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Emerging Bacterial Disease (Leaf Scald) of Sugarcane in China: Pathogenesis, Diagnosis, and Management

Muralidharan Govindaraju, Yisha Li and Muqing Zhang

Abstract

Sugarcane is the major industrial crop grown in tropical and sub-tropical regions in China. More than 100 sugarcane diseases are identified around the globe; half have been reported in China. Many varieties of sugarcane were replaced due to the infection of new pathogenic disease. Recently, leaf scald was found in China, which is one of the major sugarcane diseases also seriously affecting growth of sugarcane. Several isolates were recovered and identified using ELISA and PCR assays from the symptomatic leaf samples in Guangxi, China. The genomes of our isolates from *X. albilineans* were re-sequenced and revealed that rpf gene encoded regulation of pathogenicity factors mainly involved in the pathogenesis of sugarcane. The disease is mainly transferred through seed cane. In the past, hot water treatment was used to manage the disease. Healthy seed cane from resistant cultivars could effectively manage the leaf scald disease in sugarcane.

Keywords: sugarcane, leaf scald, *X. albilineans*, pathogenesis, bacterial disease, management

1. Introduction

Sugarcane (*Saccharum* spp.) is an industrially important crop of tropical and sub-tropical regions cultivated mainly for production of sucrose, biofuel, and ethanol [1]. China is one of the third largest sugarcane (*Saccharum officinarum* L.)-producing countries, followed by Brazil and India [2]. In the 1980s, Fujian and Guangdong were the two major sugarcane-producing provinces in China. Due to social, economic and environmental factors, major sugarcane-producing areas were moved to Guangxi and Yunnan provinces which accounts for 64 and 24% of total sugarcane-growing regions during 2015/2016. Since then, approximately 60% of the total cane yield is produced in Guangxi [2]. Sugarcane is the host of many serious plant pathogens that can mainly affect cane production and yield. In sugarcane, more than 100 pathogens have been reported to cause disease, including fungi, bacteria, phytoplasma, and virus [3]. Several of these diseases are considered to be the most severe threat to sugarcane production in Guangxi, especially leaf scald disease. Sugarcane leaf scald was first reported in the 1980s in Taiwan, China [4, 5], and recently found in 2015 in Guangxi, China. The pathogen from the recovered isolates was identified to be *Xanthomonas albilineans*.

1.1 Economic influence of leaf scald disease in sugarcane industry

Leaf scald disease (**Figure 1**) is also known as leaf burning disease, and it is caused by *X. albilineans* (Ashby) Dowson [3]. The major host plants mainly include *S. officinarum*, *Zea mays*, *Panicum antidotale*, *Bambusa vulgaris*, *Pennisetum purpureum*, and *Paspalum conjugatum*. The disease is mainly distributed in Australia, the USA, the Philippines, Myanmar, Thailand, Java, Laos, and Vietnam [6, 7]. Now, it is the most important quarantine disease in Taiwan, Guangxi, Guangdong, Yunnan, Fujian, Jiangxi, and Hainan in China [8]. Leaf scald was the reason for major losses in sugarcane at the beginning of the century when noble canes, *Saccharum officinarum*, was cultivated [9]. The major impact of the disease was reduced by the cultivation of interspecific hybrids. The susceptible cultivars were rapidly destroyed. Sugarcane cultivars resistant to leaf scald disease include F156, F160, F170, F173, and NCO310 in Taiwan, China. Other varieties resistant to this disease include Q42, Q50, Q98, Q813, POJ36, POJ2725, CP807, CP29–CP116, Co290, Co301, Co331, Co421, and B34104. The varieties susceptible to leaf scald disease are CP29–CP291, Co281, Co419, Co7301, Q44, Q63, Q66, B34104, B37161, B070, GT 46, GT 06–2081, GT 08–1589, LC 03–1137, and ROC1 [7, 10].

1.2 Symptomology

The major characteristic symptoms of leaf scald disease are divided into three phases: latent, chronic, and acute phases.

1.2.1 Latent phase

During this period, infected plants do not show any symptoms that occur in tolerant varieties and under favorable conditions for growth of plant. Stress can activate the infected plant to pass from the phase of latent into chronic or acute phase.



Figure 1.
Major sugarcane areas affected by leaf scald in China.

1.2.2 Chronic phase

The chronic phase is characterized by “white pencil line” stripe 1–2 mm wide and patches of chlorotic tissue on leaves, side shooting and burning of leaf tips. At a later stage, the margins’ stripe may become diffuse, and a red pencil line may be formed in the middle of the stripe. In dry weather, the leaf stripes dry out from the leaf apex to the margin, and finally the entire leaf become wilt. Infected plants exhibit shorter internodes, and the node of the stalk produces small tillerings, and a leaf of the tillerings shows white streaks. The disease-infected sections of stalks show reddening, and discolored vessels can pass through the internodes. Necrotic lesions may be noticed in severely affected stalks of plants (**Figure 2**).

1.2.3 Acute phase

In the acute phase of the disease, plant dies without showing any major symptoms. The infected stalks section does not show reddening of the vessels and tillerings joined into main stalk (**Figure 3**). It occurs mainly in drought condition of sugarcane growing period. In physiological water shortage condition, the leaves show the chronic streaks on re-tillering of diseased stalks [9].

1.3 Field assessment

LSD symptoms were recorded every month in the field. Disease severity was rated according to the procedure described [11]. However, all inoculated sugarcane stalks were rated individually, symptom severity ranging from 0 to 5. The ratings were used to calculate mean disease severity (DS): $DS = [(1 \times FL + 2 \times ML + 3 \times CB + 4 \times N + 5 \times D) / 5 \times T] \times 100$. However, FL = number of stalks with one or two pencil-line streaks (rating 1), ML = number of stalks with more than two pencil-line streaks (rating 2),



Figure 2.
Chronic phase of infection with pencil line stripe.

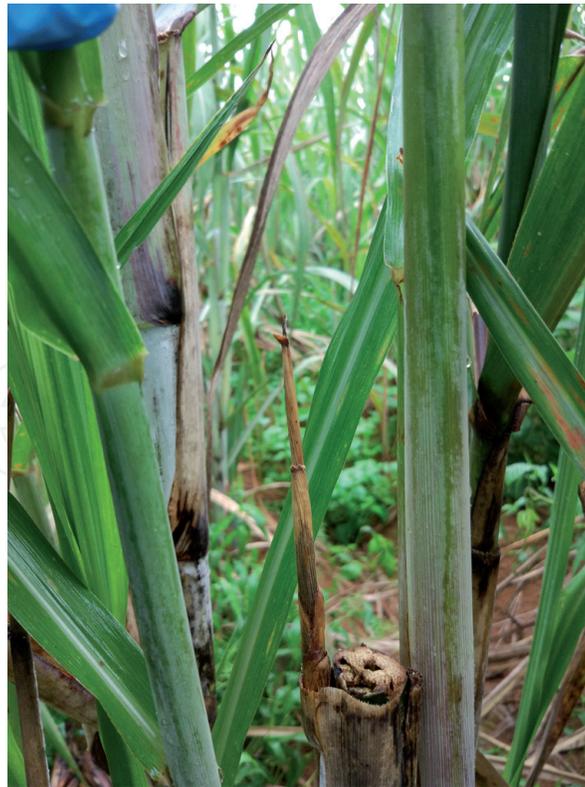


Figure 3.
Acute phase of infected plants leads to death.

CB = number of stalks with leaf chlorosis or bleaching (rating 3), N = number of stalks with leaf necrosis (rating 4), D = number of dead stalks or stalks with side shooting (rating 5), and T = total number of stalks. The rating of 5 was attributed to stalks with dead inoculated leaves 1 month after inoculation [11].

1.4 Phylogenetic

The multilocus sequence analysis (MLSA) of 119 strains of *Xanthomonas* genus is distributed into two uneven groups, with group 2 containing all but five species, namely, *X. albilineans*, *X. theicola*, *X. sacchari*, *X. translucens*, and *X. hyacinthi*, which were clustered into group 1 [12].

Three serovars associated with antigenic variations within *X. albilineans* were detected using three antisera (polyclonal antibodies) method against strains from three different geographical locations [13]. Serovars of 215 strains from 28 locations worldwide are affected by sugarcane leaf scald disease, and the distributed strains are divided into three groups according to serotype: (i) serotype 1 is the largest group, with strains from various geographic locations; (ii) serotype 2 consists of strains from tropical African countries; and (iii) serotype 3 contains strains from Caribbean islands (Fiji and Sri Lanka). This serological characterization of *X. albilineans* strains has been confirmed with monoclonal antibodies of 38 strains from different worldwide locations [14].

2. Pathogenesis

The pathogen is limited mainly to the leaf and stalk vascular bundles that are often partly or completely blocked with a gum-like substance. The organism may invade the parenchyma cells between the vascular bundles and cause reddened pockets of gum. No symptom of sugarcane plants can therefore constitute inoculum

sources for crop contamination. In addition, various epidemiological factors play a major role in field contamination. Leaf scald is spread by harvesters, hand knives, and infected setts by planting [9]. The pathogen found in the rhizosphere of infected roots has more possible transmission by root contact [15]. Leaf scald disease can also affect many other grasses which are alternate hosts for the disease. High moisture and temperature are the most favorable condition for the disease transmission.

2.1 Genes and diseases

The genome sequence of *X. albilineans* shows that the genes are effectively involved in pathogenesis. These genes include a cluster of genes called regulation of pathogenicity factors (rpf) responsible for the biosynthesis of a small diffusible signaling molecule. Diffusible signaling factor (DSF) encoded by RpfF has more similarities to long-chain fatty acyl CoA ligases [16, 17]. DSF quorum sensing and disruption of gene rpfF resulted in reduced virulence in different xanthomonads (*Xanthomonas* spp.), such as *X. axonopodis* pv. citri, *X. campestris* pv. campestris, and *X. oryzae* pv. oryzae [17, 18]. The rpf genes only control production of biofilm and other mechanisms involved in surface attachment of *X. albilineans*. A core group of genes, including *rpfF*, *rpfC* and *rpfG*, has played broader roles in gene regulation other than the transduction of DSF signals in *X. axonopodis* spp. [19]. The pathogen of leaf scald disease, *X. albilineans*, lacks type III secretion system (T3SS), which is found in most of pathogenic xanthomonads and acts as pathogenicity effectors in plant cells. *X. albilineans* also lacks all genes involved in the formation of biofilm, an important factor in virulence of plant pathogenic bacteria.

TonB-dependent transporters (TBDTs) are the important transporters involved in nutrient uptake [20] and also involved in iron or vitamin B12 uptake. These transporters are to facilitate the uptake of carbohydrates present in low amounts on the leaf surfaces [21, 22]. Genomes of several species of *Xanthomonas* are known to have high representation of TBDT genes, and it is functionally associated with pathogenicity of the bacterium [23].

During phyllosphere colonization, *Xanthomonas* encounters nutrient-limited environment on the leaf surfaces. Due to these conditions, TBDT has transported sucrose available in the phyllosphere [24]. Similarly, the genome of *X. albilineans* has 35 putative TBDT genes, and it is involved in the transport of plant cell wall derived nutrients like maltose, xylan, pectin, etc. [24–26].

2.2 Albicidin production and pathogenicity

Albicidin is a major phytotoxic compound specifically synthesized by the Gram-negative bacteria *X. albilineans* and plays an important role in pathogenicity [27]. It causes leaf scald in sugarcane [28]. The molecular target of albicidin is DNA gyrase (topoisomerase II) which is essential for DNA replication in bacteria. In planta, albicidin acts as a potent DNA gyrase inhibitor, thus blocking plastid development [29]. Albicidin (**Figure 4**) also has potent antibacterial activity which inhibits the growth of several positive and negative bacteria at nanomolar range with low minimal inhibitory concentration (MIC), e.g., *Escherichia coli* ($0.063 \mu\text{g mL}^{-1}$) [30], *Salmonella enteritidis* ($0.5 \mu\text{g mL}^{-1}$), and *Staphylococcus aureus* ($4.0 \mu\text{g mL}^{-1}$) [31]. Albicidin gives greater advantage to *X. albilineans* against other bacteria within the xylem vessels of sugarcane.

However, the entire 49-kb albicidin biosynthesis gene cluster was cloned and sequenced from *X. albilineans* (Xa23R1) [28]. This cluster is included in a genomic region of XALB1, and it contains three polyketide synthase (PKS) and non-ribosomal peptide synthase (NRPS) genes, as well as several putative modifying resistance and regulatory genes. Two additional 3-kb genomic regions of *X. albilineans* strain Xa23R1, namely, XALB2 and XALB3, were found to be involved

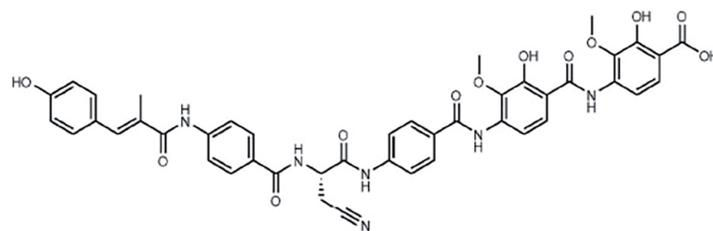


Figure 4.
Structure of albicidin.

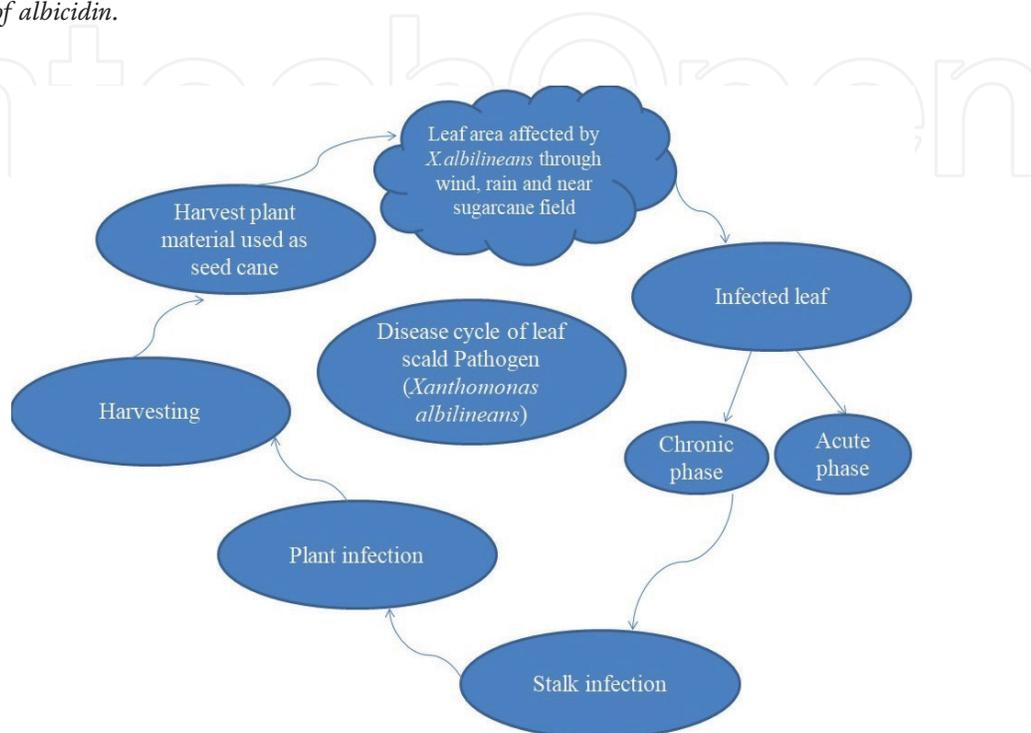


Figure 5.
Infection cycle of leaf scald pathogen.

in albicidin production. XALB2 has a single gene coding for a phosphopantetheinyl transferase required for posttranslational activation of PKS or NRPS enzymes [32]. XALB3 has a single gene coding for protein (HtpG) in albicidin production, which has not been elucidated [33].

2.3 Disease cycle

Xanthomonas albilineans can spread from inoculum sources to contaminated field of sugarcane and affect healthy sugarcane under the impact of various climatic conditions (**Figure 5**) [34]. The pathogen then colonizes the surface of the leaf, enters through the stomata, and progresses within the xylem, and symptoms may appear in infected plants. Pathogen can move into the stalk and then infect the stool showing scalding from the leaves [35]. The pathogen can be transmitted mechanically by harvesting equipment (harvesters) and the infected cane setts.

3. Diagnosis

3.1 Immunoassay

Various diagnostic methods are employed for detection of *X. albilineans*, including isolation on selective media and biochemical, immunological, and

molecular assays. ELISA was the most sensitive method and resulted in the detection of bacteria at the low titer [36]. Although isolation of *X. albilineans* with selective media is more time-consuming than other methods, it has proven to be very efficient in detecting the pathogen in symptomless and diseased plants [37]. DAC-ELISA and dot blot techniques were standardized to detect the bacterium in infected canes [38].

3.2 Molecular method

Polymerase chain reaction (PCR) protocols have been developed to detect *X. albilineans* in diseased stalks. The primer sets Ala4/L1 [39] and PGBL1/PGBL2 [40] were designed based on ITS region between the 16S and 23S rRNA genes of *X. albilineans*. PCR-based detection was further improved by *X. albilineans* specific amplification of the region between the 16S rRNA-tRNA^{Ala}-tRNA^{Ile}-23S rRNA gene by a nested PCR reaction [41]. Loop-mediated isothermal amplification (LAMP) was also employed for the detection of *X. albilineans* [42, 43]. It is an auto-cycling strand displacement DNA synthesis technique that involves the use of large fragment of DNA polymerase and a set of six primers [44], and it enables the synthesis of larger amounts of both DNA and by-products, e.g., hydroxy naphthol blue (HNB) [45, 46]. Quantitative PCR (qPCR) is a highly effective and accurate method for the detection of leaf scald disease [47].

4. Management strategy

4.1 Hot water treatment

Most of sugarcane diseases can be controlled through the use of disease-free seed cane. Hot-water treatments are used to disinfect planting material (seed cane). Before planting, soaking in ambient-temperature running water for 40 h followed by 3–4 h at 50°C is used to manage leaf scald bacteria, and it can provide 95% control efficacy [10].

4.2 Molecular approach

This approach is mainly developed to target factors that are responsible for pathogenicity other than toxins. The molecular modification in the host provides them resistance to pathogenicity factors and the factor inactivates by binding of hormones and enzymes. However, foreign genes' inactivation was carried out by gene silencing, nucleases, and coating proteins [48, 49]. The production of host cell surface components interferes with identification of the host and attachment to host cells by the pathogen.

CRISPR-Cas systems are involved in phage and plasmid defense, thus limiting HGT. The genome sequence of *X. albilineans* strain (GPE PC73) unveils the presence of two different systems named CRISPR-1 and CRISPR-2 [50]. The CRISPR-2 is associated with six cas genes (cas1, cas3, csy1, csy2, csy3, and csy4) and contains 28-bp repeats. CRISPR-2 is shared by the four strains (GPEPC73 and Xa23R1 from *X. albilineans*, GPE 39 and MUS 060 from *X. pseudalbilineans*) [50, 51]. Although, CRISPR-2 nucleic acid sequences of the repeats are 100% identical among the four strains, thus confirming the common origin of this locus [52].

Different resistant mechanisms on albicidin have been reported, including the nucleoside transporter Tsx, for which mutations have been described to import albicidin, or the endopeptidase AlbD from *Pantoea dispersa* [53–55], which cleaves albicidin

into two inactive fragments [56]. Another strategy is drug binding that counteracts the antibacterial effect of albicidin tetracycline-binding protein (TetR family) [57] or thiostrepton-binding protein (MerR family) [58]. The albicidin-binding protein AlbA from *Klebsiella oxytoca* [59] and AlbB from *Alcaligenes denitrificans* [60] provide protective effects for survival of the host strains. However, far-ultraviolet (UV) spectroscopy has indicated a mostly α -helical structure for AlbA [61], and amino acid residue His125 has played a vital role in albicidin binding [61, 62].

4.3 Genetic approach

The most potent method to prevent or manage leaf scald disease is the development and planting of resistant cultivars [3]. However, accurate determination of the resistant level of genotype against *X. albilineans* is most important in the cultivar selection program for leaf scald by artificial inoculation tests. The erratic symptom expression failed to accurately detect susceptibility, and thus multiple field trials utilizing inoculation are needed. Under this scenario, the marker-assisted selection (MAS) breeding technique, which uses DNA marker(s) linked to useful trait(s), has greater advantage in selecting clones resistant to leaf scald disease [63]. The transgenic sugarcane plants against *Xanthomonas albilineans* were produced by mediating albicidin detoxification (*albD*) gene through microprojectile bombardment [64].

4.4 Alternative control

4.4.1 Chemotherapy method

Spray of antibiotics such as streptomycin + tetracycline (60 g/ha/500 l water) at 2-week intervals was found to efficiently manage the pathogen in the field at 2 months after planting [65]. In preliminary stage, spraying of these antibiotics reduces the severity of leaf scald.

4.4.2 Biocontrol method

G. diazotrophicus may play a major role in defense against pathogens of sugarcane. It inhibited in vitro growth of leaf scald pathogen *Xanthomonas albilineans* [66, 67]. In addition, *G. diazotrophicus*-inoculated sugarcane stems were resistant to infection by *X. albilineans* [68]. Lactic acid bacteria (LAB) may be a biological alternative approach for leaf scald disease in sugarcane, and it produces antimicrobial peptides called bacteriocins and other substances, such as hydrogen peroxide, lactic acid, and reuterin, which are effective against several Gram-positive and Gram-negative bacteria. Antimicrobial peptides are alternatives for conventional pesticides and antibiotics, which are also used to treat against *X. albilineans* [69].

4.5 Farmer service program

Since 2000, healthy seed cane program has been developed by a government agency to protect the sugarcane diseases in China, further following recommendation to control or manage leaf scald disease.

1. The Pest and Disease Department plays a vital role by ensuring to provide disease-free parent seed material.
2. The government agency should arrange training program for farmers and take care of harvesting, transportation, and processing.

3. The crop agronomist should employ to cover aspects of growing the crop, such as soils, fertilizer, and herbicides and also to monitor the performance of new varieties.
4. The Inspection Department for all growers' fields should ensure that disease levels are acceptable and that remedial action is implemented where required. The monitoring of fields for pest damage is also important.
5. The efforts should be made to enhance awareness among the sugarcane farmers and strict vigilance by the quality control and regulatory authorities.

5. Conclusion

The leaf scald disease is becoming one of the serious threats to sugar industries in China. The infected plants should be removed completely to eliminate the seed cane carrying this disease in the field. Healthy seed cane was used from pathogen-free tissue culture plantlets (PTC). Better fertilization and irrigation practices may reduce the occurrence of this disease. A strict phytosanitary measure is needed to manage exchange of materials of sugarcane (seed cane) and propagation of disease.

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References

- [1] Lam E, Shine J Jr, Da Silva J, Lawton M, Bonos S, Calvino M, et al. Improving sugarcane for biofuel: Engineering for an even better feedstock. *GCB Bioenergy*. 2009;1(3):251-255
- [2] Zhang M, Govindaraju M. Sugarcane production in China. *Sugarcane-Technology and Research*, Intech. 2018. pp. 49-68. <http://dx.doi.org/10.5772/intechopen.73113>
- [3] Rott P, Davis MJ. Leaf scald In: Rott P, Bailey RA, Comstock JC, Croft BJ, Saumtally AS, editors. *A Guide to Sugarcane Diseases*. Paris: CIRAD-ISSCT; 2000
- [4] Wang JM. *Sugarcane Cultivation in China*. Beijing: China Agriculture Press; 1985
- [5] Huang HN. Distribution and classification of sugarcane diseases and control of main diseases in Guangdong province. *Sugarcane and Cane Sugar* (In Chinese). 1987;5:50-52
- [6] Saumtally A, Dookun-Saumtally A, Rao GP, Saumtally AS, Rott P. Leaf scald of sugarcane: A disease of worldwide importance. In: *Sugarcane Pathology: Bacterial and Nematode Diseases*. New Hampshire: Science Publishers; 2004. pp. 65-76
- [7] Zhang R, Shan H, Li W, Cang X, Wang X, Yin J, et al. First report of sugarcane leaf scald caused by *Xanthomonas albilineans* in the province of Guangxi, China. *Plant Disease*. 2017;101(8):1541
- [8] Li WF, Huang YK. *Diagnosis Detection and Control Technology of Modern Sugarcane Diseases*. Beijing: China Agriculture Press; 2012
- [9] Ricaud C, Egan B, Gillaspie A, Hughes C. *Diseases of Sugarcane: Major Diseases*. Amsterdam, The Netherlands: Elsevier Science Publishers B.V.; 1989
- [10] Huang Y, Li W. *Colored Atlas of Control on Diseases, Insect Pests and Weeds of Modern Sugarcane*. Beijing: China Agriculture Press; 2016
- [11] Rott P, Mohamed IS, Klett P, Soupa D, de Saint-Albin A, Feldmann P, et al. Resistance to leaf scald disease is associated with limited colonization of sugarcane and wild relatives by *Xanthomonas albilineans*. *Phytopathology*. 1997;87:1202-1213
- [12] Young JM, Park DC, Shearman HM, Fargier E. A multilocus sequence analysis of the genus *Xanthomonas*. *Systematic and Applied Microbiology*. 2008;31:366-377
- [13] Rott P, Davis MJ, Baudin P. Serological variability in *Xanthomonas albilineans* causal agent of leaf scald disease of sugarcane. *Plant Pathology*. 1994;43:344-349
- [14] Alvarez AM, Schenck S, Benedict AA. Differentiation of *Xanthomonas albilineans* strains with monoclonal antibody reaction pattern and DNA fingerprints. *Plant Pathology*. 1996;45:358-366
- [15] Klett P, Rott P. Inoculum sources for the spread of leaf scald disease of sugarcane caused by *Xanthomonas albilineans* in Guadeloupe. *Journal of Phytopathology*. 1994;142(3):283-291
- [16] Barber C, Tang J, Feng J, Pan M, Wilson T, Slater H, et al. A novel regulatory system required for pathogenicity of *Xanthomonas campestris* is mediated by a small diffusible signal molecule. *Molecular Microbiology*. 1997;24(3):555-566
- [17] Dow M. Diversification of the function of cell-to-cell signaling in

regulation of virulence within plant pathogenic xanthomonads. *Science Signaling*. 2008;1(21):23

[18] Chatterjee S, Almeida RPP, Lindow S. Living in two worlds: The plant and insect lifestyles of *Xylella fastidiosa*. *Annual Review of Phytopathology*. 2008;46:243-271

[19] Guo Y, Zhang Y, Li JL, Wang N. Diffusible signal factor mediated quorum sensing plays a central role in coordinating gene expression of *Xanthomonas citri* subsp. *citri*. *Molecular Plant-Microbe Interactions*. 2012;25:165-179

[20] Delmotte N, Knief C, Chaffron S, Innerebner G, Roschitzki B, Schlapbach R, et al. Community proteogenomics reveals insights into the physiology of phyllosphere bacteria. *Proceedings of the National Academy of Sciences of the United States of America*. 2009;106:16428-16433

[21] Lindow SE, Brandl MT. Microbiology of the phyllosphere. *Applied and Environmental Microbiology*. 2003;69:1875-1883

[22] Vorholt JA. Microbial life in the phyllosphere. *Nature Reviews. Microbiology*. 2012;10:828-840

[23] Rott P, Fleites L, Marlow G, Royer M, Gabriel DW. Identification of new candidate pathogenicity factors in the xylem invading pathogen *Xanthomonas albilineans* by transposon mutagenesis. *Molecular Plant-Microbe Interactions*. 2011;24:594-605

[24] Blanvillain S, Meyer D, Boulanger A, Lautier M, Guynet C, Denance N, et al. Plant carbohydrate scavenging through TonB-dependent receptors: A feature shared by phytopathogenic and aquatic bacteria. *PLoS One*. 2007;2:e224

[25] Pieretti I, Royer M, Barbe V, Carrere S, Koebnik R, Couloux A,

et al. Genomic insights into strategies used by *Xanthomonas albilineans* with its reduced artillery to spread within sugarcane xylem vessels. *BMC Genomics*. 2012;13:658

[26] De Bernonville TD, Noel LD, SanCristobal M, Danoun S, Becker A, Soreau P, et al. Transcriptional reprogramming and phenotypical changes associated with growth of *Xanthomonas campestris* pv. *Campestris* in cabbage xylem sap. *FEMS Microbiology Ecology*. 2014;89:527-541

[27] Birch RG. *Xanthomonas albilineans* and the anti-pathogenesis approach to disease control. *Molecular Plant Pathology*. 2001;2(1):1-11

[28] Royer M, Costet L, Vivien E, Bes M, Cousin A, Damais A, et al. Albicidin pathotoxin produced by *Xanthomonas albilineans* is encoded by three large PKS and NRPS genes present in a gene cluster also containing several putative modifying, regulatory, and resistance genes. *Molecular Plant-Microbe Interactions*. 2004;17(4):414-427

[29] Birch RG, Patil SS. Evidence that an albidin-like phytotoxin induces chlorosis in sugarcane leaf scald disease by blocking plastid DNA replication. *Physiological and Molecular Plant Pathology*. 1987;30(2):207-214

[30] Kretz J, Kerwat D, Schubert V, Grätz S, Pesic A, Semsary S, et al. Total synthesis of albidin: A lead structure from *Xanthomonas albilineans* for potent antibacterial gyrase inhibitors. *Angewandte Chemie, International Edition*. 2015;54(6):1969-1973

[31] Rostock L, Driller R, Grätz S, Kerwat D, von Eckardstein L, Petras D, et al. Molecular insights into antibiotic resistance-how a binding protein traps albidin. *Nature Communications*. 2018;9(1):3095

- [32] Huang G, Zhang L, Birch RG. Albicidin antibiotic and phytotoxin biosynthesis in *Xanthomonas albilineans* requires a phosphopantetheinyl transferase gene. *Gene*. 2000;**258**:193-199
- [33] Vivien E, Megessier S, Pieretti I, Cociancich S, Frutos R, Gabriel DW, et al. *Xanthomonas albilineans* HtpG is required for biosynthesis of the antibiotic and phytotoxin albicidin. *FEMS Microbiology Letters*. 2006;**251**:81-89
- [34] Champoiseau P, Rott P, Daugrois J-H. Epiphytic populations of *Xanthomonas albilineans* and subsequent sugarcane stalk infection are linked to rainfall in Guadeloupe. *Plant Disease*. 2009;**93**(4):339-346
- [35] Daugrois JH, Edon-Jock C, Bonoto S, Vaillant J, Rott P. Spread of sugarcane yellow leaf virus in initially disease-free sugarcane is linked to rainfall and host resistance in the humid tropical environment of Guadeloupe. *European Journal of Plant Pathology*. 2011;**129**(1):71-80
- [36] Autrey L, Dookun A, Saumtally S. Improved Serological Methods for Diagnosis of the Leaf Scald Bacterium *Xanthomonas albilineans*. In: *Proceedings of the International Society of Sugar Cane Technologists*. Sao Paulo, Brazil: Tropical Plant Pathology; 1989
- [37] Davis MJ, Rott P, Baudin P, Dean J. Evaluation of selective media and immunoassays for detection of *Xanthomonas albilineans*, causal agent of sugarcane leaf scald disease. *Plant Disease*. 1994;**78**(1):78-82
- [38] Viswanathan R, Ramesh SA. Isolation and production of antiserum to *Xanthomonas albilineans* and diagnosis of the pathogen through serological techniques. *Journal of Mycology and Plant Pathology*. 2004;**34**:797-800
- [39] Pan YB, Grisham M, Burner D. A polymerase chain reaction protocol for the detection of *Xanthomonas albilineans*, the causal agent of sugarcane leaf scald disease. *Plant Disease*. 1997;**81**(2):189-194
- [40] Pan YB, Grisham M, Burner D, Legendre B, Wei Q. Development of polymerase chain reaction primers highly specific for *Xanthomonas albilineans*, the causal bacterium of sugarcane leaf scald disease. *Plant Disease*. 1999;**83**(3):218-222
- [41] Honeycutt RJ, Sobral BW, McClelland M. tRNA intergenic spacers reveal polymorphisms diagnostic for *Xanthomonas albilineans*. *Microbiology*. 1995;**141**(12):3229-3239
- [42] Garces F, Gutierrez A, Hoy J. Detection and quantification of *Xanthomonas albilineans* by qPCR and potential characterization of sugarcane resistance to leaf scald. *Plant Disease*. 2014;**98**(1):121-126
- [43] Dias VD, Fernandez E, Cunha MG, Pieretti I, Hincapie M, Roumagnac P, et al. Comparison of loop-mediated isothermal amplification, polymerase chain reaction, and selective isolation assays for detection of *Xanthomonas albilineans* from sugarcane. *Tropical Plant Pathology*. 2018;**43**:351-359
- [44] Nagamine K, Hase T, Notomi T. Accelerated reaction by loop-mediated isothermal amplification using loop primers. *Molecular and Cellular Probes*. 2002;**16**:223-229
- [45] Fischbach J, Xander NC, Frohme M, Glökler JF. Shining a light on LAMP assays—A comparison of LAMP visualization methods including the novel use of berberine. *Biotechnology Techniques*. 2015;**58**:189-194
- [46] Goto M, Honda E, Ogura A, Nomoto A, Hanaki K. Colorimetric

detection of loop mediated isothermal amplification reaction by using hydroxy naphthol blue. *Biotechnology Techniques*. 2009;**46**:167-172

[47] Ginzinger DG. Gene quantification using real-time quantitative PCR: An emerging technology hits the mainstream. *Experimental Hematology*. 2002;**30**(6):503-512

[48] Beachy RN. Mechanisms and applications of pathogen-derived resistance in transgenic plants. *Current Opinion in Biotechnology*. 1997;**8**(2):215-220

[49] Matzke M, Mette M, Matzke A. Transgene silencing by the host genome defense: Implications for the evolution of epigenetic control mechanisms in plants and vertebrates. *Plant Molecular Biology*. 2000;**43**(2-3):401-415

[50] Pieretti I, Royer M, Barbe V, Carrere S, Koebnik R, Cociancich S, et al. The complete genome sequence of *Xanthomonas albilineans* provides new insights into the reductive genome evolution of the xylem-limited *Xanthomonadaceae*. *BMC Genomics*. 2009;**10**:616

[51] Bogdanove AJ, Koebnik R, Lu H, Furutani A, Angiuoli SV, Patil PB, et al. Two new complete genome sequences offer insight into host and tissue specificity of plant pathogenic *Xanthomonas* spp. *Journal of Bacteriology*. 2011;**193**:5450-5464

[52] Pieretti I, Cociancich S, Bolot S, Carrère S, Morisset A, Rott P, et al. Full genome sequence analysis of two isolates reveals a novel *Xanthomonas* species close to the sugarcane pathogen *Xanthomonas albilineans*. *Genes*. 2015;**6**(3):714-733

[53] Birch RG, Pemberton JM, Basnayake WV. Stable albicidin resistance in *Escherichia coli* involves an altered outer-membrane nucleoside

uptake system. *Journal of General Microbiology*. 1990;**136**:51-58

[54] Fsihi H, Kottwitz B, Bremer E. Single amino acid substitutions affecting the substrate-specificity of the *Escherichia coli* K-12 nucleoside-specific Tsx channel. *The Journal of Biological Chemistry*. 1993;**268**:17495-17503

[55] Zhang L, Birch RG. The gene for albicidin detoxification from *Pantoea dispersa* encodes an esterase and attenuates pathogenicity of *Xanthomonas albilineans* to sugarcane. *Proceedings of the National Academy of Sciences of the United States of America*. 1997;**94**:9984-9989

[56] Vieweg L et al. The albicidin resistance factor AlbD is a serine endopeptidase that hydrolyzes unusual oligoaromatic-type peptides. *Journal of the American Chemical Society*. 2015;**137**:7608-7611

[57] Ramos JL et al. The TetR family of transcriptional repressors. *Microbiology and Molecular Biology Reviews*. 2005;**69**:326-356

[58] Kahmann JD et al. Structural basis for antibiotic recognition by the TipA class of multidrug-resistance transcriptional regulators. *The EMBO Journal*. 2003;**22**:1824-1834

[59] Walker MJ, Birch RG, Pemberton JM. Cloning and characterization of an albicidin resistance gene from *Klebsiella oxytoca*. *Molecular Microbiology*. 1988;**2**:443-454

[60] Basnayake WVS, Birch RG. A gene from *Alcaligenes denitrificans* that confers albicidin resistance by reversible antibiotic binding. *Microbiology*. 1995;**141**:551-560

[61] Weng LX et al. Molecular and conformational basis of a specific and high affinity interaction between AlbA and albicidin phytotoxin. *Applied*

and Environmental Microbiology. 2005;71:1445-1452

[62] Weng LX, Xu JL, Li O, Birch RG, Zhang LH. Identification of the essential histidine residue for high-affinity binding of AlbA protein to albicidin antibiotics. Microbiology. 2003;149:451-457

[63] Costet L, Cunff L, Royaert S, Raboin LM, Hervouet C, Toubi L, et al. Haplotype structure around Bru1 reveals a narrow genetic basis for brown rust resistance in modern sugarcane cultivars. Theoretical and Applied Genetics. 2012;125:825-836

[64] Birch R, Bower R, Elliott A, Hansom S, Basnayake S, Zhang L. Regulation of transgene expression: Progress towards practical development in sugarcane, and implications for other plant species. Plant genetic engineering: towards the third millennium. In: Proceedings of the International Symposium on Plant Genetic Engineering; 6-10 December; Havana. Cuba: Elsevier Science Publishers; 1999, 2000

[65] Viswanathan R, Padmanaban P. Hand Book on Sugarcane Diseases and their Management. Coimbatore: Sugarcane Breeding Institute; 2008

[66] Blanco Y, Blanch M, Piñón D, Legaz ME, Vicente C. Antagonism of *Gluconacetobacter diazotrophicus* (a sugarcane endosymbiont) against *Xanthomonas albilineans* (pathogen) studied in alginate-immobilized sugarcane stalk tissues. Journal of Bioscience and Bioengineering. 2005;99(4):366-371

[67] Piñón D, Casas M, Blanch MA, Fontaniella B, Blanco Y, Vicente C, et al. *Gluconacetobacter diazotrophicus*, a sugar cane endosymbiont, produces a bacteriocin against *Xanthomonas albilineans*, a sugar cane pathogen. Research in Microbiology. 2002;153(6):345-351

[68] Arencibia AD, Vinagre F, Estevez Y, Bernal A, Perez J, Cavalcanti J, et al. *Gluconacetobacter diazotrophicus* elicitate a sugarcane defense response against a pathogenic bacteria *Xanthomonas albilineans*. Plant Signaling and Behavior. 2006;1(5):265-273

[69] Keymanesh K, Soltani S, Sardari S. Application of antimicrobial peptides in agriculture and food industry. World Journal of Microbiology and Biotechnology. 2009;25(6):933-944