



# NEWSLETTER

February 1986

**TAMPA BAY CHAPTER of the  
RARE FRUIT COUNCIL INTERNATIONAL, Inc.**

EDITORIAL COMMITTEE: Bob Heath  
Theresa Heath  
Arnold Stark  
Lillian Stark

NEWSLETTER MAIL ADDRESS: Arnold & Lillian Stark  
6305 Eureka Springs Rd.  
Tampa FL 33610

PRESIDENT: Arnold Stark CHAPTER MAIL ADDRESS: P.O. Box 260363, Tampa FL 33685  
(including renewals)

MEETINGS ARE HELD AT 2:00 P.M. ON THE 2nd SUNDAY OF THE MONTH.

NEXT MEETING. . . . . FEBRUARY 9, 1986

MEETING PLACE . . . . . COMMUNITY ROOM UNDER WEST RAMP, TAMPA BAY  
CENTER SHOPPING MALL, BUFFALO AND HIMES  
AVENUES NEXT TO TAMPA STADIUM. (TAKE DALE  
MABRY TO BUFFALO AVE., AT STADIUM.)

PROGRAM . . . . . CHRIS B. ROLLINS, naturalist, and Director  
of the Preston B. Bird and Mary Heinlein  
Fruit & Spice Park in Homestead, will speak  
on the subject of tropical vegetables. This  
topic will prove most fruitful to many of us,  
as we expand our common interests to include  
non-woody unusual plants.

NEW MEMBERS: Dr. & Mrs. Tran Nhu Chuong  
10506 N. Dixon Ave.  
Tampa FL 33612  
933-5622

Hard as it is to believe, your Newsletter committee made a boo-boo (please get up from the floor!) in page-numbering our last issue. We forgot that January starts the New Year (dum-de-dum-dum!) So, for those of you who save these invaluable manuscripts (would anyone even dream of not saving them - perish the thought!), please renumber the pages, in order, from 85-91 through 85-98 to 86-1 through 86-8. You may also wish to change the date of the issue from January 1985 to January 1986, and the meeting date to January 12, 1986. Our only excuse is that "time flies when you're having fun!"

\* \* \* \*

### NEW TREES

The Club has received twenty trees from a wholesale nursery in California, ten Li Chinese Jujubes, five Chocolate Persimmons, and five giant Fuyu Persimmons. These trees cost the Club \$12.50 each. If any of the Club members wish to purchase one or more of these trees at cost, contact Bob Heath. The trees were received bare root and have been planted in three gallon containers. They should be replanted in the ground before spring or left in the containers until next winter.



## THE USE OF TISSUE CULTURE FOR THE PROPAGATION OF TROPICAL FRUIT TREES

by Dr. Richard Litz

The essence of Dr. Litz's talk today was the factual aspect of plant tissue culture not only from the propagation standpoint but also for how it can be effectively utilized in conjunction with a conventional plant breeding program. Specifically, the papaya breeding program of the University of Florida involves a number of years of work by Dr. Robert Conover, who died 3 years ago. He began his work in 1974 and Dr. Litz joined his program in 1977 to utilize tissue culture systems to complement some of the intransigent breeding problems that Dr. Conover had encountered.

There is a disease which has decimated papaya production in south Florida, throughout the Caribbean region, in South America and in the Far East. It is called Papaya Ringspot Virus and causes symptoms on papaya as was presented to us by his first slide. The slide showed a grove of papayas approximately 4 months old. We could see that the leaves which had developed at the top of the plant were badly deformed, twisted and consisted primarily of the leaf veins with very little of the surface of the leaf having developed. Very little fruit will develop and that which does is markedly inferior. The next slide showed the fruit marked with a small ringed pattern on the surface which contributes the name ringspot to the disease. For many years people were confused about this particular disease because, as we know, papaya is a seed propagated plant and it is very difficult for virus to be transmitted thru seeds. The ringspot disease seems to occur spontaneously in many parts of the world where papayas are grown. Within the past two years, it has been determined that the papaya ringspot virus is the same virus which causes Watermelon Mosaic Disease, which is a disease of the cucurbit family. Since watermelons, squashes and the like are grown throughout the world, it is easy to see how the virus could spread so easily to papayas.

At the time Dr. Conover began his breeding program with papayas it had already been determined that papayas had very little resistance to ringspot virus. After a number of years of bringing papaya varieties to the Homestead Research Center from all around the world and inoculating these plants with sap from infected plants, Dr. Conover determined that there was a batch of seeds which came from the northwest corner of Columbia that did appear to have a small level of disease resistance. After a number of years of in-breeding this variety, he developed a cultivar with round fruit and precocious bearing period. It bears at virtually the first flower, very close to the ground. It has been named the Cariflora papaya. Cari from carica, the papaya species and from the Caribbean, and flora of course, for Florida. Because in working with the Papaya Ringspot Virus and the Cariflora papaya, the virus frequently killed the papaya at 3 or 4 months of age, and in order to lengthen the time that they had to work with a particular tree, they developed a method based on the culture of the shoot tip, not the meristem but the shoot tip.

His next slide showed the shoot tip from a female papaya. Papayas have very interesting sex types. There are female plants, male plants, hermaphrodite plants and neuter plants as well as some that seem to display characteristics of two types. It is not unusual, particularly in cold weather, for male plants to develop female flowers and therefore have fruit.

What they did was remove the growing shoot tip of trees in the field and place them on a tissue culture medium containing fairly high levels of auxin. Auxins are plant growth regulators which do a number of things to plant tissue. They stimulate the formation of callus which is cell proliferation, they induce the formation of roots and also stimulate the growth of the apex. After a couple of months of treatment, the culture is then changed to a medium which contains low levels of auxin, and moderately high levels of cytokinin, which are another kind of plant growth regulator. Cytokinin is a type of growth regulator which actively inhibits the growth of the shoot apex and stimulates the growth of the axillary or lateral bud.

The slide which Dr. Litz displayed next showed a mass of growing points of the plant. In the particular cluster shown, there were between 100 and 200 actively growing shoot tips, all of which have the potential of forming a mature plant. If each of these

growing shoot tips is removed from the culture and placed upon a medium with no growth regulators, then we can get a plant from each of the shoot tips. He then showed us a slide of one of the plants grown from the proliferating papaya cultures. These can be transplanted into a soil mixture with a loss of approximately 30%. The interesting thing about these plants is that although they came from the shoot tip of an actively growing mature plant, they have reverted to a juvenile form by passing through the tissue culture. This can be determined by the leaf form. Mature papaya leaves have five lobes. Juvenile leaf form is three lobed. This procedure may be used in a practical sense to propagate male plants for placing in the field. The reason for this is that the Cariflora papaya which they have developed for South Florida has both male & female forms. One of the problems which Dr. Conover had was that although he could tell very quickly what type of fruit characteristics would be bestowed by the female parent, he could never be sure what effect the male parent would have on the fruit's characteristics. So they decided to clonily propagate male plants for two or three years at a time, hold the male parent constant and each year upgrade the female parent and in this way to speed up the breeding program. At this point, shoot tip cultures are not being exploited commercially with papaya to my knowledge. The Hawaiian Sugar Growers Association has established a two acre field of clonily propagated papayas. Interestingly, they have observed that when they propagate the solo papaya, which is generally the papaya you see in supermarkets, that about 5% of the plants that are derived from tissue culture undergo a change from hermaphrodite to female. We don't know whether this is good or bad but it is certainly interesting.

Another tissue culture procedure which they have utilized with papayas is the anther culture. As we know, the anther is the male portion of a flower. The anthers are enclosed within the flower buds of the male flowers on little stalks. When the anthers are ripe in mature flowers, they release pollen which travels thru the air to the female flower to provide fertilization. As the male flower bud develops at a certain stage when the flower is approximately 1 centimeter long, a process known as meiosis occurs. Meiosis is a process in germ cells which reduces the chromosome level of the cell by half. Papayas have a chromosome no. of 18 so the process of meiosis reduces the chromosome count to 9. What they do at this point under sterile conditions is to dissect the flowers and put the anthers into a medium which in this case is a liquid medium which includes activated charcoal. The charcoal is there to absorb the toxic products that develop during growth. In a short time, the anthers begin to develop little plants as we could see on a slide next presented. Generally, they get 1 or 2 plants on each anther. These little plants as they develop only have half the chromosome number that a papaya normally has. What is the purpose of producing a plant like this that has absolutely no value since it is completely sterile? Interestingly, by treatment of the meristems of these haploid plants with colchicine, the chromosome count will double so we can produce papaya plants with a normal 18 chromosome count. Now the interesting thing about these particular plants produced this way is that all of the pollen produced will be genetically identical. This will permit them to remove all of the variability that the male parent would bestow on a subsequent generation. One of the other tissue culture systems in which they have been very interested, involves affecting very wide crosses between papayas and other species within the same genus.

In the carica family, there are about 16 or 17 species and most of these species are also susceptible to Papaya Ringspot Virus. However, there are three species which Dr. Litz had listed on his next slide which are resistant to Papaya Ringspot Virus. This resistance is a very special resistance. The Cariflora which we have been growing has a minor resistance. These three species, stipulata, pubescens, and cauliflora, are absolutely 100% resistant to this ringspot virus. They would like to transfer into papaya that resistance which is known to be caused by a single gene. This should make it a simple process to transfer that gene into the papaya species. With a straightforward breeding program, we should be able to get in a few generations a very interesting papaya. Dr. Litz then showed us some slides of these other carica species which look very much like papaya. Carica stifiolata, which grows in Central America, Carica Pubescens, native of the Andes, and Carica Cauliflora, also a native of the Andes and which grows in South Florida very well.

As the seed of a papaya develops, the embryo of the seed seems to occupy the center of the seed, surrounded by a very hard jelly like mass called the endosperm. The reason why papaya cannot be crossed with these other carica species naturally is because when the cross is made, the endosperm does not form, and since the endosperm is the food that the embryo needs to develop, the embryo will not develop and we say that the two species are sexually incompatible.

Dr. Litz next showed us a slide of the seed of a papaya crossed with *Carica Cauliflora* as the male parent. The seed was dissected so that we could see the embryo and the fact that no endosperm was apparent. Without the endosperm, the embryo will abort in approximately 60 days after pollination. In addition to a lack of endosperm, there is also poor pollination in the fruit and very few seeds develop. He showed us a slide of a fruit cut to display the small quantity of seeds at the upper end. They have attempted to avoid these problems by using tissue culture method for propagation of the crosses between papaya and the other ringspot virus immune species. One approach is to use one of the oldest tissue culture methods called embryo culture. It involves removal of the embryo from the seed of the cross and growing it on a tissue culture medium. After two or three months in the culture, one begins to see plantlets developing from the excised embryos of the hybrid plants. The main problem with this procedure is that it is extremely laborious as you need to dissect each seed separately and remove the embryo very carefully. Sometimes it works and sometimes it doesn't. They have modified the procedure somewhat by removing the intact ovules after they have made the pollination, sterilizing the ovules and placing them on a tissue culture medium. After approximately 40 days, the seed coat bursts open and releases a whole mass of embryos which develop fairly normally with cotyledons.

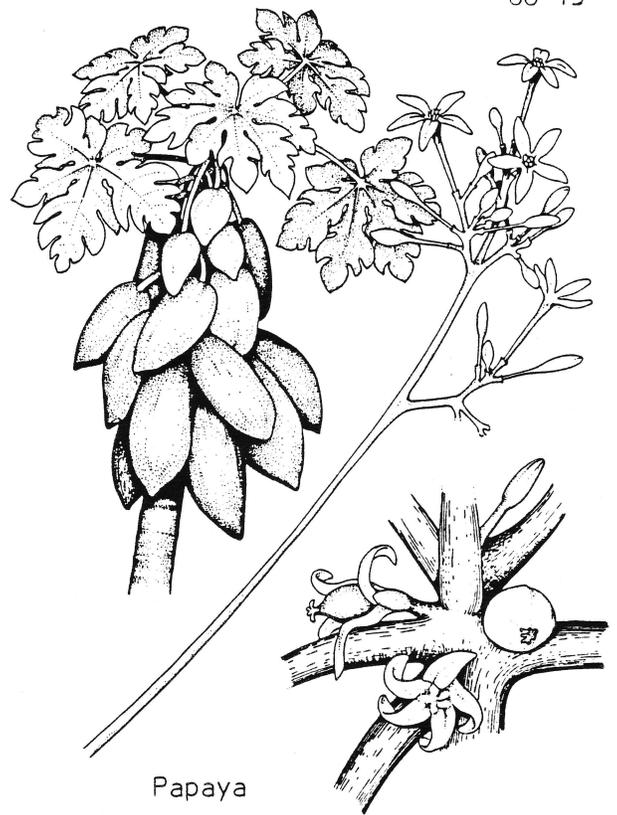
We next saw a slide that showed various stages of embryo development on the medium. At a further stage of development, we saw the plantlets with fully developed stems, root systems and leaves. At this stage, they can be transplanted into a potting mixture and continue their development in the greenhouse.

He next showed us an example of the mature tree with fruit. Unlike the papaya, it has very elongated fruit which is inedible. How discouraging, although it may be usable for cooking but for very little else. But it is resistant to the ringspot virus.

Other aspects of tissue culture which they became involved with using the papaya, involves the isolation and culture of protoplast. Protoplasts are plant cells from which the cell wall has been removed. Plants have cells which are enclosed in a cellulose wall, like tissue paper surrounding a little ball of protoplasm. The cell walls are there to keep the plant cell from bursting. In order to get involved with protoplast technology, one has to work out relatively basic systems of developing plants from the type of tissue that one is interested in. In their case, they were interested in the very young leaves of papayas. So initially, what they had to learn was how to control the development from papaya leaves. This is a relatively laborious procedure wherein you cut out small sections of papaya leaf and put them onto tissue culture medium containing perhaps 500 different combinations of auxin with or without cytokinin. Depending upon the balance of auxin with cytokinin, very different kinds of things can happen. First, if the auxin concentration is correct, a callus will form. If the auxin concentration is high, roots only will form. If the cytokinin concentration is high, something different occurs. Instead of roots developing from the callus, there are shoots and leaves developing. Dr. Litz showed us slides of both situations. In this way, it is possible to learn to control the development from a papaya leaf.

Now that we know how to control the growth of plants from papaya leaves, we cut papaya leaves in thin strips and float them on this brown solution which contains enzymes, cellulase and pectinase. These are enzymes which are produced by fungi which will break down or degrade plant cell walls and in order to keep this process from destroying the cells, it is necessary to add a lot of sugar to the mixture to keep the cells from bursting when the cell walls are removed. You do this in a flask which is shaken for maybe 3 or 4 hours, very gently. Then you pass the residue from the leaf strips thru a filter, wash it very carefully & then centrifuge it & you will collect a little pellet of papaya leaf protoplasm. The protoplast is in the upper neck of a specialized centrifuge vessel. He showed us a slide of the protoplast suspended in the solution containing a high sucrose

concentration. These are then mixed under very controlled conditions with protoplasts or other *Carica* species such as *Carica Stipulata*. He would like to make this hybridization for example, because it has never been done naturally and to get a hybrid plant. Not only do they want to use protoplasts for making these very wide inter-species crosses, but in order to do any genetic engineering with the papaya or any other plant, it is necessary to be able to expose the naked protoplast with DNA from another organism. Because the DNA from another organism is unable to pass through the cellulose outer coating of the cell, it is necessary to remove that cell wall and expose the protoplast and allow the DNA to get in and modify the plant cell. This will set the stage for genetic engineering to take place with papaya within the next ten years.



Papaya

\* \* \* \*

#### HONDURAS TOUR

Our next speaker, Chris Rollins, director of Homestead's Fruit & Spice Park, is joining forces with Tom Economou in leading a Tropical Fruit Tour of Honduras. The next tour is scheduled March 16 - 22, 1986, and costs \$639.00 per person. Other departures are scheduled for April and May. For more information, contact Tom Economou at Nature Trail, Inc., P. O. Box 450662, Miami, Florida 33145. Telephone No. (305) 285-7173.

#### JANUARY PLANT RAFFLE

Plant	Donor	Winner
Chinese Parsley	RFCI	?
Pitaya Cactus	RFCI	B. Ryland
White Sapote, Dade Var.	A. Mendez	C. Gomez-Sanchez
Abaca Pineapple	A. Mendez	Stark
Podacarpus	Connie Vernon	J. Bell
Alde	Connie Vernon	K. Netscher
Guava	RFCI	Bruce Beasor
Dorsett Apple	RFCI	A. Mendez
Pigeon Pea	Bob Heath	C. Bassham
Opuntia Cactus	Grace Calhoun	Pearl Nelson
Pomegranate	Herb Hill	?
Chayote	Priscilla Lachut	Lloyd Shipley
Black Sapote fruit	A. Mendez	John Bell
Chayote	Lachut	Glen Myrie
Chayote	Lachut	John Bell
Chayote	Lachut	Glen Myrie
Chayote	Lachut	D. Lee
Coriander	?	Bobbie Puls
Boquet Pepper	Lottice Shipley	Glen Myrie

A MESSAGE FROM THE PRESIDENT

I would like to thank Dr. Richard Litz for his interesting and informative talk. I was especially interested in hearing of all the different tropical fruits his laboratory is growing in tissue culture, for eventual propagation or genetic engineering. Perhaps in the foreseeable future, we will be able to grow all of our favorite tropical fruits without worry, for they will have had inserted into their genome genes for coldhardiness!

I have just received from Betty Dickson, our Membership Chairperson, a list of names of members whose membership has lapsed. These names are to be dropped from our mailing list, as they have not responded to a written renewal request. From past experience, I suspect a number of these will send in renewals, and we will then have to go through the effort of reinstating their names into our mailing list. This is extremely inconvenient to those people actively involved in the running of our organization, in spite of their already very busy schedule. Please prevent this unnecessary waste of time and energy by **RENEWING YOUR MEMBERSHIP PROMPTLY!** If you plan to discontinue membership, please notify the membership chairperson. Check with the membership chairperson as to when your dues are due, it is marked in the book where your nametag is kept. Our membership year end is March 31, 1986. All renewals will henceforth be on the same schedule, running from April through March. Renewing promptly will not only be of help to us, but will also prevent your missing any of our newsletters, a fate no one should suffer! Thank you.

That's it for now. See you next meeting!

\* \* \* \*

RARE FRUIT SEMINAR

The Fourth Annual International Rare Fruit Seminar is tentatively scheduled for July 19 & 20, 1986, and will be hosted by the Rare Fruit & Vegetable Council of Broward County. Those of our members who attended the last seminar in Fort Myers reported it was an event well worth experiencing. If you wish to attend, keep these dates open. We will need an official representative to the seminar. Any volunteers? Updated information will be provided as it is received from RFVCBC.

\* \* \*



"A typical RFCI member's problem  
— green lung!"

adapted from American Scientist,  
March-April, 1980

## HOSPITALITY TABLE:

Priscilla Lachut: Chayote & Dip,  
Papaya Cubes  
Marian Austin: Tangelo Muffins  
with seedless Blackberry  
Preserve  
Connie Vernon: Oatmeal & Raisin  
& Almond Cookies  
Bea Seekins: Cranberry Bread  
with Apricots & Raisins  
Bruce Beasor: Calamondin Bars  
Leland Terrell: Kumquat Chips

YOUR contribution is needed for  
the February Hospitality Table.  
Please volunteer - call Pris-  
cilla Lachut to let us know  
what you can bring to the next  
meeting.

## RECIPE OF THE MONTH

Tangelo Muffins (Marian Austin)

2 cups self rising flour  
½ cup oil  
3 Tbs sugar  
1 cup Tangelo juice \*  
3 Tbs grated Tangelo rind \*  
1 egg  
1 tsp salt (optional)  
chopped nuts (optional)

\* Orange juice and rind may be substituted.

Mix all ingredients. Fill small muffin tins 3/4 full. Bake at 450° for 12 minutes.  
Serve with butter or jelly. Yields 24 small muffins.

\* \* \* \*

A SWEET HISTORY (Excerpted from Florida Market Bulletin, 28:24, 12/85)

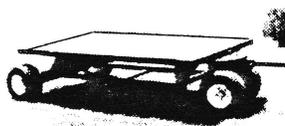
Cane sugar usage has a long history, dating back to some 8000 years, the origin being in the South Pacific. The plant was eventually moved northward to Southeast Asia and India. It was specifically mentioned by name for the first time in 325 B.C.E., by an officer in Alexander's invading army in India, where it may have been growing for several centuries previously. Sugar cane cultivation and refining spread east to China about 100 B.C.E. It was brought to the western world around the 13th century by Venetian merchants, and by 1300, Venice was the sugar capital of the world.

The Spanish introduced sugar cultivation into their colonies in the Canary Islands, from which Columbus brought cane to the Caribbean on his second voyage in 1493. By 1550, sugar had become one of the most valuable products on the island of Hispaniola (now Haiti and the Dominican Republic), and a pirate's prize. England and France established their own refineries in the 1600's to handle sugar from the West Indies.

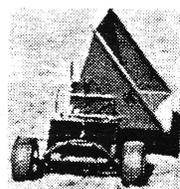
Sugar cane has been grown in Florida since the days of the Spanish occupation. Florida is now the nation's leading state in sugar cane production, with some 371,800 acres planted this year. It is an important industry, creating over 42,000 jobs, accounting for over \$1 billion of economic activity, and supplying the country with 17% of its total sugar needs.

\* \* \* \*

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## FLORIDA NATIVE FRUITING TREES No. 5

PITHECELLOBIUM *Pithecellobium dulce*

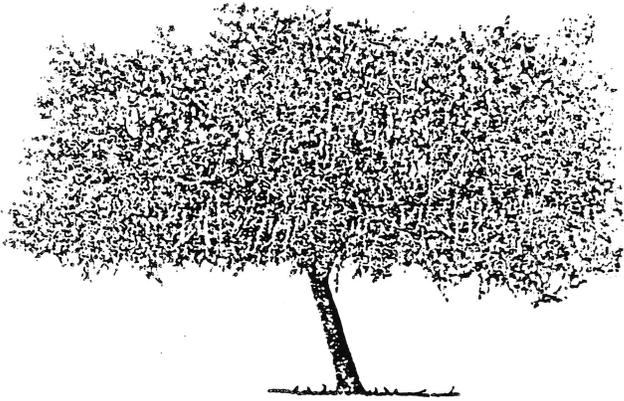
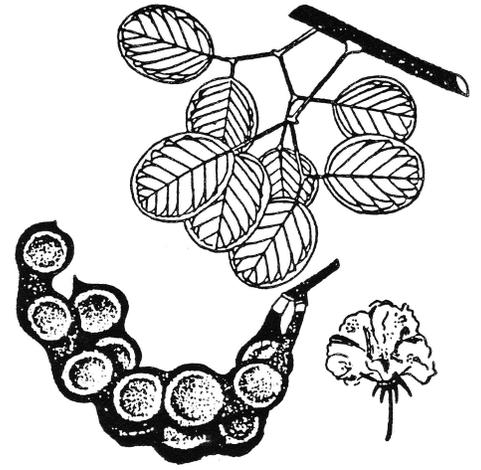
Also called Manila Tamarind, Huamuchil & Kamachile.  
Often listed as Pithecolobium.

Member Pea Family (Leguminosae).

Tropical American tree native to Phillipines, Mexico and Central America growing 50 to 50 ft. in height with spreading crown for which it is highly prized in South & Central Florida as a shade tree. It is a fast-growing tree and generally immune from pests. Twigs spiny.

Leaves light green, feathery, composed of small, one & one-half to two-inch leaflets on a common stem, and presenting a lacy appearance.

Tiny, frothy white flowers borne in round-headed clusters with many stamens, followed by reddish fruit pods about 1/2 inch wide and 4 to 7 inches long, and coiled like a ram's horn. Filled with blackish-brown edible seeds.



This species is especially suitable for inland areas in Florida where the dry, sandy soil is unsuitable for growing shade trees, especially the lush tropicals and sub-tropicals and it also is hardy in the northern part of the state. In regions with rich, moist soils it responds with vigorous growth. It should not be planted near the beach as its leaves are burned by the salt spray.

Besides the edible fruit, the tree yields good timber, a yellow dye and a mucilaginous gum.

Tampa Bay Chapter  
Rare Fruit Council International  
P O Box 260363  
Tampa FL 33685



P. JUDSON NEWCOMBE  
314 DEER PARK AVE.  
TEMPLE TERRACE, FL 33617