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Witches' broom disease of Mexican lime trees: disaster to be addressed before it will be too late

Mohsen MARDI, S. Mojtaba KHAYAM NEKOUEI, Laleh Karimi FARSAD, Frooh EHYA, Mahnaz SHABANI, Maaedeh SHAFIEE, Meisam TABATABAEI, Mohammad Reza SAFARNEJAD, Gholamreza SALEHI JOUZANI, Ghasem HOSSEINI SALEKDEH
Agricultural Biotechnology Research Institute of Iran (ABRII), P.O. Box 31535-1897, Karaj, Iran

Abstract

Witches' broom disease of lime (WBDL) associated with '*Candidatus* Phytoplasma aurantifolia' presence, is responsible for major losses of Mexican lime trees (*Citrus aurantifolia* L.). Iranian witches' broom disease of lime Network (IWBDLN) comprises of 10 mega projectswhich have been carried out since 2008 with the contribution of 15 different national institutes. Here, we will present the program and the results obtained and demonstrate how this program could contribute in controlling this destructive agent of WBDL. Furthermore, we will demonstrate how our genomics, transcriptomics, epigenomics, proteomics and metabolomics analyses of lime infected by witches' broom disease provided new insight into plant stress tolerance mechanisms. Several strategies to maximize the success of our program in controlling the disease will be presented.

Key words: Witches' broom disease of lime, '*Candidatus* Phytoplasma aurantifolia', systems biology, management.

Introduction

Witches' broom disease of lime (WBDL), associated with '*Candidatus* Phytoplasma aurantifolia', is responsible for major losses of Mexican lime trees (*Citrus aurantifolia* L.). The WBDL phytoplasma is transmitted by the leafhopper *Hishimonus phycitis* Distant (Shabani *et al*., 2011). WBDL was first observed in Oman in 1975 leading to a 98% destruction of Mexican lime trees. The disease was then observed in UAE in 1991, in India in 1999 and in Iran in 2000. From 2000 until today, 30% of the Mexican lime trees (over half a million trees/7000 hectares) in Iran have been destroyed by WBDL.

Sadly, in 2009, the agent associated with the disease expanded its territory to other plants such as grapefruit (IWBDLN 2011, personal communications) and it is anticipated that it will pose a serious threat to other horticultural products in Iran and worldwide. Although only a small part of the globe is currently affected it is critical to take it into consideration that this disease is transferred by insects and if immediate actions are not taken, there would be a huge risk of a global epidemic.

Materials and methods

IWBDLN comprises of 10 mega projects including horticulture controlling, identification and diagnosis of host/pathogen/vector, identification of resistant Mexican lime trees, identification of substituted resistant cultivars, breeding of resistant cultivars, gardens resuscitation and replacement, education, extension and notification, finance and budget, supporting, quarantine of the intact and affected areas. These activities have been being carried out since 2008 with the collaboration of 15 national institutes.

Results

Detection of witches' broom disease of lime

A novel quantum dots FRET-based biosensor for the detection of '*Ca.* P. aurantifolia' was constructed. The quantum dots (QDs) were functionalized with a specific antibody against the '*Ca.* P. aurantifolia'. The specificity and sensitivity of the constructed nanosensor kit were found to be as high as 100%. This nano-based detection kit would facilitate early detection of the disease in order to manage and control the associated agent before the disease enters the irreversible stages of infection.

Inhibition of the pathogen using nutritional and pharmaceutical compounds

Treating the infected plants with a number of nutritional and pharmaceutical compounds such as surfactin was shown to have a significant inhibitory effect on '*Ca.* P. aurantifolia' (Askari *et al*., 2011). Fortunately, based on the results obtained and the accomplishments made through the mega plans conducted by the Network, the destructive agent of WBDL is currently under control.

Systems biology analysis of Mexican lime trees interactions with '*Ca*. P. aurantifolia'

Although plant molecular breeding for inducing tolerance to various stresses can have a great impact on increasing crop productivity, it suffers from some drawbacks such as slow progress and shortage of information on the molecular events underlying the tolerance (Ghayeb *et al*., 2011). Systems biology analysis of the plant/pathogen interaction provides invaluable insights into signaling pathways and molecular mechanisms underlying plant response and tolerance to disease. The advent of omics technologies has made it possible to identify a broad spectrum of genes in living systems and discover molecular mechanisms of stress tolerance.

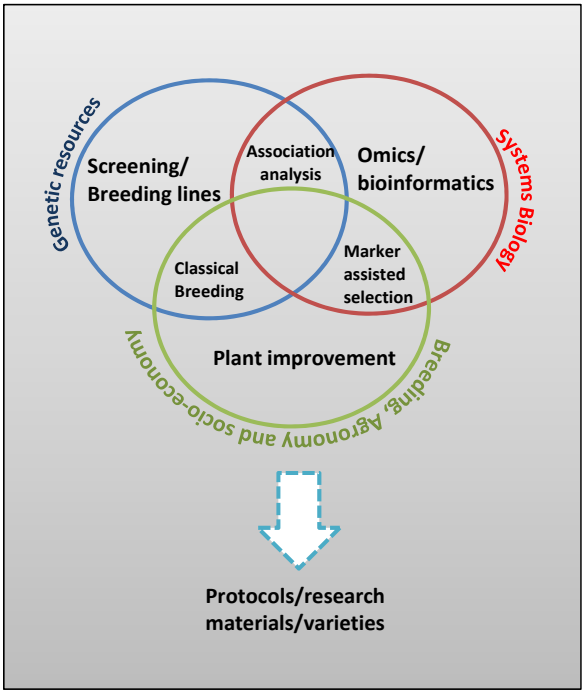

Figure 1. A strategy for developing resistant Mexican lime trees: a promising but long process.

We applied genomics, transcriptomics, epigenomics, proteomics and metabolomics approaches to analyze lime infected by witches' broom disease in order to identify new plant stress tolerance mechanisms (figure 1). The preliminary results indicated a number of candidate transcripts, microRNAs, proteins, and metabolites that might be involved in the interaction of Mexican lime trees with '*Ca.* P. aurantifolia'. We will discuss system biology based analysis of these results and novel mechanisms involved in plant pathogen interaction.

Discussion

Today, the disease has been controlled and preventive strategies have been introduced and implemented in the infected areas. However, it should be noted that its threat is always around and being on alert globally will keep new tragedies away. Owing to a comparative analysis, several mechanisms emerged as key participants in plant response to stress. Further investigation is required to elucidate the roles of these mechanisms in the susceptibility/resistance of Mexican lime tress to disease, and to determine how strategies might be developed to incorporate these genes into molecular breeding programmes.

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Corresponding author: Mohsen MARDI (e-mail: mardi@abrii.ac.ir), Head of Genomics Dept., Agricultural Biotechnology Research Institute of Iran (ABRII), Mahdasht Road, P. O. Box: 31535-1897, Karaj, Iran; Manager of Iranian Witches' Broom Disease of Lime (IWBDLN), Ministry of Jiad-e-Agriculture, Tehran, Iran.