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# Groundnut Bacterial Wilt in Asia

International Crops Research Institute for the Semi-Arid Tropics

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## Abstract

The current status of research on bacterial wilt of groundnut in Asia (specifically in China and Vietnam) is reviewed Particular emphasis is given to the available disease management options, and their packaging and validation in on-farm participatory research at hot-spot locations in China and Vietnam Recommendations are made for further collaborative research with advanced institutes to provide training to scientists and technicians from China and Vietnam in recent advances in serological and molecular techniques for identification and differentiation of races, biovars, and strains of the wilt pathogen, *Burkholderia solanacearum* Recommendations are also made for further collaborative research, and the need to interest potential donors in promoting the Groundnut Bacterial Wilt Working Group's activities The publication includes papers describing the status of the disease in China and Vietnam

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# Groundnut Bacterial Wilt in Asia

## Proceedings of the Fourth Working Group Meeting

**11–13 May 1998**
**Vietnam Agricultural Science Institute (VASI),**
**Van Dien, Thanh Tri, Hanoi, Vietnam**

*Edited by*
**S Pande, Liao Boshou,**
**Nguyen Xuan Hong, C Johansen,**
and
**CLL Gowda**

**ICRISAT**
**International Crops Research Institute for the Semi-Arid Tropics**
Patancheru 502 324, Andhra Pradesh, India

1998

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Van Dien, Thanh Tri, Hanoi, Vietnam

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# Preface

The fourth meeting of the Groundnut Bacterial Wilt Working Group was held 11–13 May, 1998 in Van Dien, Thanh Tri, Hanoi, Vietnam The meeting and field visits to bacterial wilt hot spot locations and experimental sites in Vietnam were co-sponsored by the Vietnam Agricultural Science Institute (VASI) and International Crops Research Institute for Semi-Arid Tropics (ICRISAT)

Scientists from China (4), Vietnam (25) and ICRISAT (1) participated in the meeting The present status of bacterial wilt management was discussed and accounts of current research on bacterial wilt disease in different institutions, particularly in China and Vietnam, were presented Good progress has been made, and prospects of continued collaborative research into the management of this disease are encouraging

We sincerely believe that this volume will provide a useful guide to the present status of groundnut bacterial wilt in Asia and will further strengthen coordinated work on this economically important disease

S Pande
Liao Boshou
Nguyen Xuan Hong
C Johansen
CLL Gowda

# Inaugural Session

# Welcome Address

## Nguyen Huu Nghia[1]

Distinguished guests and participants, first of all allow me on behalf of Vietnam Agricultural Science Institute (VASI) to warmly welcome all the guests and participants in the Fourth Groundnut Bacterial Wilt Working Group Meeting, especially the participants from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) and China who have come to attend this meeting

This is the Fourth Meeting of the International Working Group on Groundnut Bacterial Wilt, organized in the framework of the collaborative program of member countries of the Cereal and Legumes Asia Network (CLAN) This working group was established in March, 1990 on the basis of suggestion of member countries of the Network, who had great concern in research and control of groundnut bacterial wilt The previous meetings of this working group were organized in Malaysia (1990), Taiwan (1992), and China (1994) VASI has great honor to host and co-sponsor this important meeting

Bacterial wilt is one of the main constraints for groundnut production in many Asian countries It has attracted attention of various research and development agencies both at national and international level As you know, a lot of achievements in research and control of the disease have been obtained by Chinese research institutes in the last few years Therefore, we are glad to see the presence of Chinese scientists who are specialized in research on groundnut bacterial wilt and have not only significantly contributed to the achievement of China but have also contributed to promoting research and control of the disease in the region and CLAN member countries

In Vietnam, bacterial wilt is a serious and damaging disease that is very difficult to control Since 1990, after the establishment of the collaborative relationship between Vietnam and ICRISAT, and with the cooperation of CLAN member countries, research on groundnut bacterial wilt has made significant progress Until now, several disease-resistant, high-yielding, high quality groundnut varieties have been identified, tested in large scale production, and been accepted by Vietnamese farmers Resistance sources, especially indigenous genetic sources, have been identified for utilization in varietal improvement programs Research on the biology of the pathogen and control of the disease has also been started

1 Vietnam Agricultural Science Institute (VASI) Van Dien Thanh Tri Hanoi Vietnam



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Collaboration in research and development in this field between national and international institutions has been strengthened However, to overcome the problems caused by bacterial wilt in groundnut production, there is a lot of work to be done in coming years

Achievements obtained by other countries in the region have strongly stimulated Vietnamese scientists in research and control of the disease We hope that this meeting will be a good opportunity for Vietnamese scientists to share their experiences in research and management of groundnut bacterial wilt with international participants We also hope that Vietnamese scientists will learn from this meeting

On this occasion, allow me on behalf of VASI to express our high appreciation and grateful thanks to ICRISAT scientists and CLAN member countries for their enthusiastic and effective support and collaboration in research and development of groundnut in the past years We sincerely hope that collaboration between Vietnam, ICRISAT, and CLAN member countries will be strongly developed

Finally, I would like to wish the meeting success I wish all the guests and participants of the meeting good health and happiness

Thank you for your attention

# Welcome Address from ICRISAT

## S Pande[1]

Honorable Dr Nguyen Huu Nghia, Director General, Vietnam Agricultural Science Institute (VASI), Dr Phan Lieu, Director, Oil Plant Institute (OPI) Southern Vietnam, Dr Liao Boshou, Coordinator, Groundnut Bacterial Wilt Working Group, Dr Nguyen Xuan Hong, Cereals and Legumes Asia Network (CLAN) Coordinator, and distinguished participants, ladies and gentlemen, on behalf of ICRISAT-CLAN and on my own behalf I welcome all of you to this 3-day Working Group Meeting on Groundnut Bacterial Wilt It is my privilege to participate and represent ICRISAT at this meeting being held at one of the leading agricultural institutions in Vietnam VASI, Thanh Tri, Hanoi The choice of Thanh Tri and Hanoi, in northern Vietnam, for this meeting is most appropriate, especially considering the interest of Working Group members in interacting with many Vietnamese scientists who have recently made notable contributions to collaborative research on the management of groundnut diseases, including groundnut bacterial wilt

I am pleased to take this opportunity to spell out certain aspects of cooperation between ICRISAT and VASI The first ICRISAT mission in Vietnam, headed by the then Director General Dr L D Swindale was in 1989 and was followed by signing of the Memorandum of Understanding (MoU) for cooperation between the Ministry of Agriculture and Food Industry (MAFI), Vietnam and ICRISAT in India on 25 Sep 1989 Since then, Directors General, CLAN Coordinators, and ICRISAT scientists have made countless visits to Vietnam Some of the notable visits to strengthen groundnut research and integrated pest management research have been made by ICRISAT breeders, agronomists, entomologists, and plant pathologists We have developed and implemented collaborative workplans with Vietnam, the most recent being presented during the CLAN Coordinators' Steering Committee Meeting, 24–28 Nov 1997 in Batu, Indonesia More than 85 Vietnamese scientists and technicians have participated in various training programs at ICRISAT Germplasm exchange has been a major activity ICRISAT has supplied Vietnam with a total of 3825 germplasm accessions, breeding lines, and advanced generation lines of groundnut Also in collaboration with the National Institute

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of Vietnam ICRISAT has collected 67 accessions of groundnut germplasm, and will collect more in the near future

Collaboration with East and Southeast Asia on groundnut diseases caused by fungi, viruses, and bacteria has been encouraging, especially bacterial wilt research, which was most rewarding, having generated wilt basic and applied information on this disease An Information Bulletin on "Bacterial Wilt of Groundnut", a Technical Manual on Techniques for Diagnosis of *Pseudomonas solanacearum*, a manual for resistance screening against groundnut bacterial wilt, and high quality papers in the proceedings of the Bacterial Wilt Working Group Meetings are a few classic examples of our collaborative efforts We at ICRISAT are not able to conduct research on bacterial wilt of groundnut, and depend on the comparative advantage and expertise of groundnut scientists in China, Indonesia, Vietnam and other network member countries to collaborate in combating this disease

Groundnut bacterial wilt, caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al (formerly *P solanacearum* (E F Smith) E F Smith) has been a major constraint on crop production in the countries of South and Southeast Asia and the South Pacific The disease has been a serious constraint to groundnut production in Indonesia and China for several decades It is becoming potentially important in Vietnam, Malaysia, the Philippines, and Thailand Bacterial wilt is also a problem in some countries in Africa It is a growing threat to groundnut production in several other parts of the world, particularly in warm, humid areas The destructiveness of the disease is compounded by the wide host range of the wilt pathogen Considerable research in Indonesia and China has provided an excellent understanding of the effects of cultural practices and environmental factors on groundnut bacterial wilt, and the results have been used to formulate recommendations to reduce wilt incidence and severity

Research on genetic resistance has been given the highest priority since the early 1920s The variety Schwarz 21 was released in Indonesia in 1927, the earliest record of a disease-resistant groundnut being developed Much progress has been made in Indonesia, China, and other countries in the development of various wilt-resistant, high yielding groundnut cultivars However, there is a need for breeding wilt-resistant groundnut varieties with high yield potential and good agronomic characters, and to combine resistance to other diseases and pests Basic research on the wilt pathogen *B solanacearum* has been carried out in advanced laboratories in the developed countries as *B solanacearum* attacks many other important food crops throughout the world These studies have provided us with such diagnostic tools as monoclonal antisera and polymerase chain reaction techniques It is

expected that the results of this research will enhance our understanding in the management of the pathogen and bacterial wilt disease in groundnut

At ICRISAT Patancheru, we the groundnut scientists have the responsibility of working jointly with Asian national agricultural research system (NARS) scientists to address constraints on groundnut production in different agroecological systems in Asia This function is facilitated by CLAN, which started in 1986 as the Asian Grain Legume Network (AGLN), with ICRISAT providing a Coordinating Unit CLAN has successfully supported many collaborative activities concerning ICRISAT's mandate crops (sorghum, pearl millet, chickpea, pigeonpea, and groundnut) and its mandate eco-region (the semi-arid tropics)

The "Working Groups" concept to focus collaborative research on the challenging regional problems has been found useful and effective and has resulted in quick progress One such effective Working Group, on Groundnut Bacterial Wilt in Asia, is meeting here today

The foundation of the Groundnut Bacterial Wilt Working Group (GBWWG) was laid in 1990 after a planning meeting held in Malaysia This fourth meeting of the GBWWG is being held jointly under the auspices of VASI and ICRISAT

Groundnut bacterial wilt is an important problem in groundnut production systems in South and Southeast Asia and in similar systems elsewhere Because of the danger of its spreading to new areas where irrigation and multiple cropping are being introduced, we need to provide necessary support and funding to ensure that integrated disease management technology is soon available to farmers to minimize losses caused by this disease

We at ICRISAT sincerely hope that the recommendations of this meeting will further strengthen international collaborative research to find and provide cost effective solutions to managing bacterial wilt, the most important constraint on groundnut production in East and Southeast Asian countries

# Objectives of the Fourth Groundnut Bacterial Wilt Working Group (GBWWG) Meeting and an Overview of the Second International Bacterial Wilt Symposium

**Liao Boshou**[1]

Good morning, respected Mr Chairman, ladies and gentlemen First of all, I would like to express my warm welcome to all the participants to this meeting I am also very pleased to have this chance, for the first time, to visit Vietnam with several of my colleagues from China As you know, groundnut has become a very important oil crop and cash crop in China, Vietnam, and many other developing countries With the development of economy and society, groundnut will be more and more important because of its high nutrition, wide adaptation to various ecological conditions, high yield potential, and high output or production efficiency We take interest in noticing that groundnut production in Vietnam has increased in recent years and that a considerable portion has been exported, contributing significantly to the nation's economy

When we try to increase groundnut production, we have to struggle against many diseases, and bacterial wilt is one of them Bacterial wilt of groundnut, caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al is the most important bacterial disease of this crop throughout the world In order to improve control of this disease and coordinate concerned research in various countries, a Groundnut Bacterial Wilt Working Group (GBWWG) was established through the joint efforts of ICRISAT and the Australian Centre for International Agricultural Research (ACIAR) In March, 1990, the first Working Group (WG) meeting was held in Malaysia The second WG meeting was held in November, 1992 at the Asian Vegetable Research and Development Center (AVRDC) in Taiwan The third WG meeting was held in July 1994 at the Oil Crops Research Institute in China We greatly appreciate the efforts of ICRISAT/Cereals and Legumes Asia Network (CLAN) in organizing this meeting We are happy to see that this WG is considered one of the most successful organized by ICRISAT As most of you know, ICRISAT has been subjected to reformation in recent years and as a result many scientists have left ICRISAT, including Dr V K Mehan, the former Technical Coordinator of this WG In these circumstances, I was nominated Technical Coordinator of the WG during the CLAN Steering Committee Meeting in November 1997, in Malang, Indonesia I happened to visit ICRISAT during November and December, 1997 and had a chance to plan with some of the

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ICRISAT scientists, including Dr Pande, to hold the Fourth Working Group Meeting in Vietnam Dr Gowda, the CLAN Coordinator, visited Vietnam in late February this year and decided to hold this meeting I would like to express my thanks to the host institution, Vietnam Agricultural Science Institute (VASI), and Dr Nguyen Xuan Hong for hosting the meeting I am also very glad to see that Dr Pande a senior plant pathologist, is representing ICRISAT here

The objectives of this meeting are as follows

- Review research progress on groundnut bacterial wilt, particular in China and Vietnam, since the Third GBWWG meeting in 1994
- Evaluate the status of, and exchange information on research on groundnut bacterial wilt in concerned countries
- Discuss and recommend priorities for collaborative research at the national and international levels
- Determine the resources needed and to prepare joint proposals for funding

I am confident that we will have a good meeting here and that this meeting will contribute significant knowledge to the members of this WG to combat bacterial wilt

I would like to give you a brief introduction about the Second International Bacterial Wilt Symposium (IBWS) held in Guadeloupe, France, 22–27 June 1997 With support from the China Natural Science Foundation (CNSF) and CLAN, I was able to attend IBWS The symposium was hosted by the Institut national de la recherche agronomique (INRA) and attended by 120 participants representing 32 countries and organizations The program of the symposium was divided into eight sessions These sessions included opening and welcoming remarks, pathogen diversity, disease diagnosis, pathogenicity, host resistance I, biological control and epidemiology, host resistance II, and disease management Forty five papers were presented in the eight scientific sessions In addition, 53 poster papers were also presented There were 4 papers specifically on groundnut bacterial wilt The symposium revealed marked progress in investigating the molecular, ecological and phenotypical basis of the wilt pathogen diagnosis techniques the molecular basis and genetic enhancement of host resistance, and biological and cultural control The proceedings of the symposium, entitled "Bacterial Wilt Disease Molecular and Ecological Aspects", are being edited by P Prior (France) and C Allen (USA) and will be published by Springer Verlag in Germany in 1998

# Research on Bacterial Wilt in Vietnam

**Nguyen Xuan Hong**[1]


## *Abstract*

*In recent years, research on bacterial wilt in Vietnam has been given high priority Systematic surveys conducted in both northern and southern Vietnam have confirmed that bacterial wilt is a widespread and serious disease of groundnut It is especially severe in hilly and upland areas, and in sandy soils along river banks, where it can cause considerable plant mortality (15–50%) and yield losses Genetic resistance to bacterial wilt is a high-priority objective of several research institutions in Vietnam The varieties Schwarz 21 and Matjan, reported to be resistant in Indonesia, also have shown stable resistance in northern Vietnam The local variety Gie Nho Quan has been highly and stably resistant to bacterial wilt All isolates of* Burkholderia solanacearum *from groundnut belong to biovars 3 and 4 Research on cultural and biological control of groundnut bacterial wilt is in progress*


## Introduction

Vietnam has a tropical monsoon climate characterized by high temperatures, high humidities, and high rainfall Bacterial wilt caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al has been known to cause serious problems on a number of economically important crops in Vietnam including groundnut, potato, tomato, sesame, tobacco, and eggplant Introduction of susceptible new varieties of these crops has contributed to the increased incidence of bacterial wilt Therefore, in recent years research on bacterial wilt in Vietnam has been given increasing attention

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**Nguyen Xuan Hong** 1998 Research on bacterial wilt in Vietnam Pages 10–14 in Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

## Current Research on Bacterial Wilt in Vietnam

During 1991–97, systematic surveys jointly conducted by Vietnamese and ICRISAT scientists have confirmed that bacterial wilt is one of the most important biotic constraints to groundnut production in both northern and southern Vietnam

In northern Vietnam, bacterial wilt is more severe on spring (February to June) crops grown under rainfed conditions in hilly and upland areas, and in sandy soils along river banks The disease is more serious in autumn (July to November) in some areas where the crop is grown year after year In southern provinces, bacterial wilt occurs on groundnuts under both irrigated (winter-spring and summer crops) and rainfed (rainy-season crop) conditions Considerable plant mortality and yield losses caused by bacterial wilt (15–50%) have been observed in various areas of both northern and southern provinces

In many groundnut growing areas it was observed that the incidence of bacterial wilt was low when groundnut crops were first sown, but increased to high levels in second and subsequent sowings, leading to the abandonment or discontinuation of the crop It was also observed that continuous dry spells followed by rains could immediately induce severe wilt under field conditions Rotation of groundnut with irrigated rice appears effective in containing disease incidence and severity

Growing wilt-resistant cultivars is considered the most effective and practical approach to controling bacterial wilt Genetic resistance to bacterial wilt is a high priority objective of several research institutions in Vietnam The major research thrusts at the Vietnam Agricultural Science Institute (VASI), Hanoi, are to identify wilt-resistant genotypes and to breed for resistance A total of 715 germplasm and breeding lines were tested for reaction to bacterial wilt at hotspots and in sickplots in Vietnam Among them, 4 lines were classified as highly resistant, and 25 lines as moderately resistant (Lieu, 1997) The local variety Gie Nho Quan showed the highest level of resistance over seasons at disease hot spots in northern Vietnam, with disease incidence/plant mortality less than 2% The varieties Schwarz 21 and Matjan, both reported resistant in Indonesia, also have shown stable resistance in Vietnam These varieties gave markedly higher yields than did the control cultivars Sen Nghe An (in northern Vietnam) and Se Dia Phuong (in central coastal areas) They are being rapidly multiplied and tested on farm for possible release in the near future However, an accession of the variety Kidang (ICG 11210), which was considered resistant in Indonesia, showed high wilt incidence in northern Vietnam In southern Vietnam, research on groundnut

bacterial wilt with emphasis on host plant resistance has been intensified in recent years at the Institute of Oil Plants Several groundnut lines supplied by ICRISAT were resistant to bacterial wilt The line ICG 8666 (an accession of the variety Schwarz 21) showed the highest level of resistance, with plant mortality less than 1% over 3 seasons in Cuchi (Duc Cam et al 1995)

Research on host-plant resistance to bacterial wilt of potato also has been intensified in recent years Approximately 140 germplasm and breeding lines of potato available in northern Vietnam were screened over the past several years for resistance to bacterial wilt A total of 20 lines and varieties have shown high levels of resistance to bacterial wilt, several varieties including KT3, VT2 also gave high yields (Thanh, 1998) More recently, research on host plant resistance of tomato has been initiated at VASI, Hanoi Agricultural University, and the Institute of Vegetables and Fruits Application of molecular methods in studying *B solanacearum* isolates have been initiated at the Institute of Agricultural Genetics, Hanoi (Cuong, personal communication) Until recently, research on bacterial wilt of tobacco and eggplant was very limited, but the Department of Plant Pathology and Pesticides, Hanoi Agricultural University has initiated research on the bacterial wilt disease on these crops

## Present Staffing and Resources Available for Research

At VASI, seven scientists including phytopathologists, agronomists, and breeders are now working on different aspects of bacterial wilt A nationwide network of groundnut bacterial wilt research has been established recently, on the initiatives of VASI scientists Several hotspot locations and sickplots also have been established and are regularly used for multilocational resistance screening of groundnut germplasm and breeding lines Recently, VASI scientists, in collaboration with ICRISAT scientists, have intensified local groundnut germplasm collection and characterization More local groundnut varieties resistant to bacterial wilt are expected to be identified The Department of Plant Pathology and Genetics at VASI is currently responsible for carrying out a Ministry of Agricultural Research and Development (MARD) research project on *B solanacearum* and mechanisms of resistance to it Facilities and methodologies for pathogen isolation, biochemical tests, and artificial inoculation are also available at VASI Two researchers have completed their PhD work on identifying sources of resistance to bacterial wilt on groundnut and potato

Two scientists are engaged in research on soilborne diseases and bacterial wilt with emphasis on pathological aspects and control measures at the National Plant Protection Institute (NPPI) They also participated in surveys and research on the disease in collaboration with VASI and ICRISAT scientists

In southern Vietnam, at the Oil Plants Institute (OPI), a team of scientists is actively involved in research on groundnut bacterial wilt with emphasis on host plant resistance and integrated disease management They have established close collaboration in research on bacterial wilt

One group of researchers at the Hanoi Agricultural University is conducting extensive work on bacterial wilt of major crops in Hanoi and surrounding areas

Two scientists have recently initiated research at the Institute of Agricultural Genetics on molecular aspects of *B solanacearum*, in collaboration with the Asian Vegetable Research and Development Center (AVRDC) However, funds for this research are still awaited

## Future Plans

In Vietnam, because of the economic importance of bacterial wilt, we continue to give high priority to research on this disease The following aspects will receive attention in coming years

- Further systematic research to identify biovars, races, pathotypes, host range, and conditions favouring the multiplication and spread of the pathogen
- Rapid multiplication and introduction to large-scale production of promising bacterial wilt resistant lines or varieties of groundnut
- Breeding for bacterial wilt resistance using local and introduced sources of resistance
- Development of a farmers' participatory, community based, integrated wilt management program (using resistant cultivars, crop rotation, crop husbandry, and certain cultural practices) to alleviate severe yield losses from bacterial wilt and sustain groundnut cultivation and production
- Investigation in upland areas of rotating and/or intercropping groundnut with maize, which is a non-host crop popularly grown in Vietnam's dryland crop systems
- Increased emphasis on developing and strengthening collaborative research on bacterial wilt between Vietnamese institutions and other CLAN member countries

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# A Brief Introduction to Bacterial Wilt Research in China

**Duan Naixiong[1], Liao Boshou[1], He Li-Yuan[2], and Gao Guoqing[3]**

## *Abstract*

*Bacterial wilt (BW) caused by* Burkholderia solanacearum *is an important bacterial disease for several crops of economic importance in China, including potato, tomato, tobacco, pepper, eggplant, ginger, and groundnut The disease is generally serious in the tropical and subtropical regions of the country and less serious in the North, even though the pathogen has been observed to cause losses in some areas The* B solanacearum *isolates collected from various regions in China have been characterized and divided into five races and five biovars based on their host range and biochemical features Extensive and intensive research on bacterial wilt on the major host crops has been conducted at several institutions, including the Plant Protection Research Institute (PPRI), the Oil Crops Research Institute (OCRI), and the Vegetable Research Institute (VRI) of the Chinese Academy of Agricultural Sciences (CAAS), and some provincial institutions In recent years, the motility of some wild* B solanacearum *strains and its role in infection have been studied The molecular basis of pathogenicity has been investigated through international cooperation Proteins related to BW resistance in potato were isolated, and the gene sequence was analyzed Regenerates of potato from genetic transformation have been found with improved resistance to BW Progress has been made in varietal improvement in tomato, tobacco, potato and groundnut Cultural control practices, including rotation with paddy rice and other non-host crops, have been widely used*

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**Duan Naixiong, Liao Boshou, He Li Yuan, and Gao Guoqing** 1998 A brief introduction to bacterial wilt research in China Pages 15–19 *in* Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

## Introduction

Bacterial wilt (BW) caused by *Burkholderia solanacearum* (E F Smith) Yebucchi et al is considered one of the most important bacterial disease of plants worldwide Even though the pathogen was first described (as *Pseudomonas solanacearum*) and published by Erwin F Smith in 1896, the disease had already existed for a long period in many tropical, subtropical, and temperate zones of the world Plant species in more than 50 families have been observed to be susceptible to the pathogen Recently, 100 years after its discovery, the pathogen was re-named and classified as *B solanacearum* During the past century, over 4000 papers have been published on various aspects of BW (Kelman 1998), reflecting active research on the disease

## Host Range, Distribution and Economic Importance

In China, BW caused by *B solanacearum* is an important disease on many crops of economic importance (He 1986) The disease was first recorded in the country in the 1930s Important host plants include potato (*Solanum tuberosum*), tomato (*Lycopersicum esculentum*), tobacco (*Nicotiana tabacum*), groundnut (*Arachis hypogaea*), eggplant (*Solanum melongena*), pepper (*Capsicum annum*), ginger (*Zingiber officinale*), sesame (*Sesamum indicum*), sweet potato (*Ipomoea batatas*) mulberry (*Morus alba*) and eucalyptus (*Eucalyptus* spp)

Bacterial wilt is generally more serious in southern regions of China, where temperatures are generally high year round and annual precipitation is also generally high The disease is less important in the north, where low temperatures, especially during winter, do not favor the survival of the pathogen However, the epidemiology of bacterial wilt in different hosts is also closely related to soil type and other ecological factors For example, sesame BW was observed in Jiangxi Province but not in Hubei Province, where bacterial wilt is serious in groundnut and tomato Sweet potato BW has been observed to occur only sporadically

Currently, it is difficult to estimate the direct economic losses caused by bacterial wilt in the main host crops Potato, groundnut, tobacco, and tomato are crops widely sown in the country, and yield losses from the disease in these crops are estimated to be very large For groundnut, it is estimated that the area covered by infested fields has increased to over 300 000 ha (Liao et al 1998) and that average yield loss is 7–10%, even when wilt resistant cultivars are extensively sown

## Concerned Research Institutions in China

There are several institutions where research is currently conducted on BW in China A brief description of these institutions and the area of research on BW being conducted in them is as follows

At PPRI, Beijing, BW has been an important research priority for over two decades This laboratory has well-equipped facilities for BW research Research areas include collection of pathogen isolates, characterization of isolates based on virulence and pathogenicity, molecular mechanisms of resistance to BW, and biological control of BW of potato

At the Tobacco Research Institute, CAAS, located in Yidu, Shandong Province, the main activities are collection and evaluation of tobacco germplasm This Institute coordinates research on resistance screening and breeding for BW resistance throughout China

Research on screening and breeding for tomato, eggplant, and pepper for resistance to BW is coordinated by the Vegetables Research Institute (VRI), CAAS, Beijing

At the South Potato Center, located in Enshi, Hubei Province, potato germplasm evaluation and breeding for BW is being conducted Similarly in Fujian Academy of Agricultural Sciences, Fuzhou, research on control of tobacco BW is being conducted

Additionally, work on breeding for resistance to BW of tomato is being undertaken at Jiangsu Academy of Agricultural Sciences, Nanjing

The molecular basis of pathogenicity of *B solanacearum* is being studied at the Guangxi University, Nanning

Several other institutions are conducting research on groundnut BW The leading institution is OCRI, located in Wuhan Research has been conducted at OCRI in various aspects, including germplasm screening, breeding, pathology, and integrated control At the Crop Research Institute of Guangdong Academy of Agricultural Sciences, Guangzhou, extensive research has been done on varietal improvement for host resistance and integrated control Germplasm screening and breeding research is also being conducted in the Industrial Crops Research Institute of Guangxi Academy of Agricultural Sciences, located in Nanning, the Agricultural Research Institute of Nanchong City in Sichuan Province, and Fujian Agricultural University located in Fuzhou

## Significant Achievements

### Motility of wild *B solanacearum* strains

In PPRI, Beijing, 30 of 140 wild type strains of *B solanacearum* were observed to have motility in semi-solid motility medium (SMM) with agar after 48 hours incubation and in SMM medium without agar after 4 hours Motile wild type

strains could move dramatically toward and accumulate near root segments of host plants and some non host plants Some motile wild type strains could cause earlier and higher disease incidence in potato plants than could others It was speculated that motility was related to the infectivity of some *B solanacearum* strains

### Protein related to bacterial wilt resistance in potato

In PPRI, Beijing, a 32-kD protein (AP1) that could inhibit *B solanacearum* was purified from a BW resistant potato variety The biochemical characterization, antimicrobial spectrum, and ultrastructural localization of AP1 were investigated AP1 might be a new kind of plant-specific constitutive disease resistant protein (Yuan and He 1998)

### Transgenic potato plants

In the Biotechnology Research Center, CAAS, Beijing, transgenic potato plants with improved resistance to BW have been obtained This was the first case of improvement of resistance to *B solanacearum* through genetic engineering (Jia 1996, personal communication)

### New resistant cultivars in some vegetables and groundnut

In several institutes, new varieties of tomato and eggplant with improved resistance to BW have been developed and released for production Several new groundnut cultivars with desirable resistance have been developed

## Further Research Needs

Bacterial wilt has attracted much research effort in recent years in China The disease is still important in terms of its wide distribution and host range, the considerable economic losses it causes, and the potential for its further dissemination Further research efforts will be made to address the following

- The molecular basis of virulence or pathogenicity of *B solanacearum* will be further investigated
- Biotechnology approaches for resistance improvement will be widely used, especially for those species in which no desirable resistance sources have been identified for conventional breeding
- Varietal improvement will be an important research priority in various crops for both resistance to BW and agronomic traits
- Biological control of BW will be further investigated especially in potato

- Cultural control approaches will be investigated and applied for most host crops
- Technology transfer to farmers will be intensified

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# Groundnut Bacterial Wilt: Present Status

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# Present Status of Groundnut Bacterial Wilt in South China

**Liang Xuanqiang**[1]


## *Abstract*

*Bacterial wilt caused by* Burkholderia solanacearum *is an important production constraint in over 70% of the groundnut* (Arachis hypogea) *crops in southern China Scientists in Guandong Province have been conducting research on bacterial wilt since the 1950s, developing integrated management measures Crop rotation and field management are effective ways to control the disease, but the most useful strategy is employment of host-plant resistance Several high-yielding varieties resistant to bacterial wilt have been developed and now cover more than 70% of the diseased fields Through intensive breeding, wilt resistance and high yield have been combined with resistance to rust and root rot, in some varieties*


## Disease Distribution and Economic Importance

Groundnut is an important oil and industrial crop in South China (including Guangdong, Guangxi, Jianxi, Fujiang, and Hainan provinces), where it is cultivated on about 800000 ha Bacterial wilt (BW) caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al is a major constraint to groundnut production on over 70% of southern China's production area Yield losses of 15–25% are common The annual loss in pod yield caused by BW is estimated at over $35 \times 10^6$ kg Wilt intensity varies with season, soil type, and cultural practices In general, BW is more severe in spring than in autumn Wilt incidence in rice fields with rich clay soil on high tablelands is less than on nonirrigated uplands, poor, sandy soils, or low tablelands Good cultural practices and resistant cultivars help to reduce BW incidence

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Scientists in Guangdong have been conducting research on BW since the 1950s and have developed integrated BW management measures Additionally, they have released several cultivars with resistance to BW Improved management practices and cultivars have been widely adopted As a result, mean yield in South China has increased from 1 53 t $ha^{-1}$ in the 1980s to 2 0 t $ha^{-1}$ in 1997 Some BW management measures are described here

## Crop Rotation

Crop rotation has many advantages It can maintain and increase soil fertility and reduce soilborne insect pests and diseases, including soilborne propagules of BW To minimize BW incidence and severity, crop rotation has been widely used in South China since 1970 In South China, groundnut is cultivated in two crop-rotation systems 1 rotation with paddy rice in lowlands, usually with a cycle of 2 to 3 years (the longer the rotation cycle, the more effective the control), and 2 rotation with sweet potato, jute, maize, sugarcane, soybean, black bean, mung bean or other non BW-host crops in upland areas, always with a rotation cycle of 2 to 3 years In South China, 60% of the groundnut crop is in upland areas Rotation with BW host crops — for example tobacco, tomato, eggplant, and pepper – is discouraged

## Field Management

Such suitable cultural practices as flooding can effectively control BW In the 1995 spring season in Guangzhou, flooding a field with a wilt incidence of 42% for 30–45 days before sowing of groundnut lowered wilt incidence to 5%, an approximately ninefold reduction Improved field drainage also helps reduce BW incidence and severity if followed before the groundnut crop starts flowering Experiments conducted on the management of soil moisture showed that 60% soil moisture is an important threshold if soil moisture is above 60%, the BW pathogen can easily reproduce

Some field observations and greenhouse tests showed that soil with pH 5 0–6 8 is favorable for pathogen growth In general, alkaline soils suppress BW pathogen growth In Xixin county, groundnut is rotated repeatedly with tobacco (a host crop of BW) in purple soil with pH 8–9, but the incidence of wilt is always low (≤5%) Field sanitation practices such as burning crop residues and removing weeds — especially *Agerratum conyzoides* — also help reduce BW incidence Other agronomic practices, including soil application of organic manure seem to reduce BW incidence Chemicals, including fertilizers and nutrients, have not been found to control BW

## Host-Plant Resistance

Genetic resistance to BW is recognized as the most effective disease management strategy In the past three decades, great emphasis has been placed on managing BW through host plant resistance About 60 genotypes, including local landraces (led by Taishan Sanlirou, Taishan Zhenzhou, and Xiekangqing), exotic varieties from ICRISAT (led by Gajah, Schwarz 21, ICG 1703, PI 3933513, and CS 30), and wild *Arachis* species have been identified as sources of high, stable resistance to BW in artificial inoculation tests and multilocational sick plot tests Several high-yielding BW resistant varieties e g , Yue You 589, Yue You 92, Yue You 256, Wu You 4, Guiyou 28, and Yue You 200 have been developed through the modified pedigree method and released for general cultivation in South China These cultivars are grown on more than 70% of the diseased fields, playing an important role in controlling the disease, and significantly increasing groundnut production in South China

## Breeding for Multiple Resistance to BW, Rust, and Root Rot

Yue You 256 and Yue You 200 have a high level of resistance to BW, as well as moderate resistance to rust and high susceptiblity to root rot caused by *Fusarium* spp These two widely distributed fungal diseases cause substantial yield losses in South China Since 1988, incorporation of resistance to BW, rust, and root rot into high-yielding cultivars has received high priority From screening of about 2000 germplasm accessions, Yinduhuapi, a runner type groundnut from ICRISAT, India, was identified as possessing high levels resistance to all three diseases Through large-scale hybridization, a potential breeding line Yue You 39 with high yield and high resistance to BW, rust, and root rot was developed But Yue You 39 has some agronomically unacceptable characters such as late maturity, low shelling percentage, and too many aerial pegs Though backcrossing with high yielding, early-maturing cultivars, two varieties, Yue You 202 35 and Yue You 79, with higher yield, resistance to BW, rust, and root rot, and wide adaptation were developed and released in Guangdong province These two varieties are gradually replacing Yue You 92 and Yue You 256 in that region

Yue You 202-35 was developed from the cross Yue You 39 54 × Yue You 116, and was released by Guangdong Crop Varieties Approval Committee in 1996 During 1992–94, in advanced groundnut varieties trials, the average yield of Yue You 202-35 was 3 87 t $ha^{-1}$ over 13 locations, a net 23 4% gain over Yue You 116, the control The groundnut cultivar Yue You 202 35 is of medium duration, taking 120 days to mature It has an erect growth habit and dark green leaves The seeds contain 54 3% oil and 26% protein The reactions of Yue You 202 35 to BW, rust, and root rot are excellent (Table 1)

**Table 1 Performance of some groundnut varieties resistant to bacterial wilt released in South China[1]**

| Cultivars | Yield (t ha$^{-1}$) | BW incidence (%) | Rust (score) | Root rot incidence (%) | Resistance source used[2] | Year of release |
|---|---|---|---|---|---|---|
| Yue You 589 | 1 6 | 7 8 | 6 0 | –[3] | Taishan Sanlirou | 1970 |
| Yue You 92 | 2 6 | 4 2 | 5 0 | – | Taishan Sanlirou | 1983 |
| Yue You 256 | 2 8 | 3 4 | 3 8 | – | AH 1722 | 1987 |
| Gui You 28 | 2 2 | 5 0 | 5 5 | – | Taishan Sanlirou | 1986 |
| Wu You 4 | 2 7 | 2 3 | 4 5 | – | PI 393513 | 1988 |
| Yue You 200 | 3 1 | 4 5 | 6 5 | – | Taishan Sanlirou | 1992 |
| Yue You 202–35 | 3 8 | 4 0 | 2 5 | 2 0 | Yinduhuapi | 1996 |
| Yue You 79 | 4 1 | 4 2 | 2 5 | 2 0 | Yinduhuapi | 1997 |

1 Results from South China Groundnut Varieties Tests and Provincial Groundnut Varieties Tests
2 Resistance sources used in breeding these varieties
3 Free from root rot disease incidence

Yue You 79 was derived from a cross involving Yue You 202 35 × Yue You 116, and released by Guangdong Crop Varieties Approval Committee in 1997 During 1996/97, in Guangdong Groundnut Varieties Trials, its average yield was 4 09 t ha$^{-1}$ over 10 locations, a net increase over Yue You 202 35 It matures in 120 days in the spring season and 115 days in the summer season It has an erect growth habit and dark green leaves Disease scores of BW, rust, and root rot of Yue You 79 are similar to those of Yue You 202 35 (Table 1) Yue You 202-35 and Yue You 79 have good stability and adaptability for pod yield when sown in either rice-paddy or upland conditions

# Status of Groundnut Bacterial Wilt in Central China

**Liao Boshou[1], Duan Naixiong[1], Tan Yujun[1], and Wu Yong[2]**

## *Abstract*

*Bacterial wilt (BW) caused by* Burkholderia solanacearum *in groundnut was first reported in central China in the 1950s The disease has been a constraint to groundnut cultivation in several provinces in central China, including Sichuan, Chongqing, Hubei, Henan, Anhui, Jiangxi, Hunan, and Guizhou (Liao and Zeyong 1997) The total infested groundnut area is estimated at over 150000 hectares in the central provinces The disease is becoming more widespread because of expansion of groundnut and its continuous cultivation The disease generally causes yield losses of 10 to 30% in central China if susceptible groundnut cultivars are grown, but wilt incidence in the traditional problem areas has been significantly reduced by sowing improved resistant cultivars Therefore, as in most other diseased regions, the most important control approach for BW in the central provinces is to grow resistant cultivars, including E Hua 5, Zhong Hua 2, Lu Hua 3, Yue You 200, and 93-81 Other practical control approaches include rotation with paddy rice or with such non-host upland crops as wheat and rapeseed The Oil Crops Research Institute of the Chinese Academy of Agricultural Sciences has been the leading research agency addressing the problem in this region In the future, genetic enhancement for resistance and other traits will continue to be the most important control strategy in this region*

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**Liao Boshou, Duan Naixiong Tan Yugun and Wu Yong** 1998 Status of groundnut bacterial wilt in central China Pages 27–30 in Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

## Introduction

Bacterial wilt caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al has been an important disease of groundnut (*Arachis hypogaea* L ) for several decades in central China In the eastern part of Hubei Province, the disease was first recorded in Huangzhong Agricultural College in the 1950s, but the local farmers were well acquainted with the wilt problem earlier than this Groundnut bacterial wilt became more serious in the 1960s and was reported from all other groundnut-growing provinces of central China Progress in disease control has been made in the past two decades using integrated control approaches

### Distribution and Severity

Currently, bacterial wilt of groundnut is a serious constraint to groundnut production in several provinces in central China, including Sichuan, Chongqing, Hubei, Henan, Anhui, Jiangxi, Hunan, and Guizhou The disease has also been reported in Zhejiang and Jiangsu Provinces on the east coast, but economic losses were low there (Liao and Zeyong 1997)

The total area infested with BW in central China is over 150000 ha The disease is becoming more widespread with the expansion of groundnut cultivation in most of the provinces in central China However, wilt incidence in traditionally endemic areas has significantly decreased because resistant cultivars are being grown (Zhou and Wei 1996) The disease generally causes yield losses of 10 to 30% in susceptible groundnut cultivars At present, BW incidence in central China ranges between 2 and 12% wherever resistant cultivars are grown

## Disease Control

As in many other regions, the most important control approach for groundnut BW is to grow resistant cultivars Currently, the following resistant cultivars are being grown in the central provinces

E Hua 5 is highly resistant to BW It has small seeds, matures early, and has desirable uniformity of pod shape This cultivar was developed at the Oil Crops Research Institute (OCRI) and released in 1985 and is used in Hubei, Henan, and Jiangxi Provinces The yield of E Hua 5 is only moderate, but it has desirable tolerance to shallow soil and poor fertility, so that it can be grown in upland fields

Zhong Hua 2 is highly resistant to BW, with medium-sized seed and high seed protein content It was developed at OCRI, released in 1990, and is extensively cultivated in the central provinces However, the cultivar has poor seed dormancy, so preharvest germination usually causes up to 10% loss

Lu Hua 3 is moderately resistant to BW, with medium-sized seed It was developed at Shandong Peanut Research Institute (SPRI) Since 1986 it has been cultivated in some regions of Sichuan and Henan Provinces in central China

Yue You 200 is a highly resistant cultivar with improved yield potential and drought tolerance It was developed at the Crops Research Institute of Guangdong Academy of Agricultural Sciences and released in Sichuan Province in 1997 as Tianfu 11 93-81 (ZH 212) A highly resistant breeding line with improved yield potential and tolerance to acid soil was also developed at OCRI

Other practical control approaches include rotation with paddy rice and with some non-host upland crops such as wheat or rapeseed However, the area that can be rotated with paddy rice in central China is very small Long term rotation with non-host crops has also become less feasible in this region because of limited arable land and expanded groundnut cultivation

## Research Organizations

In central China, the OCRI of the Chinese Academy of Agricultural Sciences (CAAS), located in Wuhan, is the leading research organization addressing groundnut BW The scientists at OCRI are responsible for coordinating groundnut BW research, conducting groundnut germplasm collection and evaluation for resistance to BW, conventional breeding, and resistance improvement through biotechnology Nanchong Agricultural Research Institute in Sichuan Province is also engaged in breeding for resistance to BW

In central China, there is a network of eight sites for evaluation for resistance to BW in groundnut varietal trials

## Problems and Further Research Needs

Incidence of wilt diseases other than BW (probably caused by soilborne fungi) has significantly increased in recent years These soilborne fungal diseases have obviously complicated the further increase of groundnut productivity in regions where BW is a problem

The sizes of pathogen populations in infested fields after long term cultivation of resistant cultivars is unknown It would be interesting to investigate the dynamics of population pathogenicity in soil Farmers would take an interest in knowing whether susceptible groundnut cultivars with higher yield could be grown in diseased fields after cultivation of resistant ones for several years

The yield potential of most cultivars resistant to BW is often lower than that of susceptible ones Further breeding efforts should be made to combine resistance and high yield

Despite all of our efforts, there are areas where BW incidence is still very high because of continuous cultivation of susceptible groundnut cultivars Therefore, efforts will be made to extend BW-resistant cultivars to farmers in such areas

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# Status of Groundnut Bacterial Wilt in Northern Vietnam

**Nguyen Xuan Hong, Nguyen Van Viet, and Nguyen Thi Yen**[1]


## *Abstract*

*Groundnut bacterial wilt (BW) is a widespread disease in the major groundnut-producing areas of northern Vietnam The disease is increasingly serious in many parts of Nghe An, Ha Tinh, Thanh Hoa, and other provinces Bacterial wilt is severe not only in traditional groundnut-growing areas but also in newly cultivated areas Wilt incidence in groundnut-producing areas has increased substantially in the past few years It is highest in dryland cropping systems, especially in upland areas of hilly regions in light sandy soil along river banks and in fields where groundnuts are intercropped with dryland crops Rotation of groundnut with rice is effective in reducing wilt incidence The disease is more serious in the autumn crop Most local varieties are susceptible to disease Research on identification of wilt-resistant genotypes, development of improved varieties, and integrated management of BW is needed*


## Introduction

In northern Vietnam, groundnut is cultivated mainly in the spring season (February to June) It is the most important food legume crop of Thanh Hoa, Nghe An, Ha Tinh, and other provinces The area under groundnut increased from 102200 ha in 1992 to 123300 ha in 1995 Although soil and environment conditions are favorable for groundnut production, average yields are still low (1 09 t $ha^{-1}$) Bacterial wilt (BW) caused by *Burkholderia solanacearum* (E F Smith) Yebuuchi et al is widespread in major groundnut-producing areas and has become one of major constraints to groundnut production This report summarizes the results of surveys on status of groundnut bacterial wilt conducted by scientists of Vietnam Agricultural Science Institute (VASI) during the last year

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**Nguyen Xuan Hong Nguyen Van Viet,** and **Nguyen Thi Yen** 1998 Status of groundnut bacterial wilt in northern Vietnam Pages 31–36 in Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

## Distribution and Severity of Disease in Different Production Systems

In northern Vietnam, groundnut is concentrated mostly in Nghe An, Ha Tinh, Thanh Hoa, and Bac Giang provinces, and some areas of other provinces Groundnut is cultivated mainly under rainfed conditions in hilly and upland areas, and along river banks in dryland cropping systems Groundnut is also grown in rotation with rice in the spring season The area of groundnut has significantly increased in the last 3 years (Table 1)

Bacterial wilt has undoubtedly been present on groundnut in northern Vietnam for many years, although it was first documented in detail in 1991 (Mehan et al

**Table 1 Sown area and yield of groundnut in northern Vietnam**

| | Sown area (ha × $10^3$) | | Yield (t $ha^{-1}$) | |
|---|---|---|---|---|
| | 1992 | 1995 | 1992 | 1995 |
| **Region** | 217 3 | 259 9 | 1 04 | 1 28 |
| North | 102 2 | 123 3 | 0 74 | 1 09 |
| North Mountain and midland | 34 2 | 41 6 | 0 80 | 0 96 |
| Red River delta | 17 5 | 17 7 | 0 90 | 1 30 |
| North Central coast | 50 5 | 64 0 | 0 64 | 1 13 |
| **Province** | | | | |
| Bac Thai | 5 2 | 6 7 | 0 64 | 0 83 |
| Hoa Binh | 2 6 | 3 9 | 0 69 | 0 92 |
| Quang Ninh | 2 7 | 2 9 | 0 79 | 0 85 |
| Vinh Phu | 6 8 | 9 4 | 0 79 | 1 03 |
| Ha Bac | 8 3 | 8 6 | 1 00 | 1 07 |
| Hanoi | 2 4 | 3 3 | 0 78 | 1 09 |
| Ha Tay | 3 4 | 4 6 | 0 95 | 1 12 |
| Hai Hung | 2 3 | 2 9 | 1 02 | 1 42 |
| Nam Ha | 3 4 | 3 7 | 0 79 | 1 33 |
| Ninh Binh | 4 1 | 4 4 | 0 86 | 0 70 |
| Thanh Hoa | 11 5 | 13 6 | 0 74 | 1 11 |
| Nghe An | 21 5 | 27 0 | 0 60 | 1 22 |
| Ha Tinh | 9 8 | 13 8 | 0 58 | 1 13 |
| Quang Binh | 2 7 | 2 8 | 0 50 | 0 59 |

1991, Hong et al 1994) It is widely distributed in all areas of groundnut production Some regions are heavily infected, while in other areas the disease is sporadic It is most severe in Nghe An, Ha Tinh, Thanh Hoa, Ha Tay, Bac Giang, and Thai Nguyen provinces In general the average wilt incidence ranges from 0 2–16 9%, but in the most heavily infected fields it reaches 70–80% The disease is severe not only in the traditional groundnut producing areas (Nam Dam and Nghia Dan districts of Nghe An province, Ha Trung, Hoang Hoa, Tinh Gia districts of Thanh Hoa province, Lang Giang and Viet Yen districts of Bac Giang province) but also in newly cultivated areas in Ha Tay, Bac Ninh, Thai Nguyen, and Hoa Binh provinces

Wilt incidence in traditional groundnut-producing areas has increased very fast In fields where groundnut has been grown year after year, especially in upland areas and in the soils along the river banks, wilt incidence has significantly increased Crop rotation and host resistance have been considered important to manage BW in these areas

Wilt incidence varies greatly from place to place, depending on cropping patterns and soil conditions The disease is most severe in hilly and riverbed areas where wilt incidence ranges from 3–50% (Table 2)

**Table 2 Bacterial wilt of groundnut in different ecological conditions of northern Vietnam, 1995**

| Area | Number of locations surveyed | Wilt incidence (%) | |
|---|---|---|---|
| | | Range | Average |
| Hilly | 73 | 5–50 | 16 9 |
| River beds | 105 | 3–50 | 13 8 |
| Upland | 109 | 1–15 | 5 9 |
| Rice-groundnut rotation | 243 | 0–8 | 0 2 |

In upland areas of hilly regions, wilt incidence is highest in fields where groundnut is grown twice a year In these fields wilt incidence was 10–60% The disease was also found (3–50%) in sandy soils along river banks where only dryland crops such as groundnut, maize, tomato, and other vegetables were grown Wilt was also present (0–15%) in fields where groundnut was intercropped with vegetables, maize, or mung bean In irrigated rice-based, groundnut-growing areas, wilt incidence was lowest (0–5%) (Table 3) Thus, rotation of groundnut with

irrigated rice is the most effective way to control bacterial wilt Similar results have also been reported in China (Tan et al 1994)

**Table 3 Wilt incidence (%) of groundnut in different production systems in Vietnam, 1995**

| Location | Main cultivar | Hilly with 1 crop of groundnut | Hilly with 2 crops of groundnut | River banks | Dryland crop systems | Rice–groundnut rotation |
|---|---|---|---|---|---|---|
| Ha Tinh | Sen NA | 5–25 | – | 3–35 | 3–15 | 0–2 |
| Nghe An | Cuc NA | 5–35 | 10–45 | 5–50 | 1–10 | 0–5 |
| Thanh Hoa | Chum NA | 10–25 | 15–50 | 3–20 | 1–12 | 0–1 |
| Ninh Binh | Sen Lai | 10–25 | 15–50 | – | 1–15 | 0–5 |
| Bac Giang | Do BG | 5–20 | 10–45 | – | 1–10 | 0–5 |
| Thai Nguyen | Chay HB | 5–20 | 15–30 | 5–20 | 1–10 | 0–5 |
| Vinh Phuc | Su tuyen | 10–25 | 15–40 | 5–35 | 3–10 | 0–3 |
| Hanoi | Sen NA | 5–25 | – | – | 0–5 | 0–2 |
| Ha Tay | Tram Xuyen | 15–30 | 15–60 | 3–35 | 1–12 | 0–1 |

**Table 4 Influence of cropping season on the incidence of groundnut bacterial wilt (%) at six locations in Vietnam, 1995**

| | | Spring crop (Feb–Jun) | | Autumn crop (Jul–Nov) | |
|---|---|---|---|---|---|
| Location | Cultivar | Average | Highest | Average | Highest |
| Ha Tinh | Chum Nghe An | 20–25 | 70 | 30–35 | 90 |
| Ha Tinh | Cuc Nghe An | 15–20 | 50 | 20–25 | 80 |
| Bac Giang | Do Bac Giang | 15–20 | 70 | 20–25 | 90 |
| Bac Giang | Su Tuyen | 15–25 | 75 | 25–30 | 95 |
| Ha Tay | Sen Lai | 10–15 | 60 | 20–25 | 80 |
| Ninh Binh | 4329 | 5–7 | 40 | 15–20 | 70 |

Bacterial wilt is widespread in all groundnut-producing areas year-round However, it is more serious in autumn (Jul–Nov) in areas where the crop is grown year after year Spring crop incidence reached a maximum of 40–75% in a 1995 survey, maximum incidence in the autumn season was even higher, reaching 70–95% (Table 4)

The majority of currently grown groundnut varieties are highly susceptible to BW Only two genotypes, Tram Xuyen and 4329, have shown moderate levels of susceptibility to wilt under high disease pressure (Table 5) In recent years, germplasm accessions have been extensively evaluated for resistance to bacterial wilt (Hong et al 1994) but more research on identification of wilt-resistant groundnut genotypes is needed

**Table 5 Bacterial wilt reaction of major groundnut varieties popularly grown in northern Vietnam (1994/95)**

| Varieties | Wilt incidence in hot–spot locations (%) | Disease reaction[1] |
|---|---|---|
| Sen Nghe An | 70 2 | S |
| Sen Lai | 50 7 | S |
| Do Bac Giang | 66 2 | S |
| Su Tuyen | 52 8 | S |
| Tram Xuyen | 42 3 | MS |
| Chum Nghe An | 68 2 | S |
| V 79 | 53 9 | S |
| 4329 | 41 9 | MS |

1 S = susceptible MS = moderately susceptible

## Conclusion and Suggestions

Bacterial wilt is one of the most important constraints to groundnut production in northern Vietnam In recent years, yield losses caused by the disease in farmers' field have significantly increased Major groundnut varieties popularly grown in northern Vietnam are susceptible to the disease Further genetic evaluation to identify sources of resistance to BW is needed Research on development of improved varieties with multiple disease resistance to BW and other major diseases should be given high priority More systematic research needs to be undertaken on integrated management of groundnut BW, using host plant resistance and cultural practices, particularly crop rotation, in order to alleviate losses caused by the disease in major groundnut–growing areas of northern Vietnam

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# Bacterial Wilt of Groundnut in Southern Vietnam

**Nguyen Van Tung[1], V K Mehan[2], Phan Lieu[1], and Ngo Thi Lam Giang[1]**


## *Abstract*

*Bacterial wilt (BW) caused by* Burkholderia solanacearum *is becoming increasingly prevalent and severe in the groundnut* (Arachis hypogaea L ) *crop of southern Vietnam Scientists at the Oil Plant Institute in Hanoi have carried out a detailed survey of BW in southeastern Vietnam Incidence and severity was low in fields under groundnut-rice-rice cropping systems and moderate to severe (up to 40% wilting) under groundnut-groundnut-rice systems The disease was more serious in the summer-autumn season because of high soil temperature and moisture Nine resistant ICRISAT breeding lines had good wilt reactions over 2 years, but their shelling percentage was lower than that of local varieties Integrated disease management systems are needed to control fungal diseases and BW*


## Introduction

Bacterial wilt (BW) is a serious constraint to groundnut production in many regions of Vietnam Depending upon the agro-ecological condition and crop rotations, the severity of BW varies across a region

In collaboration with ICRISAT, the Oil Plant Institute (OPI) in Hanoi has carried out a detailed survey to determine the status of this disease in some groundnut-growing districts in the southeast region of Vietnam Also during these surveys, disease was monitored on bacterial wilt-resistant groundnut lines sent from ICRISAT The results of this survey are presented in this paper


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## Survey of Bacterial Wilt Incidence

During the winter-spring season of 1995/96, an expert from ICRISAT (Dr V K Mehan) directly collaborated with us in carrying out the survey in four districts In each district, 5 to 6 villages and 5 to 50 groundnut fields were surveyed Data collection included cropping system, crop growth stage, BW incidence, and crop cultivars grown in each district (Table 1)

### Trang Bang district

In this district, BW incidence was 0–28% (mostly 16–18% on 60–70 day-old crops) in Loc Hung Village, and 0–9% in Gia Loc village where farmers follow a groundnut-groundnut-rice (G G-R) cropping system Incidence was 6–12% in fields with a groundnut-fallow-vegetable (G-F-V) cropping system

Low incidence (0–3%) of BW was recorded in 35–50 day old crops in fields with a G-G-R cropping system in An Tinh and Don Thuan villages In fields in Loc Hung and Don Thuan villages where a groundnut-rice-rice (G-R-R) rotation was followed, BW occurred at low incidence (0–4%) It was seen in a few fields (incidence <1% in 35–50 day-old crops) in An Tinh village

### Go Dau district

In Go Dau district, BW was very severe in Phuoc Dong village It was prevalent in almost all fields with a G-G-R cropping system, with incidence of 11–43% (mostly 25–30%) in 65–75 day old crops, and 2–8% in 35–60 day old crops Some apparently healthy plants also showed latent bacterial infection in their roots Incidence commonly reached 30–40% in the winter-spring season, and 40–50% in the summer-autumn season Because of the increasing severity of BW in this village, many farmers are now switching to maize and vegetables in the summer-autumn season Bacterial wilt was observed in all fields with G-G-R cropping systems surveyed in Hiep Thanh village, with incidence of 2–12% The disease occurred in most fields in Phuoc Thanh village (incidence 3–10% in 60–85 day old crops, and 0–5% in 35–55 day old crops) In Thanh Phuoc and Bau Don villages, bacterial wilt was present at low incidence in 30–55 day old crops, in fields with G R-R (0–2%) and G-G-R (2–5%) It occurred at moderate incidence (8–10%) in 70 day old crops with a groundnut groundnut-vegetable (G-G-V) rotation in Thanh Phuoc village

### Cu Chi district

Bacterial wilt was observed at 1–4% in 50–65 day-old crops in G G R, and 2–8%

**Table 1. Bacterial wilt on groundnut in southern Vietnam, winter–spring 1995/96, season**

| District | Village | Cropping system[1] | Crop age (days) | No of fields surveyed | BW incidence (%) |
|---|---|---|---|---|---|
| Trang Bang | Loc Hung | G–G–R | 27–70 | 30 | 0–28 |
| | | G–R–R | 25–50 | 15 | 0–4 |
| | Gia Loc | G–G–R | 45–60 | 30 | 0–9 |
| | | G–F–V | 45–85 | 10 | 6–12 |
| | Don Thuan | G–G–R | 50–60 | 10 | 1–3 |
| | | G–R–R | 45–70 | 25 | 0–4 |
| | An Tinh | G–G–R | 50–60 | 5 | 0–3 |
| | | G–R–R | 35–50 | 15 | 0–1 |
| Go Dau | Phuoc Dong | G–G–R | 65–75 | 10 | 11–43 |
| | | G–G–R | 35–60 | 20 | 2–8 |
| | Hiep Thanh | G–G–R | 40–65 | 10 | 2–12 |
| | Phuoc Thanh | G–G–R | 35–55 | 10 | 0–5 |
| | | G–G–R | 60–85 | 10 | 3–10 |
| | Thanh Phuoc | G–G–R | 40–70 | 10 | 2–5 |
| | | G–R–R | 40–55 | 8 | 0–2 |
| | | G–G–V | 10 | 2 | 8–10 |
| | Bau Don | G–R–R | 40–55 | 10 | 0–2 |
| | | G–G–R | 30–50 | 5 | 0–4 |
| Cu Chi | Trung Lap Ha | G–R–R | 50–65 | 8 | 1–4 |
| | | G–R–R | 35–40 | 5 | 0 |
| | | G–G–R | 60–75 | 7 | 2–8 |
| | Phuoc Thanh | G–G–R | 55–60 | 10 | 2–4 |
| | | G–R–R | 55–65 | 5 | 0–2 |
| | Phuoc Hiep | G–R–R | 30–55 | 15 | 0–5 |
| Duc Hoa | Hoa Khanh Dong | G–G–R | 40–55 | 10 | 1–3 |
| | | G–R–R | 40–55 | 7 | 0–2 |
| | | G–F–G/R | 70–80 | 8 | 7–8 |
| | Duc Lap Thuong | G–G–R | 40–60 | 20 | 0–6 |
| | Duc Hoa Thuong | G–G–R | 50–75 | 15 | 1–6 |
| | | G–R–R | 35–48 | 10 | 0 |
| | Duc Lap Ha | G–G–R | 40–80 | 15 | 0–8 |
| | | G–R–G/R | 50–75 | 5 | 4–8 |

1 G = groundnut R = rice V = vegetable F = fallow

in fields in G-R R cropping systems in Trung Lap Ha village of Cu Chi district In the latter cropping system, a number of plants in some fields showed initial disease symptoms The disease was not visible in 35–40 day old crops

In Phuoc Thanh village, bacterial wilt incidence was 2–4% in G-G-R cropping systems, and 0–2% in 35–40 day-old crops in G-R R cropping systems

### Duc Hoa district

Bacterial wilt incidence was 0–6% in fields with G-G-R cropping system surveyed in Duc Lap Thuong and Duc Hoa Thuong, and 0–8% in Duc Lap Ha village It was not observed in 35–48 day-old crops in black soil fields in Duc Hoa Thoung village In Hoa Khanh Dong village, BW incidence was low in 40–55 day old crops in fields with G G-R (1–3%) and G-R-R (0–2%) cropping systems

Bacterial wilt occurred at moderate incidence in 50–80 day-old crops with a G F-G rotation in Duc Lap Ha (4–8%) and Hoa Khanh Dong (7–8%) villages

## BW Incidence and Yield of ICRISAT Groundnut Lines

From the several groundnut lines that were identified by ICRISAT as sources of resistance to BW, OPI selected nine lines and evaluated them for agronomic and other characteristics, including BW reaction, in two seasons Mean duration of these varieties (90 days) was at par with a local variety (Table 2) Among these lines, ICGV 8666 had the highest resistance (BW incidence=0 1%) In other characteristics, such as shelling and oil content, it also corresponded to a local variety (Table 2)

In the winter-spring season of 1997 98, we carried out trials at five locations to compare the performance of ICG 8666 and ICM 8645 with that of a local variety (Table 3) These two resistant genotypes were liked by the farmers

## Conclusions

- Bacterial wilt is becoming increasingly prevalent and severe, as a result of continuous cropping and irrigation practices
- Incidence and severity was low in G R R cropping system, and moderate to severe (30–40%) in fields with a G-G-R cropping system
- Incidence and severity were higher in the summer-autumn season, mainly due to high soil temperature and moisture content
- Farmers approved of the resistance, duration, pod yield, and other traits of ICG 8666, 8645, 8675 However, there is a need to improve the shelling percentage of these varieties

**Table 2. Bacterial wilt incidence and yield of some selected groundnut varieties, summer–autumn 1995 and winter-spring 1995/96, in Trang Bang district, Vietnam**

| Varieties | BW incidence (%) | Shelling (%) | Strong seed (%) | 100–seed mass (g) | Oil content (%) | Pod yield (kg ha⁻¹) S–Autumn | Pod yield (kg ha⁻¹) W–Spring |
|---|---|---|---|---|---|---|---|
| ICG 8625 | 1 1 | 65 | 87 | 57 | 43 0 | 935 | 3000 |
| ICG 8626 | 2 2 | 70 | 79 | 59 | 45 8 | 1230 | 3067 |
| ICG 8632 | 1 1 | 67 | 88 | 54 | 44 8 | 1080 | 3300 |
| ICG 8645 | 1 6 | 71 | 87 | 52 | 43 4 | 1180 | 2783 |
| ICG 8654 | 4 3 | 62 | 88 | 54 | 44 6 | 1050 | 2983 |
| ICG 8662 | 3 2 | 70 | 90 | 49 | 44 1 | 1020 | 2667 |
| ICG 8666 | 0 1 | 69 | 90 | 57 | 44 4 | 1280 | 3200 |
| ICG 8675 | 1 9 | 69 | 92 | 50 | 45 1 | 1230 | 3133 |
| Local control | 57 8 | 71 | 86 | 50 | 44 1 | 800 | 2617 |
| CV (%) | 28 7 | | | | 0 3 | | 9 |
| LSD (0 05) | | | | | 0 2 | | 482 |

**Table 3 Yield of groundnut varieties ICG 8666 and ICG 8645 in winter–spring 1997/98 (means of five locations)**

| Varieties | Shelling (%) | Sound kernels (%) | 100–seed mass (g) | Pod yield (kg ha⁻¹) |
|---|---|---|---|---|
| ICG 8666 (VD–9) | 74 4 | 86 8 | 46 0 | 2901 |
| ICG 8645 (VD–10) | 74 2 | 84 3 | 46 1 | 2888 |
| Local control | 78 6 | 84 0 | 42 2 | 2534 |
| CV (%) | 1 6 | 2 2 | 1 6 | 5 |
| SE (±) | 0 6 | 0 9 | 6 9 | 66 |

## Suggestions

- Diseases such as pod rot and damping-off, in addition to BW, play very important roles in groundnut production in southern Vietnam Therefore, integrated disease management (IDM) systems must be established
- In order to find valuable components for building IDM systems, support from ICRISAT and other organizations will be needed

# Technology Development for Disease Diagnosis and Control

# Latent Infection of *Burkholderia solanacearum* in Groundnut and its Role in Resistance Enhancement

**Liao Boshou, Shan Zhihui, Li Dong, Lei Yong,** and **Tan Yujun**[1]


## Abstract

*Bacterial wilt (BW) caused by* Burkholderia solanacearum *is an important disease of groundnut* (Arachis hypogaea L ) *in China, Indonesia, Vietnam, and some other countries Sowing resistant groundnut cultivars is the most efficient and practical way to control the disease Generally, success of breeding for resistance to BW of groundnut and other crops was judged by the percentage of surviving plants of newly bred lines relative to that of the susceptible line under inoculation and infection pressure However, latent colonization of* B solanacearum *without obvious wilting symptoms has recently become common in some resistant cultivars Latent colonization could reduce root proliferation, tolerance to drought, and ultimately, pod yield of infected plants Specific experiments involving artificial inoculation and enzyme-linked immunosorbent assays (ELISA) were conducted to investigate the characteristics of reaction to latent colonization and their variation among resistant groundnut genotypes Frequencies of latent colonization varied among genotypes Low latent colonization was detected in only a few resistant lines, these lines also possessed stable resistance to BW across seasons and locations Genotypes with high latent colonization frequencies were more sensitive to drought stress in late growth stages, even when wilt incidence was low Hypocotyl tissues supported the highest bacterial colonization frequency Improved techniques to detect latent colonization and evaluate resistance have been developed Latent infection is a new trait to be addressed in breeding for BW resistance*


## Introduction

Bacterial wilt (BW) caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al in groundnut (*Arachis hypogaea* L ) has been an important constraint to groundnut

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production in China, Indonesia, and Vietnam for decades and is becoming increasingly widespread in several other countries (Mehan et al 1990, Mehan and Liao 1994, Liao et al 1994) In China, it is believed that in recent years, groundnut fields infected with *B solanacearum* have increased to over 300000 ha The spread of BW has been caused by expansion in area of groundnut cultivation, use of susceptible cultivars in the new growing regions, and lack of rotation with non host crops (Liao et al 1994) It has been well proven that genetic improvement for host resistance is the most important component of any integrated control program for groundnut BW The use of resistant cultivars is the most practical and feasible control method for farmers in many regions (Mehan and Liao 1994) Thus, germplasm screening and breeding for resistance have been the research priorities for alleviating the disease in the past few decades

Even though wilt incidence has been significantly reduced by sowing resistant cultivars, groundnut production and productivity in diseased areas are still relatively low Generally, resistant cultivars have low yield potentials and undesirable reactions to drought and other stresses Latent infection of *B solanacearum* without obvious wilting symptoms in resistant groundnut cultivars can reduce root proliferation, symbiotic nitrogen fixation, tolerance to drought, and yield (Liao et al 1994) These effects appeared to be associated with low productivity of the crop in diseased areas Several solanaceous species, such as tobacco, potato (Ciampi and Sequeria 1980, Ciampi et al 1980), and tomato (Prior et al 1990), are also known to be latently infected by the pathogen without symptoms However, in previous breeding programs, latent infection and its effects were seldom considered Obviously, further breeding efforts should be made to overcome the latent infection problem In this paper, latent infection related to breeding for resistance to *B solanacearum* in groundnut is described, based on the available research results

## Traditional Criteria for Testing Resistance to Bacterial Wilt

The major objective of plant breeders has been to combine favorable agronomic characteristics with acceptable resistance to abiotic and biotic stresses Traditionally, when evaluating resistance to BW in several crops infected by *B solanacearum*, the proportion of wilted to non wilted plants has been the selection criterion, and all components of the interaction between plant, pathogen, and the environment have been taken into account (Prior et al 1994) In previous breeding programs in China and other countries, resistance to BW in groundnut was defined only as high plant survival percentage under specified infection pressure (Mehan et al 1990, Liao et al 1994) When the

survival percentage is over 80%, the genotype is said to be resistant, over 90%, it is highly resistant (Li and Tan 1984, Mehan et al 1990) This definition appeared to be simple and practical in conventional breeding The resistant cultivars were once considered pathogen-free based on the lack of visual occurrence of wilt

In terms of the interaction of plant and pathogen, resistance is defined as any measurable mechanism able to overcome completely or to limit the development of a pathogen or its effects (Prior et al 1994) This definition emphasized the positive control of a pathogen by plant metabolism Tolerance is defined as the overall ability of a plant to withstand the development of a pathogen without major losses in yield (Prior et al 1994) Accordingly, where infection or colonization of *B solanacearum* in groundnut genotypes without wilting symptoms is concerned, the genotypes may be classified as resistant to, or tolerant of the disease But if the yield losses or other undesirable influences on the symptomless plants are considered, the above definitions for resistance or tolerance must be modified Visual occurrence of wilt symptoms is not enough for evaluating resistance to BW

## Influence of Latent Infection of *B solanacearum* on Groundnut

Even though groundnut plants with high survival percentage in naturally diseased fields or artificial inoculation traditionally were thought to be free from *B solanacearum*, latent infection by the pathogen has been found in some of these genotypes Latent infections of *B solanacearum* are also known to occur in cultivars of potato (Ciampi and Sequeria 1980, Bowman and Sequeria 1982), tomato (Prior et al 1990), eggplant, and pepper (Prior et al 1994) including some resistant genotypes Latent infection was thought to be a generalized and common trait of *B solanacearum* pathogenesis (Prior et al 1994)

In repeated artificial inoculation experiments (Liao et al 1997, unpublished data), groundnut seed emergence was reduced by 20–40% by soaking the seeds in a bacterial suspension It was assumed that the invasion of *B solanacearum* into groundnut seeds might encourage the infection of other bacteria or fungi in soil, even if the wilt pathogen could not kill the seeds directly Liao et al (1997) found reduced dry root mass, root volume, number of nodules, pod number plant$^{-1}$, and yield plant$^{-1}$ in resistant groundnut cultivars artificially inoculated with bacteria, compared to noninoculated plants of the same cultivars grown under the same conditions It is interesting to note that there is variation among groundnut genotypes in their reaction to latent infection (Table 1)

**Table 1 Percentage reduction in root mass, root volume, nodules, and yield per plant in groundnut lines artificially inoculated by *B solanacearum* and tested in China**

| Genotype | Root mass | Root volume | Nodules plant$^{-1}$ | Pods plant$^{-1}$ | Yield plant$^{-1}$ |
|---|---|---|---|---|---|
| Xiekangqing | 3 57 | 3 53 | 5 81 | –38 49 | –45 26 |
| Zao 18 | –9 63 | –19 72 | –16 23 | –23 19 | –29 36 |
| Taishan Zhenzhu | –10 97 | –11 23 | –30 82 | –23 80 | –18 44 |
| ZH 212 | –34 83 | –40 19 | –27 93 | –29 96 | –58 76 |
| ZH 112 | –8 00 | –21 73 | –53 81 | –33 98 | –37 72 |
| Yueyou 200 | –4 76 | –1 09 | –18 56 | 1 98 | 5 96 |
| 89–15048 | –6 89 | –7 69 | –6 47 | –13 81 | 4 80 |
| Luhua 3 | 5 08 | –1 92 | 6 65 | –0 50 | –8 30 |
| 93–76 | –7 57 | –6 74 | –8 16 | –9 12 | –13 04 |
| Yueyou 92 | 4 04 | 6 72 | 9 82 | –12 50 | 1 85 |
| Taishan Sanlirou | –23 14 | –13 68 | –36 86 | –9 11 | –5 89 |
| Guiyou 28 | –15 05 | –12 16 | –11 77 | –12 48 | –1 16 |
| E Hua 4 | –7 04 | –11 11 | –33 20 | –1 85 | –5 95 |
| Gouliaozhong | –11 92 | –10 06 | –11 76 | –7 07 | –2 15 |
| Jiangtainzhong | 6 25 | 6 87 | 1 93 | 6 98 | 3 25 |
| 85–1526 | –15 38 | –16 67 | –4 55 | –33 64 | –29 41 |
| Zhonghua 2 | –10 46 | –25 08 | –0 13 | –20 63 | –25 68 |
| Zhonghua 4 | 2 04 | –2 27 | –20 18 | –5 01 | 5 76 |
| 91–074 | –5 68 | 2 73 | 3 65 | 0 88 | 5 40 |
| 87–77 | –16 82 | –26 94 | –6 95 | –40 56 | –31 53 |

In naturally diseased field plots, wilt was noticed in resistant plants at the late growth stages, when the plants were stressed by drought In these wilted plants, vascular bundles were discolored or destroyed due to infection and colonization by *B solanacearum* The latent infection in resistant cultivars was thought to reduce the absorption ability and drought tolerance in later growth stages It was noteworthy that some of the released resistant cultivars yielded well in fields free from *B solanacearum* but yielded relatively poorly in infested fields even though wilt incidence was not high Latent infection might be a reason, along with the poor soil fertility of the diseased fields Latent infection should be considered in further efforts to breed for BW resistance

## Techniques for Detecting Latent Infection

To enumerate culturable populations or total populations of bacteria in plant tissues, new methods were developed Counts of culturable bacteria were obtained after growing them on a suitable medium Additionally, serological methods were developed, providing means for rapid detection and identification of bacteria Research in this field has mainly been conducted to obtain race- or biovar-specific antibodies for use in the detection of *B solancearum* from soil or plant tissues, particularly in detecting latent infection

In order to investigate latent colonization by *B solanacearum* and its variation among different groundnut genotypes, a special project was started, supported by the Chinese Natural Science Foundation Experiments consisting of artificial inoculation and enzyme-linked immunosorbent assays (ELISA) were conducted to detect latent colonization in resistant groundnut genotypes Polycolonal antibodies were produced by immunizing a female rabbit with an isolate of *B soalancearum* collected from the natural disease nursery in Hong An (Shan et al 1997) Seeds of 30 resistant genotypes were artificially inoculated by soaking them in a bacterial suspension for 30 minutes and then sowing them in sandy soil At 50 to 60 days after sowing, inoculated plants of each genotype were sampled and cut into root, hypocotyl, base stem, and midstem portions These samples were then tested by ELISA Frequencies of latent colonization varied among different resistant genotypes A highly resistant line, 89-15048, which had survival percentage of 100% for several seasons, showed a latent colonization frequency less than 20%, while those in other resistant and susceptible genotypes ranged from 30% to 80% The resistant germplasm accessions Xiekangqing and Taishan Sanlirou, extensively used as breeding parents in China, possessed colonization frequencies of 35% and 46%, respectively, in this experiment It was noteworthy that not all inoculated seeds were infected or colonized, even in susceptible lines This phenomenon needs further investigation

The tap-root tissues of tomato plants were highly colonized by *B solanacearum* (Prior et al 1994), but in groundnut, tissues from the hypocotyl had highest colonization frequency in seed inoculation tests Infection of roots occurred in naturally diseased fields only after emergence The base stem of groundnut was colonized by the bacteria, but colonization of the midstem was detected in only a few samples These results were similar to the results obtained after inoculation of different plant parts of tomato However, in tomato, the proportion of invaded midstems without wilt symptoms varied according to the degree of resistance, i e , lines with more resistance had lower levels of stem invasion (Prior et al 1994)

## Implication of Latent Infection in Groundnut Breeding

Breeding for BW resistance and its application in managing the disease are likely to remain the major components of control strategies In general, groundnut breeding for resistance to BW has progressed very slowly, especially when attempts have been made to combine resistance with high yield potential and resistance or tolerance to other stresses This slow progress might be related to lack of awareness in earlier breeding programs about the role of latent infection Therefore, there is a need to modify germplasm evaluation methods and selection criteria in breeding for wilt resistance to overcome the effects of latent infection Disease incidence based on visual wilting symptoms will remain the basic criterion for evaluating resistance, particularly under field conditions Plants colonized at low levels could be selected within a population of symptomless plants to obtain more resistant cultivars A combination of wilt incidence and colonization values could optimize breeding programs and result in selection of wider resistance with greater adaptation to different environments (Prior et al 1994)

In groundnut, more than 90 accessions belonging to different botanical types have been identified as resistant to BW worldwide (Liao et al 1994) Resistance has also been found in wild species of *Arachis* (Yeh 1990) and interspecific hybrid derivatives (Zhang 1996, personal communication) Compared to several solanaceous crops such as potato, tomato, and tobacco, sources of resistance to BW in groundnut are much more diverse However, there is a need to screen these diverse sources of resistance to groundnut BW for latent infection by *B solanacearum* isolates In this direction, improved techniques for detecting latent colonization in groundnut tissues by ELISA appear to be useful in BW resistance breeding programs

In preliminary investigations, groundnut genotypes with high latent colonization frequencies were more sensitive to terminal drought stress, even when wilt incidence was low Hypocotyl tissue had the highest bacterial colonization frequency, and should be used for evaluating reaction of latent colonization in groundnut lines used in breeding for BW resistance

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# Pathological Aspects and Cultural and Biological Control of Groundnut Bacterial Wilt in Vietnam

**Nguyen Xuan Hong, Nguyen Thi Yen,** and **Vi Anh Tuan**[1]

## *Abstract*

*In recent years, extensive research at the Vietnam Agricultural Sciences Institute has explained much about pathogenicity of* Burkholderia solanacearum, *which causes bacterial wilt (BW) of groundnut, and how to control the disease Vietnamese isolates of* B solanacearum *vary greatly in their ability to cause wilt, and groundnut genotypes also vary in their reaction to the pathogen Rotation of groundnut with rice or sugarcane significantly reduces disease severity, but this type of rotation is not always feasible Some* Pseudomonas species *and* Bacillus subtilis *have shown potential as biological control agents in seed treatment More research on biological control is needed*

## Introduction

Groundnut bacterial wilt caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al is widespread in the major groundnut production areas of Vietnam The disease has become increasingly serious since it was first documented in 1991 (Mehan et al 1991) In recent years, major research efforts in Vietnam have been focused on systematic surveys to assess economic importance, distribution, and severity of the disease in major groundnut growing areas and identification of sources of resistance among local and introduced germplasm breeding lines, and varieties Several groundnut cultivars resistant to bacterial wilt have been identified and widely tested for their possible release in near future However, research on the pathogen and other management measures has made limited progress

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**Nguyen Xuan Hong, Nguyen Thi Yen** and **Vi Anh Tuan** 1998 Pathological aspects and cultural and biological control of groundnut bacterial wilt in Vietnam Pages 53–57 in Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

This paper reports the preliminary results of research on pathology and cultural and biological control of groundnut BW conducted recently at Vietnam Agricultural Science Institute (VASI), Hanoi

## Races, Biovars, and Pathogenecity of *Burkholderia solanacearum*

In Vietnam, isolates of *B solanacearum* from groundnut belong to biovars 3 and 4 Among 25 isolates collected from different ecological areas of northern and central Vietnam, 14 belonged to biovar 3 and 11 belonged to biovar 4 Isolates of both biovars were found in the same endemic region (Table 1)

**Table 1 Races and biovars of selected *Burkholderia solanacearum* isolates in Vietnam[1]**

| Isolate Source | Dulcitol | Mannitol | Sorbitol | Maltosa | Lactosa | Cellobiosa | Biovar | Race |
|---|---|---|---|---|---|---|---|---|
| HT–1 Ha Tinh | + | + | + | – | – | – | 4 | 1 |
| HT–2 Ha Tinh | + | + | + | – | – | – | 4 | 1 |
| HT–3 Ha Tinh | + | + | + | + | + | + | 3 | 1 |
| HL–3 Nghe An | + | + | + | + | + | + | 3 | 1 |
| HL–7 Nghe An | + | + | + | – | – | – | 4 | 1 |
| XH–3 Nghe An | + | + | + | – | – | – | 4 | 1 |
| XH–5 Nghe An | + | + | + | – | – | – | 4 | 1 |
| HTR–4 Thanh Hoa | + | + | + | – | – | – | 4 | 1 |
| QO–1 Ha Tay | + | + | + | – | – | – | 4 | 1 |
| VY 10 Bac Giang | + | + | + | – | – | – | 4 | 1 |
| HB–1 Bac Giang | + | + | + | + | + | + | 3 | 1 |
| AK–13 Ha Tay | + | + | + | – | – | – | 4 | 1 |
| BD–1 Binh Dinh | + | + | + | + | + | + | 3 | 1 |
| BD–2 Binh Dinh | + | + | + | + | + | + | 3 | 1 |
| BD–3 Binh Dinh | + | + | + | + | + | + | 3 | 1 |
| BD–4 Binh Dinh | + | + | + | + | + | + | 3 | 1 |

1 + = positive reaction – = negative reaction

Isolates of *B solanacearum* from groundnut are highly virulent on eggplant, potato, tomato, and sesame but less virulent on tobacco (Table 2) All isolates grow well on SPA medium at 30–35°C and release brown pigment in this medium Under greenhouse conditions at temperatures of 28–32°C, they cause a hypersensitive reaction on tobacco leaves 18 to 24 after inoculation by

injection Injection of leaf axils of young, susceptible groundnut plants with a pathogen suspension (108 CFU $mL^{-1}$/mL) 3 weeks after sowing caused wilt symptoms in 3 to 4 days

**Table 2 Reactions of six crop species to groundnut isolates of *Burkholderia solanacearum* in Vietnam**

| Crop species | Period of incubation (days) | Wilt intensity (%) |
|---|---|---|
| Eggplant (*Solanum melongena*) | 34 | 100 |
| Potato (*Solanum tuberosum*) | 45 | 80 |
| Tomato (*Lycopersicum esculentum*) | 45 | 90 |
| Sesame (*Sesamum indicum*) | 45 | 70 |
| Tobacco (*Nicotianna tabacum*) | 8 | 20 |
| Sweet potato (*Ipomea batatas*) | -1 | 0 |

1 = Data not available

Isolates of *B solanacearum* from different areas of Vietnam vary widely in their pathogenicity Isolates from hot spot locations, such as Ha Tinh and Xuan Mai are more virulent than those from other provinces (Table 3)

**Table 3 Pathogenicity of 6 isolates of *Burkholderia solanacearum* on selected groundnut genotypes following seed inoculations, Vietnam Agricultural Science Institute, 1998**

| | Wilt intensity (%)[1] | | | | | |
|---|---|---|---|---|---|---|
| Genotype | DA | XM | HT | BD | QN | TTH |
| ICGV 86143 | 100 | 100 | 100 | 48 | 43 | 30 |
| ICG 3704 | 76 | 92 | 96 | 72 | 28 | 33 |
| ICG 8666 | 18 | 17 | 22 | 21 | 7 | 8 |
| ICG 5273 | 14 | 33 | 35 | 22 | 10 | 12 |
| Gie Nho Quan | 4 | 3 | 5 | 0 | 0 | 0 |

1 DA = Dong Anh (Hanoi) BD = Binh Dinh XM = Xuan Mai (Ha Tay) QN = Quang Ninh HT = Ha Tinh TTH = Thua Thien (Hue)

Some groundnut varieties, such as ICG 8666 (Schwarz 21) and Gie Nho Quan showed stable resistance to all isolates of *B solanacearum* tested, whereas the

variety ICGV 96143 gave differential reactions to different isolates On the same groundnut variety and under the same conditions, one isolate of the pathogen can be more virulent and cause higher wilt intensity than others Research on the possible pathotypes of the *B solanacearum* in Vietnam is in progress

Preliminary research conducted at VASI has indicated that highly-virulent isolates of *B solanacearum* from groundnut are more tolerant and can survive under stress conditions longer than less-virulent isolates

Different methods of inoculation significantly affect wilt severity in groundnut genotypes In comparison to seed inoculation, stem inoculation caused much more severe wilt on the genotype ICG 5273 but less intense wilt on ICG 3704 These data showed that different mechanisms of resistance to groundnut BW exist among test genotypes (Table 4)

**Table 4 Reaction (% wilted plants) of selected groundnut cultivars to stem and seed inoculation with a highly virulent isolate (HT) of *Burkholderia solanacearum***

| Groundnut cultivar | Stem inoculation | Seed inoculation |
|---|---|---|
| ICG 8666 (Schwarz 21) | 20 0 | 22 4 |
| ICG 5273 (Matjan) | 60 0 | 26 5 |
| ICG 3704 | 64 3 | 100 0 |

## Cultural Control Measures

Rotation of groundnut with rice has proved most effective in reducing the inoculum in soil and wilt incidence (Hong et al 1997) However, in upland and riverbed areas, groundnut is grown as a dryland crop In these areas, groundnut cannot be rotated with rice Therefore, rotation of groundnut with alternate nonhost crops would be an important means to contain the disease Intercropping groundnut and nonhost crops also reduces wilt incidence

In northern Vietnam, surveys in different dryland cropping systems on upland (hilly) and riverbed areas indicated that rotating or intercropping groundnut with maize significantly reduced wilt incidence Sugarcane also was a good crop for rotation with groundnut In some areas that are heavily infested with the pathogen, rotation of groundnut with sugarcane for 3 years significantly reduced BW incidence

## Biological Control

In recent years, scientists at VASI have initiated experiments using antagonistic bacteria to control BW One strain of *Pseudomonas fluorescens* (No 4) and another strain of *Pseudomonas* sp are highly effective in suppressing BW These possible biocontrol agents were used for seed treatment (Table 5) More recently, preliminary research conducted in Hanoi Agricultural University (HAU) indicated that *P fluorescens* and *B subtilis* could significantly suppress *B solanacearum* Further research on biological control of BW will be conducted at VASI and HAU

**Table 5 Effect of seed treatment with antagonistic bacteria on wilt intensity of groundnut variety ICG 3704, in sick plots at Vietnam Agricultural Science Institute, 1997**

| Treatment | Wilt intensity (%) |
|---|---|
| No seed treatment | 86 2 |
| Seed treatment with *Pseudomonas fluorescens* | 15 0 |
| Seed treatment with *Pseudomonas* sp (*P aeruginosa* ?) | 15 8 |

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# Integrated Disease Management with Special Reference to Bacterial Wilt of Groundnut

**S Pande, C Johansen,** and **J Narayana Rao**[1]


## *Abstract*

*Groundnut is one of the most important oilseed crops in Asia, Africa, and the Americas Because of the tremendous diversity of its uses, and of environments in which groundnut is cultivated, it is infected by over 100 plant pathogens Bacterial wilt (BW), caused by* Burkholderia solanacearum, *is a major constraint to groundnut production in China, Indonesia, Vietnam, Malaysia, and Uganda Much progress has been made in understanding the epidemiology of races and biovars of* B solanacearum *that attack groundnut Effective greenhouse and field screening techniques have been developed, resistance sources identified, and resistant cultivars developed Integrated BW management in groundnut involves wilt-resistant cultivars, rotation with nonhost crops, and crop sanitation, but these practices are not yet sufficiently refined to be used by resource-poor farmers in South and Southeast Asia Therefore, more on-farm participatory research by farmers is needed to devise appropriate packages of these strategies for BW endemic areas*


## Introduction

The cultivated groundnut (*Arachis hypogaea* L) is a self-pollinating annual herbaceous legume It is a unique plant because it flowers above ground and produces pods containing one to five edible kernels below ground It is grown on over 18000000 ha throughout the world India, China, USA, Sudan, Senegal,

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Nigeria, and Indonesia are the seven largest producers of groundnut (Porter et al 1984) It is a valuable source of protein for human and animal nutrition, and provides a high-quality cooking oil The crop is important to the economies of farmers in several developing countries Vietnam is one country where the crop is gaining importance and has become an essential component of cropping systems and the farm economy

Crop management practices for groundnut vary from labor-intensive practices with few or no other inputs in some developing countries of Africa and Asia to almost total mechanization in the USA Accordingly, disease management practices vary widely There may be minimal disease management, some use of chemicals, or total reliance on host plant resistance (HPR) Madden (1987) defined integrated pest management as "a holistic, multidisciplinary management system that integrates control methods on the basis of ecological and economic principles for pests that coexist in an agroecosystem" This notion certainly describes disease management within groundnut production systems worldwide Cultural practices, host plant resistance, and fungicide usage are generally integrated into management plans designed to minimize initial levels of disease and obstruct disease progress In this paper, specific examples of integrated management of BW are discussed Emphasis has been placed on groundnut production systems in Asia and current strategies for disease management in the principal groundnut countries in South and Southeast Asia In addition, future needs are considered at the conclusion of the paper

## Economic Losses

Bacterial wilt caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al is a major constraint to groundnut production over large areas of China, Indonesia, and Vietnam (Table 1) Yield losses are usually in the range of 10–40%, but losses can reach 100% when highly susceptible cultivars are grown in heavily infested fields In China, it is estimated that the annual loss of groundnut pods from wilt exceeds 50000 t Severe losses are also reported in parts of Malaysia, Fiji, Papua New Guinea, and Uganda (Opio and Busolo-Bulafu 1990, Mehan et al 1994)

## Geographical Distribution

The global distribution of groundnut BW is given in Table 2 In China, the disease occurs in 16 groundnut-growing provinces, ranging from 19°N to 39°N It is most severe in the southern and central provinces, where it is estimated that over 200000 ha of groundnut fields are infested with the wilt pathogen (Mehan et al 1994)

**Table 1 Estimated annual groundnut yield losses caused by bacterial wilt in Asia[1]**

| Country | Yield loss (%) | Reference |
|---|---|---|
| China | 10–100 | Tan et al 1994 |
| Indonesia | 5–65 | Machmud and Rais 1994 |
| Malaysia | 0–20 | Lum and Hamidah 1994 |
| Philippines | ~ 30 | Natural 1994 |
| Thailand | >50 | Butranu et al 1994 |
| Vietnam | 5–80 | Hong et al 1994 |

1 Based on surveys of farmers fields and research station data

In Indonesia, the incidence and severity of BW vary depending on locations and seasons, the disease is most severe in West Java, South Sulawesi, and South Sumarta It is also economically important in central and eastern Java, Bali, and North Sulawesi In Vietnam, the disease can be severe in at least six major groundnut-growing provinces In Malaysia, BW is severe in Serdang and Kelantan areas and less severe in the Kedah area

Bacterial wilt has been reported in Thailand, Sri Lanka, Papua New Guinea, and India (Hayward 1990) and in several African countries (Ethiopia, Libya, Madagascar, Mauritius, Somalia, South Africa, Uganda, and Zimbabwe), as well as in Australia, Taiwan, and Japan, (Table 2, Mehan et al 1986), however, information is sparse, and the present status of the disease in many of these countries is not known The disease has been reported in the USA (Gitaitis and Hammons 1984), and it is an important constraint to groundnut production in central and northwestern regions of Uganda (Opio and Busolo-Bulafu 1990)

## Etiology and Biology

The taxonomy, characteristics, natural and inoculated host range, and geographical distribution of the causal agent of BW, *Burkholderia solanacearum*, are reviewed by Bradbury (1986) *Burkholderia solanacearum* is an aerobic, non-fluorescent, non-spore-forming, rod-shaped, gram-negative bacterium, approximately 0 5 W 1 5 5m (Kelman 1954, Hayward 1964) Key diagnostic features are its cultural characteristics on tetrazolium agar medium, where virulent isolates, that are mainly non-flagellate and non motile, form irregularly round, fluidal, creamy white colonies with light pink centers (Kelman 1954) Extracellular slime formation is a common attribute of all virulent isolates of *B*

**Table 2. Global distribution and importance of groundnut bacterial wilt[1]**

| Country | Province/Region | Importance |
|---|---|---|
| China[2] | Liaoning, Shandong, Hubei, Jiangsu, Anhui, Henan, Zhejiang, Hunan, Sichuan, Guizhou | Growing importance disease incidence <20% observed |
| | Hubei, Jiangxi, Fujiang, Guangxi, Guangdong, Hainan | Important, disease incidence >30% observed |
| Indonesia | Irian Jaya, Kalimantan | Important, disease incidence 10–30% observed |
| | Java, Sumatra, Sulawasi, Lombok, Bali | Very important, disease incidence >30% observed |
| Vietnam | Northern Vietnam Vinh Phu | Growing importance, disease incidence <10% observed |
| | Northern Vietnam Bac Thai, Ha Bac, Ha Tinh, Thanh Hao, Nghe An (Nghe Tinh) Southern Vietnam Long An, Tay Ninh | Important, disease incidence >30% observed |
| Malaysia | Kedah, Ipoh | Growing importance, disease incidence <10% observed |
| | Kelantan, Terengganu, Selangor | Important, disease incidence >30% observed |
| India | Andhra Pradesh, Meghalaya, Tamil Nadu, Karnataka | Not considered important, but disease observed or reported |

1 Groundnut bacterial wilt has also been reported in the Philippines Thailand Sri Lanka and Papua New Guinea (Hayward 1990) It has also been reported from Mauritius Madagascar Libya Somalia Ethiopia Zimbabwe South Africa Swaziland Taiwan Japan and Australia (Mehan et al 1986) and from Georgia and North Carolina in USA (Gitaitis and Hammons 1984) It is an economically important disease in Uganda (Opio and Busolo Bulafu 1990)

2 Based on information from provincial surveys

*solanacearum* Avirulent isolates usually bear one to four polar flagella and are highly motile The bacterium produces a brown diffusible pigment on some media, including tyrosine Acid production from carbohydrates varies greatly between biovars and strains Optimum temperature for growth varies between 25 and 35°C Further information is available in a recent review (Mehan et al 1994)

*Burkholderia solanacearum* has a very wide natural host range which includes many crop plants and weeds that are found in groundnut production systems (Bradbury 1986) Susceptible legumes include phaseolus bean, lupins, pea, soyabean, faba bean, cowpea, *Trifolium* spp and *Stylosanthes* spp The species is highly heterogenous (Bradbury 1986) Isolates are classified into five races based on host range (Buddenhagen and Kelman 1964, He et al 1983), and into five biovars based on biochemical characteristics (Hayward 1964, He et al 1983) Races and biovars are informal groupings at the intraspecific level Although the two systems of classification are largely independent, each system has contributed considerably to understanding the complex pathogenicity of *B solanacearum* (Mehan et al 1994) Race 1 isolates cause wilt in groundnuts and in many other leguminous and solanaceous plants Biovar 1 isolates cause wilt of groundnut in the USA, biovar 3 and, to a lesser extent, biovar 4 isolates cause wilt of groundnut in Asia and Africa (Hayward 1991) Biovars 2 and 5 have not been reported from groundnut

Serological techniques based on both polyclonal and monoclonal antibodies have been developed for detection and identification of *B solanacearum* (Alvarez et al 1993, Robinson 1993) Molecular techniques are also being developed to enhance detection of the bacterium and to better understand its considerable genetic diversity (Cook et al 1989, Seal 1994) *Burkholderia solanacearum*-specific DNA sequences have been identified and oligonucleotide primers constructed for those regions that helped in detecting single cells of the bacterium in hosts by the polymerase chain reaction (PCR) (Seal 1994) A rapid identification test was also developed for distinguishing biovars 3, 4, and 5 based on restrict on fragment length polymorphism DNA probes Further work is focusing on development of an immunocapture PCR test for *B solanacearum*

Bacterial wilt is most important on groundnut in the warm, humid, and subhumid tropics, especially in South and Southeast Asia (Hayward 1991, Mehan and Liao 1994) It has also caused sporadic damage in wetter areas of the semi arid tropics In general, isolates of *B solanacearum* from groundnut are reported to be more virulent on groundnut than are isolates from other hosts (Mehan and Liao 1994), however, strains from groundnut differ greatly in their pathogenicity on the host (Tan et al 1992) Further information is needed on the geographic distribution of races that infect groundnut before specific linkages with environmental conditions can be made to explain why wilt is severe in some zones but not in others

## Symptoms and Disease Diagnosis

Wilt symptoms can be observed 2 to 3 weeks after sowing Very young seedlings may wilt, but it is more common for wilting to commence at flowering (Mehan et

al 1994) *Burkholderia solanacearum* invades groundnut through wounds or through natural openings in roots The bacteria enter the water-conducting tissues, multiply, and block the vessels to cause wilting of the plant Infection of young plants results in rapid wilting of stems and foliage, but leaves remain green until final rotting occurs When older plants or cultivars that are not highly susceptible are infected, wilting proceeds more gradually, usually starting with the lateral branches Infected plants have discolored and rotted roots and sometimes rotted pods

The diagnostic characteristics of this disease are dark brown discoloration in the xylem and pith, and streaming of bacterial ooze from the cut ends of infected stems and roots when these are immersed in water

## Epidemiology

The disease is soilborne, and its long-term survival is favored by continuous cropping of groundnut and other host plants and the presence of weed hosts (Mehan and Liao 1994) The bacterium is mainly disseminated through water and infested soil Soil temperatures above 25°C together with high soil moisture favor the development of wilt (Wang et al 1983) Wilt peaks when the soil temperature is over 30°C for 10 days (Tan and Liao 1990) Under continuously wet conditions, wilt develops and spreads, but severe wilt symptoms may not appear for some time Infected plants wilt rapidly if they are subjected to a dry period A clear relationship between soil type and groundnut wilt has not been established In Indonesia, wilt is predominantly a problem of heavy clay or loam soils (Machmud 1986), whereas in China wilt is more common in sandy soils (He 1990) Wilt is less prevalent in soils with high organic matter, and preliminary information suggests that alkaline soils are wilt-suppressive (Yeh 1990) Machmud and Middleton (1990) found seed transmission of 5 to 8% in freshly harvested seed, but this was greatly reduced by drying Seed transmission is of obvious quarantine significance, and more research is needed to determine the extent of transmission (Mehan and Liao 1994)

## Disease Management

The best approach to management of wilt is to combine appropriate cultural control practices with the sowing of resistant varieties Crop sanitation measures such as burning crop residues, removing weeds, and cleaning implements after cultivation also help to reduce carry-over and spread of the disease Adjustment of sowing date to avoid periods of high temperature and soil moisture has had limited success (Kelman 1953) Rotation with immune or highly resistant crops, including

rice, maize, sugarcane, soyabean, and sorghum, is useful (Wang and Hou 1982, He 1990) Chemical control is not economically feasible Varieties with high levels of resistance and good agronomic qualities have been bred in China and Indonesia (Liao et al 1990, Yeh 1990, Machmud 1993) and Mehan et al (1994) list 54 resistant genotypes Additionally, several wild *Arachis* spp are highly resistant (Yeh 1990) Considerable progress has been made in the management of the BW of groundnut

## Agronomic and Cultural Management

### Crop rotation

As groundnut BW is mainly soilborne, rotation of groundnut with crops that are immune or highly resistant to *B solanacearum* and with nonhost crops such as rice, maize, soybean, and sugarcane, are effective measures Rotation of groundnut with rice or sugarcane for 2 to 3 years can greatly reduce wilt incidence and severity (Wang and Hou 1982, He 1990, Tan and Liao 1990, Machmud 1993) Groundnut-rice rotation systems are successfully used in several regions of China (He 1990) Rotation with wheat, sorghum, and cotton is also effective in reducing wilt incidence In rainfed uplands, rotation of groundnut with maize and sorghum or intercropping groundnut with maize are useful ways to contain the disease

Although crop rotations for shorter periods with immune crops have proved effective in containing the disease, leaving a gap of at least 3 to 4 years between groundnut crops is more effective, especially in soils that are heavily infested with the pathogen Long gaps are not practical where groundnut is important major crop (e g , in Vietnam) Little is known about how various cropping systems affect soil microorganisms in general or the survival of the wilt pathogen in particular

Continuous cultivation of highly resistant groundnut cultivars could either reduce or maintain the *B solanacearum* population, or it could select for more virulent strains

### Flooding and fallowing

Flooding groundnut fields for 15 to 30 days before sowing also reduces wilt incidence (Li et al 1981, He 1990) In areas where groundnut is grown under irrigation in the dry season, it should be possible to control or greatly reduce disease levels by dry season fallowing, because the bacterium is highly susceptible to desiccation The effects of fallowing can be enhanced by cultivation to improve soil drying and reduce weed growth Improved soil drainage helps in reducing wilt incidence and severity

### Sowing date

Depending upon the length of the growing season and cultivars grown, the sowing date can be adjusted to avoid periods of high temperature or ample soil moisture conditions that favour bacterial infection and disease development A few attempts have been made to minimize crop losses by altering sowing dates, but these have had limited success (Kelman 1953) In the Hubei Province of China, early sowing in mid-April results in much lower incidence and severity of BW than does sowing in June (Mehan et al 1994)

### Crop sanitation

Crop sanitation (e g , burning crop residues, removing weeds, and cleaning farm tools after operations in infested fields) should help reduce disease levels Experimental evidence on the effects of crop sanitation is not available

### Soil amendments by fertilizers and manures

Soil application of urea, mineral ash, and organic manure can be useful in reducing wilt incidence (Chang and Hsu 1988, Yeh 1990) These fertilizers are likely to stimulate soil microbial activity against the wilt pathogen, and some of their components may enhance host resistance More research is required to elucidate the mechanisms by which soil amendments control wilt, specifically from other groundnut-producing countries of South and Southeast Asia

### Chemical soil treatment

Soil treatment with sulfur, lime, and other chemicals, including fungicides and antibiotics, has not proved useful in controlling groundnut BW Applying 300 kg $ha^{-1}$ of chloropicrin 10 days before sowing can effectively control the disease (Wang and Hou 1982), but this treatment is expensive

## Seed Transmission and Plant Quarantine

The bacterium, *B solanacearum* is transmitted through infected groundnut seed (Machmud and Middleton 1990) Seed transmission is reported to be at the rate of 58%, particularly in freshly harvested seed Thus, infected seed is likely to provide a primary source of inoculum, particularly for disease-free areas There is rapid loss of viability of the bacterium as groundnut seeds dry out to a moisture content below 9% (Zhang et al 1993) Seed transmission is of obvious quarantine significance, and more research is needed to determine the degree to which *B solanacearum* can be seed-transmitted in groundnut

## Management Using Biological Agents

Biological control of *B solanacearum* has been extensively investigated and found promising under laboratory and greenhouse conditions However, most biological control agents used to control BW, including several antagonistic *Pseudomonas* (*P cepacia*, *P fluroresoens*, *P glumae*, and *Bacillus* spp ), have not proved highly effective against *B solanacearum* in the field (Trigalet et al 1994) Of considerable interest is the more recent work involving some genetically engineered hrp avirulent mutants of *B solanacearum* (Trigalet and Trigalet-Demery 1990) It would be useful to develop such biocontrol agents into effective commercial products for seed treatment However, it may take a long time before approaches to biological control reach the stage of commercial application

## Host-Plant Resistance

Sowing disease resistant cultivars is the cheapest and most effective method by which to combat BW of groundnut Much progress has been made in developing screening techniques, identifying stable sources of resistance and breeding resistant cultivars (Mehan et al 1994)

Worldwide, some 90 germplasm accessions and improved cultivars of groundnut have been reported resistant to BW Most of these resistant genotypes are from China and Indonesia (Mehan et al 1994) Resistant lines have also originated in or been collected from Argentina, Brazil, India, Israel, Peru, Uganda, and USA (Machmud 1993) Most of the screening efforts have been made in China and Indonesia, where more than 6000 germplasm accessions have been tested

Among the available resistant genotypes, 31 belong to the spanish type (var *vulgaris*) including more than 20 improved cultivars bred in China and Indonesia A few valencia (var *fastigiata*) lines resistant to foliar diseases have also been found resistant to BW in China and Indonesia (Yeh 1990, Mehan and Liao 1994) Resistance to BW in these genotypes (e g , NCAc 17127 and PI 393641) might be less stable than that in the resistant spanish lines Recently, these valencia lines showed varying levels of resistance at different locations in China Only three resistant genotypes belong to the virginia type (var *hypogaea*), and they are important in breeding programs (Duan et al 1993)

In recent years, 50 "Chinese dragon" type genotypes have been identified as resistant to BW (Duan et al 1993) All of the resistant dragon lines were collected from heavily infested locations in southern China No resistant material of this type has been obtained from the northern provinces The dragon lines are landraces that have long been cultivated in China (Sun et al 1963),

arachaeological evidence indicates that they had been cultivated for at least 500 years before the virginia type groundnuts were introduced toward the end of the 19th Century As resistant genotypes are far more common in the dragon type than in the other three botanical types, there may have been selection for resistance down through the centuries in which BW has been a serious problem in southern China The characterization of some 200 dragon genotypes indicates considerable variation in many characters Genetic diversity for resistance among dragon lines merits further investigation

Screening on a limited scale has led to the identification of some resistant accessions of wild *Arachis* species Resistance has also been found in some interspecific hybrid derivatives that also have resistance to foliar fungal diseases (Mehan and Liao 1994)

Efforts to breed groundnuts with resistance to BW have been concentrated mainly in Indonesia and China In Indonesia, Schwarz 21 and its derivatives (Gajah and Kidang) have been extensively used as resistance donors in breeding programs since the early 1950s In China, two wilt resistant lines with good general combining ability, Xiekangqing and Taishan Sanlirou, have been used as resistance donors (Liao et al 1990) As most resistance donors used in breeding programs belong to ssp *fastigiata*, the genetic base of wilt resistance is narrow

In Indonesia, two wilt-resistant cultivars, Schwarz 21, released in 1925, and Gajah released in 1952, are widely grown in Java where BW previously caused heavy yield losses in susceptible cultivars In the 1950s, three wilt resistant cultivars (Kidang, Banteng, and Macan) were developed from crosses between Schwarz 21 and some introductions from Japan, Israel, and USA Wilt resistant cultivars such as Anoa, Rusa, Pelanduk, Tupai, and Tapir, released in 1982/83, were developed from crosses between Gajah or Kidang and introductions from Honduras and USA Some of these cultivars (e g , Pelanduk and Tupai) are now grown by farmers in Indonesia (Machmud 1993)

In China, breeding efforts in the 1960s led to the release of two moderately resistant cultivars, Yue You 589 and Sueitien Later, an exotic resistance source was crossed with a local cultivar, and two important resistant and high yielding cultivars, Yue You 92 and Yue You 256, were bred in Guangdong (Liao et al 1990) In the last decade, several high-yielding, wilt-resistant cultivars were released to farmers These include Jinyou 3121, Guiyou 28, Lutlua 3, El hua 5, and Zhong Hua 2 (Liao et al 1990) El Hua 5 is currently grown on some 18000 ha in most of the wilt-affected areas in central China Zhong Hua 2, an early maturing, wilt-resistant cultivar with wide adaptation, is becoming popular with farmers in

central China A new wilt-resistant breeding line, Yue You 200, with high yield potential, is now at the demonstration stage and will be released soon in Guangdong Province, China (Zhang et al 1994) Some recently bred cultivars that have multiple resistances to wilt and rust have also been developed in China

More than half of the global groundnut germplasm collection has still to be screened for resistance to BW, and new sources of resistance could still be identified Special emphasis should be placed on combining wilt resistance with high yield potential and resistance to foliar fungal diseases

## Integrated Disease Management

In general an integrated approach to BW control should involve wilt-resistant groundnut cultivars, rotation with non-host crops, and crop sanitation Rotation of groundnut with rice in lowland areas or in upland areas equipped with an irrigation system, or rotation with a non-host coupled with the use of resistant groundnut cultivars and the use of herbicides in irrigated areas, are important and effective means of reducing disease incidence in infested soils It is not possible to adopt crop rotations in upland and marginal lands where groundnuts are grown under rainfed conditions In such areas, use of resistant cultivars and seed from disease-free areas is advocated However, more on farm research is needed to devise appropriate packages of these strategies for BW-endemic areas

## Future Research Needs

Although BW has been extensively studied worldwide for several decades, efficient and complete control measures are not yet available to resource-poor farmers in Asia or elsewhere Substantial progress has been made in the development and release of resistant cultivars, but resistant varieties alone have not solved the problem completely It is expected that highly resistant and high-yielding cultivars endowed with other desirable characteristics will be made available to farmers in the near future, specifically in regions where the disease is a major constraint to crop production High priority should be given to participatory on farm validation of the components of disease management and their integration Extension programs that include supplying seed of newly developed resistant cultivars to farmers should be mandatory for any research and development program

To enhance and diversify genetic resistance to BW, it is imperative to assemble various germplasm accessions and varieties with putative resistance in countries where the disease is endemic Sources of resistance from different botanical accessions will be useful in increasing the diversity and stability of resistance

There is a need to establish an International Groundnut Bacterial Wilt Nursery (IGBWN) to determine the stability of wilt resistance through multilocational testing Concerted efforts are also needed to foster the exchange of wilt resistant germplasm and breeding lines Priority should also be given to testing resistant lines, including possible differentials, for their reaction to various groundnut adapted strains of pathotypes of the wilt pathogen from different regions of the world

Crop rotations with non-host crops are often not acceptable to farmers, especially if such rotations are prolonged, and this highlights the need for complementary control measures Therefore more research is needed to devise appropriate integrated packages involving HPR, crop rotation, sanitation, and agronomical soil amendments specifically for local disease-endemic areas It is likely that control methods involving biological agents will need local adaptations in the future This prospect offers a new research avenue

There is a need to increase and strengthen the collaboration in research on bacterial wilt through the Groundnut Bacterial Wilt Working Group It is expected that this collaboration will provide solutions to location- and region-specific problems and will contribute to improved disease control

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# Germplasm Screening and Breeding for Resistance to Bacterial Wilt in China

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## *Abstract*

*Bacterial wilt (BW) caused by Burkholderia solanacearum is an important constraint on groundnut production on >300000 ha in China This is about 8% of the total groundnut area sown each year Wilt incidence and yield loss are estimated to be 10–30% in susceptible groundnut cultivars It is well known that genetic resistance is the most important component of integrated management of groundnut BW Therefore, between 1974 and 1978, about 1700 germplasm accessions were screened at hot spot locations in China Two lines, Xiekangqing and Taishan Sanlirou, were identified as highly resistant to BW Since then, these lines have been extensively used in breeding for BW resistance In addition to these lines, during 1986–93, over 3000 germplasm accessions were also screened and over 50 dragon type lines and 3 virginia lines were identified as resistant Furthermore, of the 1100 local groundnut germplasm screened during 1994–97, 13 lines were identified as resistant These resistant genotypes (~ 70 including checks) were tested across locations in China, and resistance in most of these genotypes was stable Seventy-eight wild* Arachis *accessions were also evaluated under natural infection conditions, and 24 showed good levels of resistance to BW Twelve resistant genotypes were also identified from the lines received from ICRISAT during 1994–97 Fifteen resistant lines were transferred from China to ICRISAT in 1995 Several new breeding lines, including Yue You 200, Yue You 79, and 93-81, have been developed A few resistant lines were also selected from an interspecific hybrid (Baisha 1016×W* Arachis chacoense*) at Henan Academy of Agricultural Sciences Efforts to enhance resistance by genetic transformation are in progress Further research efforts will be made for enhancing yield potential, resistance to other wilt pathogens, and drought tolerance of BW-resistant groundnut cultivars*

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## Introduction

Groundnut is an important oil and cash crop in China In the mid 1990s, groundnut was annually harvested from an area of about 3 8 million ha, with production of around 9 to 10 million tons Bacterial wilt caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al has been an important constraint to groundnut production in China for decades In recent years, the disease has spread to more than 300000 ha, nearly 8% of the annually sown area of groundnut in the country Incidence is about 10–30% in susceptible cultivars and less than 10% in resistant cultivars currently grown in the major groundnut-growing regions of China In the past three decades, extensive research has been done to identify the components of integrated control approaches for bacterial wilt Despite this effort, no effective chemical means to minimize BW are yet available Long-term rotation is the only effective cultural control practice, but is seldom feasible because of the limitations of arable land However, use of BW-resistant groundnut cultivars is the most effective and practical method for control in farmers' fields Hence, groundnut germplasm screening and resistance enhancement have been emphasized in recent years In this paper, progress made in genetic improvement of host resistance to BW in China since 1994 is reviewed

## Germplasm Screening

### Screening methods

In large-scale germplasm screening for resistance to BW, groundnut lines are usually evaluated in single-row plots without replication, under natural disease pressure at hot-spot locations Genotypes that show high survival percentage are selected and further tested under natural disease pressure in three to four replications with a susceptible control sown at regular intervals In China, disease nurseries have been established at such hot-spot locations as Hong An, Hubei Province by the Oil Crops Research Institute (OCRI), Chinese Academy of Agricultural Sciences (CAAS), and in Guangzhou, Guangdong Province In the Hong An nursery, groundnut has been continuously grown for over 30 years, and wilt incidence in susceptible groundnut genotypes is consistently over 90%, indicating a desirable disease pressure for meaningful evaluation of groundnut genotypes

Artificial inoculation is routinely followed to enhance infection and confirm resistance Infection pressure in artificial inoculation is kept similar to that available under natural conditions Soaking seeds in bacterial suspension for 30 minutes before sowing has been the most effective method of inoculation However, the seed

inoculation method also reduced seed emergence When sample size was small, stem and root inoculation techniques were also used to evaluate and confirm BW resistance Multilocational tests were generally needed to verify the stability of resistance Field resistance can also be evaluated at hot-spot locations in Sichuan, Guangxi, Jiangxi, and other provinces (Tan and Liao 1990)

### Germplasm screening before 1994

During 1974–77, about 1700 groundnut germplasm accessions collected in China were systematically tested in Hong An, Hubei Province This was the first large-scale screening for resistance Two groundnut accessions – Xiekangqing and Taishan Sanlirou – were identified as resistant and have been extensively used in breeding in China During 1986–93, over 3000 groundnut germplasm accessions were screened, and over 50 dragon type lines and 3 virginia lines were identified as resistant A total of 70 lines were resistant, and their resistance was generally stable across locations and seasons in China (Duan et al 1993)

### Germplasm screening, 1994 to date

About 1100 accessions of groundnut germplasm were screened for their reaction to bacterial wilt between 1994 and 1997 Most of these accessions were breeding lines developed by various research institutes in China From these screenings, 13 resistant lines were identified Seventy-eight wild accessions were evaluated under natural disease pressure, and 24 showed reasonable levels of resistance However, the resistance in wild *Arachis* spp needs further evaluation and confirmation

Twelve resistant genotypes were repeatedly identified among germplasm lines introduced from ICRISAT during 1994–97 The resistance of this material was desirable (Table 1), but most of them were small-seeded with lower yield potential than the local resistant germplasm

In 1995, 15 groundnut accessions with desirable resistance to bacterial wilt developed or identified in China (Table 2) were sent to the ICRISAT genebank These lines are available to interested researchers

## Breeding for Resistance

### Breeding methodology

Conventional hybridization has been applied effectively in breeding for resistance to BW groundnut Resistance to BW in groundnut is simply inherited and could be easily transferred to different cultivars Selection for resistance has been conducted in the $F_2$ through $F_6$ generations An effective disease-screening nursery

**Table 1 Reaction of groundnut germplasm lines from ICRISAT to bacterial wilt in China, 1994–96**

| Line | Resistance (%) 1994 | 1995 | 1996 | Mean | Reaction |
|---|---|---|---|---|---|
| ICG 1608 | 80 0 | 93 3 | 88 4 | 87 4 | R |
| ICG 1610 | 76 9 | 90 9 | 81 7 | 83 2 | R |
| ICG 3121 | 100 0 | 70 0 | 97 0 | 89 0 | R |
| ICG 5313 | 95 0 | 100 0 | 97 0 | 97 3 | HR |
| ICG 6417 | 92 9 | 91 7 | 92 0 | 92 2 | HR |
| ICG 7919 | 85 0 | 100 0 | 89 7 | 91 6 | HR |
| ICG 8684 | 100 0 | 84 6 | 87 9 | 90 8 | HR |
| ICG 8701 | 89 7 | 92 9 | 91 4 | 91 2 | HR |
| ICG 8704 | 76 9 | 93 8 | 85 5 | 85 4 | R |
| ICG 8706 | 96 6 | 86 7 | 89 5 | 90 9 | HR |
| ICG 8707 | 71 4 | 88 8 | 80 4 | 80 2 | R |
| ICG 8719 | 88 5 | 93 8 | 91 8 | 91 4 | HR |

1 R = resistant HR = highly resistant

**Table 2 Reaction of local groundnut lines to bacterial wilt in China, 1995**

| Lines | Reaction | Sources |
|---|---|---|
| Xiekangqing | R | Local variety |
| Taishan Zhengzhu | R | Local variety |
| E Hua 5 | R | Improved cultivar |
| Zhong Hua 2 | R | Improved cultivar |
| Yue You 92 | R | Improved cultivar |
| Gui You 28 | R | Improved cultivar |
| Yue You 200 | R | Breeding line |
| Jiangtianzhong | MR | Local variety |
| Gouliaozhong | MR | Local variety |
| Luhua 3 | MR | Improved cultivar |
| Kangqing 2 | R | Breeding line |
| 89–15048 | R | Breeding line |
| 91–054(ZH 112) | R | Breeding line |
| 91–076 | R | Breeding line |
| 91–081(ZH 212) | R | Breeding line |
| Zao 18 | HS | Breeding line |

1 R = resistant MR = Moderately resistant HS = highly susceptible

is available for resistance selection, and artificial inoculation is used as and when needed Survival percentage is currently used as the resistance selection criterion Multilocational tests are conducted to determine stability, yield, and adaptation of selected lines (Chen and Hong 1995)

### Resistance donors

In China, several resistant germplasm accessions have been used in breeding Xiekangqing and Taishan Sanlirou have been most extensively used as resistance donors Improved resistant cultivars, including E Hua 5, Lu Hua 3, and Yue You 92, were bred from Xiekangqing, while Zhong Hua 2 and Zhong Hua 4 were bred from Taishan Sanlirou Induhuapi has been effectively used as a resistant parent in Guangdong Province

It is interesting to note that only germplasm lines belonging to ssp *fastigiata* in the cultivated *Arachis* have been successfully used as resistance donors in breeding programs Apparently, early maturity and tolerance to acid soil and poor soil fertility in spanish or valencia types are most desirable in different agro-ecological conditions and wilt-prone areas in China Generally, the resistant genotypes identified from spp *hypogaea* are thought to be promising in increasing yield levels of BW resistant lines, however, some of their characters may inhibit integration of wilt resistance with desirable agronomic traits necessary for adaptation in the disease-prone areas Large pods are usually important for high yield potential, but in most cases this trait is associated with a sensitive reaction to calcium deficit in acid soils, a condition predominating in most regions where *B solanacearum* causes serious wilt in groundnut Therefore, direct use of large-podded genotypes in breeding seems difficult Efforts are currently in progress to use some of the resistant high-yielding genotypes of ssp *hypogaea* in BW resistance breeding programs in China and elsewhere (Liao and Xu 1997, Liao et al 1990)

### Status of resistant cultivars and breeding lines

Since 1994, several improved groundnut cultivars or promising breeding lines have been developed or released in China Yue You 200, a breeding line developed at Guangdong Academy of Agricultural Sciences (GAAS), promising both for resistance to BW and high yield in central China, was released as Tianfu 11 in Sichuan Province Yue You 202–35 also was developed at GAAS and released in Guangdong Province in 1997 (Liang et al 1997) It is resistant to both bacterial wilt and rust (*Puccinia arachidis*), with a relative high oil content (54 3%) A promising breeding line, Yue You 79, with desirable resistance to both BW and rust, has been developed at GAAS

At OCRI, two resistant lines have been developed Line 93-81 is highly resistant, with tolerance to acid soil Another resistant line, 93-76, with desirable pod shape and uniformity, as well as drought tolerance, has been identified (Cui 1998, personal communication) Many advanced breeding lines are in the pipeline under further selection

Wan Hua 2 was released in Anhui Province in 1996 It was described as resistant to BW In Fujian Province, Quan Hua 10, with moderate resistance to BW, was released in 1995 (Zhou and Wei 1996)

### Resistant lines developed from interspecific hybridization

An advanced breeding line, 9102, was developed at Henan Academy of Agricultural Sciences (HAAS) located in Zhengzhou (Zhang Xingyou 1997, personal communication) It was developed from a cross between Baisha 1016, a spanish-type cultivar, and *A chacoense* The groundnut line 9102 is resistant to BW in Guangxi and Hubei Provinces It matures early, has high seed oil content (56%), and produces high yields

### Biotechnology for resistance enhancement

Even though resistance to BW in groundnut has not been found negatively correlated with agronomic traits, integrating BW resistance with high yield potential has progressed relatively slowly In order to enhance yield potential, stability of resistance to BW, an anti-bacterial gene and transformation system like those employed successfully in potato for resistance to *B solanacearum* are being used in transforming groundnut at OCRI

## Further Research Needs

Genetic enhancement for resistance will be the most important research area in further addressing the BW problem in China The following are possible subjects for future research efforts

- Screening more desirable resistant resources
- Investigating genetic diversity of wilt resistance in groundnut
- Enhancing yield potential of wilt resistant groundnut cultivars
- Enhancing resistance to other wilt pathogens and drought tolerance of BW-resistant cultivars
- Enhancing quality of BW-resistant cultivars

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# Germplasm Evaluation and Breeding for Groundnut Bacterial Wilt Resistance in Vietnam

**Nguyen Van Lieu , Tran Dinh Long , and Nguyen Xuan Hong**[1]


## Abstract

*Bacterial wilt caused by* Burkholderia solanacearum *is one of the most serious diseases of groundnut in Southeast Asia, particularly in Vietnam Before 1991, evaluation of groundnut germplasm for bacterial wilt (BW) resistance had never been done in Vietnam The first systematic screening for BW resistance was done in 1992–95, when a total of 715 groundnut accessions were screened in sick plots at hot-spot locations in northern Vietnam Only 4 1% of the screened lines were resistant to BW Of those, only 0 6% were highly resistant Most available accessions were susceptible to the disease Groundnut line Gie Nho Quan possessed the highest resistance to BW, with a survival rating of 97% This variety also had good agronomic characters, and therefore is a good candidate to be cultivated in infested areas, and to be used as a BW resistance gene donor in breeding for BW resistance in Vietnam Several other genotypes, such as Schwarz 21 (KPS 13), Matjan (KPS 18), and ICGV 87157 are promising resistant cultivars The pod yield of these cultivars is substantially higher than that of the local control, Do Bac Giang Breeding for BW resistance has been initiated and some progress made*


## Introduction

Bacterial wilt of groundnut caused by *Burkholderia solanacearum* (E F Smith) Yabuuchi et al is the only economically important bacterial disease of groundnuts

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**Nguyen Van Lieu Tran Dinh Long and Nguyen Xuang Hong** 1998 Germplasm evaluation and breeding for groundnut bacterial wilt resistance in Vietnam Pages 82–87 in Groundnut bacterial wilt proceedings of the Fourth Working Group Meeting 11–13 May 1998 Vietnam Agricultural Science Institute Van Dien Thanh Tri Hanoi Vietnam (Pande S Liao Boshou Nguyen Xuan Hong Johansen C and Gowda C L L eds ) Patancheru 502 324 Andhra Pradesh India International Crops Research Institute for the Semi Arid Tropics

(Hong and Mehan 1993) It develops favorably in warm, humid regions of the world where groundnuts are grown Southeast Asia is the most favorable region for BW China and Indonesia are two other countries with large groundnut growing areas infected by BW These two countries have paid much attention to BW research The most remarkable achievement in BW research was the development of groundnut variety Schwarz 21, which was bred in Indonesia in 1925 (Mehan et al 1986) Soon after this discovery, several highly resistant varieties were bred in Indonesia, China, and other countries and have contributed to reducing yield losses in groundnut Until 1994, it was estimated that more than 6000 groundnut accessions had been evaluated for BW resistance in China and Indonesia alone, and about 140 accessions were resistant to BW From those genetic resources, many highly BW resistant varieties with good agronomic characteristics have been released for direct production or to be used in BW resistance breeding programes

Bacterial wilt of groundnut has been found in Vietnam since 1962 (Ung Dinh 1962, Dang 1968) Scientists who carried out preliminary experiments to find BW resistant genotypes among local varieties did not have any success From 1970 to 1990, research on BW was negligible A comprehensive research program, including germplasm evaluation and breeding for BW resistance, has been conducted since 1991 (Hong and Mehan 1993)

## Materials and Methods

### Germplasm evaluation

Sick plots were sown at hot-spot locations to identify BW resistance In these sick plots, densities of bacteria exceeded $10^6$ CFU 100 $g^{-1}$ soil Sowings for resistance screening were carried out twice a year In the spring season, sowings usually started 10 March, and in the autumn season, sowing time was around 20 July Seeds of tested entries were soaked in a bacterial suspension ($6 \times 10^8$ CFU 1 $mL^{-1}$) for 20 minutes before sowing

Experiments were laid out in an augmented design without replication A susceptible control (ICG 3704) and one local control (Do Bac Giang or Sen Nghe An) were sown after every 10 test entries Each test entry was sown in a 1 $m^2$ plot area with 60 to 80 plants $m^{-2}$ These experiments were repeated for at least three seasons From 1992 through 1995, a total of 715 groundnut accessions (including 596 exotic lines) were screened for BW resistance in sick plots Seventy days after sowing, the percentage of wilted plants was counted to assess BW incidence Test lines were categorized as, 1 highly resistant (≤10% wilted plants), 2 resistant

(11–30%), 3 moderately susceptible (31–50%), 4 susceptible (51–90%), and 5 highly susceptible (>91%)

### Breeding for bacterial wilt resistance

Bacterial wilt resistant lines were evaluated for agronomic characteristics Germplasm lines or bred varieties with such desirable agronomic characteristics as high pod yield or good potential to resist drought and/or some main foliar diseases, were further selected evaluation for stability of yield in multilocational trials Remaining lines and varieties were kept in the gene bank for future use In multilocational trials, these lines were sown in randomized complete block designs and scored for pod yield and seed characteristics

## Results and Discussion

### Germplasm evaluation

Out of 715 lines, only a few (~ 4%) were found to be resistant to BW in the 1992–95 germplasm evaluation (Table 1) Most of the resistant accessions belonged to the spanish type, few were virginia or valencia types

Frequency of occurrence of resistance in short-duration lines was lower than in medium- and long duration lines

**Table 1 Germplasm evaluation for bacterial wilt resistance, 1992–95, Vietnam**

| Disease category | Number | Percentage | Most promising lines in category |
|---|---|---|---|
| 1 Highly resistant | 4 | 0 6 | Gie Nho Quan, NcAc 17127, PI 393531 |
| 2 Resistant | 25 | 3 5 | Schwarz 21, ICGV 87206 Matjan |
| 3 Moderately susceptible | 69 | 9 6 | SeKontum, do Lao Cai, NcAc 17090 |
| 4 Susceptible | 522 | 73 0 | Sen Nghe An, Do Bac Giang, Tram Xuyen |
| 5 Highly susceptible | 95 | 13 3 | No2, ICG 3704, UPL–PN2 |
| Total | 715 | 100 0 | |

**Table 2 Bacterial wilt resistant performance of Gie Nho Quan groundnut variety under artificial inoculation[1] with different isolates of *Burkholderia solanacearum***

| | Plants surviving 21 days after treatment (%) | | | |
|---|---|---|---|---|
| Genotype | QO[2] | $HT_1$ | $HTr_4$ | HB |
| Gie Nho Quan | 100 0 | 92 0 | 100 0 | 96 0 |
| Sen Nghe An | 70 0 | 31 2 | 69 2 | 50 0 |
| ICG 3704 | 46 7 | 0 0 | 42 3 | 20 0 |

1 Inoculation method seed treatment + root cutting
2 Q0 $HT_1$ $Htr_4$ HB Isolates of *B solanacearum*

**Table 3 Bacterial wilt resistant performance of Gie Nho Quan groundnut variety in natural sick plots at four locations in Vietnam during spring season, 1996–97**

| | Plants surviving at 70 days after sowing (%) | | | |
|---|---|---|---|---|
| Genotype | Thach Binh (Ninh Binh) | Thanh Liem (Ha Ham) | Agr Uni N°3 (Thai Nguyen) | Xuan Mai (Ha Tay) |
| Gie Nho Quan | 98 8 | 99 0 | 99 2 | 98 9 |
| Schwarz 21 (KPS 13) | 84 0 | 85 2 | – | 87 5 |
| Matjan (KPS 18) | 80 0 | 85 0 | – | 88 4 |
| Sen Nghe An | 69 2 | 56 5 | 65 0 | 31 5 |
| V 79 | 57 7 | – | 61 8 | 46 2 |
| ICG 3704 | 42 3 | 23 5 | 34 5 | 0 8 |

The significant finding of our resistance screening was the identification of a highly resistant landrace, Gie Nho Quan This landrace, when inoculated with different isolates of *B solanacearum*, was highly resistant in comparison to other genotypes (Table 2) It was evaluated along with other available BW-resistant sources in sick plots at hot-spot locations (Table 3) The data presented in Tables 2 and 3 clearly indicate that Gie Nho Quan was the most resistant groundnut genotype Its resistance was more stable over locations, than that of other varieties Its resistance level was higher than those of the two traditional sources of BW

resistance, Schwarz 21 and Matjan Gie Nho Quan is considered to be the best source of resistance to BW of groundnut in Vietnam The characteristic features of Gie Nho Quan are spanish type, dark-green leaf color, 40–42 cm stem height, small seeds and pods, pink-white testa color, high shelling percentage (77 8%), short duration, moderate foliar disease resistance, and drought tolerance Pod yield of this variety was not high, but could be acceptable when grown in BW infested areas

### Breeding for bacterial wilt resistance

The selected resistant groundnut lines were evaluated in multilocational trials for pod yield and other important characters, such as 100-pod mass, 100-seed mass, shelling percentage, testa colour, and duration After three years of multilocational testing, we identified four good varieties (Table 4) All four were of medium duration with exportable testa colour All of them except Gie Nho Quan had higher yields than the local control These varieties will be further tested in farmers' fields for BW resistance

Several crosses have been made since 1996, and selection for resistant genotypes is in progress

**Table 4 Main characteristics of some promising resistant groundnut varieties with good yield potential in Vietnam**

| Variety | Duration (days) | Mature pods plant$^{1}$ | 100seed mass (g) | Shelling percentage | Pod yield mean(kg ha$^{1}$) |
|---|---|---|---|---|---|
| Schwarz 21 (KPS 13) | 126 | 9 1 | 54 8 | 70 5 | 241 |
| Matjan | 126 | 9 3 | 58 2 | 72 1 | 253 |
| ICGV 87157 | 130 | 10 2 | 52 3 | 70 2 | 278 |
| Gie Nho Quan | 120 | 14 7 | 40 1 | 77 8 | 206 |
| Do Bac Giang | 123 | 8 6 | 51 2 | 73 2 | 228 |
| Local control | | | | | |
| CV (%) | | 6 7 | 4 3 | 1 8 | 46 |
| LSD 0 05 | | 1 2 | 3 9 | 2 5 | 20 |

## Conclusions

- Most of evaluated groundnut accessions were susceptible to BW in North Vietnam
- The resistance potential of spanish types was higher than the potential of virginia and valencia types

- Short-duration accessions were more susceptible to BW than were medium- and long-duration accessions
- Gie Nho Quan, a local landrace, is potentially one of the most valuable source of resistance in Vietnam It can be used for growing in BW infested areas and making crosses to create resistant varieties
- Schwarz 21, Matjan, and some other BW resistant varieties gave good yields and therefore were selected for on-farm testing in BW infested areas

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# Recommendations

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# Recommendations

During the Fourth Groundnut Bacterial Wilt Working Group (GBWWG) Meeting, further research priorities and related workplans were intensively discussed among the participants It was believed that most of the recommendations formed in the Third GBWWG Meeting held in Wuhan, China in July 1994 still stand Following are the revised recommendations based on the discussions

## Disease Monitoring

Systematic surveys have been conducted in the past in all member countries, and substantial information is available on the disease distribution across different regions and cropping systems However, high-yielding varieties, which may or may not be resistant to bacterial wilt, have been extensively used or distributed recently, especially in the non-traditional groundnut-growing areas of China and Vietnam It is therefore agreed that annual surveys should continue in all countries, and information should be gathered on the changing scenario of BW with the change in varieties Also, it was suggested to conduct targeted surveys of disease-endemic areas using Geographical Positioning System (GPS) and Geographical Information Systems (GIS)

## Basic Research on *Burkholderia solanacearum*

Basic research on *B solanacearum* strains affecting groundnut is highly desired Research activities for disease diagnosis, pathogen detection, and determination of pathogenicity at hot spots in China and Vietnam will be conducted and strengthened through collaboration with advanced laboratories in developed and developing countries Wherever possible, use of both molecular and conventional approaches will be followed

Efforts should be made to develop more sensitive and specific monoclonal antibodies to detect the individual biovars and pathotypes of *B solanacearum* in different plant parts (seed, plant tissues, and soil)

Strains highly virulent on groundnut, reported from China and Vietnam, should be sent to an advanced research laboratory (such as Rothamsted Experimental Station in UK) for maintenance, characterization, and documentation Priority in this effort should be given to strains collected from member countries



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## Germplasm and Information Exchange Among Member Countries

A Working Group news sheet will be edited by the Technical Coordinator (TC) and circulated among members of the Working Group Two to three issues will be produced annually Groundnut genotypes will be exchanged among member countries The exchange will be carried out bilaterally or through ICRISAT

## Genetic Enhancement for Host–Plant Resistance

Host–plant resistance is an important component of disease management More coordinated research is needed to evaluate wilt resistant landraces, especially those collected from humid areas of China and Vietnam These landraces should be assembled at ICRISAT and screened at disease hot spots in China and Vietnam There is a concerted need to collect promising germplasm from disease-endemic areas, and conserve it at ICRISAT Sources of stable resistance to BW are available in member countries, and most of these sources are available at the ICRISAT gene bank Researchers are encouraged to test these sources systematically at hot spots in their countries This was suggested as an alternative to an International Groundnut Bacterial Wilt Nursery (IGBWN) Results of screening should be reported in GBWWG meetings

The Group strongly felt the need to give high priority to breeding high-yielding cultivars combining resistances to BW and major fungal foliar diseases, specifically late leaf spot and rust Members also wanted to monitor the incidence and severity of web blotch, which has been recently found to be a disease of increasing potential in southern China and northern Vietnam (specifically on newly developed BW-resistant cultivars) These foliar diseases are serious constraints to groundnut production in warm, humid environments

The Group also felt the need to emphasize understanding of the mechanism of resistance (such as the role of incubation period and latent infection) and its use in breeding for disease resistance

Further screening of groundnut germplasm lines to identify additional and diverse sources of resistances to BW will be continued in China, Indonesia, and Vietnam Breeding for BW resistance combined with multiple resistance to fungal foliar diseases (late leaf spots and rust) and other wilt diseases, with drought tolerance and improved quality will be highly emphasized Further investigation will be made of the genetics and mechanisms of resistance to BW in groundnut Priority will be given to improving methods for resistance evaluation, including techniques for artificial inoculation and for detecting latent infection

## Interaction between *B solanacearum* and other soilborne pathogens

The Group strongly felt the need to start systematic studies on the interaction of the groundnut wilt pathogen with such soilborne fungi as *Fusarium* spp *Aspergillus flavus*, and *Sclerotium rolfsii*

This relationship is quite commonly observed, and it is felt that bacteria and fungi together are causing enormous damage, especially in droughts and other rain free periods in upland areas The Group also felt a need to develop and/or assemble a package of practices available to combat soilborne diseases and include them in on-farm studies on groundnut BW management

## Integrated Disease Management and Transfer of Technology

Cultural control will be further studied in terms of mechanisms of crop rotation influencing the pathogen population and pathogenicity Rotation and field management practices are still recommended to farmers wherever feasible It was felt that substantial information is available on host-plant resistance and cultural practices that reduce disease incidence, but these combinations are not yet sufficiently verified and validated in on-farm research so that these practices can be used by farmers Therefore, concerted efforts are needed to initiate farmers' participatory research to devise appropriate location-specific packages of these strategies More research efforts will be made for biological control by using avirulent strains of *B solanacearum* strains Seed transmission will be further monitored

## Workshop and Training Activities

In-country workshops were recommended for China and Vietnam These in country workshops will be held for more extensive information exchange and technology transfer The activity will be supported mainly by individual member countries with limited support from CLAN An in country workshop is tentatively planned for 1999 or 2000 in southern Vietnam

## Funding Proposal Development

It was agreed that all national programs of the member countries should support research on groundnut BW wherever this disease is a constraint to crop production At present, national programs are the main sources of research funding International project and fundings are important for BW research, especially for international cooperation Proposals written by member countries will be prepared and submitted to donors for potential funding

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