GRAPEVINE BREEDING AND GENETICS

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Keywords: biotechnology, botany, conventional breeding, DNA fingerprinting, embryo rescue, genetic engineering, genetic map, genetic resource, genomic, in vitro selection, micropropagation, marker-assisted selection, molecular marker, mutation, polyploidy, quality, resistance, tissue culture, tolerance, yield.

Contents

Summary

The grapevine is one of the most important economic fruit crops that are widely grown in almost all continents. The fruits produced worldwide are mainly processed into wine. In addition, significant portions have also been used for fresh consumption, dried as raisins or processed into juice. The objectives of grapevine breeding vary according to its usage and often are region-specific. However, most breeding programs aim at combining high yield and high fruit quality with improved resistance to multiple diseases and pests, and/or increased adaptation to adverse environments. These desirable traits can be exploited from the vast genetic resources of the *Vitis* genus. Different methods have been used to incorporate useful traits including conventional breeding, mutation and polyploid breeding, and biotechnological approaches. Integration of these tools will allow breeders to meet with the increasing demands for novel grapevine varieties with improved yield and quality in an era of limited resources, increasing health and ecological awareness, as well as increasing environmental pressures.

1. Introduction

The grapevine is an economically important woody perennial fruit crop, cultivated in most of the continents of the world. Approximately 71% of the world’s harvest is processed into wine, 27% is consumed fresh and 2% is dried for raisins. In 2009, grapevine cultivation covered approximately 8 million hectares with ca. 67 million metric tons of fruit produced annually. Italy, China, United States, France and Spain are the leading grapevine producers in the world. Other than these five countries, Turkey, Iran, Argentina, Chile, India, Australia and South Africa also produce grape in large quantities.

V. vinifera L. is the most widely cultivated *Vitis* species due to its adaptability to a wide range of temperate to subtropical or tropical conditions. It probably originated in the Mediterranean Basin and the Middle East, where it was domesticated 5,000 years ago. *V. vinifera* has given rise to over 14,000 cultivars grown in most of the cultivated areas worldwide. It is estimated that more than 90% of the world's grapevines are either *V. vinifera* or *V. vinifera* hybrids. *V. vinifera* cultivars are usually of outstanding quality but they are susceptible to a variety of biotic and abiotic stresses. Therefore, grapevine improvement is necessary to achieve specific goals and local adaptation. Numerous breeding programs have been conducted in different parts of the world using various breeding methods that will be mentioned in this chapter. The advantages, specific problems and challenges, as well as the achievements associated with each method will also be discussed. Continued advances in technology at the genomic level will increase the understanding of grapevine genetics and genes which underlie its important traits. Therefore, integration of these new and classical technologies into grapevine breeding programs will further ensure outstanding breeding successes in the future.

2. Botany and Taxonomy of Grapevines

The grape family, *Vitaceae* consists of 17 genera and more than 1,000 species. *Vitis* species are widely distributed in the tropics and subtropics, and have been extended into the temperate zones of the world. Variation in chromosome numbers is observed among different species/genus of *Vitaceae*. Some genus like *Ampelocissus*, *Parthenocissus* and *Ampelopsis* have a chromosome number of $2n = 40$. Species of the genus *Cissus* have a chromosome number of $2n = 24$. However, most genera of *Vitaceae* have a chromosome number of $2n = 38$. *Vitis* is the only food-producing genus in the family and is divided into 2 subgenera based on morphological criteria and chromosome number: *Muscadinia* Planch (muscadine grape), $2n = 40$, and *Euvitis* Planch (bunch grape), $2n = 38$. *Muscadinia* consists of three species including *V. munsoniana* and *V. rotundifolia* which are native to southeastern America and *V. popenoei*, native to Central America. The species of *Muscadinia* have also been considered within a separate genus, and their placement is still one of many ongoing debates. *Euvitis* species are interfertile and separated by geographic, phonologic and ecologic barriers, composing the American group (ca. 30 species), the Eurasia group (1 species) and the Asiatic group (more than 30 species). Most American species including *V. aestivalis*, *V. berlandieri*, *V. longii*, *V. cinerea*, *V. labrusca*, *V. monticola*, *V. riparia*, *V. rupestris*, *V. smalliana* and *V. shuttleworthii* have been used as parents in the breeding programs and/or rootstocks that exhibit resistance to several diseases and other pests, and tolerance to various soil and climatic conditions (Table 1). Native species in eastern Asia, China, Japan and south into Java constitute the Asiatic group. In China, 40 species, 1 subspecies and 13 varieties of Chinese wild grapes were found. Among these, *V. amurensis* is perhaps most commonly known. It has been frequently used to transfer cold hardiness and disease resistance (Table 1). In some areas of northeastern China and Japan, the edible fruits of this species are used as fresh fruit, juice, wine and jelly. Although, only one species (*V. vinifera*) was found in the Eurasia group, this species and its hybrids constitute over 90 percent of cultivated grapes. *V. vinifera* consists of three subspecies, a cultivated subspecies, ssp. *sativa* D.C. (spp. *vinifera*), and two wild subspecies, ssp. *sylvestris* Gmel. and ssp. *caucasica* Vav.

Species	Resistance to biotic stresses							Tolerance to abiotic stresses						Growth			Used for breeding			Other characteristics	Geographic location
	Downy mildew	Powdery mildew	Black rot	Anthracnose	Pierce's disease	Root-knot nematode	Phylloxera	Drought	Lime	Cold	Hot	Salinity	Iron	Grafting	Rooting	Vigor	New cultivars	Rootstocks	Interspecific hybrids		
V. rotundifolia	+	+	+	+	+	+	+				+						++	+	+		se USA
V. rupestris	+	+	+				+	+	-					+	+	+	++	+++	+++		Originally c TX, AR, MS, TN, KY, WV nw MD, sw PA; now rare mostly s MO, n AR
V. riparia	+	+	+				+		-	+		+		+	+		++	+++	+++	Flower & ripen early, high sugar & acid in fruit	All ne USA into Canada, south to n LA, VA
V. monticola							+	+	+											Poor growth & wood production	sc TX
V. vulpina	+		+		+		+		-	+						+	-	+	+		se USA
V. aestivalis	+	+			+		-				+				-		-	-	++	Desirable fruit characteristics	New England to GA, west to AR & Mississippi river
V. lincecumii	+			+			+	+			+										AR, LA, OK, TX
V. bicolor										+											ne North America from Canada to n GA
V. candicans	+	+	+		+	+	+		-		+	+		-	-		-	+	-		AL, AK, LA, OK, TX
V. cinerea	+	+	+				+		-					-	-		-	+	++		c & se USA
V. berlandieri	+	+					+	+	+			+	+	-	-		+	+++	-		Texas, n Mexico
V. labrusca	+	+	+/-	+	-		+			+							+++	++	+++	Large berries, strong distinctive flavor	East coast from ME to SC, west to OH, MI south to LA, AL
V. acerifolia	+	+					-	+		+		+	-		+		+	++	-	Mild flavor, seeds germinate at once	e NM & CO, KS, OK, n TX
V. × champinii					+	+	+	+				+	-	-			+	+	-	Natural hybrid (*V. candicans* × *V. rupestris*)	sc TX
V. amurensis	+	+		+				-		+							+	-	++		China, Japan, Korea, Russia

AL, Alabama; AR, Arkansas; CO, Colorado; GA, Georgia; KS, Kansas; KY, Kentucky; LA, Louisiana; MO, Missouri; MS, Mississippi; OH, Ohio; OK, Oklahoma; MD, Maryland; ME, Maine; MI, Michigan; PA, Pennsylvania; SC, South Carolina; TN, Tennessee; TX, Texas; VA, Virginia; WV, West Virginia; c, central; e, east; n, north; s, south; w, west.

Table 1. Characteristics and usage of some *Vitis* species.

The unique characteristics of *Muscadinia* are non-shedding bark, prominent lenticels, short small clusters, thick-skin berries that detach one by one as they mature, and simple tendrils with no fork. In addition, they have a unique fruity aroma. Their seeds are oblong without beaks. It was found that a bunch of *V. munsoniana* and *V. rotundifolia* consists of 6-24 berries, which do not ripen at the same time. By contrast, *Euvitis* has shedding bark, forked tendrils, mostly elongated flower clusters and berries adhering to the stem at maturity. Their seeds are pyriform, with long and short beaks.

Muscadinia requires approximately 100 days on the vine for the fruits to mature and a long growing season. They have higher resistance levels to most of the diseases than *Euvitis*. In addition, they have been cultivated commercially since the middle of the 18^{th} century. Most grape products like wine, juice and jam have been made from *V. rotundifolia* because of its unique fruity aroma. The interspecific crosses between *Euvitis* and *Muscadinia* are difficult to achieve due to chromosome number and genomic differences. Most of F_1 hybrids are completely or highly sterile, but some hybrid combinations can survive. However, the species of *Euvitis* are easily intercrossed and produce vigorous and fertile F_1 progeny.

Flowering of grape requires two consecutive growing seasons. The latent primary buds will be induced during the summer. However, initiation and development of the flower will not take place until the following spring. In addition, flowering in grapes is controlled by the gibberellin: cytokinin balance. Moreover, the external factors such as high light intensity, short term exposure to high temperature and optimum levels of soil moisture and macronutrients, are also factors which promote their flowering. There are three types of flower depending on the characteristics of the species and cultivars (Figure 1).

Perfect or hermaphroditic flowers have both functional pistils and stamens. The female flower has functional pistils. However, it has stamens that produce pollen, but is generally sterile. The male flower has an undeveloped pistil, but contains only a small rudimentary ovary which cannot be fertilized. These flower characteristics can be used to differentiate the wild and cultivated forms of grapevine. Male and female flowers are found on separate vines in the wild forms that are dioecious (most of them have male flowers), while the cultivated forms usually have hermaphroditic flowers.

In Chinese wild grapes, it was found that most types are male or female. Hermaphroditic flowers are found in only a few species of *V. quinquangularis*, *V. amurensis* and *V. davidii*. In perfect flowers, the pistil is normally surrounded by five stamens, although the number may vary from five to more on individual flowers of the same inflorescence. The other distinct characteristic between wild and cultivated forms is their seeds. Seeds obtained from wild grapes are round in shape with short beaks, while those from cultivated forms have long beaks.

In dioecious grapes, fertilization is most likely to occur by means of wind or insect pollination, whereas, in the hermaphroditic flowers, fertilization happens via self-pollination. The ovary and seeds of fertilized grapevine flowers develop into a fleshy fruit called a berry, typically known as a grape. Berries of *Euvitis* develop in clusters of up to 100 or more, but in *Muscadinia* small clusters of three to five berries are usually formed. Each berry contains one to four seeds. Grape berries are non-climacteric fruits whose ripening is associated with an accumulation of soluble solids and berry pigmentation (anthocyanins). During ripening berry softness increases and hexoses accumulate while malic and tartaric acids decline.

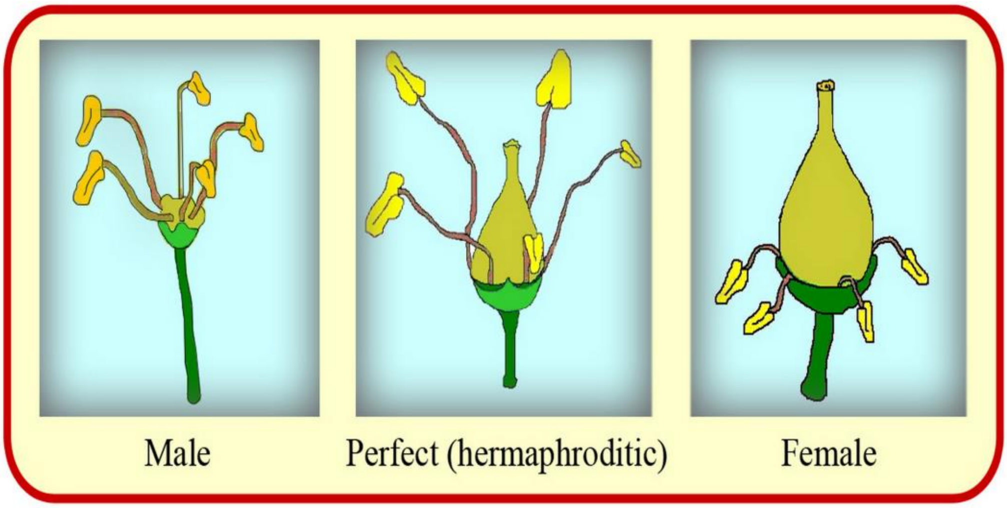

Figure 1. Three types of grapevine flowers.

3. Grapevine Genetic Resources

There are several species of wild and cultivated grapevines in the world, and each species consists of numerous varieties. In different countries more than 10,000 named varieties of grapevine were found. Moreover, the list of new varieties is continually being added to every year. Grapevine germplasm collections are crucial for maintaining the genetic resources of *Vitis*. Genetic resources can be divided into 4 types; wild species, old traditional cultivars, new cultivars and breeding lines. The wild species are potential sources for various traits (Table 2).

Both *Euvitis* and *Muscadinia* species are good sources of resistance. Several American species are resistant to phylloxera (*Dactylosphaera vitifoliae*; mainly *V. riparia*, *V. rupestris* and *V. berlandieri*) and are used extensively in breeding programs. Among these three species, only *V. berlandieri* is adapted to highly calcareous soils, and it is used to develop root stocks resistant to phylloxera and lime-induced chlorosis.

Wild Chinese species including *V. bryoniifolia*, *V. davidii* and *V. piasezkii*, and Asiatic species *V. amurensis* are good sources of powdery mildew (*Uncinula necator*), and/or downy mildew (*Plasmopara viticola*) resistance. *Muscadinia* is a useful source of resistance to phylloxera, nematodes, Pierce's disease (PD; *Xylella fastidiosa*) and fungal diseases including powdery mildew and downy mildew. Several sources of tolerance to abiotic stresses (cold, drought, salinity, lime etc.) are also observed in different *Vitis* spp. (Table 2), for example, cold tolerance is found in the northern species *V. riparia*, *V. labrusca* and *V. amurensis*.

While southern species, *V. lincecumii*, *V. bourquiniana* and *V. rotundifolia*, provide tolerance to hot conditions. However, these wild species have small berries with excessive seeds and strong pungent flavors. Therefore, extensive backcrosses are often needed to eliminate some unfavorable characteristics of the wild species. In addition to the wild *Vitis* spp., many traditional or new cultivars are also good sources of desirable traits (Table 2).

Desired properties	**Disease/ nematode/ insect/ stress**	***Vitis* species/ varieties**
Disease resistance		
Fungal resistance	Powdery mildew (*Uncinula necator*)	*V. aestivalis*, *V. amurensis*, *V. cinerea*, *V. berlandieri*, *V. labrusca*, *V. riparia*, *V. rotundifolia*, *V. rupestris*, *V. davidii*, *V. piasezkii*, *V. quinquangularis*, *V. romanetii*, 'Nistru'
	Downy mildew (*Plasmopara viticola*)	*V. aestivalis*, *V. amurensis*, *V. bryoniifolia*, *V. labrusca*, *V. lincecumii*, *V. flexuosa*, *V. piasezkii*, *V. pseudoreticulata*, *V. riparia*, *V. rupestris*, *V. rotundifolia*, *V. romanetii*, *V. yenshanensis*
	Black rot (*Guignardia bidwellii*)	*V. candicans*, *V. cinerea*, *V. riparia*, *V. rupestris*, *V. rotundifolia*, 'Chancellor', 'Léon Millot', 'Seyval'
	Anthracnose (*Elsinoe ampelina*)	*V. amurensis*, *V. munsoniana*, *V. labrusca*, *V. rotundifolia*, *V. smalliana*, *V. simpsoni*, *V. shuttleworthii*, *V. davidii*, *V. piasezkii*, *V. pseudoreticulata*, *V. quinquangularis*, *V. romanetii*, 'Muscat Hamburg', 'Muscat Onitskanskii', 'Suruchenskii', 'Bangalore Blue', 'Gros Colman'
	Botrytis bunch rot (*Botrytis cinerea*)	*V. riparia*, *V. rupestris*, *V. vinifera*
	Rust (*Physopella ampelopsidis*)	*V. caribaea*, *V. rotundifolia*, *V. simpsoni*, *V. shuttleworthii*
	Rotbrenner (*Pseudopezicula tracheiphila*)	*V. cinerea*, *V. vinifera*
Bacterial resistance	Pierce's disease (*Xylella fastidiosa*)	*V. aestivalis*, *V. candicans*, *V. rotundifolia*, *V. smalliana*, *V. simpsoni*, *V. shuttleworthii*, *V. × champinii*, *V. vulpina*, 'Norris', 'Lake Emerald', 'Blue Lake'
	Crown gall (*Agrobacterium tumefaciens*)	*V. amurensis*, *V. labrusca*, 'Chardonnay', 'Fateasca Regala', 'Muscat Ottonel', 'Rhine Riesling', 'Crimean Comichon'
Viral resistance	Grapevine fanleaf virus (GFLV)	*V. arizonica*, *V. candicans*, *V. riparia*, *V. rotundifolia*, *V. rufotomentosa*, *V. slavinii*, *V. vinifera*
Nematode resistance	Root knot nematode (*Meloidogyne* spp.)	*V. candicans*, *V. × champinii*, *V. rotundifolia*
	Dagger nematode (*Xiphinema index*)	*V. cinerea*, *V. rotundifolia*, *V. rufotomentosa*
Insect resistance	Phylloxera (*Dactylosphaera vitifoliae*)	*V. berlandieri*, *V. cinerea*, *V. × champinii*, *V. rotundifolia*, *V. riparia*, *V. rupestris*
	Aphid (*Aphis illinoisensis*)	*V. cinerea*
Abiotic stress tolerance	Cold	*V. adstricta*, *V. amurensis*, *V. acerifolia*, *V. labrusca*, *V. riparia*, *V. vinifera* ssp *sylvestris* var tipica, var balcanica, var aberrans, *V. vulpina*, *V. yenshanensis*, 'Italian Riesling'
	Hot	*V. candicans*, *V. lincecumii*, *V. bourquiniana*, *V. rotundifolia*

	Drought	*V. acerifolia, V. arizonica, V. × champinii, V. monticola, V. berlandieri, V. rupestris, V. vinifera*
	Salinity	*V. acerifolia, V. berlandieri, V. riperia, V. candicans, V. × champinii*
	Iron	*V. berlandieri, V. vinifera*
	Lime	*V. monticola*

Table 2. Sources of resistance/tolerance and other desirable traits.

Germplasm collections have stored at least 10,000 grapevine varieties. However, because synonyms (many names for the same varieties) and homonyms (convergence of name for different varieties) occur in grapevine, a more accurate estimate of the number of varieties might be closer to 5,000. The true number of varieties and the relationships between them remain to be determined possibly by extensive DNA profiling of the grape varieties in different collections and the development of a common database. Germplasms are being maintained in the field as well as in tissue culture or cryopreservation (maintained at -196ºC). It was found that selection of the superior accessions and evaluation of the germplasms are probably one of the first steps for grapevine breeding.

During the past ten years, there has been increasing interest in grape germplasm resources and genetic diversity analysis. It helps to protect some varieties of grapes, especially wild grapes like *V. vinifera* ssp. *sylvestris*, which is an ancestor of cultivated varieties, and facilitates their utilization in grapevine breeding programs. *Sylvestris* grapes can climb forest trees at about 20-30 m of height and produce small bunches of fruits. In addition, they resist/tolerate both biotic and abiotic stress factors including drought, lime, pests and diseases. Nowadays, information on grape database and germplasm collections can be found on the website (Table 3).

Databases	**Source**
Bulgarian *Vitis*	http://bulgenom.abi.bg/Genotyping%20and%20Mapping%20 Crops%20Selected.htm/
European *Vitis*	http://www.eu-vitis.de/
Greek *Vitis*	http://gvd.biology.uoc.gr/gvd/index.htm/
Italian *Vitis*	http://www.vitisdb.it/
Swiss *Vitis*	http://www1.unine.ch/svmd/
Vitis International Variety Catalogue	http://www.vivc.de/
Germplasm collections	**Source**
Conservation des ressources phyto-génétiques	http://www.bdn.ch/culture/reben_startseite/
Deutsche Genbank Reben	http://www.deutsche-genbank-reben.jki.bund.de/
INRA Daomaine de Vassal - Centre de resources génétiques de la vigne	http://www1.montpellier.inra.fr/vassal/
The International Grape Genome Program (IGGP)	http://www.vitaceae.org/
The NCBI taxonomy for *V. vinifera*	http://www.ncbi.nlm.nih.gov/Taxonomy/Browser/wwwtax.cgi? mode=Info&id29760&lvl=3&keep=1&srchmode=1&unlock
The Institute for Genomic Research (TIGR)	http://www.tigr.org/tigr-scripts/tgi/T_index.cgi?species=grape

The Genomic-Info Research Unit (URGI) of INRA	http://urgi.infobiogen.fr/GnpMap2/mapping/searchMap.do
The French National Resources Centre for Plant Genome (CNRGV)	http://cnrgv.toulouse.inra.fr/ENG/
USDA, ARS Cold Hardy Grape Collection	http://www.ars.usda.gov/Aboutus/docs.htm?docid=6245/
USDA, ARS National Germplasm Repository at Davi	http://www.ars.usda.gov/main/site_main.htm?modecode =53-06-20-00/

Table 3. Grape database and germplasm collections.

4. Inheritance and Genetic Analyses of Grapevines

In grapevines, inheritance and genetic analyses have been carried out only for a limited number of traits because it has a long life cycle, a large number of chromosomes, partial sterility of ovules, and low seed germination. The following are examples of the traits that have been studied. Downy mildew resistance in *Vitis* is based on two genetic systems: (1) a single gene for the hypersensitive reaction of stomatal tissues, for which resistant species are homozygous dominant, and the susceptible *V. vinifera* is homozygous recessive, (2) polygenes for inhibition of the mycelium growth in plant tissues. Broad sense heritability for downy mildew resistance was estimated at 0.83 to 0.94, suggesting minimum environmental effects, while narrow sense heritability was 0.26 to 0.39. For powdery mildew resistance, a polygenic system has been suggested. The narrow sense heritability estimate for this trait ranged from 0.31-0.51, suggesting quite a high contribution of additive genetic variance. *Botrytis* resistance is associated with the synthesis of phytoalexin stilbene. This resistance had a narrow sense heritability and broad sense heritability of 0.23 to 0.26 and 0.82 to 0.92, respectively. It was found that resistance to anthracnose (*Elsinoe ampelina*) is controlled by two dominant genes for susceptibility (An_1 and An_2) and a single dominant resistance gene (An_3).

The narrow sense heritability for anthracnose resistance was measured as 0.79, indicating that anthracnose resistance gene(s) were highly heritable. The resistance to black rot (*Guignardia bidwellii*) was reported to be either controlled by 2 dominant genes or quantitatively controlled. Previous research suggested that Pierce's disease resistance required 3 dominant genes, Pd_1, Pd_2 and Pd_3. Resistance to grapevine fanleaf virus can be obtained by using host plants resistant to the virus or plants resistant to the dagger nematode vector, *Xiphinema index*. The resistance to nematode was controlled by either a single dominant gene or by two genes, one dominant and one recessive, while the resistance to the virus was a recessive trait controlled by at least 2 genes. Resistance to phylloxera, an insect pest, is controlled by multiple genes. Those from *V. rotundifolia* appear to be dominant. *V.* × *champinii* and *V. candicans* confer resistance to the root knot nematode (*Meloidogyne* spp.), for which the resistance is mediated by a dominant gene. The heritability of resistance to this pest was estimated at 0.39, suggesting that additive effects were important.

Two types of seedlessness are found in grapevine: (1) stenospermocarpic, the seeds abort while still small and soft, (2) parthenocarpic, the seeds do not develop at all. The former is the one utilized in table grape improvement. Seedlessness is related to three independent recessive genes regulated by a dominant inhibitor locus, *SdI* (Seed Development Inhibitor). Two pairs of genes affect fruit color with epistatic action: *B*, a dominant gene for black fruit, and *R*, a dominant gene for red fruit. The white-fruited grapes are recessive for both genes. Similarly, the composition of fruit anthocyanins is controlled by 2 genes: *G* for diglucosides or *g* for monoglucosides, and *O* for triphenols or *o* for diphenols. For fruit aroma, muscat flavor is controlled by 5 dominant complementary genes, methyl anthranilate is controlled by 3 dominant complementary genes, while volatile ester levels are determined by 2 genes. Several quality traits have generally high heritabilities: cluster compactness (0.55), berry weight (0.49), skin texture (0.75) and pulp texture (1.0).

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Wikipedia: The Free Encyclopedia. Developed under the auspices of the Wikimedia Foundation, Inc., [http://en.wikipedia.org] [This website provides comprehensive definitions of technical words.]

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Biographical Sketches

Piyada Tantasawat was born in Bangkok, Thailand, and obtained her B.Sc. in Agriculture (Horticulture) at Kasetsart Univetsity with First Class Honors. She worked as a research assistant at Kasetsart University for nearly a year before receiving a Fulbright Scholarship to pursue her M.Sc. at Cornell University where she obtained her Ph.D. in Plant Breeding in 1997. After three years of postdoctoral research in plant genetic engineering and biochemistry at Cornell University, she returned to Thailand and was appointed to the staff of the School of Crop Production Technology, Institute of Agricultural Technology, Suranaree University of Technology (SUT), Thailand. Her major scientific interests are breeding for plant pest resistance utilizing conventional, mutation, and biotechnological approaches, plant tissue culture, and plant resistance mechanisms. She has worked on grapevine, mungbean, sunflower, cucumber, tomato and orchid improvement, and published over 40 papers that are related to these interests.

Oythip Poolsawat was born in Nakhon Nayok, Thailand. She has a B.Sc. (Second Class Honors) and a Ph.D. in Crop Production Technology from SUT. She is currently a post-doctoral fellow at SUT. She is interested in the resistance mechanisms and breeding of grapevines for resistance to diseases, especially anthracnose.

Wirot Chaowiset was born in Suphanburi, Thailand, and has a B.Sc. and an M.Sc. in Crop Production Technology from SUT, Thailand. His thesis study was on physiology of *Pueraria mirifica* and the effect of the active ingredients in *P. mirifica* on the relaxation of blood vessels in rats. After graduation (2007), he worked as a research assistant in the sunflower breeding project. Currently, he is a post-graduate researcher at SUT. He has performed research on many crops, including tomatoes, cucumbers, grapevines and orchids etc. He is interested in plant physiology, mechanisms of resistance, and crop improvement via tissue culture and other techniques.