA in 2006, 1.2% of global GDP and 4.3% of agricultural GDP (in comparison 2% and 7.4% for cashew)

**2. Producers.** There are three categories of producers and associated production systems for pineapple in Benin.

1. Farmers producing pineapple in near optimal conditions with an intensive cultivation system including irrigation to compensate water deficit in the area. The production is intended for national, regional and international markets.
2. Farmers using semi-intensive conditions with structural handicaps regarding the areas under cultivation. They don't reach the optimal level of intensification and the production is for the national market.
3. Farmers with extensive production systems who plant small areas and the production is generally for self-consumption.

<sup>1</sup> Dohou Videgnon B., *Programme National de Développement de la Filière Ananas*.

In order to guarantee consumers' security the European Union defined quality standards for exported pineapple are adhered to. Therefore the product is followed from the farm to the consumers table, ensuring "traceability". Most of the small producers are not able to guarantee quality pineapple. Accordingly, pineapple for export is supplied only by the few modern farmers. Smallholders represent the whole group of small-scale and family producers, they are not tied to an arrangement with a company, and their number fluctuates with pineapple

production. Their production is absorbed by the local market and by processors and exporters who turn to them whenever they need to increase their production volumes. Sometimes, they practise handmade (artisanal) pineapple processing<sup>2</sup>. Frequently but not systematically, they find the export market closed due to the low quality of their product. Moreover, when offered access to this market the price received is often too low and the payment cycle too long.

It is clear that there is an absence of a functioning and regularized model that could create an environment of trust through transparency and price information. It is also important to note that Benin's pineapple sector is particularly well suited for the small-scale farmer. The initial investment is minimal; it requires primarily labour and farm tools. Suckers<sup>3</sup> are also readily available on other farms and can be purchased throughout the year. Finally, the proximity of pineapple-growing regions to urban centres facilitates access to the necessary agrochemicals. Even if they dominate pineapple production and play important social and economic roles, smallholders face many problems and often have limited access to inputs, mechanical equipment, and training. "We do everything by hand"<sup>4</sup>, complained one smallholder during the interview. Small pineapple producers, mainly on the Allada plateau, are gathered in Economic Interest Groups (IEG). But these producers organizations are less viable because of internal conflicts. The regional and national professional organizations Fédération Nationale des Organisations des Professionnelles de l'Ananas du Bénin (FENOPAB), Association des Producteurs des Fruits au Bénin (APFB), Union des Producteurs du Sud- Bénin (UPS-Bénin), Réseau des Producteurs d'Ananas du Bénin (RePAB) and Comité paysan de Gestion des Exportations d'Ananas (COGEX-ANA) provide some support to Benin producers.

**3. Varieties and Cultivation.** Two cultivars of pineapple are produced in Benin: 'Smooth Cayenne' (known locally as "Sweet Cayenne") and Pérola (known locally as Abacaxi, Pain de sucre and sugar loaf). The cultivation cycle lasts 15 to 24 months according to the production zone. Pineapple cultivation is divided into the following stages: soil preparation, suckers sorting, seeding, manure or fertilizer input, hormone input and ethephon

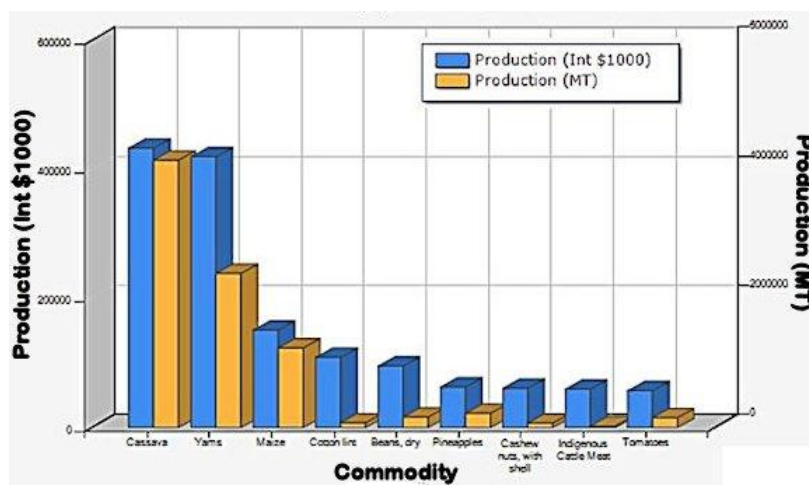


Fig. 2. Benin's top commodities by value in 2010 (Source: FAOSTAT, 2012.).

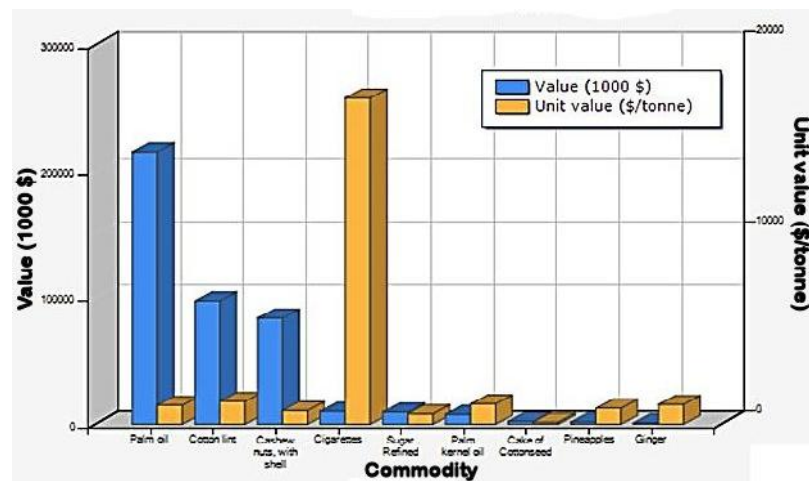


Fig 3. Benin's top exports in 2010 (Source FAOSTAT, 2012).

Fédération Nationale des Organisations des Professionnelles de l'Ananas du Bénin (FENOPAB), Association des Producteurs des Fruits au Bénin (APFB), Union des Producteurs du Sud- Bénin (UPS-Bénin), Réseau des Producteurs d'Ananas du Bénin (RePAB) and Comité paysan de Gestion des Exportations d'Ananas (COGEX-ANA) provide some support to Benin producers.

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<sup>2</sup> Interview with a smallholder

<sup>3</sup> A secondary shoot produced from the base or roots of a woody plant that gives rise to a new plant.

<sup>4</sup> Interview with a smallholder



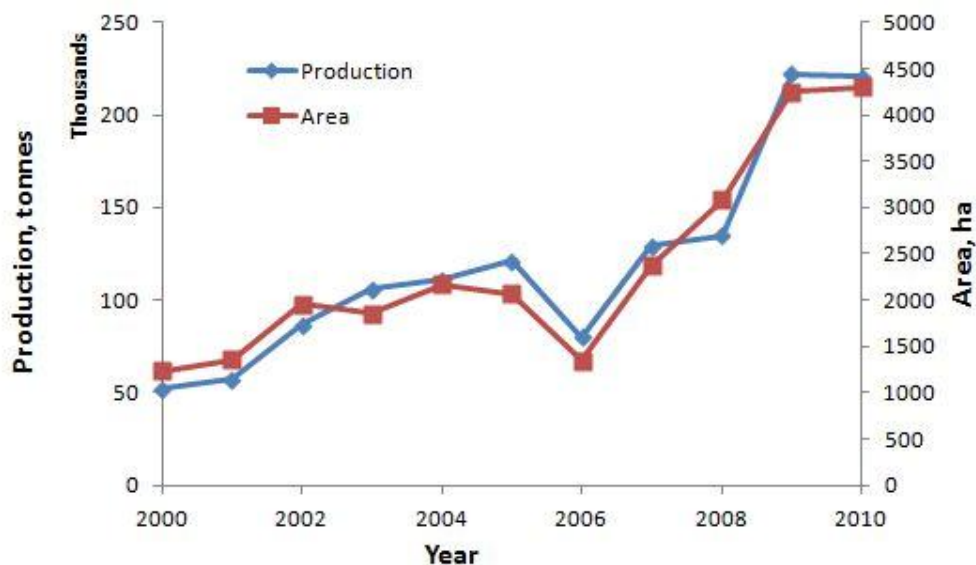


Figure 2. Pineapple production and area planted to pineapples from 2000 through 2010 (Source: FAOSTAT, 2012).

Table 1 – World’s top 20 pineapple producers including Benin.

Rank	Area	Production (Int \$1000)	Production (MT)	Flag
1	Philippines	618330	2169230	
2	Brazil	604306	2120030	
3	Costa Rica	563467	1976760	
4	Thailand	548616	1924660	
5	China	433005	1519072	Im
6	India	404879	1420400	
7	Indonesia	396322	1390380	
8	Nigeria	299868	1052000	Im
9	Mexico	200029	701746	
10	Viet Nam	136023	477200	Im
11	Malaysia	118599	416070	
12	Colombia	113451	398010	
13	Venezuela	105866	371400	Im
14	Peru	88525	310566	
15	Kenya	77598	272231	
16	Bangladesh	66841	234493	
17	Benin	62938	220800	Im
18	Guatemala	59232	207800	Im
19	Democratic Rep. of Congo	57165	200548	
20	United States of America	48457	170000	F

Source: FAO, 2012, F-FAO estimate, Im-FAO data based on imputation methodology, P-Provisional official data.

### III. Valorisation of the Production: Commercialization, Processing and consumption

**1. Primary Pineapple Commercialization.** Traders, essentially women, buy fruits on farms or production zones markets. Then they transfer fruits from production zones to consumption centres. Measurement standards are pile and the *bachée*, which is a filled back of a pick-up (car) sheeted by a tarpaulin. The price is mostly fixed by traders because of the perishable nature of pineapple and the incapability of growers to stock their products. The main transport mean remains sheeted vehicle. Part of Beninese fresh pineapple is exported to European Union based on contract between potential buyers and the Association of Producers. Indeed, the quality of Beninese

pineapple is very appreciated, hence an important export potential. Only the *Cayenne lisse* variety is exported to European Union. *Pain de sucre* fruits are only sold in the local market. The European Union (mainly France and Belgium), and Switzerland represent the main destination for Beninese pineapple exports (Table 2). Also, it is important to note that a certain number of small producers and processors have targeted the burgeoning and promising segment of organic pineapple and Fair Trade. This strategy enables them to sustain exports towards Europe. On the other hand, 74% of Beninese pineapple is exported to neighbour countries, essentially to Nigeria (62%). But significantly lower quantities are exported to Burkina-Faso and Niger (10% in total).

Table 2 - Evolution of Beninese exports (2001-2009; Source : FAOSTAT, 2012 – A. Gnimadi).

	2001	2002	2003	2004	2005	2006	2007	2008	2009
Quantity (tonnes)	657	964	895	1 273	1 117	1 336	1 876	1 854	2 133
Value (1000 \$)			77	89		2 255	2 159	1 833	1 798
Unit value (\$/tonne)						1 688	1 151	989	843

From the description above, it is possible to deduct that Beninese exports are still modest and the country is not constant on export markets, so Benin is a very small actor far from the main world suppliers. This is mainly due to the fact that there is no major operator in Beninese production chain and also the low level of infrastructure, very important support for the competitiveness of quality fruit export. Some experiments with organic pineapple cultivation are carried out in Benin in cooperation with foreign partners like Costa Rica and Switzerland. Moreover, certain small producers and processors have targeted this segment as well as the Fair Trade.

**2. Pineapple processing.** Processing units mushroomed in 1994 following the devaluation of currency Franc CFA. Obtained products include: juice (fresh or pasteurised), syrup, jam and dried pineapple. Fresh juice processors are of a large number and are concentrated in villages and towns. Several processors (more than 100) produce pasteurised juice, and most of them are based in Cotonou (the biggest city) and Porto-Novo (the Capital City). There are also several semi-industrial and industrial factories. Here are the main constraints in pineapple processing sub-sector: high cost of packing (essentially imported), difficulties in raw material supply, and questionable quality of the juice. Dried pineapple processing is not so developed and is produced by only 2 units in Abomey and Allada. As raw material, they use rejected fruit (not suitable for export) after sorting. Processors participate to national and regional workshops in order to promote Beninese products.

**3. Distribution, consumption of pineapple and its by-products.** Pineapple by-products that are distributed include: juice (fresh or pasteurised), syrup, jam and dried pineapple. But fresh pineapple represents by far the most sold product, followed by fresh and pasteurised juice. Fresh juice is consumed locally and is dominated by informal sector. The other products are rarely bought at national and regional level. There is a real national potential market for pineapple and its derived products in Benin. Pineapple is consumed in diverse forms: fresh fruit or beverage. Fresh pineapple is sold alongside roads and on markets. Itinerant vendors also exist. An important part of Beninese pineapple is exported to neighbour countries like Togo, Niger and Nigeria. Processed products are distributed in restaurants and supermarkets of major urban centres. However, pure fresh pineapple juice and dried pineapple are exported to France by CSFT (Centre de Séchage des Fruits Tropicaux – Centre for Drying Tropical Fruits, in Allada and Abomey) within the Fair Trade network. These products are used in blends (for flavour), yoghourts, biscuits, jam, specific dishes and desserts. The potential of local and extern market of pineapple is still relatively unknown, therefore under-exploited. The attempt to export Beninese pasteurised juice failed: it is uncompetitive because of production costs. Products that compete with pineapple are other fruits, particularly orange and mango. Local demand of pineapple falls when these fruits come out. On the other hand, this demand is the highest during the Lent. Competing products for juices include local processed sweet beverages like Coca-Cola, Fanta, Fizzi, Sprite (produced local breweries Societe Beninoise de Brasserie - SOBEBRA). Other types of sweet beverages are imported from the sub-region (Nigeria, Togo, Ivory Coast, etc.) and from Europe.

#### IV. Pineapple Sector Potential in Benin

There are very suitable climatic and soil conditions for pineapple cultivation in eight of the 12 of Benin's Departments: Atlantique, Littoral, Ouémé, Plateau, Mono, Couffo, Zou and Collines. The number of households involved in the pineapple industry is estimated at more than 10,000 in 2002 (ABePEC, 2009). There is a significant potential for fruit and pineapple production and export in Benin. Beninese pineapple production has an average increase of 13% a year. However, this is in contrast with the weak position on value export markets. The local potential market for fresh pineapple consumption is estimated at about a quarter of the production. The processing market represents only a tiny part of the total production (Table 3), despite the potential and the proven success of processed products (juices, dried pineapple, syrups, marmalades and jams) on local, regional and international markets. The sub-sector of processing comprises individual actors, micro, small and medium-sized enterprises; their access to international markets still remains limited.

Table 3. Repartition of pineapple by destinations (Source : Projet d'Appui au Programme National de Développement de la Filière Ananas).

N°	Destination	Quantity (t)	%
1	Fresh consumption at local level	28,800	24
2	Processing by local plants	2,400	2
3	Export towards Nigeria	74,400	62
4	Export towards Burkina-Faso	7,200	6
5	Export towards Niger	4,800	4
5	Exportation towards European Union	2,400	

At the international level, the value of the World pineapple imports is estimated at more than 7 million tonnes a year, of which 20% is fresh fruit and 80% represents processed pineapple. The total volume of EU imports is increasing steadily (more than 800,000 tonnes in 2009). Processed pineapple in the world trade is dominated by Asian countries, which are far from the major markets so they have to build an efficient industry of fruit processing. Fresh fruit represents 20% of world market, but seems to be more remunerative than processed fruit. Africa and Latin America are competing in this market with Costa Rica controlling the North America and Europe market with the volume of 1,511,460 tonnes (FAOSTAT 2012). The success of Costa Rica is due the great success of 'MD-2', which was first introduced to world markets by Del Monte in 1996. This sweeter and more savoury Hawaii-bred variety of pineapple has benefited from an extensive combination of research and development, supply chain improvement, and marketing by large multinational corporations, the likes of Del Monte and Dole.

#### V. Public Support and Other Supporting Initiatives

**The Government.** State elaborates and insures the implementation of Agriculture Policy through ministries and their different technical services: Ministry of Agriculture, Livestock breeding and Fishing (MAEP); Ministry of Industry and Trade; Ministry of Economy and Finances; Ministry of Transport and Public Works. Public supporting structures are dedicated to standards and technical regulations, research, quality control, and trade promotion. The extension services are handled at department level through the CerPA (Regional Centres for Agriculture Promotion). CeRPA Atlantique/Littoral plays the main role, given the importance of these Departments in the pineapple industry (80% of Beninese production). Research is carried out at the National Institute of Agriculture Research (INRAB), with a research station in Niaouli (near Allada), in the heart of the main production zone. But according to many actors (producers) and previous studies, the support of these structures is not so perceptible. Moreover, the INRAB has only one (1) researcher for pineapple and this one doesn't have adequate means and resources to constitute a real research team.

**International partners.** These partners include United Nations Development Programme (UNDP); The World Bank; FAO; United Nations Industrial Development Organization (UNIDO); International Institute for Tropical Agriculture (IITA); European Union; German Technical Cooperation (GTZ); Agence Francaise de Développekent (AFD); and many NGOs and initiatives.

## VI. Perspectives

**SWOT Analysis of the sector.** The strengths, weaknesses, threats and opportunities of the pineapple sector in Benin are listed below (Table 4).

Table 4 – Strengths, weaknesses, threats and opportunities (SWOT Analysis) of the Benin pineapple sector.

Strengths	Weaknesses
<ul style="list-style-type: none"> <li>- Producers have experience in pineapple cultivation (at least 18 years);</li> <li>- Two companies have experience in dried pineapple and pasteurized juice production (near 15 years);</li> <li>- Processors have experience in fresh juice production (near 15 years);</li> <li>- High quality of pineapple from Benin;</li> <li>- Manufactured pineapple juice is natural and chemicals free.</li> </ul>	<ul style="list-style-type: none"> <li>- Weak access to suitable equipments;</li> <li>- Dried pineapple has no market at national level yet;</li> <li>- Difficult access to specific inputs;</li> <li>- Difficult access to packing;</li> <li>- Absence of conservation facilities for products;</li> <li>- Lack of technical knowledge of producers;</li> <li>- Lack of organisation of the producers;</li> <li>- Pineapple juice is more expensive than competing products (soft drink);</li> <li>- Organizational weakness of the whole sector.</li> </ul>
<p><b>Opportunities</b></p> <ul style="list-style-type: none"> <li>- Involvement of State in the organisational process of the sector;</li> <li>- Producers are active, and gathered in associations and Economic Interest Groupings (EIG);</li> <li>- Availability and access to land;</li> <li>- Possibility to produce organic pineapple;</li> <li>- Local market open to Beninese pineapple;</li> <li>- Existence of broader markets (Malanville, Sèmè-Kraké, Hillacondji) though which Beninese pineapple could be transited to neighbour countries.</li> </ul>	<p><b>Threats</b></p> <ul style="list-style-type: none"> <li>- Difficult access of Beninese pineapple to Nigerian market (giant neighbour);</li> <li>- Police troubles and worries during transport;</li> <li>- Water deficit not compensate by irrigation;</li> <li>- Importers impose their will within the sector or they are dishonest;</li> <li>- Lack of control of the market demand;</li> <li>- Consumers' snobbism.</li> </ul>

**2. Actions to be taken at each level to develop the sector.** The actions to be taken at each level to promote and develop the pineapple sector Benin in are listed below.

### 1) Production level

- Reinforce counseling support to producers so that they can produce quality fruits of EU standards;
- Facilitate access to seeds and other inputs;
- Reinforce the management of pineapple producers associations.

### 2) Processing level

- Reinforce managerial and technical capacities of stakeholders in processing industry;
- Facilitate access to high-performance equipments;
- Facilitate access to low-cost (cheaper) packing;
- Support the implementation of infrastructures of conservation for products.

### 3) Commercialization level

- Reinforce managerial and technical capacities of traders;
- Reinforce the raising of derivate products like dried pineapple and organic pineapple;
- Facilitate access to other transport means like shipping;
- Reinforce the capacity and competence of quality control structures.

### 4) Overall sector level

- Facilitate access to loans;
- Cultivate an enhance the notion of quality among stakeholders;
- Develop research-action in the sector in order to provide the producers with “technological packages”;
- Enable visits to neighbour countries like Ghana or Ivory Coast to see and learn how these countries managed to build a successful pineapple industry based on small producers (mainly Ghana);
- Define and implement a global policy to promote and support the pineapple sector in Benin.

## CONCLUSION

The pineapple sector is very important for Benin's economy, mainly because all attempts to revive the cotton sector, the first source of income, have no encouraging results. So, all eyes are turned towards the pineapple sector and expectations are very high. Even if the sector has experienced a significant development and Beninese entrepreneurs have proved their capability, many failures and weaknesses such as funding, research, processing, transport and access to international markets, and other technical supports hold the sector back. Despite its important socio-economic role, pineapple production has some negative impact on the environment, as shown in the Department of Atlantique. The country has huge unused agricultural potentials suitable for pineapple cultivation. It is important that besides the Government, the private sector, NGOs and international partners are trying to provide to the industry a new stimulus, through many initiatives. However, it remains to be seen how smallholder and individual producers will be able to get their fair share from these initiatives. Their fate is of great concern and the authorities will have to remain vigilant and ensure that they are not left by the wayside. The donor community also has a role to play by encouraging the experiment in Benin of best practices (in the fields of technical supports, research, funding, contract farming) that have elsewhere proved successful in fostering the overall growth of a thriving fruit export industry while improving the livelihoods of smallholder farmers.

## ACKNOWLEDGEMENT

The paper is based on information the author collected during field visits and interviews in Benin (2008, 2011) and two studies by Tossou Ch. C., Floquet A. & Sinsin B. (2009) and Gnimadi A. (2008). Other sources like books and Websites are also consulted. The author would like to warmly thank Mr. Jean Gnikobou, owner of the farm "Le Paysan" in Zinvie (Commune of Abomey-Calavi, Southern Benin)

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Fig. 7. Left to right, 'Cayenne lisse' and 'Perola' (Zinvié, South Benin) - Photo: F.-T Adossou (2000).



Fig. 8. Pineapple plantation, Zinvié, South Benin. Photo: F.-T. Adossou (2000).



Fig. 9. Farmer, Zinvié, South Benin. Photo: F.-T. Adossou (2000).

## News from Brazil

### In Vitro Culture of Pineapple Apical Meristems for Viral Removal

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#### INTRODUCTION

Pineapple is one of the most accepted tropical fruits in the world. Brazil, as one of the centers of origin and genetic diversity of the species, and has been a leader in genetic improvement and germplasm conservation of this important fruit (Cabral et al., 2004).

Among the diseases of major impact on the Brazilian pineapple industry fusariosis is the main one. Several measures have been taken to control it, including the development of resistant hybrids such as the new cultivars Imperial, Vitória and Ajubá (Cabral and Matos, 2005; Ventura et al., 2007; Cabral and Matos, 2008).

Pineapple wilt caused by a viral complex (*Pineapple mealybug wilt associated virus*, PMWaV 1-3), transmitted by the mealybug *Dysmicoccus brevipes* (Sether et al., 2002 and 2005), is another disease that has caused high economic losses to susceptible cultivars in the main production areas of Brazil and the world. In addition to the direct losses of plant vigor and production, the indirect effects must be considered. In some cases contaminated plants do not show symptoms. These appear only when the plant infected by the virus is also colonized by the mealybug vector. Plantlets produced by uncertified biofactories represent a risk factor for introduction of contaminated plants into still clean production areas. This risk is amplified by the possible presence of virus strains without known symptoms.

Commercial pineapple fields are usually planted using slips or suckers, harvested directly in other commercial fields and hence with serious risk of dissemination of the wilt virus, even when this planting material is treated chemically for mealybug control. A molecular diagnosis technique to detect the viral complex in pineapple plants has been established by Embrapa, allowing the indexation of elite materials from the genetic improvement program (Andrade et al., 2010). Once the virus is confirmed to be present, it is necessary to have available and be able to apply a strategy of cleaning and removal of that pathogen, a procedure especially important for getting healthy mother plants as the source for production of certified plantlets.

Meristem cultures and thermotherapy have been the main techniques used for removal of viruses in different plant species. In pineapple plants, however, trials carried out in Hawaii using *in vitro* culture and thermotherapy of axillary buds, showed that the application of high temperatures was not efficient and the culturing of axillary buds presented discrete results which were directly dependent on the size of the explants used (Sether et al., 2001).

In this context, the objective of this work was to test a new strategy based upon the development of a methodology for the removal of the pineapple wilt virus from apical meristems excised from *in vitro* plants.

#### MATERIAL AND METHODS

In this trial were used plants of the pineapple hybrid 'Ajubá' grown in a greenhouse and infected by the pineapple wilt virus as shown by previous indexation using the method proposed by Andrade et al. (2010). Each axillary bud of the plants was individually identified. The axillary buds from the upper third of the stem were removed, cleaned and each one placed separately into trial tubes containing MS culture medium free of plant regulators (Souza et al., 2009). In the first phase, the axillary buds with a size of about 5 to 7 mm, after removal of all tissue possible around them, were incubated in a growth chamber at a temperature of  $27 \pm 1^\circ\text{C}$ , photon flux of  $22 \mu\text{E m}^{-2}\text{s}^{-1}$  and photoperiod of 16 h. In the second phase, individual plants originated from the buds had their

apical meristems excised using a stereoscopic microscope in a laminar flux chamber. The explants were reduced to a size of about 1 mm and then placed into a MS culture medium with 0.5 mg/L BAP, 30 g/L of sucrose and 2.0 g/l of Phytigel, and grown under the same environmental conditions used in the first phase. After the meristem explants developed into complete plants these were again indexed using as control a group of plants from the first trial phase, i.e. plants obtained *in vitro* which have not been subjected to the removal of the small-size meristems.

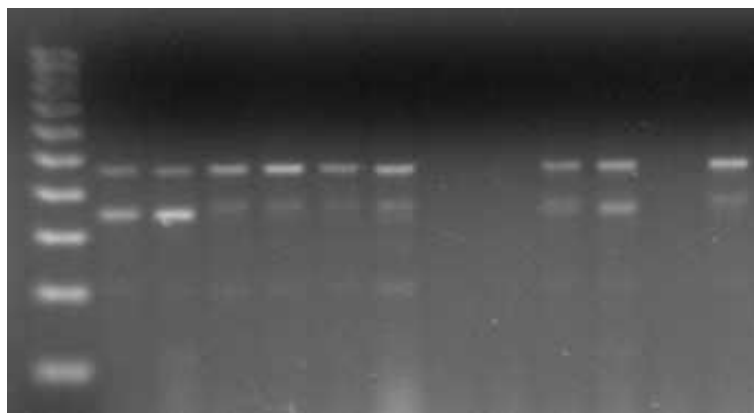
## RESULTS AND DISCUSSION

The buds introduced in the first phase took about 60 days for swelling and plant development. However, plants were allowed to grow to a larger size in order to enable the excision of the apical meristems with a higher chance of success, as this is a rather delicate procedure, especially in the case of a monocotyledonous plant *in vitro*. Almost no references could be found in literature on this kind of procedure, except for Albuquerque et al. (2000), who used this type of explant to remove the fungus *Fusarium subglutinans* f. sp. *ananas*, responsible for fusariosis of pineapple. The apical meristem is rather watery and difficult to identify, whereas axillary buds of pineapple stems and crowns are much easier to obtain and are commonly used for propagation.

Plant development was extremely slow in the second phase, probably due to the small size of the meristems used. A similar behavior has been observed for other plant species whose meristems are cultured *in vitro* for clonal cleaning with results being dependent on the meristems size. Biswas et al (2007) reported that strawberry meristems should be 0.3 to 0.5 mm tall to get success in virus removal. For cassava, Souza et al. (2009) recommended the use of a meristematic explant with no more than two foliar primordia, which represents a meristem size of about 0.2 to 0.3 mm, whereas for banana, another monocotyledonous plant, the appropriate size is about 1 mm.

The indexation results obtained showed that the simple procedure of tissue culture is not enough for virus removal. All six control plants (100%) from the first trial phase still presented the virus (Fig. 1). Among plants cultured over two phases, only 50% still showed the presence of the virus, a result considered rather positive, as none additional procedure, such as thermotherapy or others, had been applied. As the upper two thirds of the adult pineapple plant stem may have about 20 buds to be introduced *in vitro*, a recovery of ten virus free plants would assure an adequate source for micropropagation of healthy plants.

The passage of a plant through tissue culture procedures can improve significantly its performance in the field, with yield gains due to the improved health status and uniformity of the planting material. However, just this technique may not assure the clones to be free of viruses. The present study has been continued by our research group with other sets of pineapple plants and cultivars in order to validate the technique reported in this paper.



**Figure 1** – *in vitro* indexation of ‘Ajubá’ pineapple plantlets for PMWaV by RT-PCR. Lines 1-6 are from plantlets originated from lateral buds removed from infected plants; Lines 7-12 are from plantlets originated from apical meristems about 1 mm tall excised from *in vitro* plants. M, marker of 100pb molecular weight

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## **The New Face of Pineapple Genetic Improvement at Embrapa Cassava & Fruits**

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Pineapple is one of the most consumed tropical fruits in the world and has conquered the pleasure of so many different people due to its unequaled aroma and flavor. Its shape has earned it the title king of fruits. Brazil, as center of origin and diversity of the species, has large tradition in its consumption, cultivation and also in research on this crop. Embrapa Cassava & Fruits has a solid genetic improvement program that has generated hybrids with good commercial potential such as the Fusarium-resistant hybrids ‘Imperial’, ‘Vitória’ and ‘Ajubá’, as result of more than two decades of work carried out by its scientists. This program continues to grow and to develop new materials. However, in the past few years a new face of this improvement program has appeared and its first products should be available later this year: the ornamental pineapples. In the rich germplasm bank with over 600 accessions of high genetic variability, wild genotypes with great ornamental potential have been identified and an intense work on their characterization has been done during the past few years. Among these there are many that have small fruits, curved peduncles and different colors, which are rather interesting characteristics to be explored as ornamental ones.

Many genotypes have shown potential for the flower business as pot plants, floral peduncles, for landscaping and for green leaf cuttings. Depending on this use their characteristics to be studied are different. The pineapples to be used as floral peduncles should have long peduncles, small fruits and a well-balanced crown/fruit ratio. For the foreign market these peduncles have to be straight and at least 40 cm long. Surveys done with Brazilian consumers and florists show they have preferences different from those in other countries. Brazilians appreciate tortuous peduncles as they result in more dynamic floral arrangements. For sales in pots the plants must be compact, with small fruits and short leaves and peduncles. On the other side, the use in landscaping is free, but some plants of larger size are considered to be more appropriate for large spaces. The leaves are excellent for arrangements and last for more than 30 days in floral setups. Hybrids of great beauty with varied shapes, colors and architecture have been generated in the improvement program.

Ornamental pineapples from Brazil have been commercialized in Europe for more than a decade, and with great success due to the expression of the exotic appeal of its fruit and peduncle ready to be inserted in nice floral arrangements. However, only two varieties are known, *Ananas comosus* var. *bracteatus* and var. *erectifolius* and novelties are always welcome. In Brazil the market is still small and the use of ornamental pineapple is inexpressive. The var. *erectifolius* is well-known by the native people that used this material in the handcrafting of cords and other things. It has its origin in the Amazon region and its production system is known by growers and exporters.

Hence there is a demand for new varieties obtained by controlled hybridizations focused on innovative characteristics. And this is the goal of the program at Embrapa Cassava & Fruits to exploit the ornamental potential of this Brazilian plant species. An interesting aspect of this program has been the positive and solid partnership with private companies that know the market and the product, such as ABX Tropical Flowers for Export. This company started the production of ornamental pineapples in 2005 at a farm localized in the municipality of Ceará-Mirim, only about 50 km from Natal, the capital of Rio Grande do Norte. In 2006 the company started exportating to the Netherlands and from then the exports have been rather regular reaching weekly amounts of five thousand peduncles of the two ornamental varieties. The company knew about Embrapa's program and accepted a partnership to evaluate the new ornamental hybrids using the cultivation system and the postharvest handling practices already known for the traditional varieties.

Another partnership was set up with the associate companies Topplant/BioClone, both of large experience in plantlet production of fruit species, pineapple included. TopPlant was founded on March 2002, specialized in the plantlet production of melons, water melons, papaya, passion fruit and legumes in general, using modern propagation techniques like grafting and micrografting under protected cultivation. And BioClone is a company incubated by the PROETA program of Embrapa, since 2008, specialized in micropropagation of pineapple, banana, sugar cane and tropical flowers. Both companies are localized in the municipality of Icapuí, Ceará state, region of Mata Fresca, in a central position in relation to the fruit production regions in the states of Ceará and Rio Grande do Norte. The interest of both companies is to sell ornamental pineapples on the national market, where the state of Ceará is already a well-established flower production pole. The products of major interest for these companies are potted pineapples and cloned plantlets of new varieties.

On the other side, and not less important, a partnership was set up with the José Carvalho Foundation whose social mission is to give support to rural communities through small grower associations. One of the campi of this Foundation is located in the town of Entre Rios, Bahia, where the Tina Carvalho Agrotechnical School has as its main task the education of the children and young people from the associated families. Our partnership has the goal to develop a production system suited for small growers as a new alternative of income source and family life standard improvement.

In summary, these well-succeeded alliances with rather diverse private partners with different goals – from overseas market to distant and local inland markets, have shown both the elasticity and versatility of uses and economic applications of the colorful ornamental pineapples and the effectiveness of market-driven partners and partnerships as the best way for the conversion of a research product into a market product.



Fig 3. ABX Tropical Flowers fields with ornamental pineapple hybrids produced by Embrapa Cassava & Fruits to be offered to the markets in 2012.

## **In Search of an Organic Pineapple Production System for the Region of Chapada Diamantina, State of Bahia, Brazil**

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The production of organic foods has increased strongly in the world over the past years. It is estimated that more than 35 million ha are now under organic production system (IFOAM, 2012). There are several reasons for its use, such as higher income for the grower, human awareness about contamination risks by agrochemicals and increasing demand for healthy foods without negative effects of pesticides. In addition, the pesticides may contaminate the workers when used incorrectly. Furthermore, they may lead to soil and water contamination and reduce biodiversity in the areas of conventional production.

Currently, organic food production in Brazil is receiving governmental support through policies that encourage consumption, research and production (MAPA, 2012). However, there is rather limited information available about organic production systems of several crops in the Brazilian agribusiness sector, including pineapple. Information on organic pineapple production in the world is also limited, in spite of the importance of this fruit as one of the most consumed in hotels, open markets, grocery stores and restaurants.

Organic foods appeal to health, and due to that they attract consumers that pay more for products without the risk of being carriers of agrochemical residues. Thus, the offer of organic foods in hotels, restaurants and grocery stores is a distinction that should raise the income of these establishments and thereby stimulate the organic food production. In this context, Embrapa Cassava and Fruits is executing a project on organic pineapple production in the municipality of Lençóis in the region of Chapada Diamantina, state of Bahia, Brazil, located at 400 m above sea level. The climate in the region is characterized by rainy summers, relatively cold and dry winters, and average yearly rainfall of around 950 mm. The soil is classified as a red latosol, with average aluminum content of  $3\text{cmol}_c\text{dm}^{-3}$ .

To run the project, Embrapa Cassava and Fruits established a partnership with the Bioenergia company, an organization with the goal to process juice from organic fruits of citrus, guava, mango, passion fruit, pineapple and species of *Spondias*. Bioenergia has a production area of 1,200 hectares and also intends, in the near future, to buy organic fruits from smallholders who will receive technical assistance from Bioenergia's experts on organic fruit production.

The experimental area available for studies on organic pineapple production is three hectares. The project goal is to evaluate the performance of several pineapple varieties under an organic growing system, as well as to study the effects of levels of organic manure, soil cover crops and planting densities on plant growth, yield and soil proprieties.

The first step for setting up the trials has been to obtain healthy planting material of the varieties Fantástico, Imperial, Vitória, MD-2 and Pérola. The first three are new and have resistance to *Fusarium gutiforme*, causal agent of the fusariosis disease, the most serious constraint of the pineapple crop in Brazil. Healthy planting material has been produced by the stem sectioning technique. The soil characteristics are undergoing improvement by growing cover crops such as sorghum (*Sorghum* spp.), millet (*Pennisetum americanum*), sun hemp (*Crotalaria juncea*), jack beans (*Canavalia ensiformes*), velvet bean (*Mucuna atterima*) and stylosanthes (*Stylosanthes guianensis*). These species have been planted as single or blend (1. sorghum; 2. millet; 3. sun hemp; 4. velvet bean; 5. jack bean; 6. stylosanthes; 7. blend sorghum + velvet bean; 8. native vegetation).

Another experimental test is underway to evaluate growth of pineapple varieties in response to levels of organic manure produced on farm. The rates of organic manure studied are: 10, 20, 30, 40 and 50 t/ha. This study aims at optimizing the use of organic manure for organic pineapple production based on the nutritional requirement and the availability of nutrients in the manure. The variables to be evaluated are: yield, fruit weight, nutrient uptake and root development.

Planting density of the varieties mentioned above will be studied for both fresh fruit production and fruit processing to juice. As fruit size is important for fresh fruit sale, high planting densities that may be appropriate for larger juice yield, may not be the best option for fresh fruit yield and economic returns. It is planned to study the following planting densities: 26,315; 31,250; 38,460; 47,619 and 51,332 plants/ha.

We expect to develop an organic pineapple production system for some of the varieties to be studied, especially for those resistant to fusariosis. In addition, important knowledge on interactions of genetic (varietal), crop management and environmental factors should be obtained.

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## News from Costa Rica

### Thermographs to Monitor Physiological Events in the Pineapple Crop

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#### INTRODUCTION

Many researchers (Sanford, 1962; Py et al., 1987; Malézieux et al., 1994; Bartholomew et al., 2002) have documented the influence that temperature has on the growth response of the pineapple crop. Temperature affects the length of the vegetative and reproductive cycles, and the expression of diverse undesirable physiological phenomena such as natural induction of flowering and our popular "water blow" and "sun blow" ("golpe de agua" and "golpe de sol", most likely what is called translucence, a disorder of fresh fruit where all air spaces are filled with liquid; in fruit processing such fruit are often referred to as "sinkers" because they will not float on water) and sunburn.

Most producers of pineapple have weather stations on their farms to monitor weather and climate changes and the analysis and correlation of different productive outcomes associated with this. Thus, for example, many companies in Costa Rica invest in solar screens and apply kaolin clays at certain times to mitigate the negative effect of the sun and temperature on pineapple fruit the quality .

While these weather stations provide a wealth of extremely valuable weather data, for security reasons the communication interface is located in green areas close to administrative areas. Such a location is generally incompatible with climatic realities of important areas of the pineapple farm.

For this reason and because for many companies, screening, prevention, projection of cultural practices and investigation of phenomena such as natural flowering and sunburn of the fruit, is a priority, we propose the following technology and methodology for monitoring the temperature of specific pineapple fields.

We present an example of temperature differences in the Canton of Los Chiles, measured by an automatic weather station located in the vicinity of the administrative office and 3 Sensitech TempTale®4 thermographs located in three pineapple fields in different physiographic conditions.

#### METHODOLOGY

Average temperatures were measured with Sensitech TempTale®4 thermographs with a measurement range of -30 ° C to 70 ° C. The temperature records of these thermographs were compared with temperature records from a micro climate station, located in a more secure administrative area.

#### Thermographers conditioning and placement for use in open field

Because the thermographs are not water proof, they must be protected from ingress of moisture by a plastic film cover (ten layers of plastic have given excellent results). The user or researcher must decide where to locate the datalogger. In this case, it was placed halfway up the plant, using the 'D' leaf as a reference, and the datalogger was secured to the adaxial leaf surface with a plastic tape (Figure 1).

#### Comparison of temperature data collected with thermographs vs. micro climate station

It is very important to have a weather station in the pineapple farm that provides records of solar radiation, precipitation, relative humidity and rainfall. However, when temperature is a climate variable with transcendent influence on the physiological behavior of pineapple, it is important to have more tools to maximize the accuracy of the captured information under particular conditions.

To compare the data statistically, we stratified the data into blocks by time of day (Table 1). The sampling time and programming of measurement intervals, produced 1755 temperature records, which data were then categorized by block in a box-plot using INFOSTAT software (Figure 2).

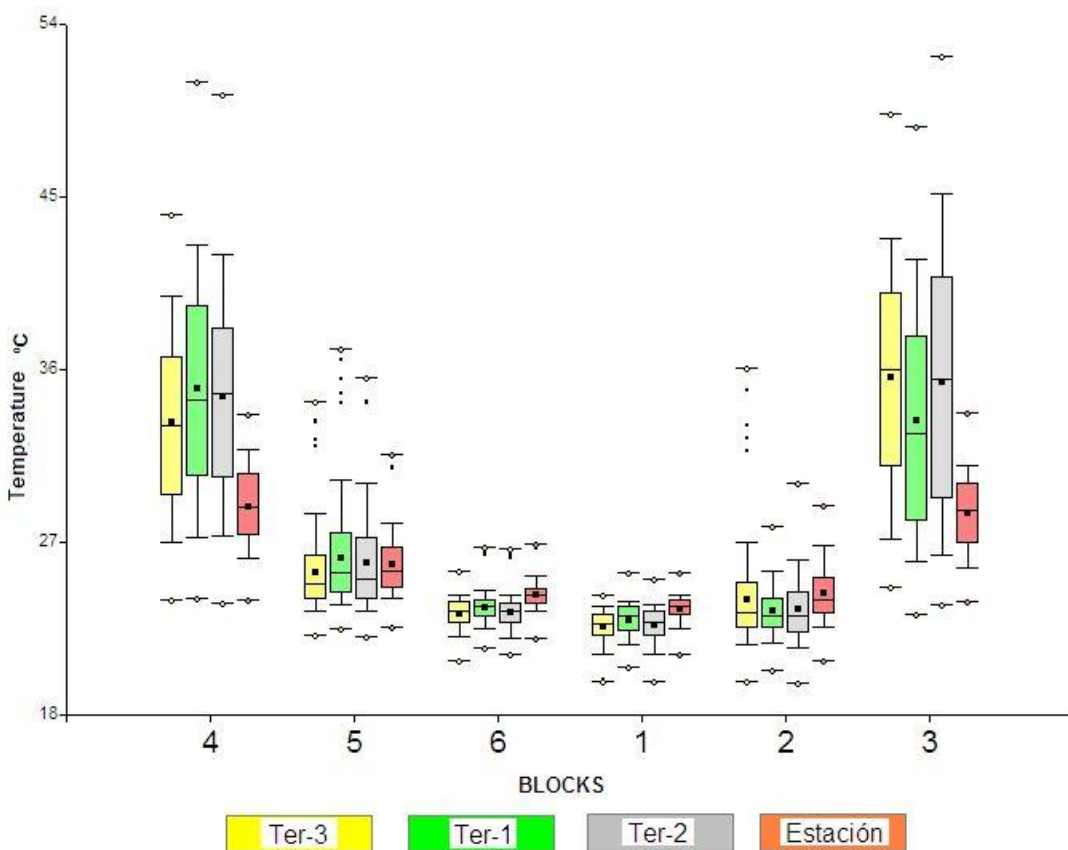


Figure 2. Temperature records categorized by block so the data could be compared statistically, from 3 thermographs placed in pineapple fields of distinct physiographic features and data from a weather station located in administrative areas. Data are from September to November 2011 in Los Chiles, Costa Rica and the time blocks were: 1, 12:00 – 3:30a.m.; 2, 4:00 - 7:30 a.m.; 3, 8:00 – 11:30 a.m.; 4, 12:00 p.m. – 3:30 p.m.; 5, 4:00 – 7:30 p.m.; and 6, 8:00 – 11:30 p.m.

The records for the weather station and the thermographs were very comparable during time blocks 1, 2, 5, and 6 (Figure 2). The small differences in mean temperature between the thermographs and the weather station, which were not significant, could be due to the physiographic locations of the recording instruments. The large differences in mean temperature between the weather station and the thermographs for time blocks 3 and 4 correspond to temperatures between 8:00 am to 3.30 pm. The lack of any statistical difference between the data collected by the weather station and the thermographs during these two periods is likely due to the extreme temperature changes that occurred in the plant canopy. At 8:00 a.m. the canopy temperature would be similar to air temperature. Due to significant heat storage in the non-transpiring pineapple canopy (Ekern, 1965), temperatures in the canopy, where ventilation is restricted, would increase throughout most of the day while a shielded temperature sensor at a weather station would rise with the corresponding air temperature. Higher resolution data, e.g., hourly means, could reveal differences in temperature between physiographic locations. However, even the block data suggests the need to better consider the characteristics of the pineapple canopy. Such data could provide answers about natural flowering behavior, "sun blow", "water blow" and "sunburn", maturation problems, "red eye", effectiveness of paraquat during the demolition or removal of the plantation and to analyze the behavior of some pests that attack the pineapple crop.

#### Possible applications of the methodology

According to the results of this evaluation and analysis, some applications that can be given to the tool could be the following:

1. Monitoring canopy temperatures in natural flowering times.
2. Using it as a tool for research or monitoring in sunburn control.

3. Using it as a tool for quantification and determination of the degree days, which greatly influence the of the "forcing-harvest" stage (Fournier et al., 2010; Malézieux et al., 1994). The versatility of this and similar equipment, and its low cost, allow more accurate monitoring of temperature in areas and provide greater numbers of replications that can improve calculation accuracy.
4. Explain the behavior of some pests affecting the crop at different times of the year, predict the incidence and plan preventive treatments.
5. Explain the growth rate of the crop in different seasons or environmental conditions, and adjust it based on nutrition programs.
6. Making maps with interpolations of temperature per lot per month on the farm, to infer certain responses on the crop growth or the incidence of pests.
7. The tool and methodology lends itself to any assessment, treatment or research, where the temperature has a direct or indirect effect on the response variables studied.

More details and recommendations on monitoring and management can be found at [www.proagrocr.com](http://www.proagrocr.com).

### **ACKNOWLEDGEMENT**

The author thanks Dr. Duane P. Bartholomew for his contributions in the expansion and correction of technical aspects of this work.

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### **Del Monte Receives Approval to Expand Area Planted to GM Pineapple**

The Costa Rican National Bio safety Committee of the State Phytosanitary Service (SFE) reportedly gave Del Monte permission to grow between 80 and 120 ha of transgenic pineapples of the rose variety on its farm in Buenos Aires de Puntarenas. Friends of the Earth Cardiff raises the typical objections to a GM crop. Ed. Note: For details, see links in **Web Sites of Possible Interest** below.

## News from Cuba

### Rapid Introduction of 'MD-2' Pineapple Using Micropropagation

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For many years, the main pineapple cultivar grown in Cuba was 'Spanish Red' and some small area planted with Smooth Cayenne. As a result of grower demand, a project for the rapid expansion of production of 'MD-2' vitroplants in Cuba resulted from a coordinated effort between Cuba and Republica Bolivariana de Venezuela governments using tissue culture technology.

The production of 'MD-2' was expanded rapidly in Ghana with the employment of the modern tissue culture laboratory. Micropropagation is an effective tool for the commercial propagation. However, tissue culture was costly to provide all the planting material needed to meet current and projected grower demand.

Centro de Bioplantitas has efficient and viable protocols (conventional micropropagation and Temporary Immersion Bioreactors) for the rapid micropropagation of pineapple vitroplants. In the Bioplant Export Laboratory, at the present time there are 250,000 'MD-2' explants in the multiplication phase. While in the acclimatization and nursery phases there are 320,000 plantlets in adaptation for transplanting to field (Fig 1). Field plantings of pineapple were into double-row beds at a population of approximately 60,000 plants ha<sup>-1</sup> with 96% survival after three month in field.



Fig 1. Plants produced in the laboratory, acclimatization phase, nursery and planted in the field.

### Genetic Characterization of the Cuban Pineapple Collection by RAPD

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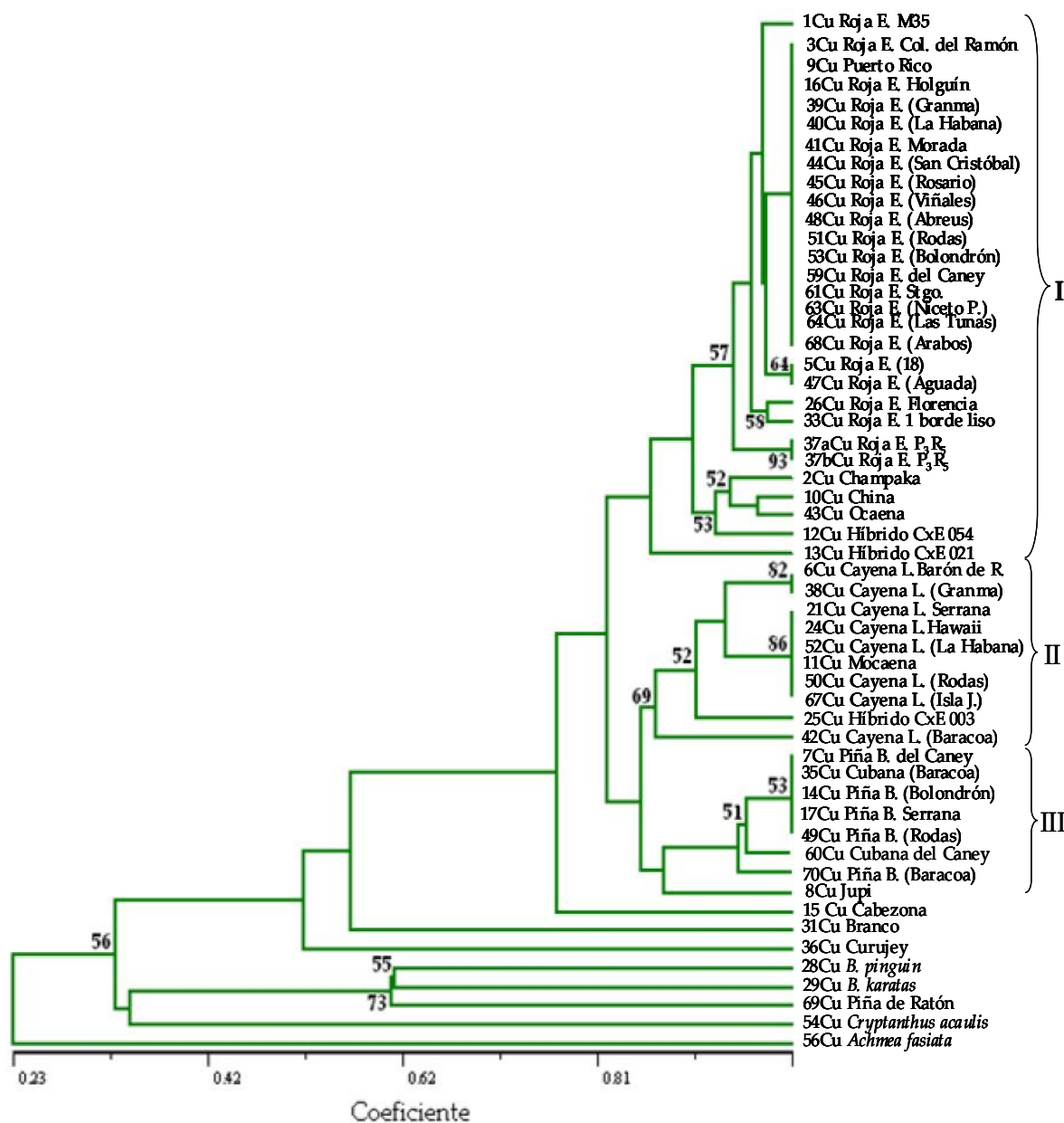
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In order to genetically characterize the Cuban pineapple (*Ananas comosus* (L.) Merrill) germplasm collection, some genomic experiments were developed at Agronomic Faculty in Agrarian University of Havana,

Cuba, in collaboration with Canarian Institute for Agronomic Researches and Experimental Station of “La Mayola” in Spain. We carried out the molecular characterization of this collection with RAPD markers, from 55 pineapple accessions and specimens from closely related species. In the present work, a molecular characterization of this bank was undertaken by RAPD, using seven decamer oligonucleotide primers to amplify the collection. A total of 57 polymorphic RAPD bands were generated with probed combinations. All primers yielded polymorphic bands in numbers ranging from 5 to 14. Primer OPA-16 produced the highest number of polymorphisms, followed by OPF-06 and CS-12. When different pineapple genotypes were compared, their genetic similarity indexes exhibited an average of 0.76, ranging from 0.11 to 0.98. Most cultivars clustered into three horticultural groups: Spanish, Cayenne and Pernambuco, although some isolated cases fell outside these clusters. We conclude that the genetic diversity of the collection is low; a problem that may be solved by incorporating accessions carrying genes conferring resistance to the main biotic and abiotic factors that affect crop yields from other centers of origin.



**Figure 1.** Dendrogram showing diversity of Cuban pineapple germplasm collection by RAPD marker (Simple Matching coefficient and UPGMA).

## Relationship Between Ethylene-Polyamine Levels and Stress-Induced Flowering of *in vitro* Cultured Pineapple Plants Under Summer Conditions

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### Abstract

The histological changes during the floral induction of *in vitro* cultured pineapple plants (*Ananas comosus* (L.) Merr) cv. Smooth Cayenne were used to study the implications of polyamine-ethylene metabolism under non-inductive natural conditions during the Cuban summer. Pineapple plants were studied throughout the course of floral initiation and development after treatment with two applications of 350 mg L<sup>-1</sup> ethephon (480 g L<sup>-1</sup>) at 0 and 48 h. Ethephon increased the level of endogenous ethylene (C<sub>2</sub>H<sub>4</sub>) in treated plants while no increase was observed in control plants. A rise in free polyamine was linked to important cytological events during floral initiation. Maximum accumulation of putrescine occurred at 12 and 48 hours after treatment while the spermidine levels in treated plants were less than in control plants. Spermine was not detected in treated plants and its concentration was very low in those that were not treated. Floral differentiation indicated by cell proliferation and the start of stem enlargement began 72 hours after treatment.

### INTRODUCTION

When plants are stressed, they generate stress substances that regulate gene expression to adapt to the stress conditions. The stress substances include reactive oxygen species, jasmonic acid, salicylic acid, ethylene, and abscisic acid (Liu and Zhang 2004; Hey *et al.* 2010; Takeno, 2012). Among these stress substances, salicylic acid and ethylene have been reported to induce flowering. Ethylene induces flowering in the Bromeliaceae, including pineapple. However, this is an exceptional case, and ethylene inhibits flowering in many plant species (Takeno, 2012).

The physiological study of the floral transition has led to the identification of several putative floral signals such as sucrose, cytokinins, gibberellins, and reduced N-compounds in general, and ethylene is translocated in the phloem sap from leaves to shoot apical meristems (Corbesier and Coupland 2006, Davis 2009).

While flowering can be induced in pineapple by ethylene, the leaf basal-white tissue produces ethylene (Min and Bartholomew, 1997, Bartholomew *et al.*, 2003) but no relationship between leaf ethylene production and flowering has been established. Use of ethylene and ethylene-releasing chemicals such as ethephon [(2-chloroethyl) phosphonic acid] has become a common practice for flowering induction among pineapple growers (Randhawa *et al.*, 1970; Reid and Wu, 1991; Manica *et al.*, 1994). Induction under natural conditions is stimulated by shortened day-length and cool night temperatures (Van Overbeek and Cruzado 1948; Gowing 1961, Friend and Lydon 1979, Friend 1981) and is affected by plant weight (Cunha 2005, Van de Poel *et al.* 2009).

Flowering induction of pineapple is associated with developmental stage, weather and nutritional conditions, hormonal behavior and silencing of the ACACS2 gene (Rebolledo *et al.*, 1997, Trusov and Botella, 2006, Avila, 2006, Liu *et al.* 2011). Phytohormones need to be coherently integrated, in a full context of signal convergence. However little is known if other pathways such as polyamines are implicated in the complex interaction among the genetic and environmental factors during pineapple floral induction. In this research, the histological changes of the inflorescence and the polyamine-ethylene metabolism of pineapple plants cv. Smooth Cayenne were studied during the summer when natural conditions for induction of flowering are less propitious.

### MATERIALS AND METHODS

Pineapple plants (*Ananas comosus* (L.) Merrill) cv. Smooth Cayenne Serrana, from *in vitro* culture, were grown on ferralitic red compacted soils, and were irrigated every seven days and fertilized based on established procedures (Isidrón, 2002). After twelve months growth plants were selected for induced flowering. To get 100% induction, no nitrogen was supplied for one month, and irrigation was withheld for the two weeks prior to induction. Induction was accomplished with 50 mL of a solution containing 350 mg.L<sup>-1</sup> ethephon (480 g.L<sup>-1</sup> as active ingredient), 2% urea and 0.5 % CaCO<sub>3</sub> (m/v) into the shoot apex of each plant. Plants were treated at 0 h and 48 h later. Untreated plants were used as the control. Samples were obtained from three portions of the D-leaf

(Devadas, 2005) basal white tissue, each 12 hours for 72 hours after first application. Fresh tissues were used for C<sub>2</sub>H<sub>4</sub> determination while tissues used for endogenous polyamine analysis were stored at -80 °C.

### **Polyamines analysis**

Pineapple stem apices were homogenized in chilled mortars at a ratio of 0.5 g fresh tissue per mL<sup>-1</sup> of 5% (v/v) of HClO<sub>4</sub>. The homogenates were centrifuged at 48,000 x g for 20 min, after which aliquots from the supernatant were collected for analysis. Aliquots containing polyamines were benzoilated as previously described by Flores and Galston (1982) and quantified by injecting 20 µL of the sample into a Pharmacia LKB high-performance liquid chromatograph with 1.7 diamine-heptan as the primary standard. The mobile phase was 52% acetonitrile in water delivered at 20 °C at a flow rate of 1.5 mL/min. The eluate was monitored with a UV detector at 254 nm.

### **Ethylene determination**

Samples containing 2 g of fresh tissue were transferred into a 10 mL *vacutainer* sealed with a serum cap, and incubated at 25 °C in the dark or in room light for 2 h. After incubation, a 1.0 mL gas sample was withdrawn from the headspace and injected into a gas chromatograph (Clarus 500 Perkin Elmer) equipped with a DB-1.40 m, 0.32 mm D.I., 1.0 µm stationary phase column and a flame ionization detector at 250°C temperature. The Oven temperature was 35 °C (30 min) @ 4°C min<sup>-1</sup> 50°C, @ 45° C min<sup>-1</sup> 200°C (1.0 min). Nitrogen (N<sub>2</sub>) 8.0 psi 14 cm/s<sup>2</sup> was the carrier gas. A known concentration (100 ppm) of ethylene gas (Scott Specialty Gases PA, USA) was used as standard.

### **Determination of inflorescence developmental stage**

Six apices were immediately put into formalin acetic alcohol (FAA) solutions for anatomical analysis following the protocol of Johansen (1940). Photomicrographs were obtained with a Nikon microphot-FX.

## **RESULTS**

### **Endogenous changes in PAs during inflorescence initiation**

The endogenous level of putrescine was twice that of control plants after 12 h and over three times that of control plants after 48 h (Table 1). Spermidine levels in treated plants were mostly less than levels in untreated plants after 12 h from floral induction (Table 1). Spermine was not detected in treated plants and its concentration was very low in those that were not treated.

### **Changes in endogenous ethylene production after forced induction**

Ethylene production by treated and control plants was similar at 0 and 12 h but production by treated plants was higher than for control plants at and after 24 h (Table 2) The highest levels produced by treated plants were measured at 60 and 72 h.

### **Morpho-histological changes in pineapple meristem**

Anatomical changes in the apical bud during the floral-differentiation process were similar to those described by others ( Gifford, 1969, Bartholomew, 1977, and Wee and Rao, 1979). The vegetative stage showed classical characteristics, with a slightly flattened dome, compacted, surrounded by bracts, and the terminal portion mass consisted of undifferentiated cells. At 48 h after the induction treatment there was incipient vacuolization of the cells that integrate the zone identified as corpus, which clearly showed a higher content of plasto-chromes. At 72 h the primordial leaves were most separated and bud apex was broadened.

## **DISCUSSION**

Under the unfavorable inductive conditions of summer, application of ethephon solution at 0 and 48 h promoted a peculiar behavior of putrescine with two peaks of accumulation at 12 and 48 h, presumably in response to ethylene release from the ethephon . However, there was no clear relationship between ethylene production and putrescine levels. The relationship between polyamines and ethylene has been studied by several authors (Lee and Chu 1992; Lutts and Bouharmont 1996; Turano *et al.*, 1997; Tamai *et al.*, 1999; Locke *et al.*, 2000; Cassol and Matto, 2003) and the results of others show that higher ethylene production is not always

accompanied by a parallel increase in putrescine levels in plants. The trend in putrescine levels in untreated pineapple plants under unfavorable inductive conditions in the summer was similar to trends in ethephon treated plants, although values were lower and much more stable. It indicates that the environmental factors promoted these performances in putrescine and ethylene concentrations, both of which increased when ethephon was applied to strengthen natural induction. Nevertheless Spd y Spm showed lowest concentrations.

The activity of s-adenosylmethionine decarboxylase presumably did not change as ethylene produced from ethephon application was released by a pathway that did not involve the methionine cycle, while putrescine remained higher as signal associated with positive response to abiotic stress. In contrast to Yamaguchi *et al.* (2007) and Shi *et al.* (2011), who reported that spermine (instead of putrescine) would be the polyamine with a protective role under dehydration stress, in our experiments both spermidine and spermine (data not shown) levels decreased. These data imply that putrescine (but not spermine) could be the polyamine responsible for the better performance observed under our experimental conditions. It is important to note that differences between reports could arise due to differential stress treatment or growing environment.

Polyamines have been globally associated to plant responses to abiotic stress. Particularly, putrescine has been related to a better response to cold and dehydration stresses. Furthermore, the increment in putrescine upon cold treatment correlates with the induction of known stress-responsive genes, and suggests that putrescine may be directly or indirectly involved in ABA metabolism and gene expression (Alet *et al.*, 2011). Endogenous hormonal balance between gibberellins and ABA too reach increase only in ABA when pineapple plant in this experiment was treated with ethephon in comparison to control (Avila, 2006, data no shown).

At 48 h after treatment, the shoot apex widened, the floret primordia were formed and the leaf primordial grew very slowly. After 72 h and as a consequence of mitotic activity, the bud with leaf primordial increase the separation, process which take place giving way to the axis continue to develop the cells that form the dome begins to project and expand over a syncline and anticline growth is evidenced by a bulge emerging feature of the first stage of differentiation.

The histological behavior is in correspondence with previous hormonal analysis. Putrescine high levels signaling stress generated for ethylene release from ethephon assimilation occurred previous to 48 h when is appreciated the first changes on shoot apex showing the inflorescence initiation. Second ethephon application strengthened the floral stimulus and it secured the irreversibility of bud differentiation, whereas 72 h the greater part of treated plants showed histological performance former described.

In this study, the two applications of ethephon increased ethylene production and initiated the floral induction process. The ethylene must induce activation at the molecular level that involves the transcriptional activation of specific genes and the result of these genes activation is then transmitted to cauline apical meristem where morphogenesis takes place. Three genes for ACC synthase have been cloned so far in pineapples and two of them have been characterized (Cazzonelli *et al.*, 1998; Botella *et al.*, 2000). ACACS1 was shown to be expressed in fruits and in wounded leaves (Cazzonelli *et al.*, 1998), while ACACS2 expression is proposed to be associated with flowering (Botella *et al.*, 2000, 2005).

The exact molecular mechanism of ethylene and polyamine action on pineapple plant responses to induce and to face abiotic stress flower promoter under non-inductive natural conditions started to be elucidated, particularly on plants coming from in *vitro* culture. Identification of more factors involved in hormone-mediated crosstalk between abiotic stress signaling and flowering merits extensive future study.

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## TABLES

Table1. Effects of ethephon solution on polyamine concentrations in the shoot apex of pineapple plants. Each data point represents the mean of three replicates (n=3).

	Putrescine (nmol/gFW)						
	0h	12h	24h	36h	48h	60h	72h
Untreated	53	120	60	49	130	100	45
Treated	52	255	40	95	465	45	96
	Spermidine (nmol/gFW)						
	0h	12h	24h	36h	48h	60h	72h
Untreated	72	50	66	30	38	65	48
Treated	71	10	20	28	12	14	12

Table2. Effects of ethephon solution on ethylene production of D-leaf basal white tissue of pineapple plants. Each data point represents the mean of three replicates (n=3).

	Ethylene (umol/gFW/h)						
	0h	12h	24h	36h	48h	60h	72h
Untreated	0.06	0.08	2.90	4.60	2.40	5.22	2.53
Treated	0.06	0.08	6.23	6.49	6.65	12.85	13.60

## Effect of Substrate Volume and Foliar Fertilization on Morpho-Physiological Changes in 'MD-2' Pineapple Plantlets

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### Abstract

Some producing countries, and in particular Cuba, do not have good quality propagation material needed when planting in new areas. Research protocols followed by the Laboratory for tissue and cell culture at Bioplantitas Center have made it possible to increase the quantity of high quality seeds required for the introduction of new pineapple cultivars such as 'MD-2'. The effect of amounts of substrates (222.6 cm<sup>3</sup> and 356.3 cm<sup>3</sup>, Fig. 1) and foliar fertilization doses (0.0, 0.5, 1.0, 1.5, 2.0 and 2.5 mL/plant) applied every seven days (Multi-N-P-K (29.5 kg/ha) + urea (20 kg/ha) + Mg (15 kg/ha) + Boro (2.0 kg/ha) + Fe (2.0 kg/ha) were evaluated. Data were collected on morpho-physiological changes in 'MD-2' pineapple plantlets at 90 days after transplanting. There was no significant effect of substrate volume on plantlet percent survival or growth. However, percent survival, number of leaves, "D" leaf length and width, plant length, relative foliar area, fresh mass and photosynthetic activity, transpiration and stomatal conductance of plantlets were greatest when plantlets were fertilized with 0.5, 1.0 or 1.5 mL/plant). Plantlets fertilized with 2.0 and 2.5 mL/plant had the lowest values for all variables, maybe for the negative effects of the high concentrations of the nutrients in the leaves of the vitroplantitas in both substrates evaluated.



Fig 4. Pineapple plantlets in volumes of 356.3 (left) or 222.6 (right) cm<sup>3</sup> of substrate in the nursery stage.

## Effects of Culture System on Morphological Change in ‘MD-2’ Pineapple Plantlets

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### Abstract

Pineapple is a crop of great commercial importance; micropropagation protocols have enabled the production of high quality seed and the accelerated introduction of new varieties. But the knowledge that has been achieved through biotechnological techniques is not an efficient method for the introduction of promising new crops such as the introduction of ‘MD-2’ pineapple. The present study is to evaluate different forms and plantation marks on the morphological change in plantlets of *Ananas comosus* (L.) Merr. MD-2 hybrid produced by Bioplants Center. Results show that this cultivar is highly susceptible to fungal diseases, especially *Phytophthora* spp. when environmental and climatic conditions are ideal for disease development. In the other hand, the plants where the framework was used in a double row planting, with plastic mulch and planted above stonemason, achieved the best values in all variables (percent survival, number of leaves, leaf length and width "D" "plant length and leaf area relative). Plants that were planted in single row, without mulch and the bottom of the groove showed the lowest values in all variables.

### INTRODUCTION

Pineapple (*Ananas comosus* (L.) Merr.) is one of the main fruits of the world and is grown to meet nutritional needs of the population and canned and fresh fruit sales are an important source of foreign exchange earnings. Leal et al. (1996) believed it was essential that the varieties of pineapple grown to suit local markets be based on germplasm from breeding programs with wide diversity to avoid the risk of genetic erosion in the species. In addition, the small number of varieties used commercially carries a high risk to the productivity from an introduced biotic. Bioplants Center has been introducing Smooth Cayenne cultivars produced in different countries and some that have been cultivated by farmers in the country for several decades.

In Cuba, and perhaps in other pineapple-producing countries, the deficiency in propagation material is a problem that arises when you want to promote new areas or introduce a new variety. The introduction of the ‘MD-2’ cultivar on a commercial scale is imperative; this variety has characteristics of great economic importance, such as high yields and fruit quality (Fournier and Mare-Alphonsine, 2007; Bartholomew, 2009). However, it is necessary to the implementation of a meristem bank to establish seed production schemes using biotechnology techniques that allow for the evaluation of cultivar to the conditions in Ciego de Avila, Cuba.

Bioplants Center has developed a novel protocol based on the use of liquid medium and temporary immersion technology coupled with the implementation of a semiautomated system, which allows reducing the time required to generate sufficient quantities of plantlets for the creation of basic seed banks that allow the development of pineapple plantations with quality seed. Generated technologies have been endorsed in the pilot

plant of the institution to produce about one million plantlets per year in a small growth chamber (Escalona *et al.*, 1999).

The success of pineapple planting depends on the successful completion of all tasks from seed selection to harvest and post-harvest in a timely manner. However, little is known about the pineapple plantlets in different forms and plantation marks on agronomical change in pineapples plantlets (*Ananas comosus* (L.) Merr.) MD-2 hybrid under environmental conditions of Ciego de Avila, Cuba.

## MATERIALS AND METHODS

This research was carried out between December 2006 and 2007 at the Experimental Station "Dr. Tomas Roig" of Bioplants Center. Pineapple plantlets (*Ananas comosus* (L.) Merr.) 'MD-2' were micropropagated following the protocol of Daquinta and Benegas (1997) using crown buds as explants. The buds were disinfected with sodium hypochlorite and cultured *in vitro* for up to ten subcultures.

### Culture conditions in the acclimatization phase.

Plantlets of 'MD-2' hybrid propagated following the instructions for the crop provided by the Technology Transfer Unit of Bioplants Center (2009) from the *in vitro* rooting phase were transferred to the *ex vitro* acclimatization phase. They were dipped in Previcur Energy® (Bayer Cropscience; 3.0 mL.L<sup>-1</sup>) for 5 minutes and then planted in different volumes of substrates. The substrate used was sieved red ferralytic soil mixed (1:1,v:v) with filter cake (derived from sugarcane plants at end of industrial process) (Hernández *et al.*, 1999). The light intensity over the plantlets was gradually increased to natural light levels over a period of 30 days to harden them. The plantlets were maintained in this condition for six months with an automatic fertirrigation system before they were transferred to field conditions (Fig 1).



Fig 5. Culture of 'MD-2' plantlets in the nursery phase.

The characteristic soil in the field was a red ferralytic and the irrigation drop (4.0 L/mnts) was used for four months. After this time the irrigation system was changed to a mobile system (CASTELVETRO-MO. OC MIS manufacture in Italia) with frequency of irrigation every 7 days. The experimental treatments were made up as follows:



Double rows planted on a ridge with plastic mulch (55,000 plants/ha)

Single rows planted on a ridge with plastic mulch (33,000 plants/ha)

Single rows on level land without plastic mulch (33,000 plants/ha)

Each experimental treatment was 300 plantlets with 100 plants in each of three replicates arranged in a completely randomized design. From January until September (each 30 days), data collected in the experiments were: survival (%), number of leaves, plant length (cm), length and width of leaf “D” (cm), relative leaf area (cm<sup>2</sup>), number of roots, and fresh weight (g). At each sampling date, leaf number, “D” leaf length and width and plant height data were collected on 50 randomly selected plants while fresh weight, root number and leaf area data were collected on 20 such plants. Analysis of variance was conducted using SPSS Program. Duncan’s multiple range tests were used for mean separation at the  $p < 0.05$  level.

## RESULTS AND DISCUSSION

The plants in double or single rows planted on a ridge with plastic mulch, had survival percentages of up to 95% (Fig 2) and survival percentages were not significantly different during the experiment. Both treatments had significantly higher survival percentages than was found for treatment 3. In May, a drastic decrease in mortality was observed, which was related to the rainy season, which in one seven-day period registered 183.4 mm of rain in only 5 days, it caused the appearance of fungous disease (Fig 2).

If no protection measures are taken, especially when humidity is high, rains are frequent and temperatures are high, it is known that ‘MD-2’ is highly susceptible to *Phytophthora cinnamomi* or *P. parasitica*. Under such conditions, losses can be as high as 80 to 90% soon after planting in the field (Taniguchi, 2007). This author, evaluating three races of *Phytophthora* (*cinnamomi*, *nicotiana* and *palmivora*) in ‘MD-2’, demonstrated that *Phytophthora nicotiana* resulted in the highest mortality (100%) followed by *Phytophthora cinnamomi* (42%). The results of this experiment demonstrate the great susceptibility of ‘MD-2’ plants produced by micropropagation techniques to this pathogen when there is excess of humidity in the field (Fig 3).

The growth responses of plants in treatments 1 and 2 were similar for all variables evaluated through the month of April; in May and beyond, growth of plants in treatment 2 was significantly less than that for treatment 1 (Table 1). The leaves are the main organ responsible for the formation of photosynthates for the normal development of seedlings, but in treatment 3, plants grew slowly and eventually died due to fungal infections. The reduction in growth and increased losses were associated with increased rainfall after May, which is the beginning of the rainy season in Cuba.

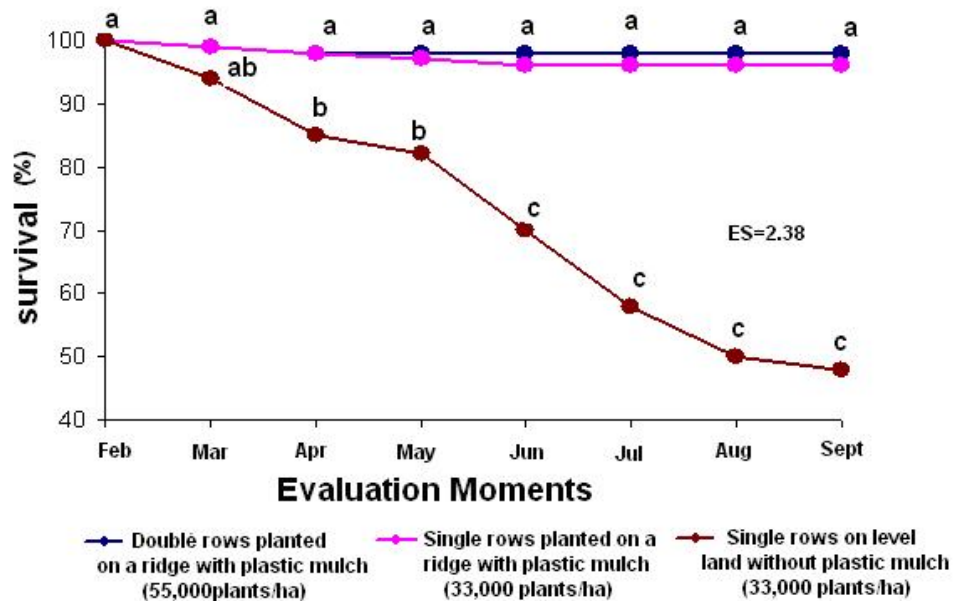


Fig 2. Effect of different treatments on the survival (%) of 'MD-2' pineapple plantlets. Means followed by different letters in rows are significantly different using ANOVA, Duncan's test,  $p < 0.05$ .



Fig 3. Pineapple plants infested with *Phytophthora* spp. planted in single row without plastic mulch and planted in the bottom of the groove.

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Table 1. Effect of different forms and plantation marks on morphological variable in ‘MD-2’ pineapple plantlets during field conditions.

Treatments	Month of evaluation	Number of leaves	Plant length (cm)	Length of leaves "D" (cm)	Width of leaf "D" (cm)	Relative leaf area (cm <sup>2</sup> )
Double rows planted on a ridge with plastic mulch (55,000 plants/ha)	February	14 c	25,2 b	27,1 c	1,5 c	90,2 b
	March	14 c	28,3 b	27,9 c	2,1 c	110,1 b
	Apryl	14 c	32,1 b	30,3 c	2,3 bc	130,4 ab
	May	17 c	35,3 a	36,1 c	2,5 bc	150,3 a
	June	18 bc	35,1 a	36,0 b	2,8 ab	160,2 a
	July	22 b	38,0 a	37,3 b	3,0 a	170,1 a
	August	23 a	40,6 a	38,9 ab	3,2 a	175,5 a
	September	25 d	45,8 a	45,2 a	3,5 a	180,6 a
	Single rows planted on a ridge with plastic mulch (33,000 plants/ha)	February	12 d	25,7 b	25,8 c	1,5 c
March		12 d	25,8 b	27,3 c	1,7 c	85,3 b
Apryl		12 d	26,3 b	27,9 c	1,7 c	86,8 b
May		14 d	27,5 b	28,2 c	1,8 bc	88,3 b
June		14 c	28,3 b	28,9 c	2,0 b	90,6 b
July		15 c	30,1 b	29,5 c	2,2 b	92,2 b
August		16 b	30,9 b	30,2 c	2,3 b	95,3 b
September		18 b	32,4 b	31,0 c	2,4 b	105,2 b
Single rows on level land without plastic mulch (33,000 plants/ha)		February	8 e	24,3 c	25,2 c	1,3 d
	March	8 e	25,2 b	25,9 c	1,3 d	75,3 c
	Apryl	8 e	26,8 b	26,8 c	1,2 d	76,3 c
	May	8 e	27,7 b	27,3 c	1,2 d	78,8 c
	June	5 e	0 d	0 d	0 e	0 d
	July	0 e	0 d	0 d	0 e	0 d
	August	0 e	0 d	0 d	0 e	0 d
	September	0 e	0 d	0 d	0 e	0 d

Means followed by different letters in column are significantly different using ANOVA, Duncan's test, p< 0.05.

## News From Indonesia

### Changes in Soil Organic Carbon after Application of Several Cover Crops Residues in Pineapple Field in Indonesia

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Although the benefits of growing cover crops as rotation crops in the pineapple field for improving both physical and chemical soil properties has been known well, however, detailed study of the decomposition rate under humid tropical climate is still rare. An experiment was conducted to investigate the changes of soil organic carbon and its components (humic and fulvic acid) after application of several cover crop residues, growth during land preparation of pineapple crop.

#### MATERIAL AND METHODS

The study was conducted in PT Great Giant Pineapple, Lampung, Sumatra Island, Indonesia. The altitude was 45 m above sea level with the geographic position was 4° 59' SL and 105° 13' EL. The rainfall was around 2500 mm/year, with air temperature minimum of about 23 °C and maximum of about 33 °C. The soil properties prior to planting were: soil texture, sandy clay loam (50% sand and 35% clay); bulk density, 1.09 g cm<sup>-3</sup>; pH, 3.93; C-organic, 1.04%; CEC 4.18 meq/100 g; N-total 0.08%.

A completely randomized block design was used with seven treatments and 3 replications. Treatment plots were 10 m x 10 m plots with three replicates. The cover crops were (1) *Calopogonium mucunoides*, (2) *Centrosema pubescens*, (3) *Pueraria javanica*, (4) *Sesbania grandiflora*, (5) *Pennisetum purpureum*, and (6) *Sesamun indicum* plus an untreated control.

The soil was prepared by plowing twice with a disc-harrow. All the cover crops were sown from seeds except *Pennisetum* which was grown from stem cuttings. After three months, the cover crops were harvested, cut into pieces and mixed into the soil around 20-cm depth with hoes.

The soil was sampled every month to monitor the changes of organic carbon. Total organic carbon was analyzed using Walkey and Black methods; humic and fulvic acids were extracted using 0.1 N NaOH, and the determination of C content of the humic and acid fulvic acids was analyzed using the method of Tatsukawa (1966).

#### RESULTS AND DISCUSSION

Cover crop (Fig 5) biomass after three months was highest for *Pennisetum* and lowest for *Centrosema* and *Pueraria* (Table 1). The soil organic carbon was unchanged in the control but increased with time up to 12 weeks for most of the cover crop treatments (Fig. 1). In the first week, organic C was higher in the *Pennisetum* treatment than in the other treatments. The highest organic-C was achieved at four weeks in the *Pueraria javanica* treatment, at eight weeks for the *Pennisetum* and *Sesamun* treatments, and at 12 weeks for the *Sesbania sp.* treatment. The decomposition rate of *Pennisetum* was fastest, however, the degradation was also fastest. On the other hand, in the *Sesbania* treatment, the organic-C increased slowly but remained high and well above other treatments after 12 weeks. The C/N ratios followed a similar pattern to that for organic carbon and there was no evidence of a large species effect on the C/N ratios (Fig 2).

The pattern for soil humic acid was almost the same as that for organic carbon, which is increase after two weeks and decrease after 12 weeks (Fig 3). Humic acid levels initially were depressed in the control and *Sesbania sp.* Treatments. The highest levels of humic acid occurred in the *Centrosoma* treatment at 12 weeks after mixing into the soil. The fulvic acid content was high in the first weeks after the crop residue was mixed into the soil, and then decreased with time (Fig 4).

Table 1. Amount of biomass of each cover crops after three months.

Cover crop	Wet biomass, (t/ha)	Dry biomass (t/ha)
<i>Sesbania</i>	36.07	8.0
<i>Centrosema</i>	14.21	2.62
<i>Colopogonium</i>	15.41	3.08
<i>Puerera</i>	19.19	2.76
<i>Pennisetum</i>	124.5	43.24
<i>Sesamun</i>	20.91	3.97

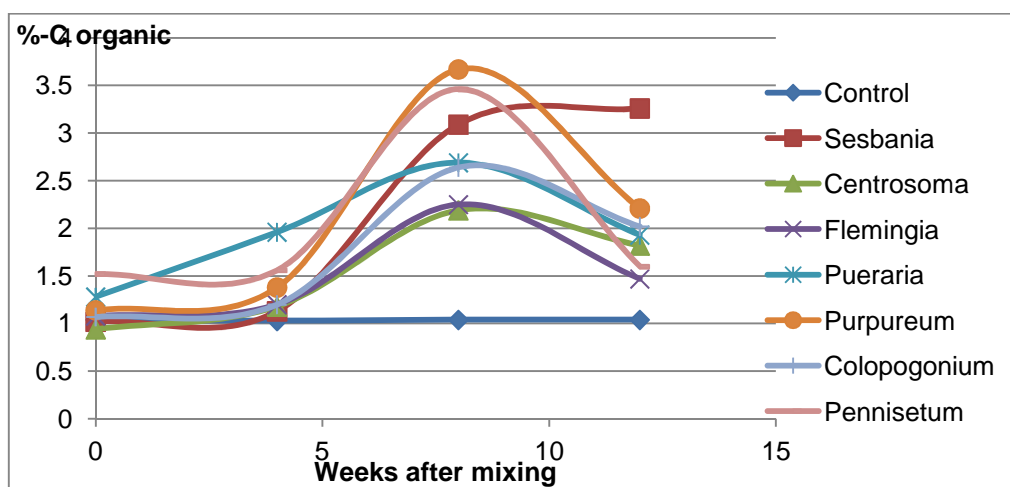


Fig 1. Soil organic carbon of each cover crop application.

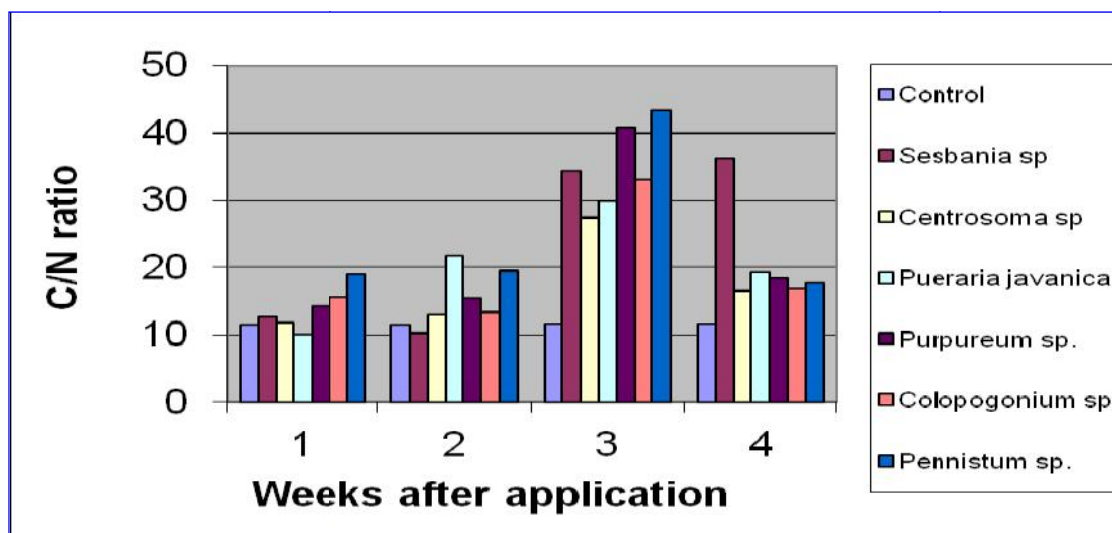


Fig 2. Carbon: nitrogen (C/N) ratio of each cover crop application.

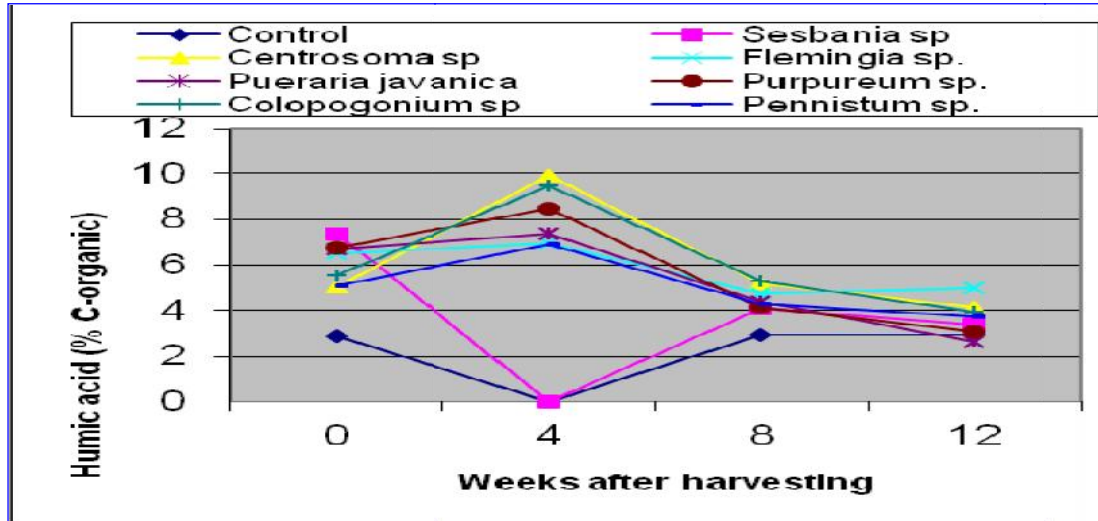


Fig 3. Humic acid content after harvesting of each cover crop application.

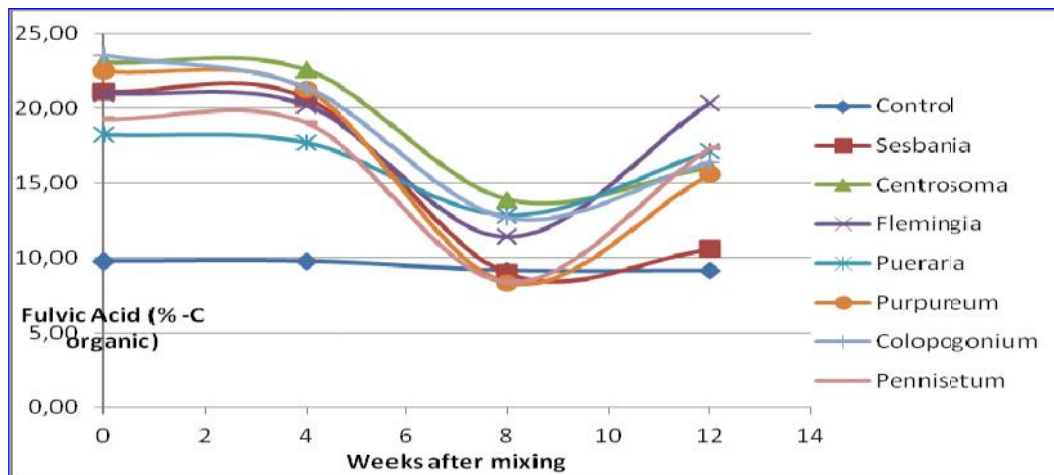


Fig 4. Fulvic Acid content after harvesting of each cover crop application.



## Symphylids Control in Pineapple Fields in Indonesia

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### Introduction

In recent years, poor plant roots have been a major problem on our Plantation (Dudy, 2006). Surveys on the plantation site by destructive sampling of pineapple plants demonstrated that one cause of this problem is the existence of symphylid on plant roots in both symptomatic and healthy plants. Symptomatic pineapple plants showed reddish leaves and witches broom in its roots (Fig 1).

To overcome this problem we conducted several field experiments ranging from symphylid population monitoring, measurement of crop losses due symphylid attack, and symphylid population management. Based on population surveys, symptomatic plants had an average 1.9 symphylids in its roots distributed on 40.74 % of samples while healthy plants had an average of 2.0 symphylids in its roots distributed on 17.2% of the samples. Plants show symptoms mainly during the initial growth of the plant when young roots start to grow. In our field we classified initial growth of plants as the first 5 months after planting. The roots of pineapple plants cannot regenerate or produce again if damaged or attacked by pests and diseases (Umble et al. 2006).

The life cycle of symphylids is influenced by environment temperature and moisture. Inoculation of symphylids in a green house experiment showed symphylids were more active when the monthly temperature averaged 28.62 °C than when at 32.9 °C. We also did surveys of potential host weeds for symphylid in our fields. Symphylids were found in root and soil around the root zones of *Lantana camara* and *Paspalum conjugatum*. From 53.33% sample *Lantana camara* had an average 9.75 symphylids per weed while 46.67% of the *Paspalum conjugatum* samples had an average 8.57 symphylids per weed. Still in progress is a project to survey other major weeds in our fields to identify other possible symphylid hosts.



Figure 1. Healthy pineapple plants on left, plants with reddish leaves due to root damage on the right and below are roots showing the typical “witches broom” symptom that is characteristic of symphylid feeding.

With a severe infestation, there is no effective treatment to control the population. However, control should be done to reduce/avoid greater losses and to ensure plant growth. Symphylids can move to depths of 90 cm below the soil surface and so can avoid tillage or pesticide applications. Symphylids will return to the soil around the root zone if the effect of pesticides has been reduced (Umble et al. 2006). In our field trial and survey, we placed bait in the root zone approximately 15-25 cm below soil surface. The depth was not only more representative of the population in pineapple fields compared to 35 and 45 cm (Fig 2), but also was economical for our labor to collect the bait samples.

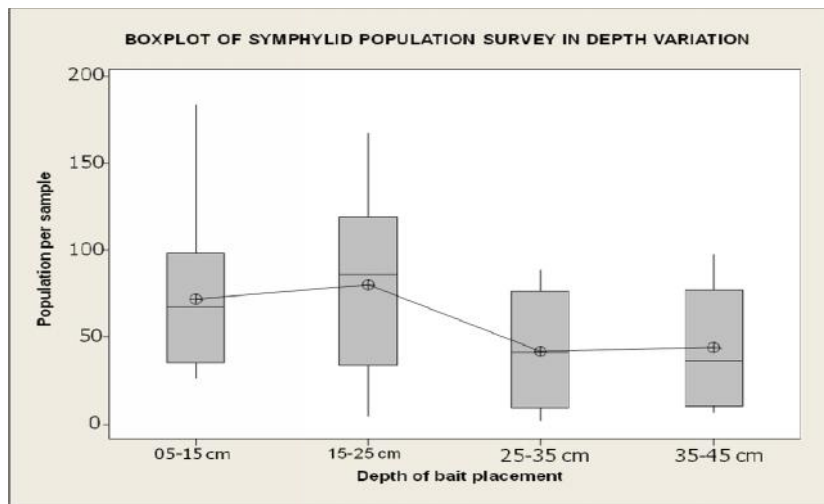


Fig 2. Boxplot of symphylid population survey results with soil depth.

Potato slices have been used as bait material for symphylids (Umble et al, 2003). In our fields, we have an abundance of papaya and cassava leaves. A trial was done to compare potato with papaya and cassava leaves. Papaya leaves were as effective as potato slices and both were more effective than cassava leaves (Fig. 3). Based on the result of this study, we used papaya leaves as alternative bait material beside potato slice.

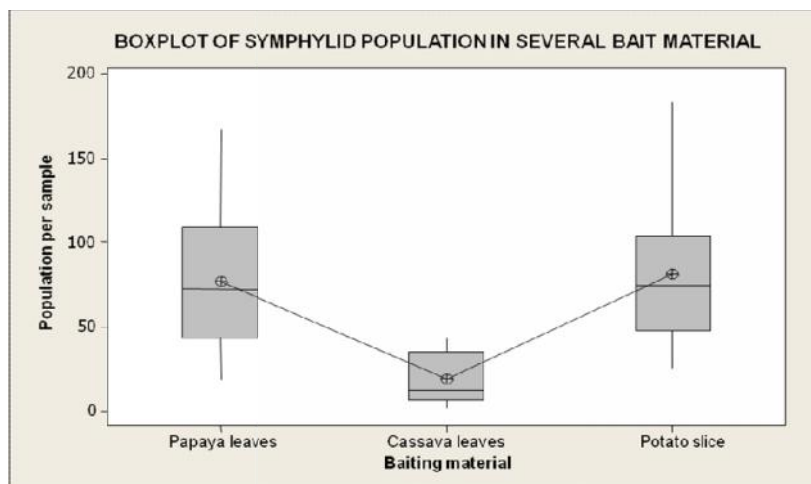


Fig 3. Boxplot of symphyliid population sampled using several bait materials.

Insecticides used to control symphyliid population because there has been no specific pesticide for Myriapoda. However insecticides are used in our plantation appear to show less satisfactory results. In this study, we compare pesticides effectiveness in controlling simphyliid at high population levels.

#### MATERIAL AND METHODS

A field trial was conducted at the pineapple plantation of Great Giant Co. to compare insecticide treatment applied manually as granules in leaf axils or as a broadcast spray. Applications in  $\text{kg ha}^{-1}$  of active ingredient were, manually: 1) carbosulfan, 2.5; 2) carbofuran, 1.5; 3) bifenthrin, 0.125; and by spray: 4) chlorpyrifos, 2.4; 5) bifenthrin, 0.1; and 6) fipronil, 0.25. ‘Smooth Cayenne’ pineapple plants at 4 months were used in this study. Monitoring was by the baiting method (Marie-Alphonsine et al. 2010) with baits placed at depths of 25 cm, 35 cm and 45 cm with Randomize Block Design to represent the population in the field. Symphyliid populations were observed at 2, 4, 6 and 10 weeks after treatment. Our tolerant population is 5 symphyliids per sample using the bait method.

#### RESULTS AND DISCUSSION

Based on observations at 2, 4, and 6 weeks after application (WAA), there were no statistically significant differences between treatments. At 10 WAA, the symphyliids per sample for the treatments were fipronil, 1.5; bifenthrin, 3.7; and carbosulfan, 10.9. Based on the results of this study we issued a recommendation to spray  $0.25 \text{ kg ha}^{-1}$  a.i. of fipronil when a symphyliid population outbreak occurred in our field and apply  $0.1 \text{ kg ha}^{-1}$  a.i. granular bifenthrin in the leaf axils after chopping when the symphyliid population reaches threshold levels.

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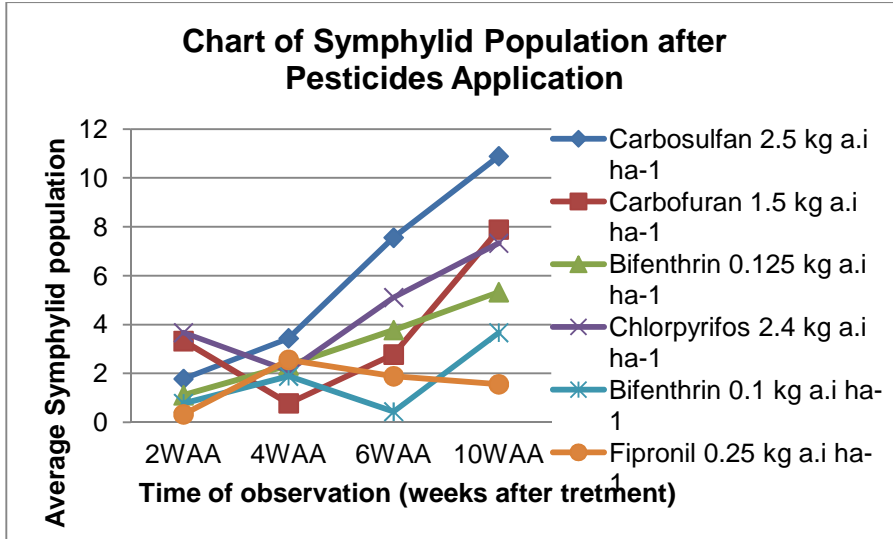


Fig 5. Symphylid populations two to ten weeks after pesticides application (WAA).

## News from Malaysia

### Vanillin Enhances *Agrobacterium*-Mediated Transformation of 'N36' Pineapple

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#### Abstract

The effect of vanillin on *Agrobacterium*-mediated transformation of pineapple v N36 was examined. Vanillin in concentrations ranging from 0-500  $\mu$ M was included in the medium during co-cultivation of the longitudinal-section pineapple plantlets with *Agrobacterium tumefaciens* strains LBA4404 that harboured the hygromycin phosphotransferase gene (*hpt*). The transformants were selected in 20  $\mu$ g/ml hygromycin, and confirmed by GUS histochemical assay and polymerase chain reaction. The highest GUS assay, 50% positive blue colour, was observed in 500  $\mu$ M vanillin. Findings suggested that vanillin might substituted for acetosyringone for *Agrobacterium*-mediated transformation of pineapple plantlets.

**Keywords:** hygromycin, GUS assay, cefotaxime, cell culture

#### INTRODUCTION

Pineapple is the third most important fruit crop in the tropics and subtropics, after banana and citrus (Rohrbach et al., 2002). Many efforts have been made to produce improved cultivars of pineapple via hybridization but only a few new hybrids have been released worldwide. Conventional breeding is difficult due to the high level of genomic heterozygosity and apparent genome instability (Kato et al., 2004). Biotechnology approaches showed it is possible to solve agronomic and post harvest problems such as herbicide-tolerance (Sripaoraya et al., 2001), precocious flowering and chilling induced internal fruit browning (Ko et al., 2006).

Genetic transformations of pineapple have been achieved by bombardment of leaves (Ko et al., 2006) and *Agrobacterium*-mediated transformation on various types of pineapple explants (Firoozabady et al., 2006). *Agrobacterium*-mediated transformation is considered an easy method that produces a high success rate and bypasses the callus culture phase before or after transformation. The method is also an inexpensive procedure that can produce single or a few copies of integrated transgene and enables the transfer of large, up to 150 kb, segments of DNA into the host genome (Wang et al., 2009).

It has been known for decades that phenolic compounds play an important role in inducing the *Agrobacterium-vir* gene, which leads to the transfer of T-DNA into the host plant. However, some explants of monocot species can be efficiently transformed without the aid of external *vir* inducing chemicals. For example, the pre-cultured immature embryos and embryogenic calli of wheat co-cultured under desiccation conditions were efficiently transformed (Cheng et al. 2003). The successful transformation may be due to the difference in the inoculation and co-cultivation duration, competence of target tissues, pH during co-cultivation and *Agrobacterium* strain (Karami et al., 2009).

To date, acetosyringone is the only phenolic compound that has been extensively used as a *vir* gene inducer. However, high concentrations of acetosyringone caused chimeras in transgenic pineapple (Firoozabady et al., 2006). Other phenolic compounds may be less potent than acetosyringone yet still be effective *vir*-inducing chemicals. Vanillin is a phenolic compound with the molecular formula  $C_8H_8O_3$  with aldehyde, ether, and phenol functional groups. Vanillin improved *Agrobacterium*-mediated transformation of the unicellular green alga (Cha et al., 2011). Therefore, in the present study the effect of vanillin on *Agrobacterium*-mediated transformation of pineapple was investigated. The finding would provide a new alternative chemical used in *Agrobacterium*-mediated transformation especially pineapple.

## MATERIALS AND METHODS

### Plant material

Previously established *in vitro* 'N36' pineapple plantlets, a hybrid of 'Gandul' (Singapore Spanish) and Sarawak ("Smooth Cayenne") (Aziz et al., 2011) were used. The plantlets were proliferated in MS medium containing 30 g L<sup>-1</sup> sucrose, 1 mg L<sup>-1</sup> BAP and 0.5 mg L<sup>-1</sup> IBA. To determine the effect of vanillin on pineapple, three weeks old plantlets were cultured on phytohormone-free MS medium added with vanillin concentrations of 0, 0.1, 0.2, 0.3, 0.4 or 0.5 mM for four weeks. Subsequently, the fresh weights were measured at harvest. Five replicates were made for each vanillin concentration. All cultures were incubated at 25 °C under a photoperiod of 16 h day white light provided by a fluorescent lamp.

### Agrobacterium strain and vectors

*Agrobacterium tumefaciens* strain LBA4404 carrying the pCAMBIA 1304 vector (Cha et al., 2011) was used. The bacteria was grown on LB medium (in g/L 1.0 NaCl, 1.0 peptone, 10.0 agar and 0.5 yeast extract) containing 5mM glucose, 100 µg/L streptomycin, 50 µg/mL kanamycin and incubated in the dark at 27 °C for two days. For transformation, 5 mL of bacteria grown overnight in broth culture was mixed with 45 mL LB broth containing 5mM glucose, 100 µg/L streptomycin and 50 µg/mL kanamycin. The mixture was incubated at 27 °C with shaking until the optical density at 600 nm reached 0.8 to 1.0. Cells were harvested by centrifugation at 5000 rpm for 10 min, re-suspended in MS medium at 0.8-1.0 OD<sub>600</sub> and used for transformation.

### Transformation procedure

Three week old pineapple plantlets approximately 0.6 cm in diameter and 8 cm tall were used for the transformation, which was done according to the method by Wang et al., (2009). The plantlets were longitudinally-dissection to give two explants per plantlet. The explants were immersed in 50 ml MS medium containing bacteria cells (OD<sub>600</sub> at 0.8-1.0) for 10 min, dried on sterilized tissue paper and transferred onto co-cultivation medium. The medium consisted of MS basal salt with, in g/L, 30 sucrose, 1.0 BAP and 0.5 IBA and 0, 0.1, 0.2, 0.3, 0.4 or 0.5 mM vanillin with 50 explants for each vanillin concentration. The cultures were incubated in the dark for 3 days (Wang et al., 2009) and then transferred to continuous light until new shoot tips emerged. The shoot tips were separated and transferred onto fresh co-cultivation medium with 20 mg/L hygromycin and 250 mg/L cefotaxime added to eliminate the bacteria. The putative transformed-plantlets were grow to a height of 0.5 cm and randomly-chosen for confirmation of transformation.

### Analysis of Transformation

GUS histochemical assay (Jefferson et al., 1987 with minor modifications) of the putatively transformed plantlets or shoots was performed by immersing them in 500 µL GUS assay buffer (2 mM X-gluc, 0.1 M pH 7.0 phosphate buffer, 1 mM K<sub>3</sub>Fe(CN)<sub>6</sub>, 10mM Na<sub>2</sub>EDTA, and 0.1% Triton X-100) incubated at 37 °C for 48h. Chlorophyll was cleared using 70% (v/v) ethanol for 1–2 h and tissues were then re-suspended in 40% (v/v) glycerol.

For PCR analysis, total genomic DNA of putatively transformed and non-transformed tissues was extracted using the SDS method (Edward et al., 1991). PCR was carried out in a 30µL reaction mixture comprising 200 ng DNA, 10X reaction buffer, 2 mM MgCl<sub>2</sub>, 1.25 mM dNTP mixture, 2.5 µM of each primer, 1 U *Taq* DNA polymerase (Geneaid). The Hpt-F and Hpt-R primers (Cha et al., 2011) were used to amplify fragments of the *gfp-gusA* gene fusion. Amplification was carried out in a thermal cycler (Eppendorf). The conditions for amplification of the *hpt* gene were: initial denaturing at 94 °C for 4 min, and 40 cycles of 94 °C for 45s denature and 72 °C for 1 min 10s (annealing and extension) followed by 1 cycle at 72 °C for 7 min. Amplified products were separated on 1 % agarose gel in 1 x TAE buffer.

## RESULTS AND DISCUSSION

### Effect of vanillin on pineapple plantlets

All pineapple plantlets survived four weeks of culture with added vanillin. The fresh weight of pineapple plantlets increased with increasing vanillin concentration in the medium (Fig. 1). The plantlets looked healthy with no necrotic or chlorotic spots observed on the vegetative organs. To our knowledge this is the first report

on the effect of vanillin on *Agrobacterium*-mediated transformation of pineapple. Vanillin had no negative effect on the growth of pineapple plantlets, which was also true for soybean and *Nannochloropsis* (Patterson, 1981; Cha et al., 2011). In contrast, the phenolic compounds caffeic acid, chlorogenic acid, t-cinnamic acid, p-coumaric acid, ferulic acid, gallic acid, p-hydroxybenzaldehyde, 5-sulfosalicylic acid are toxic.

### ***Agrobacterium*-mediated Transformation**

Most of the explants died after four days of co-cultivation with *Agrobacterium*, especially in the control. In general, more than 40 % of explants survived in 0.2 mM or more vanillin (Fig. 2A) for at least two weeks. The number of surviving-explants during the co-cultivation period increased as the vanillin concentration in the co-cultivation medium increased. The highest percentage of surviving-explants (55.5%) was observed with 0.5 mM vanillin. Two weeks after exposure to light, new shoot tips were observed emerging at the basal-wounded site of the surviving-explants. However, there was only 26 to 38 % shoot regeneration for the surviving-explants (Fig 2B), with one to three shoot tips per explant. Explants that did not produce new shoots died.

GUS assay results showed that all samples tested developed homogenised blue-coloration and the colour density increased with the vanillin concentrations used (Fig. 3). Based on these results and assuming only transformed cells successfully produced shoot tips, the percentage transformation was estimated (Fig. 2C). Only 4% transformation was observed in the control and in 0.1 mM vanillin while 12 to 14% transformation was recorded in vanillin concentration above 0.2 mM. PCR analyses showed that the *hpt* gene (~678 bp) was present in the transformed plantlets but not in non-transformed plants (Fig. 4).

In *Agrobacterium*-mediated transformation, phenolic compounds play an important role in inducing *Agrobacterium vir* gene expression, which leads to the transfer of T-DNA into the host plant. The host-released phenolic compounds involves in the expression induction of *vir* genes also has been review and identified (Karami et al., 2009). The present protocol demonstrated that vanillin can be used as an alternative of acetosyringone in plant tissue transformation. This study indicates that adding and increasing concentration of vanillin until will enhance the growth and increase the transformation efficiency of pineapple N36.

### **ACKNOWLEDGEMENT**

Authors wish to thank Department of Biological Sciences, Universiti Malaysia Terengganu for providing the facilities and funding the study.

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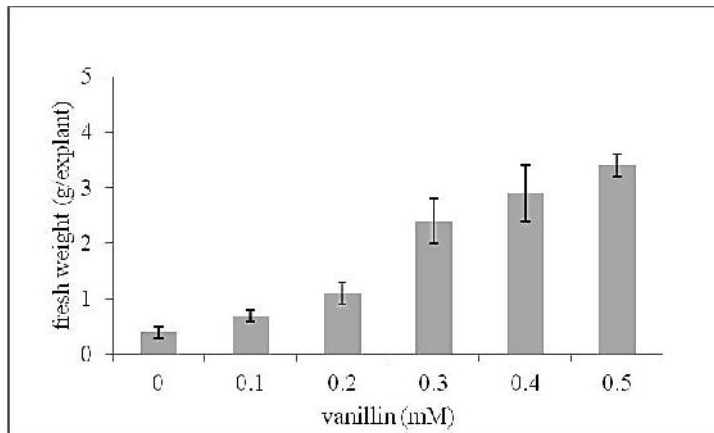


Fig 1. The fresh weight of pineapple cav. N36 plantlets after four weeks cultured in various concentration of vanillin.

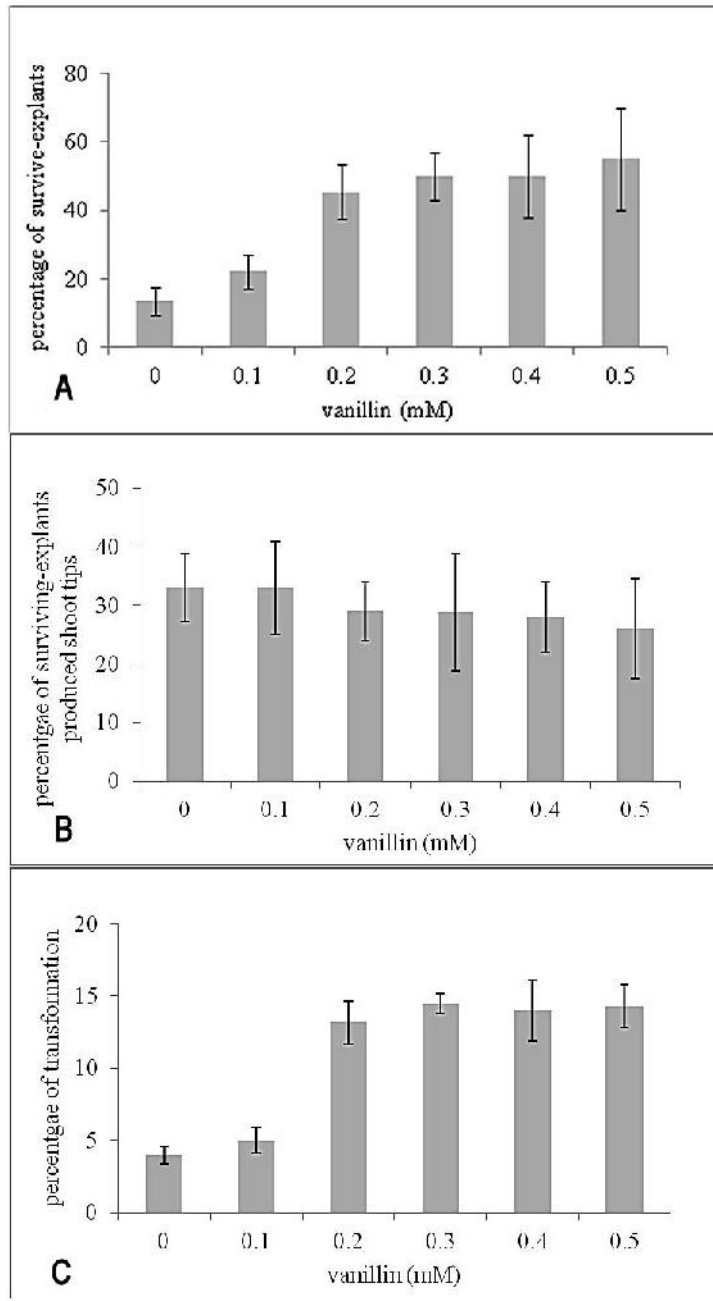


Fig 2. Percentage of survive-explants (A) and percentage of surviving-explants produced shoot tips (B) and percentage of transformation (C) of pineapple after four weeks co-cultivated in various vanillin concentrations.

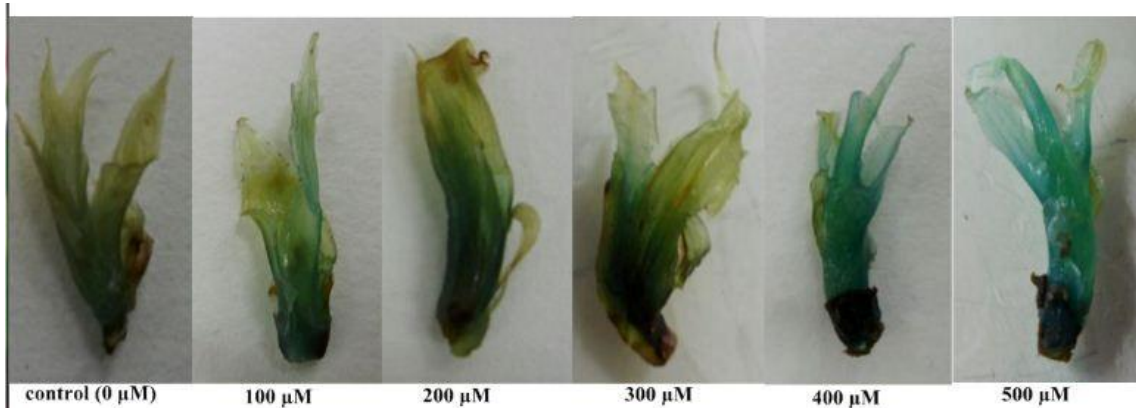


Fig 3. Homogenous blue coloration GUS assay developed on transformed pineapple plantlets treated with different concentrations of vanillin during the co-cultivation with *Agrobacterium*.

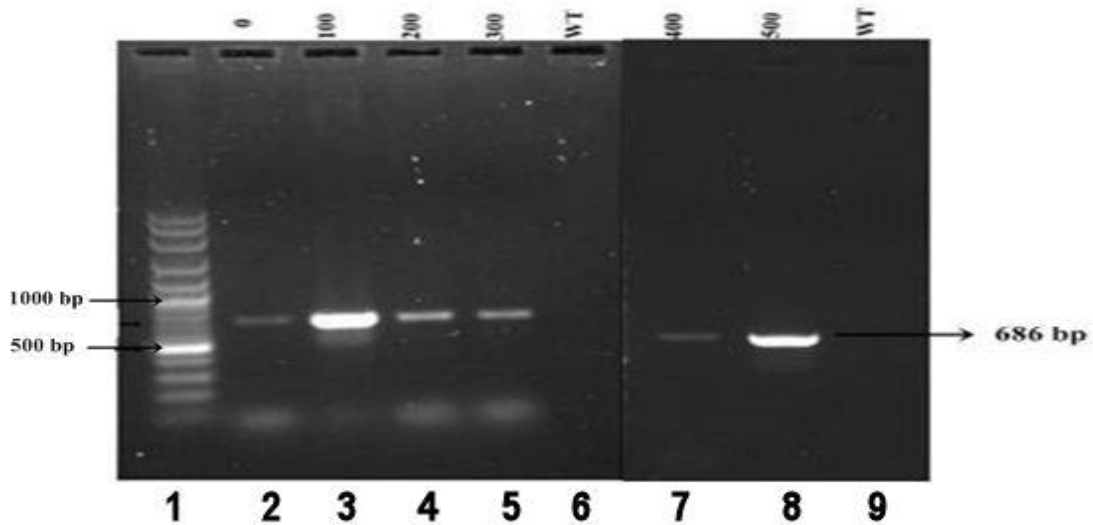


Fig 4. PCR analyses show the present of htp (~700 bp) in transformed pineapple plantlets co-cultivated in different concentration of vanillin, and no band produced in the non-transformed. Lane 1 = markers, lane 2 = 0 mM vanillin, lane 3 = 0.1 mM vanillin, lane 4 = 0.2 mM vanillin, lane 5 = 0.3 mM vanillin, lane 6 & 9 = non-transformed, lane 7 = 0.4 mM vanillin and lane 8 = 0.5 mM vanillin.

## News from South Africa

### Adult Beetle Kairomones as Attractants for the Monitoring and Control of *Popillia bipunctata* (Coleoptera: Rutelinae), a Whitegrub pest of Pineapples in South Africa.

Graham Petty. 13 Somerset Place, Lambert Road, Port Alfred, 6170, Republic of South Africa. E-mail: [pettys@telkomsa.net](mailto:pettys@telkomsa.net).

Pineapples in the Eastern Cape Province of South Africa are infested and damaged by the larvae / whitegrubs of more than twelve species of scarabaeid beetles. *Popillia bipunctata* (Fabricius) (Coleoptera : Scarabaeidae: Rutelinae) was first collected and identified as one of these pests, in the Bathurst – Grahamstown area, in about 2000 and has subsequently caused severe and widespread pineapple damage.

A number of different floral volatiles /kairomones were evaluated as attractants in bucket-funnel insect traps at three Bathurst district pineapple farms. Evaluations were done during the 3-month summer beetle-flight periods of 2003/2004 and 2004/2005. Evaluated kairomones included: cinnamyl alcohol, nerolidol, beta-phenethyl butyrate, geraniol and beta-ionone. Eugenol, as a synergist was mixed with each of these volatiles, and composed 10% of total volume.

For each kairomone mixture, eight different types of trap were used and their relative efficacy assessed. Trap variation was in design, colour (all sulphur-yellow, all dark green or a combination of both) and type of kairomone dispenser. The bucket-funnel traps all had vertical fins in the funnel top section. Fins were either yellow or green, as were the lower sections of the traps.

#### RESULTS

The numbers and percentages of beetles caught by the five kairomones during a three month period are shown in the following table.

Kairomone (+10% eugenol)	Beetles per trap	Percentages of total
Cinnamyl alcohol	2 760	46
Nerolidol	1450	24
Phenethyl butyrate	667	11
Geraniol	608	10
Ionone	485	8
<b>TOTAL BEETLES</b>	<b>5970</b>	

The following was **concluded** from these results and the number of beetles caught in the different trap types:

- Most effective kairomone was Cinnamyl alcohol-Eugenol mixture, followed by Nerolidol.
- Traps should be in place in December, January and February.
- A sulphur yellow bucket – dark green fins combination was more effective than any mono-colour trap, or inverting the combination.
- The commercial vial-wick kairomone dispenser was more effective than other types of commercially available dispensers.

#### Application of a kairomone trapping system

One of two different objectives may be achieved:

- With a low number of traps, widely dispersed, the threat posed by the beetles can be monitored and timely action taken before eggs are laid and grubs become a problem.
- With sufficient number of traps per unit area it should be possible to reduce beetle numbers below economic threshold.

## Services

The listings below are provided as a convenience to readers and should in no way be construed as an endorsement of those providing commercial or professional services. Those offering specialized services to pineapple growers or researchers are invited to contact the editor for possible inclusion in the listings below.

### Commercial Services

- **Centro de Bioplantas.** Dr. Justo L. Gonzalez Olmedo, Director of Foreign Affairs Office, Centro De Bioplantas. Universidad De Ciego De Avila, Carretera a Moron Km 9. Cp69450. Cuba. Centro De Bioplantas offers certificates of authenticity for pineapple material propagated in their tissue culture facility. Web site: <http://www.Bioplantas.cu>.
- **Maintain CF 125** continues to be available for use in pineapple plant propagation anywhere in the world. Supplies can be obtained from N. Bhushan Mandava, Repar Corporation, 8070 Georgia Ave., Suite 209, Silver Spring, MD 20910. Tel: (301) 562 – 7330; Fax: (202) 223 – 0141; On the web at [www.reparcorp.com](http://www.reparcorp.com); E-Mail: [mandava@compuserve.com](mailto:mandava@compuserve.com).
- **Thai Orchids Lab.** Dr. Paiboolya Gavinlertvatana. Horticulture/ agriculture/ forestry tissue culture laboratory with exports to Australia, U.S.A., Africa, and Asia. CO2 & MD2 pineapple available (open to acquiring additional varieties) or confidential exclusive contract propagation. Phone: +1.617.910.0563 Website: <http://www.tolusa.com/>.
- **Vitropic**, Zone d'Activités Economiques des Avants, 34270 Saint Mathieu de Trévières France; Tel: + 33 (0)4 67 55 34 58; Fax: + 33 (0)4 67 55 23 05. E-mail : [vitropic@vitropic.fr](mailto:vitropic@vitropic.fr). Web site: [www.vitropic.fr](http://www.vitropic.fr). Vitropic proposes the best individuals from the CIRAD FHLOR selected clones including: Cayenne Group, Queen Group, Perolera Group, MD2, Ornamentals pineapples. The range is continuously extending, do not hesitate to ask for more information.

### Professional Services

- **Mr. Wilbert Campos Alvarado.** M.Sc. Tropical Soils & Crop Mgmt. E-mail. [wcamposa@gmail.com](mailto:wcamposa@gmail.com). Phone: (506) 8815-7271. Apdo. Postal 536-7210, Guapiles, Costa Rica. Experience in all stages of production (soil preparation, plant nutrition, diseases, pest control, PGR use, etc) postharvest treatment and packing plant management of pineapple for the fresh fruit market. Also have several years experience in pineapple R&D and technical services in Costa Rica, Guatemala, Ecuador and Ghana. As a manager of operations I have experience in budgeting, farm planning -market aimed and my work is based in the corner stones of productivity, quality and return on investment.
- **Ing. Alejandro Chavarría.** APDO 4437-56 Pital, San Carlos. Alajuela, Costa Rica. Tel: (506) 88-20-79-55 / (506) 24-73-40-00. E-mail: [alechava@hotmail.com](mailto:alechava@hotmail.com). I have worked like an International Pineapple Consulting in México, Costa Rica and Brazil. Experienced in project feasibility, plantation design, agricultural machinery, all aspects of farm crop management, post harvest management and establishment of good agricultural practices.
- **Dr. Mark Paul Culik.** INCAPER, Rua Alfonso Sarlo 160, CEP 29052-010, Vitoria, ES, Brazil; Tel: 27-3636-9817; E-mail: [markculik3@yahoo.com](mailto:markculik3@yahoo.com). Experience: PhD in Plant and Soil Sciences with more than 25 years of agricultural pest management experience in crops ranging from apples to papaya and pineapple, identification of pests and beneficial arthropods ranging from mites to fruit flies, and current work on scale insects, including pineapple mealybugs. Areas of specialization: Entomology, Insect and Pest Identification, Integrated Pest Management.
- **Mr. L. Douglas MacClure.** 360 Hoopalua Dr., Pukalani, Hawaii, U.S.A. E-mail: [norfolkldm@aol.com](mailto:norfolkldm@aol.com). Experience: More than 39 years with Maui Pineapple Company heading plantation and diversified agriculture operations and started the Royal Coast Tropical Fruit Company in Costa Rica. Collected and summarized production information in Asia and Central America. Also consulted on pineapple for companies and growers in El Salvador, Australia, Thailand and Indonesia.
- **Mr. Graham J. Petty** 13 Somerset Place, Lambert Road, Port Alfred, 6170, Republic of South Africa. Phone: +27 (0) 46 624 4868; E-mail: [pettys@telkomsa.net](mailto:pettys@telkomsa.net). Experience: M.Sc. (Agric) Pretoria : Pr. Sci. Nat. . Researcher and advisor to the South African Canning Pineapple Industry on matters of Pest Management in pineapple culture, for 34 years. Economic entomology and management of biological control agents have received particular attention
- **Ing. Jhonny Vásquez Jiménez,** MSc. San Carlos, Costa Rica. E-mail: [jvasquez@proagrocr.com](mailto:jvasquez@proagrocr.com), (506) 89103878, (506) 24756795. Advice on the agricultural management of pineapple. Design and conduct research experiments for companies and formulators of new production technologies in the area of nutrition, plant pathology, weeds and other disorders.

- **Mr. José R. Vásquez**, MBA with emphasis in Agribusiness (E-mail: jrva46@excite.com, jrva46@gmail.com). Phone: 504 2668 1191; 504 94899901. Experience: Environmental and Sustainable Agriculture. Pineapple and melon production, from seed propagation, planting, field maintenance, forcing, harvesting, post-harvest management and commercialization.
- **Dr. José Aires Ventura**. Incaper, Rua Afonso Sarlo 160 (bento Ferreira), 29052-010, Vitoria-ES, Brazil. E-mail: [ventura@incaper.es.gov.br](mailto:ventura@incaper.es.gov.br); Tel.: 55-27-31379874. [www.incaper.es.gov.br](http://www.incaper.es.gov.br). Area of Specialization: Plant Pathology (research in pineapple diseases management; Fusarium diagnosis, diseases resistance).

## Book Reviews and Web Sites

### Book Reviews

No reviews were provided for this issue.

### Web Sites of Possible Interest

- COLEACP PIP Guide to good crop protection practices for pineapple. [http://pip.coleacp.org/files/documents/GBPP-ananas%20bio%2004-2011-02-1-UK\\_0.pdf](http://pip.coleacp.org/files/documents/GBPP-ananas%20bio%2004-2011-02-1-UK_0.pdf)
- Costa Rica: Green light for Del Monte's transgenic pineapples. [http://www.freshplaza.com/news\\_detail.asp?id=88734](http://www.freshplaza.com/news_detail.asp?id=88734)
- Genetically transformed pineapple. <http://www.google.com/patents/US5952543?printsec=abstract#v=onepage&q&f=false>
- FAO Manual Pineapple: Post-Harvest Operations. [http://www.fao.org/fileadmin/user\\_upload/inpho/docs/Post\\_Harvest\\_Compedium\\_-\\_Pineapple.pdf](http://www.fao.org/fileadmin/user_upload/inpho/docs/Post_Harvest_Compedium_-_Pineapple.pdf)
- Infonet-biovision. Crops, fruits, vegetables. <http://www.infonet-biovision.org/default/ovvImg/-1/crops>.
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- Organic pineapple farming in Ghana – A good choice for small holders. <http://www.pegnet.ifw-kiel.de/activities/research/results/kwp-1671.pdf>
- Re vista mensual sobrel a actualidad ambienta I ISSN 1409-214X N° 1 58 Noviembre 2006. Piña en Costa Rica: producción y ambiente. Pp. 20. <http://www.ambientico.una.ac.cr/158.pdf>.
- Stop Del Monte GM pineapple. <http://www.foecardiff.co.uk/content/stop-del-monte-gm-pineapple>.
- The International Tropical Fruits Network (TFNet, <http://www.itfnet.org/>) announced the return of its newsletter, Tropical Fruit Net. Starting in 2012, Tropical Fruit Net will be produced every other month and will include the latest news, projects, features, and events. Contributions are welcomed from individuals, especially from TFNet member countries, on articles related to production, market and consumption of tropical fruits.
- Training guides for MD-2 pineapples. [http://pdf.usaid.gov/pdf\\_docs/PNADP290.pdf](http://pdf.usaid.gov/pdf_docs/PNADP290.pdf) ; [http://pdf.usaid.gov/pdf\\_docs/PNADQ034.pdf](http://pdf.usaid.gov/pdf_docs/PNADQ034.pdf), [http://pdf.usaid.gov/pdf\\_docs/PNADQ035.pdf](http://pdf.usaid.gov/pdf_docs/PNADQ035.pdf)
- Transgenic plants with modified carotenoid levels. <http://www.ipaustralia.com.au/applicant/del-monte-fresh-produce-company/patents/AU2003293407/>
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## New References on Pineapple

The list below includes papers related to various aspects of pineapple culture, physiology, processing, preservation or byproducts that were published or located since the last issue of the newsletter was printed. Some papers may seem relatively unrelated to pineapple but the list follows the principle of inclusion to provide the widest possible content. Often, abstracts of the papers listed below can be found on-line and of course all abstracts of paper published in *Acta Horticulturae* are available from [info@ishs.org](mailto:info@ishs.org).

Abdullah, H., 2011. Quality maintenance of pineapple in postharvest handling. *Acta Horticulturae* 902:403-408.

Abraham, E., Deepa, B., Pothan, L.A., Jacob, M., Thomas, S., Cvelbar, U., and Anandjiwala, R., 2011. Extraction of nanocellulose fibrils from lignocellulosic fibres: a novel approach. *Carbohydrate Polymers* 86:1468-1475.

Abrahamian, P. and Kantharajah, A., 2011. Effect of vitamins on in vitro organogenesis of plant. *American Journal of Plant Sciences* 2:669-674.

- Adebisi-Adelani, O., Oyedele, O.O., Olajide-Taiwo, L.O., Olajide-Taiwo, F.B., and Fabiyi, A.O., 2010. Research-extension-farmer collaborative linkage on horticultural technologies in South West Nigeria: a case of NIHORT adopted villages. *Middle East Journal of Scientific Research* 6:157-161.
- Adisak, J. and Jintana, J., 2011. Chemical properties and sugar contents in 'Smooth Cayenne' pineapple fruit during maturation. *Acta Horticulturae* 902:423-426.
- Agbangba, E.C., Olodo, G.P., Dagbenonbakin, G.D., Kindomihou, V., Akpo, L.E., and Sokpon, N., 2011. Preliminary DRIS model parameterization to access pineapple variety 'Perola' nutrient status in Benin (West Africa). *African Journal of Agricultural Research* 6:5841-5847.
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Tables and graphs: Submit tables in Word format or as spreadsheet files. When submitting graphs, provide the original file or submit as a graphics file (jpg, png or other format).

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