J Gen Plant Pathol (2014) 80:504–509
DOI 10.1007/s10327-014-0550-2

PRESIDENTIAL ADDRESS

Genetic diversity of *Ralstonia solanacearum* and disease management strategy

Kenichi Tsuchiya

Received: 14 July 2014 / Accepted: 15 July 2014 / Published online: 12 August 2014

Introduction

Ralstonia solanacearum (Smith) (Yabuuchi et al. 1996) is the causal organism of bacterial wilt of more than 200 species representing 50 families of plants, including economically important crops such as potato, tomato, eggplant, and banana. Bacterial wilt is one of the most important diseases causing a major constraint on farmers. The disease has been widely recognized in tropical, subtropical and warm temperate regions of the world (Hayward 1991, 1994a).

Isolates of *R. solanacearum* differ in host range, geographical distribution, pathogenicity, epidemiological characters, and physiological properties. To describe this intraspecific variability of *R. solanacearum*, researchers have devised many classification schemes to identify this bacterium and understand its evolution (Horita and Tsuchiya 2009).

For more than 5 decades, *R. solanacearum* has been mainly classified according to race and biovar, based on differences in host range and biochemical properties, respectively (Denny and Hayward 2001; Hayward 1964, 1994b).

Consequently, over the last 3 decades, molecular biological studies have been introduced to classify and identify *R. solanacearum* strains from diverse origins. In various genetic analyses, genetic variability between and within the population of *R. solanacearum* has been assessed. On the basis of results obtained by molecular analyses, *R. solanacearum* has become defined as a species complex (Gillings and Fahy 1994; Poussier et al. 2000b; Villa et al. 2005), and a new classification system has been suggested (Fegan and Prior 2005; Remenant et al. 2011).

In this paper, I discuss three topics: (1) current classification schemes of *R. solanacearum*, (2) the genetic diversity of the pathogenic strains, in particular, those of potato and Zingiberaceae plants in relation to continuously present or newly emergent diseases in Japan, and (3) perspectives on disease control strategies.

Current classification of *R. solanacearum* strains

For more than 5 decades, *R. solanacearum* has been mainly classified based on a binary system, namely race and biovar, one emphasizing pathogenicity (host range or hosts primarily affected) and the other using selected biochemical characters (metabolism of different carbon sources) (Hayward 1991), respectively. Five races (Buddenhagen and Kelman 1964; Denny and Hayward 2001; He 1986) and six biovars (Hayward 1964, 1994b) have been described and designated so far.

Race 1 affects tobacco, tomato, many solanaceous and other weeds and certain diploid bananas; race 2 affects triploid bananas (causing Moko disease) and *Heliconia*; race 3 affects potato and tomato; race 4 affects ginger; and race 5 affects mulberry. On the other hand, six biovars (1, 2, N2, 3, 4, and 5) have been differentiated based on the metabolism of six kinds of sugars and three kinds of hexose alcohols. Although this dichotomous classification scheme has proven to be useful in assessing geographical and

This article is an abstract of the Presidential Address presented at the 2014 Annual Meeting of the Phytopathological Society of Japan in Sapporo. The content basically follows that of a review paper for the 100th anniversary (DOI 10.1007/s10327-014-0537-z).

K. Tsuchiya (✉)
Graduate School of Agriculture, Kyushu University, Fukuoka 812-8581, Japan
e-mail: kentsuch@agr.kyushu-u.ac.jp

pathological differences within *R. solanacearum* strains worldwide, it does not completely represent the diversity within *R. solanacearum* strains so that we can understand the distribution and population dynamics of this bacterium in global environments. Therefore, genetic variability among the strains has needed be assessed accurately and in detail using a more useful system.

Palleroni and Doudoroff (1971) clarified that DNA–DNA homology between *R. solanacearum* strains each belonging to a distinct biovar is below the same species level (<70 %) and suggested the possibility that this bacterium originally consisted of multiple species. Accordingly, various molecular biological techniques (e.g., RFLP, 16S ribosomal RNA gene sequences, rep-PCR) have been applied to classify and discriminate *R. solanacearum* strains since 1989.

An analysis of restriction fragment length polymorphisms (RFLP) identified the hypersensitive response and pathogenicity (*hrp*) locus and additional loci, thus revealing two evolutionary divisions, division I (strains mainly originating from Asia and Australia) and division II (strains mainly originating from South and Central America), which correlate nicely with the biovar distribution (Cook et al. 1989). Moreover, based on the sequence data from analyses of bacterial rDNA (16S, ITS), pathogenicity-related genes (e.g., *hrpB*, *egl*), and housekeeping genes (e.g., *mutS*, *gyrB*), *R. solanacearum* strains revealed to be divided into four major clades regardless of the respective genes used (Castillo and Greenberg 2007; Fegan et al. 1998; Poussier et al. 2000a, b; Villa et al. 2005; Wicker et al. 2012). Fegan and Prior (2005) called these major clades phylotypes (I–IV) and suggested that each phylotype should be treated as a specific taxonomic group that corresponds almost completely to separate species or subspecies. According to this new classification scheme, the concept of division and subdivision was reorganized (Fegan and Prior 2005). Strains in phylotype I originated in Asia, whereas strains in phylotype II are predominantly from the Americas, and phylotype III comprises strains from Africa and nearby islands. Phylotype IV contains strains isolated primarily from Indonesia and has also been found in some Asian countries (including Japan) and Australia. Each phylotype is correlated with geographic origin, specific races and biovars.

In addition, each phylotype comprises a number of sequevars (Fegan and Prior 2005; Wicker et al. 2012), which are primarily determined on the basis of the differences of the conserved region of the endoglucanase gene (*egl*) sequence; 52 sequevars have been suggested so far (Fegan and Prior 2005; Wicker et al. 2012). Since the phylotype and sequevar classification scheme is not inconsistent with previous classification systems, that is, having an additive or complementary nature, this system has been well received and become popular worldwide. Furthermore, due to the development of a multiplex-PCR method to differentiate each phylotype based on differences in the nucleotide sequences of the rDNA-ITS region, progress has been made on the discrimination of each phylotype of worldwide strains, including Japanese strains (Fegan and Prior 2005; Horita and Tsuchiya 2012; Wicker et al. 2012).

Genetic diversity of *R. solanacearum* strains in Japan

So far in Japan, more than 46 species of 24 families of plants have been reported as hosts, and new hosts continue to be found each year (Horita and Tsuchiya 2009, 2012). Distribution of races 1, 3, and 4 and biovars N2, 3, and 4 have been clarified (Horita and Tsuchiya 2001; Horita et al. 2005, 2010; Morita et al. 1996; Tsuchiya 2008; Yano et al. 2005).

On the other hand, Japanese strains can be divided into 2 phylotypes (I and IV) and 11 sequevars, and phylotypes I and IV consist of 9 and 2 sequevars, respectively (Horita and Tsuchiya 2012; Waki et al. 2013). Strains in phylotype II or III have not been isolated so far in the country. Phylotype of the Japanese strain is closely related to biovar and its original host. Biovar N2 strains from potato belong to phylotype IV, whereas biovar 3 and biovar 4 strains from potato and other kind of plant isolates are all included in phylotype I (Horita and Tsuchiya 2009). Phylotype I strains have been isolated from a wide range of crops and areas, whereas phylotype IV strains have been isolated only from potato cultivation areas. Similarly, some of the sequevars are correlated with specific race, original host and/or geographic origin (Horita and Tsuchiya 2012; Waki et al. 2013).

Genetic and pathological characters of potato strains of *R. solanacearum* in Japan

In Japan, potato bacterial wilt was first reported in the 1950s, in the Hokkaido region, the northern district of Japan. Since the 1960s, it occurred mainly in Nagasaki Prefecture, Kyushu District, warm, temperate, southwestern regions, which suggested that bacterial wilt disease had been widespread and continuous in potato- cultivating fields for over 40 years, and a recent outbreak occurred in Okinawa Island, the southernmost prefecture (Horita and Ooshiro 2002; Katayama and Kimura 1986; Suga et al. 2013).

The Japanese potato strains belong to three biovars (N2, 3, 4) and two phylotypes (I and IV). Both biovar 4 (phylotype I) and biovar N2 (phylotype IV) strains are common and widely isolated from Hokkaido to Okinawa. On the

other hand, biovar 3 (phylotype I) strains are found in limited areas (Nagasaki and Okinawa Prefectures) in Japan (Horita et al. 2010). From a plant quarantine point of view, phylotype II/biovar 2 (American strains) strains have not yet been reported in Japan (Elphinstone 2005; Nouri et al. 2009; Swanson et al. 2005), since only domestic seed potatoes have been distributed for cultivation and the import of the seed potatoes from foreign countries has been strictly prohibited by a plant protection law (Horita et al. 2012).

The Japanese potato strains belonging to either phylotype I and phylotype IV have been divided into 17 types based on differences in their pathogenicity to eggplant, *Solanum integrifolium* (rootstock for eggplant), tomato, and others (Suga et al. 2013). Although phylotype IV has only been isolated from potato in Japan, those strains belonging to phylotype IV have affected tomato cultivation along with phylotype I in Korea, Indonesia, and Australia (Jeong et al. 2007; Wicker et al. 2012), so we may need to pay attention to infection of tomato plants by phylotype IV strains in Japan as well. Also phylotype IV strains showed high virulence to the breeding lines carrying bacterial wilt resistance conferred from the wild potato *Solanum phureja* (Boshou 2005), which is resistant to phylotype I strains (Suga et al. 2013). Although phylotype IV strains have not been commonly used as targets for breeding for bacterial wilt resistance in Japan, further investigations using suitable *R. solanacearum* strains (include phylotype IV strains) will be needed before the practical introduction of new potato varieties to the field and to explore other new genetic resources for breeding for resistance to Japanese potato phylotype IV strains.

Characteristics and dissemination of emergent *R. solanacearum* strains of ginger bacterial wilt in Japan

Bacterial wilt of Zingiberaceae plants caused by race 4 has become very destructive in areas of Asia–Pacific region, including Hawaii, and several Asian countries in the last decade (Hayward 1994a).

In Japan, bacterial wilt of Zingiberaceae plants was first reported in fields cultivated with *Curcuma alismatifolia* in Kochi Prefecture in 1995 (Morita et al. 1996). Subsequently, the disease has occurred in ginger and mioga fields throughout neighboring areas of the same prefecture (Tsuchiya et al. 2005; Yano et al. 2005). Since 2005, the pathogen has been spreading to ginger fields in prefectures outside Kochi such as Tochigi (Waki et al. 2013). Biovar classification of the isolates from Kochi Prefecture revealed that all of the strains were identical to biovar 4; however, the ginger strains from Tochigi Prefecture belonged to biovars 3 and 4 (Waki et al. 2013).

Genetic diversity analyzed by rep-PCR revealed that the Japanese race 4 strains are subclassified into two DNA fingerprint types; type I showed high similarity to that of Zingiberaceae strains from Thailand, and type II strains are homogeneous to ginger strains from China and Australia (Tsuchiya et al. 2005). Based on these results, two PCR primer sets were designed to discriminate each strain representing type I or type II DNA fingerprints, respectively (Horita et al. 2004). Most strains from Japan (Kochi, Tochigi, and some prefectures in Kyushu District) were included in type I, whereas some of the strains (Kochi and Kagoshima prefectures) were type II.

Phylogenetic analysis based on the *egl* sequences, Zingiberaceae plant strains in phylotype I derived from Japan and foreign countries were further divided into several major groups, each corresponding closely to six sequevars (Waki et al. 2013). These groups were closely correlated with the host species and/or geographic origin.

On the other hand, since a few exceptional Japanese ginger strains are an unknown sequevar, the pathogen originated from different Asian countries. Disseminated by latently infested seed rhizomes, it was dispersed within Japan, causing disease epidemics through separate routes.

Zingiberaceae plants strains representing type I DNA fingerprints were strongly pathogenic to ginger, mioga, and curcuma. By contrast, the rest of the ginger strains representing type II fingerprints were strongly pathogenic to ginger only (Tsuchiya et al. 2005; Yano et al. 2011). In cross-inoculation tests with Japanese strains of *R. solanacearum*, isolates from other than Zingiberaceae plant were not pathogenic to ginger; however, the Zingiberaceae plant isolates in type I were strongly virulent to potato, but weakly virulent or avirulent on other Solanaceae crops (e.g., tomato, tobacco, eggplant, pepper) and peanuts (Yano et al. 2011). On the other hand, the pathogenic characteristics of the Japanese strains in type II were the same as those of the ginger strains from China and Australia (both are in the same DNA type II), which were strongly pathogenic to most Solanaceae crops, but weakly virulent or avirulent on tobacco and peanuts. These findings thus suggested that the DNA-based groupings of the strains are closely correlated with the pathogenic characteristics such as host range and virulence on specific plants), as well as becoming to latent host plants during domestic dissemination of the pathogenic strains.

Disease management strategies for control of bacterial wilt

Despite decades of efforts by many national and international organizations, bacterial wilt has continued to be a considerable problem throughout the world. As satisfactory

and long-term control strategy of bacterial wilt will be achieved through integrated pest management (IPM) by effectively combining cultural control (crop rotation, intercropping, soil amendment, solarization etc.), chemical control (soil fumigants), biological control (antagonistic microorganisms), and host resistance (resistant cultivar, induced resistance). For example, more effective protection than usual is expected by combining commercial high-grafted seedlings (grafting at a higher position onto resistant rootstock with susceptible scion), in which bacterial multiplication and movement are limited in the rootstocks and infection and wilting are suppressed in the scions, with anaerobic/reductive soil disinfestation method (Kajihara and Nakaho 2014; Momma 2013). Biofumigation with an organic amendment using plant products (Ooshiro 2008) and "plant activators" that induce SAR (systemic acquired resistance) in plants (Takahashi et al. 2006) should also become alternatives to chemical fumigants. Furthermore, the isolation and application of plant growth-promoting endophytic bacteria (PGPEB) as biocontrol agents against *R. solanacearum* and inducers of a disease suppression mechanism associated with the systemic induction of resistance via the SA (salicylic acid)-dependent pathway was recently reported (Hoang et al. 2011).

Conclusion

A phylotype/sequevar classification system of *R. solanacearum* species complex has several advantages. (1) It can clarify the phylogenetic relationship among the strains and can differentiate the strains more precisely and exactly than traditional conventional methods (race and biovar system) by using the DNA sequences deposited in a public database. (2) Many strains that differ in host, geographic origins and phenotypic characters, some of which are difficult to collect and test in the same laboratory, can be compared and discriminated at one time under common criteria. (3) It is basically compatible with traditional classification system. Thus, this system has been well received and become popular worldwide, and it will contribute to the analysis of genetic diversity among strains as well as help establish methods for genetic diagnosis.

However, the system does have some critical problems. (1) Phylotype and sequevar are informal classifications at the species and subspecies level, and the former infrasubspecific system (race and biovar) (each phylotype may be treated as one formal taxonomic group in the near future). (2) The basic criteria to establish new sequevar still need to be clarified. (3) The correlation of the sequevars with other phenotypic characters (e.g., host range, virulence, biovar) has not yet been assessed precisely, except for some special cases. Further systematic analysis of these points is needed.

Control of bacterial wilt requires an integrated management program; it is important to combine various control techniques effectively.

Regarding the emergent occurrence of potato and ginger bacterial wilt, there is a concern that some strains might have been disseminated via infected, generally asymptomatic planting material not only on a global scale but also a domestic scale. Thus, seeds and plants for planting are generally the key subject of rigorous inspection to ensure material is pathogen-free as well as to eliminate the pathogens in infested fields.

Efficient countermeasures must therefore be independently considered and implemented according to the local situation (i.e., whether bacterial wilt disease has previously been recorded, the host cultivation history, pathogenic characterization of the pathogen).

Newly introduced (or imported) seed rhizomes should be subject to a quarantine check, and the host cultivation history of the areas where rhizomes are produced (i.e., whether bacterial wilt disease has previously been recorded there) should be investigated as in the case of potato. It is necessary to investigate methods ranging from visual inspection of material to sampling, reinforcing these with simple and accurate diagnostic measures or the imposition of post-entry quarantine measures to ensure freedom from the disease in domestic seed materials.

Acknowledgments I express my deepest gratitude to all my colleagues, staff and students for their enthusiastic collaboration and cooperation in this research. The research was supported in part by Grant-in-Aid for Research projects from the Ministry of Agriculture, Forestry and Fisheries of Japan, the Ministry of Education, Culture, Sports, Science, and Technology of Japan, and the Japanese Society for the Promotion of Science.

References

Boshou L (2005) A broad review and perspective on breeding for resistance to bacterial wilt. In: Allen C, Prior P, Hayward AC (eds) Bacterial wilt disease and the *Ralstonia solanacearum* species complex. APS Press, St. Paul, pp 225–238

Buddenhagen I, Kelman A (1964) Biological and physiological aspects of bacterial wilt caused by *Pseudomonas solanacearum*. Annu Rev Phytopathol 2:203–230

Castillo JA, Greenberg JT (2007) Evolutionary dynamics of *Ralstonia solanacearum*. Appl Environ Microbiol 73:1225–1238

Cook D, Barlow E, Sequeira L (1989) Genetic diversity of *Pseudomonas solanacearum*: detection of restriction fragment length polymorphisms with DNA probes that specify virulence and the hypersensitive response. Mol Plant Microbe Interact 2:113–121

Denny TP, Hayward AC (2001) Gram-negative bacteria: *Ralstonia*. In: Schaad NW, Jones JB, Chun W (eds) Laboratory guide for identification of plant pathogenic bacteria, 3rd edn. APS Press, St. Paul, pp 151–174

Elphinstone JG (2005) The current bacterial wilt situation: a global overview. In: Allen C, Prior P, Hayward AC (eds) Bacterial wilt disease and the *Ralstonia solanacearum* species complex. APS Press, St. Paul, pp 9–28

Fegan M, Prior P (2005) How complex is the "*Ralstonia solanacearum* species complex"? In: Allen C, Prior P, Hayward AC (eds) Bacterial wilt disease and the *Ralstonia solanacearum* species complex. APS Press, St. Paul, pp 449–461

Fegan M, Taghavi M, Sly LI, Hayward AC (1998) Phylogeny, diversity and molecular diagnostics of *Ralstonia solanacearum*. In: Prior P, Allen C, Elphinstone J (eds) Bacterial wilt disease: molecular and ecological aspects. Springer, Berlin, pp 19–33

Gillings MR, Fahy P (1994) Genomic fingerprinting: Towards a united view of the *Pseudomonas solanacearum* species complex. In: Hayward AC, Hartman GL (eds) Bacterial wilt: The disease and its causative agent, *Pseudomonas solanacearum*. CAB International, Wallingford, pp 95–112

Hayward AC (1964) Characteristics of *Pseudomonas solanacearum*. J Appl Bacteriol 27:265–277

Hayward AC (1991) Biology and epidemiology of bacterial wilt caused by *Pseudomonas solanacearum*. Annu Rev Phytopathol 29:65–87

Hayward AC (1994a) The host of *Pseudomonas solanacearum*. In: Hayward AC, Hartman GL (eds) Bacterial wilt: The disease and its causative agent, *Pseudomonas solanacearum*. CAB International, Wallingford, pp 9–24

Hayward AC (1994b) Systematics and phylogeny of *Pseudomonas solanacearum* and related bacteria. In: Hayward AC, Hartman GL (eds) Bacterial wilt: The disease and its causative agent, *Pseudomonas solanacearum*. CAB International, Wallingford, pp 123–135

He LY (1986) Bacterial wilt in the People's Republic of China. In: Persley GJ (ed) Bacterial wilt disease in Asia and the South Pacific. Australian Centre for International Agricultural Research (ACIAR) Proc No. 13, Canberra, pp 40-48

Hoang HL, Furuya N, Takeshita M, Tsuchiya K (2011) Biocontrol of bacterial wilt of tobacco via induced resistance by endophytic bacteria. Phytopathology 101:S72

Horita M, Ooshiro A (2002) Genetic diversity of *Ralstonia solanacearum* strains isolated from potato and balsam pear in Okinawa Island. Bacterial Wilt Newsl 17:23

Horita M, Tsuchiya K (2001) Genetic diversity of Japanese strains of *Ralstonia solanacearum*. Phytopathology 91:399–407

Horita M, Tsuchiya K (2009) Current status and future prospects of the classification system for the bacterial wilt pathogen *Ralstonia solanacearum* species complex (in Japanese). Jpn J Phytopathol 75:297–306

Horita M, Tsuchiya K (2012) MAFF Microorganism genetic resource manual No. 12 (ver. 2) *Ralstonia solanacearum* (in Japanese). National Institute of Agrobiological Sciences, Tsukuba, pp 1–32

Horita M, Yano K, Tsuchiya K (2004) PCR-based specific detection of *Ralstonia solanacearum* race 4 strains. J Gen Plant Pathol 70:278–283

Horita M, Tsuchiya K, Ooshiro A (2005) Characteristics of *Ralstonia solanacearum* biovar N2 strains in Asia. J Phytopathol 153:209–213

Horita M, Suga Y, Ooshiro A, Tsuchiya K (2010) Analysis of genetic and biological characters of Japanese potato strains of *Ralstonia solanacearum*. J Gen Plant Pathol 76:196–207

Horita M, Suga Y, Ooshiro A, Tsuchiya K (2012) Genetic and biological characters of potato isolates of *Ralstonia solanacearum* in Japan (in Japanese). Plant Protect 66:274–276

Jeong Y, Kim J, Kang Y, Lee S, Hwang I (2007) Genetic diversity and distribution of Korean isolates of *Ralstonia solanacearum*. Plant Dis 91:1277–1287

Kajihara H, Nakaho K (2014) High grafting tomatoes to control bacterial wilt caused by *Ralstonia solanacearum* in long-term culture from summer to autumn (in Japanese). Plant Protect 68:75–78

Katayama K, Kimura S (1986) Ecology and protection of bacterial wilt of potato: 1. Ecology and strains of *Pseudomonas solanacearum* (in Japanese with English summary). Bull Nagasaki Agric Forest Exp Stn 14:1–30

Momma N (2013) Mechanisms for suppression of *Fusarium oxysporum* f. sp. *lycopersici* by anaerobicity-mediated biological soil disinfestation (ABSD) (in Japanese). Plant Protect 67:210–213

Morita Y, Yano K, Tsuchiya K, Kawada Y (1996) Bacterial wilt of *Curcuma alismatifolia* caused by *Pseudomonas solanacearum* (in Japanese). Proc Assoc Pl Protec Shikoku 31:1–6

Nouri S, Bahar M, Fegan M (2009) Diversity of *Ralstonia solanacearum* causing potato bacterial wilt in Iran and the first record of phylotype II/biovar 2T strains outside South America. Plant Pathol 58:243–249

Ooshiro A (2008) Control of bacterial wilt by *Geranium carolinianum* L (in Japanese). Plant Protect 62:90–95

Palleroni NJ, Doudoroff M (1971) Phenotypic characterization and deoxyribonucleic acid homologies of *Pseudomonas solanacearum*. J Bacteriol 107:690–696

Poussier S, Prior P, Luisetti J, Hayward C, Fegan M (2000a) Partial sequencing of the *hrpB* and endoglucanase genes confirms and expands the known diversity within the *Ralstonia solanacearum* species complex. Syst Appl Microbiol 23:479–486

Poussier S, Trigalet-Demery D, Vandewalle P, Goffinet B, Luisetti J, Trigalet A (2000b) Genetic diversity of *Ralstonia solanacearum* as assessed by PCR-RFLP of the *hrp* gene region, AFLP and 16S rRNA sequence analysis, and identification of an African subdivision. Microbiology 146:1679–1692

Remenant B, de Cambiaire J-C, Cellier G, Jacobs JM, Mangenot S, Barbe V, Lajus A, Vallenet D, Medigue C, Fegan M, Allen C, Prior P (2011) *Ralstonia syzygii*, the blood disease bacterium and some Asian *R. solanacearum* strains form a single genomic species despite divergent lifestyles. PLoS One 6:e24356

Suga Y, Horita M, Umekita M, Furuya N, Tsuchiya K (2013) Pathogenic characters of Japanese potato strains of *Ralstonia solanacearum*. J Gen Plant Pathol 79:110–114

Swanson JK, Yao J, Tans-Kersten J, Allen C (2005) Behavior of *Ralstonia solanacearum* race 3 biovar 2 during latent and active infection of geranium. Phytopathology 95:136–143

Takahashi H, Ishihara T, Hase S, Chiba A, Nakaho K, Arie T, Teraoka T, Iwata M, Tugane T, Shibata D, Takenaka S (2006) Beta-cyanoalanine synthase as a molecular marker for induced resistance by fungal glycoprotein elicitor and commercial plant activators. Phytopathology 96:908–916

Tsuchiya K (2008) Occurrence and spread of bacterial wilt diseases of Zingiberaceae plants caused by foreign strains (in Japanese). Plant Protect 62:72–75

Tsuchiya K, Yano K, Horita M, Morita Y, Kawada Y, d'Ursel CM (2005) Occurrence and epidemic adaptation of new strains of *Ralstonia solanacearum* associated with *Zingiberaceae* plants under agro-ecosystem in Japan. In: Allen C, Prior P, Hayward AC (eds) Bacterial wilt disease and the *Ralstonia solanacearum* species complex. APS Press, St. Paul, pp 463–469

Villa JE, Tsuchiya K, Horita M, Natural M, Opina N, Hyakumachi M (2005) Phylogenetic relationships of *Ralstonia solanacearum* species complex strains from Asia and other continents based on 16S rDNA, endoglucanase, and *hrpB* gene sequences. J Gen Plant Pathol 71:39–46

Waki T, Horita M, Kurose D, Mulya K, Tsuchiya K (2013) Genetic diversity of Zingiberaceae plant isolates of *Ralstonia solanacearum* in the Asia-Pacific region. JARQ 47:283–294

Wicker E, Lefeuvre P, de Cambiaire J-C, Lemaire C, Poussier S, Prior P (2012) Contrasting recombination patterns and demographic histories of the plant pathogen *Ralstonia solanacearum* inferred from MLSA. ISME J 6:961–974

Yabuuchi E, Kosako Y, Yano I, Hotta H, Nishiuchi Y (1996) Transfer of two *Burkholderia* and an *Alcaligenes* species to *Ralstonia* gen. nov.: proposal of *Ralstonia pickettii* (Ralston, Palleroni and Doudoroff 1973) comb. nov., *Ralstonia solanacearum* (Smith 1896) comb. nov. and *Ralstonia eutropha* (Davis 1969) comb. nov. Microbiol Immunol 39:897–904

Yano K, Kawada Y, Tsuchiya K, Horita M (2005) First report of bacterial wilt of mioga (*Zingiber mioga*) caused by *Ralstonia solanacearum* in Japan (in Japanese with English summary). Jpn J Phytopathol 71:179–182

Yano K, Kawada Y, Horita M, Hikichi Y, Tsuchiya K (2011) Phylogenetic discrimination and host ranges of *Ralstonia solanacearum* isolates from Zingiberaceae plants (in Japanese with English summary). Jpn J Phytopathol 77:88–95