The Tristeza Disease

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Many centuries ago a single microscopical entity cried for independence and freedom to conquer. Built like a chromosome and possessing the incredible power of self multiplication, this thread-like particle abandoned its abnormal cell to become a famous world-wide virus, responsible for the most destructive disease of citrus. Carried by men and aphids, it spread first through the Southeast Asia regions, then to South Africa and from there it reached South America in the first decades of this century. About this same time the virus was introduced in the United States. It is believed that with time the tristeza virus will take over all existing citrus trees in the entire world.

Entering many citrus areas, the invader has caused extensive destruction to orchards budded on the intolerant sour orange rootstock. Certainly, an accumulated total of more than 20 million citrus trees have already been killed and many thousands are declining.

The total number of citrus trees presently existing in the world is around 400-500 million plants and perhaps 50 percent are carrying the tristeza virus. Among the healthy trees, over 100 million are budded on tristeza intolerant rootstocks, mainly sour orange.

In a general view, the citrus areas of the world may be currently divided today into 3 regions or classes in relation to tristeza.

Region I - Areas having practically all citrus trees infected with the virus:

Southeast Asia
Australia
Africa South of Sahara
Four South American countries (Argentina, Brazil, Paraguay, and Uruguay)

The black aphid *Toxoptera citricidus* Kirk, the most efficient vector of tristeza, occurs in fairly high populations apparently in all of these areas. The total citrus presently existing in this region is estimated at around 200 million trees, all of them, of course, budded on tristeza tolerant rootstocks.

Region II - Areas not yet infected by the virus, but where most trees are susceptible to the tristeza disease:

Mediterranean countries (except Spain and perhaps Morocco)
Mexico and certain areas of the United States
Central America
South American countries (except Argentina, Brazil, Paraguay, Uruguay and Peru)

Sour orange is used extensively in the citrus areas of these countries, the number of tristeza intolerant trees amounting to more than 100 million. Isolated declining trees have been found infected with the virus within this region, but no evidence of field spread was noticed. Black aphid has not been reported here, but other aphids exist. It is totally unpredictable how much longer these citrus areas will remain untouched by tristeza.

Region III Citrus areas where tristeza virus is known to be spreading slowly:

Peru - a large part of the coastal area of this country is already infected and the virus has been encountered in the Amazon area of the Chanchamayo River. The black aphid is present in Peru.

United States - slow spreading of the virus is occurring in Florida and California. Tree losses have been quite limited, particularly in Florida, because most orchards are budded on tolerant rootstocks.

Spain - spreading of the virus is occuring in the famous orchards of the Valencia area. A high proportion of the trees in this country is budded on sour orange rootstock.

Morocco - a number of declining trees was found recently in this country, exhibiting symptoms resembling those of tristeza. Further studies may indicate a slow spread of tristeza in Morocco (personal communication)

Dissemination of the tristeza virus in these areas, except for Peru, is made by *Toxoptera aurantii, Aphis gossypii, Myzus persicae* and other aphids. These are inefficient vectors, but the rate of spread of the virus frequently accelerates with the increase of inoculum in an area. Replantings are being made on tolerant rootstocks.

Since the great potentialities of the virus for causing destruction were fully realized, extensive investigations and efforts have been dedicated to the study of tristeza and its control. As a result, man has learned to grow citrus in the presence of the virus. Sour orange and several other citrus types have had to be abandoned as rootstocks for budding most commercial citrus varieties. To live with tristeza is not an easy task. Several important varieties are affected by the virus independent of the rootstock and the productivity and vigor of the tree are reduced. These trees exhibit more or less severe stem pitting symptoms according to the virulence of the infecting virus strain. Fortunately, pre-immunization of mother trees of partially intolerant varieties with selected mild strains is proving to be successful in preventing greater damage in the orchards.

Existing in a number of million entities in each tree, virus mutants occur rather frequently. One of these mutants particularly destructive to sweet orange varieties and to the Rangpur lime rootstock, was recently found in Brazil, in the Capao Bonito area of the State of Sao Paulo. Mutants capable of severely affecting other scion and rootstock varieties may eventually appear in this area or others of the world. Continuous research is the only guarantee that citrus growers can have of a successful citrus industry in the areas where tristeza virus has become endemic.

The Tristeza Virus and Disease Symptoms

Scientists realized that the decline disease invading citrus orchards of the Americas was caused by an infectious pathogen sometime in the beginning of this century. By 1942 the disease was named tristeza by Sylvio Moreira in Brazil. The virus nature of the disorder was revealed in 1946 by Fawcett and Wallace in the United States and by Meneghini in Brazil. The first electron micrograph of the virus showing its thread-like particle about 2,000 mμ in length and 10-12 mμ in diameter was made by Kitajima and co-workers in 1964 in Brazil.

Research conducted in various citrus areas of the world has shown that the tristeza virus must exist in a multiplicity of strains ranging from extremely mild to very severe, according to the degree of symptom expression on infected test plants. Cross-protection tests with selected virus sources has resulted in a certain protective effect. However, virus strain mixtures appear to occur within a given host plant. Virus strains with increasing severity have been named T_1, T_2, T_3, ..., T_n and apparently the more severe strains are always accompanied by the milder strains. For instance, a tree infected with T_3 also yields T_2 and T_1. It has been suggested that these strain components may occur in trees in different proportions inducing variable degrees of symptom expression among infected trees and within branches of a tree. The explanation for the presence of virus strains within a certain tree, according to Grant and Higgins, appears to be related to the failure of any 1 strain to become thoroughly systemic, thus affording susceptible sites for the development of other strains.

All virus strains of the tristeza complex probably have been introduced in most citrus producing areas. However, strain components and disease expression vary from country to country. This behavior is being attributed mainly to climatic conditions, differences in host reaction and, to a larger extent, differences in the ability of aphid species to extract and transmit strains of the tristeza complex.

There are 3 major recognized disease symptoms caused by the tristeza virus: stem pitting, typical tristeza decline, and seedling yellows. Tristeza stem pitting occurring in grapefruit and many other citrus types is characterized by the development of longitudinal pits or furrows in the xylem surface of trunk and branches. Diseased trees show poor, bushy growth, decreased vigor and sometimes distorted fruits. 'West Indian' lime, extensively used as indicator plant for tristeza, develops vein flecking, vein clearing and eventually leaf cupping in addition to stem pitting. Stem pitting is induced in intolerant hosts by very mild, mild and severe tristeza strains.

Typical tristeza decline of infected sweet orange or mandarin trees budded on sour orange rootstock have many features. Vein yellowing, weak flushes, dieback, thickening of the bark at the union and honeycombing from the surface of the wood of the sour orange stock are some of the more characteristic symptoms of the disease. According to Schneider, the virus causes sieve-tube necrosis in the sour orange stock just below the union, thereby girdling the tree. In consequence, there is no downward passage of carbohydrates, the reserves of starch in the stock are consumed and the roots decay, followed by the decline and death of the scion. The decline may progress quickly in 2 to 4 months, affecting all the foliage, and thereafter death of the tree; or it may take 12 months or more to succumb. Mild and severe strains of the virus cause this type of tristeza.

Seedling yellows is a reaction of certain citrus types having extremely intolerant tissues, to the artificial infection with severe strains of the virus. It does not occur naturally in the field. Young seedlings of sour orange, lemon and some other citrus, when inoculated with severe tristeza by budding, develop the yellows reaction characterized by severe stunting and yellowing of the leaves. New leaves are often small, yellow and distorted with corky eruptions along the midrib and veins. The diseased seedlings may or may not recover their normal appearance.

Most authors regard tristeza as a single virus with numerous strains, the difference in symptomatology attributed to different host reactions to the various strains, as explained above. However, some authors consider that "2 distinct viruses are associated with the tristeza virus complex", but no one has been able so far to separate these 2 components.

Host Reaction to Infection

Reactions of the various species and varieties of citrus and citrus relatives to tristeza infection depend largely on the capacity of the plant to allow multiplication of the virus and to the degree of tolerance of the tissue to presence of the virus. Accordingly, citrus types and relatives may belong to 1 of the following classes:

1. Extremely intolerant types

Citrus types and relatives included in this class have tissues extremely sensitive to infection by the tristeza virus. Common virus strains cannot establish themselves in the plant, probably because invaded cells die readily before multiplication can take place. Grown as seedling plants or scions they behave as resistant types, but a few cells are certainly killed every time an aphid inoculates the virus. Thus the hypersensitivity of these cells protects the whole plant.

Extremely mild strains may, however, become systemic in plants of this class and then slight stem pitting may occur.

If extremely intolerant types are used as a rootstock, and large amounts of the virus descends from the top with the sap, many phloem cells are killed below the union and tristeza symptoms are shown by the tree. Young seedlings of these varieties exhibit seedling yellows reaction when bud-inoculated with severe strains of the virus, provided enough inoculum is introduced to cause sieve-tube blockage.

Sour orange and lemons are the best representatives of this class. Citremon, certain varieties of shaddock *(C. grandis)* and possibly *Severinia buxifolia* also belong to this class.

They can be successfully used as rootstock only for citrus types belonging to this same class.

2. Intolerant types

Types of this class have tissues intolerant to the virus, particularly to severe strains. Mild and very mild strains of the virus induce stem pitting symptoms on them, while more severe ones kill the cells and do not become systemic in plants of these types. If used as rootstocks and inoculated with large amounts of severe strains of the virus, trees develop typical tristeza symptoms. This type of behavior is shown by grapefruit, most shaddock cultivars, some tangelos and possibly other citrus types. Severity of stem pitting varies within varieties of these types, suggesting genetic differences in the degree of susceptibility to the virus. Some of the more intolerant ones may also show seedling yellows symptoms.

3. Partially intolerant types

Citrus types included here have tissues that allow multiplication of the tristeza virus complex and are affected by the virus. They all show stem pitting symptoms that are more or less numerous according to the severity of the infecting virus strain and the degree of susceptibility of their tissues. There is a wide range of susceptibility among types belonging to this class. Trees budded on some of the more intolerant types of this class decline and die. Among types representative of this class, according to decreasing degree of intolerance are Acid limes, Papeda group, certain tangelos, several lemon hybrids, Limequats and kumquats, calamondin, citrons, sweet limes, certain citranges, some tangors, and certain sweet orange varieties.

Some of the types included in this class have a good performance as rootstocks in spite of a certain amount of stem pitting, for example 'Morton' citrange.

4. Tolerant types

Plants of citrus types belonging to this class allow the multiplication of the tristeza virus strains and are little or not affected by their presence. Examples of this class are tangerines, most sweet oranges, most tangelos, most tangors, rough lemon, 'Rangpur' lime and other mandarin limes, and *Citrus volkameriana.*

Tangerines are among the more tolerant types and rarely exhibit stem pitting. Sweet orange cultivars have varying degrees of tolerance and may show stem pitting in the presence of severe strains of the virus. Citrus types listed in this class when used as rootstock, generally show good performance.

5. Resistant or immune types

Trifoliate orange and some of its hybrids have been found to have tissues apparently resistant or immune to tristeza. The virus cannot multiply in the tissues of these plants and it is not recovered from seedlings sometime after inoculation. Used as rootstocks, they are not affected by the virus coming down with the sap to any visible degree.

Rootstocks for Tristeza Infected Areas

Sour orange and other intolerant rootstock varieties have had to be abandoned in the tristeza-infested areas. They have been replaced mainly by rough lemon, 'Rangpur' lime, sweet orange, trifoliate orange and trifoliate orange hybrids and 'Cleopatra' mandarin. None of these rootstocks have, however, all the desirable characteristics shown by sour orange. In addition, mutants or variant strains of the virus may affect trees on these so-called tolerant rootstocks. The decline of sweet orange trees on trifoliate orange in Argentina is attributed to a new strain of the tristeza virus. In Brazil, the Capao Bonito variant affects trees of sweet orange and tangerines on 'Rangpur' lime rootstock. The search for new rootstock types that are tolerant to tristeza is a continuous need in the areas where the virus has become endemic.

Several hundred citrus types and relatives have been tested as rootstocks for sweet orange in Brazil. Table 1 lists 301 of these types, of which nearly 60 percent have been found intolerant to the virus. They include several species and varieties of *Citrus* and other genera, grouped as follows:

Type	*Tolerant*	*Intolerant*
Lemons and lemon hybrids	1	34
Acid limes	1	16
Sweet limes	0	9
Citrons	0	5
Rough lemons	3	1
Mandarin-limes	14	0
Tangors	9	1
Tangerines	34	0
Tangelos	17	11
Grapefruits	0	13
Shaddocks	0	22
Sour oranges	0	21
Sweet oranges	35	0
Trifoliate orange and trifoliate hybrids	18	3
Miscellaneous	8	25
TOTAL	140	161

Most of these types exhibit stem pitting in variable amounts except for sour oranges and true lemons which are extremely intolerant to the virus, the tangerines which are highly tolerant and trifoliate orange and some of its hybrids which are resistant. The presence of stem pitting indicates that the vigor and productivity of the plant is affected to a certain degree by the presence of the virus. However, certain types that are slightly to moderately pitted will give fairly good performance as a rootstock, such as 'Morton' citrange and *Citrus taiwanica* (Florida selection).

New citrus hybrids, especially those being produced and tested in California, may be found very valuable for the future of the citrus industry in the tristeza-infested areas.

FINAL REMARKS

A tremendous amount of effort and research work has been dedicated to the study of the tristeza disease. Rapid progress toward a comprehensive knowledge of the disorder has resulted from this intensive research on an international scale. Many scientists deserve mention for their great contribution to the understanding and control of the tristeza disease. Among them, I ask permission to point out the names of H. J. Webber, C. W. Bennett, T. J. Grant, H. Schneider, J. M. Wallace, E. O. Olson, W. P. Bitters, P. A. Norman, W. C. Price, L. C. Knorr, E. P. DuCharme, M. Cohen, and J. F. L. Childs from the United States; S. Moreira, A. S. Costa, M. Meneghini, A. A. Bitancourt, V. V. Rossetti, E. W. Kitajima, D. C. Giacometti, and G. W. Muller from Brazil; Lilian Fraser and L. L. Stubbs from Australia; A. P. D. McClean, R. E. Schwarz, and P. C. J. Oberholzer from South Africa; and K. Mendel and I. Reichert from Israel.

The tristeza problem is not coming to an end. On the contrary, much more work will be necessary from these and many other scientists throughout the world to control the great invader, the tristeza virus.

Table 1.

Reaction of citrus types and citrus relatives to tristeza virus infection when used as rootstock for sweet orange and their stem pitting expression.

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Lemons and Lemon Hybrids		
Acid		2
American		3
Amber	I	0
Armstrong		0
Bernia	I	0
Camargo		2
Cidra		2
Cowgill		0
De ba Ahmed		
Dehra Dun		
Deodoro		0
Des 4 saisons		0
Doce		0
Eureka		0
Genoa		0
Gigante		2
Harris		3
Harvey		2
Kulu		2
Kusner		
Lisbon Thornless		0
Lisbon Tetraploid		0
Morocco		
Meyer	I	
Periforme	Q	0
Perrine		2
Ponderosa	I	2
Punjab		
Siciliano		0
Vicosa		0
Villafranca		0
Woglum	I	0
PI-10785		
PI-126539		
PI-136469		
Acid Limes		
Abacaxe		3
Beledy		3
Cristal		2
Galego		3
Galego Thornless		1
Galego do Norte		2
Ingles		2
Kadu Mul		0
Kalpi		3
Acid Limes (cont.)		
Key		3
Kirk		
Marfim		2
Philipine	I	
Rio Claro	T	1
Seda		2
Selvagem		2
Tahiti		1
Sweet Limes		
Columbia		1
Dourada		1
Francana		1
Palestine Thornless		3
Persia		1
Sweet lemon		1
Teheran		1
Umbigo		1
Vermelha de Goias	(slow decline)	1
Citrons		
Commercial	I	2
Comprida		3
Diamante		2
Redonda		2
Sweet		2
Rough Lemons		
Brazilian	T	0
Florida	T	0
Deodoro		
South Africa	T	0
Mandarin Limes		
Cravo	T	0
Cravo doce	T	0
Japanshe citroen	T	0
Kusaie	T	0
Ling Ming	T	0
Otaheite lime	T	0
Philippine Red	T	0
Pook Ling Ming	T	0
Rangpur	T	0
Rangpur India C-26-1	T	0
Rangpur Otaheite	T	0

Table 1. (cont.)

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Mandarin Limes (cont.		
Rangpur D-33(40)	T	0
Rangpur Red Ling Mung	T	0
Rangpur Rose	T	0
Tangors		
Baia x Mexirica	T	0
Docinha S.J.R.		3
Mo	T	1
Murcott Honey	T	1
Reticulata	T	1
Sabara	T	0
Tangerona	T	0
Temple	T	0
Umatilla	T	0
653	T	0
Tangerines		
Batangas	T	0
Campiona	T	0
Chao Chou Tien Chieh	T	0
Clementine	T	0
Cleopatra	T	0
Cravo	T	0
Dancy	T	0
Kara (Frost)	T	0
King	T	0
King tetraploid	T	0
King of Siam	T	0
Kinnow	T	0
Kumembo	T	0
Mandarin 10630	T	0
Mandarin 117477	T	0
Mandarin 114212	T	0
Mandarin 114412	T	0
Mexirica do Rio	T	0
Nobilis (CPB-10642)	T	0
Oneco	T	0
Osceola	T	0
Pau	T	0
Ponkan	T	0
Satsuma, Owari	T	0
Satsuma, Wase	T	0
Suen Kut	T	0
Sun Chu Shu Kat	T	0
Sunki	T	0
Swatow 10031	T	0
Swatow 10032	T	0
Swatow 14054	T	0

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Tangerines (cont.)		
Weshart	T	0
Wilking	T	0
Willow leaf tetraploid	T	0
Tangelos		
Minneola	T	0
Orlando	T	0
Pina		2
Sampson	T	0
San Jacinto	T	
Seminole	T	0
Sunshine	T	0
Suwanee	T	0
Thornton		0
Umatilla	T	
Yalaha	T	2
Watt		1
Webber	T	3
Wekiwa		
Williams	T	0
Tresca gft x Dancy 126-5-17	T	
Tresca gft x Dancy 126-5-19		
Tresca gft x Dancy 126-6-18	I	
PI-18-H-6	T	
PI-18-I-8	T (?)	
PI-18-D-14		
PI-18-E-7		
PI-18-T-2		
PI-16-1-4		
PI-18-1-13	T	
PI-16-A-1	T	
PI-16-N-7	T	
PI-52018-W-2F		
Grapefruits		
Duncan		3
Foster		3
Leonardy		2
MacCarty		3
Marsh seedless	I	3
Pernambuco	I	2
Red Blush	I	3
Red Mexican	I	3
Royal	I	3
Ruby Red		3
Thompson	I	3

Table 1. (cont.)

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Grapefruits		
Triumph		3
Vicosa		2
Shaddocks		
Asia		
Chinesa		3
Cuban		2
Flemmings		
Hawaiian		
India Red		3
Inerme		1
Kao Panne		1
Melancia		2
Nakorn		
Lemon		
Ogami		2
Periforme		2
Pink		2
Rangpur x Sour		
Shatenyan		3
Siam		2
Siamese		3
Tau Yau		0
Thong Dee		
Vermelha		2
Zamboa		3
Sour Oranges		
Algiers Seville	I	0
Arancia Bizzaria	I	
Azeda thornless	I	0
Azeda Sao Paulo	I	0
Bergamia	I	0
Bigaradier	I	0
Stow Bittersweet	I	0
Daidai (PI-117471)	I	
Dummitt Bittersweet	I	0
Dummitt	I	0
Egyptian	I	0
Lanceta Amarga	I	0
Viradouro	I	0
Myrtifolia	I	0
Oklawaha	I	0
Paraguay	I	0
Rehovoth Palestine	I	0
Sauvage	I	0
Sour (U.S.D.A.)	I	1
Spain	I	0
Tunis	I	0

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Sweet Oranges		
Baianinha	T	2
Barao	T	2
Blood Oval	T	2
Buckeye Navel	T	1
Cadenera	T	
Caipira	T	1
Enterprise	T	
Florida sweet sdlg	T	1
Golden Nugget Navel	T	
Hamlin	T	1
Hart's late	T	1
Homosassa	T	1
Jaffa	T	2
Lamb Summer	T	3
Lue Gim Gong	T	1
Mediterranean Sweet	T	3
Mortera	T	2
Natal	T	1
Navelencia	T	3
Parson Brown	T	2
Pera	T	3
Pineapple	T	2
Piralima	T	2
Rehovot	T	1
Robertson navel	T	1
Rubi Blood	T	2
Saint Michel	T	1
Sanguinea	T	
Seleta Amarela	T	1
Shamouti	T	2
Surprise navel	T	2
Trovita	T	2
Valencia Late	T	
Washington navel	T	1
Westin	T	3
Trifoliate Orange and Trifoliate Hybrids		
Carrizo citrange	T	0
Citrumelo PI-4200	T	
Citrumelo PI-4475	T	0
Citrumelo PI-4477	T	0
Citrumelo PI-4482	T	
Citrumelo PI-4604	T	
EEL-203 Citremon	I	2
EEL-150 Citrangequat	I	2
Morton citrange	T	2
Rusk citrange	T	2
Sacaton citrumelo	T	0
Saunders citrange	T	2

Table 1. (cont.)

Citrus types and citrus relatives	*Reaction**	*Stem pitting***
Trifoliate Orange and Trifoliate Hybrids (cont.)		
Savage citrange	T	3
Thompson citrangequat	I	2
Trifoliate (small flower)	T	0
Trifoliate (large flower)	T	0
Trifoliate tetraploid	T	0
Troyer citrange	T	0
Uvalde citrange	T	0
Winter Haven citrumelo	T	0
Cunningham citrange	T	0
Miscellaneous		
Citrus celebica	I	2
C. excelsa	I	3
C. hystrix	I	
C. ichangensis	I	2
C. junos	T	3
C. karna	T	
C. longispina	I	
C. macroptera	I	
C. mitis (Calamondin)	I	3
C. macrophylla	I	3
C. tachebana Skek	T	0
C. taiwanica (Florida)	T	2
C. volkameriana	T	1
C. webberii	I	3
Citropsis sp.	I	
Severinia buxifolia	I	0
Severinia sp.	I	0
Microcitrus sp.	I	3
Micromelum tephrocarpa	I	
Poorman orange (grapefruit?)	I	
Calashu	T	
Kansu	I	
Cowgill Marcott	T	
Limue Khargi	I	
Misri Batabi	I	
Natsu mikan	I	
Satsumelo 10-V-3	T	
Nagami kumquat	I	
Meiwa kumquat	I	
Nippon kumquat	I	
Lakeland limequat	I	
Tavares limequat	I	
Lemonquat	I	

* Reaction of stocks to tristeza virus: I = Intolerant
T = Tolerant
Q = Questionable

** Stem pitting expressed in the scions of the various citrus types, was rated:
0 = none
1 = slightly pitted
2 = moderated pitted
3 = severely pitted