

Identification and Controlling of Stem Bulging of Passion Fruit (*Passiflora Edulis*) in Sri Lanka

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Abstract— Stem bulging is one of the major biological constrains of passion fruit cultivation especially in Low country wet zone areas in Sri Lanka. Green bark split as a result of bulging, discoloration of bark and also stained internal tissues are the symptoms of this stem bulging. A survey was conducted in those infected areas to find out the causal factors of passion fruit stem bulging. Soil samples were collected from disease infected and disease free locations and analyzed for soil pH, major nutrients and some micro nutrients. Examination of the internal tissues of stem showed at first inspection, looked like fungal hyphae. Microscopic observations were consistently indicated the *Fusarium* species association with infected tissues. Also, Passion fruit stem bulging samples were taken and dispatched to the CABI (Centre for Agriculture and Bioscience International) for further diagnosis of causal organism. The field trail - 1 was conducted at Agro ecological zone WL1a to observe the disease transmission of stem bulging from infected plants to healthy plants. The field trail – 2 was conducted at Agro ecological zone WL1a based on fungicides, insecticides and foliar fertilizer to control the stem bulging of passion fruit. The experiment was laid out in a Randomize Complete Block Design and five treatments were used with 3 replicates. The treatments were, T1- Foliar application of insecticide - Thiamethoxam 25WG 3g/10l, T2- Foliar application of fungicide - Thiophanate methyl 70 WP 6g/ 10 l, T3- Alternative foliar application of Thiamethoxam 35 WG + Thiophanate methyl 70 WG, T4- Alternative foliar application of Thiamethoxam 25 WG + Thiophanate methyl 70WP + Albert solution (commercial formulation of nutrient mixture) 5g/10 l and T5- Control. Results numerated that, stem bulging can be transmitted from infected plant to healthy plants as a disease. *Fusarium* species were consistently found in bulged area of the samples tested. The lowest disease incidence was observed in Thiophanate methyl 70 WP treated plots of field trials indicating *Fusarium* species

may be the causal factor of the problem. Insecticide applications were failed in controlling stem bulging in the field conditions. According to the CABI report No: IMI 504215 & IMI 504216, the isolation made from internal tissues and two fungal cultures were subjected for molecular identification and *Gibberella fujikuroi* and *Fusarium* sp. were identified as associate fungi of stem bulging of passion fruit.

Keywords— Stem bulging, Passion fruit, *Gibberella fujikuroi*, *Fusarium* sp.

I. INTRODUCTION

Stem bulging is one of the major biological constrains of passion fruit cultivation especially in Kalutara and Ratnapura districts (Agro ecological zone WL1a) in Sri Lanka since 1980s. Since then, this problem has gradually spread. Symptoms were observed in most of the farmer fields with different severity levels. The main symptoms observed in the affected vines were presented in Passion fruit stem branches. Green bark split as a result of bulging, discoloration of bark and also stained internal tissues. However initial examination showed no sign of fungal fruit bodies or insects.



Fig.1: Passion fruit stem bulging

Attempts have been made to develop some solutions for this problem. It was suspected that the symptoms resemble damaged caused by Boron deficiencies. However, application of Boron as a soil treatment (2g of borax per vine) at planting failed to manage stem bulging problem of passion fruit. Preliminary field observations numerated that, stem bulging can be transmitted from diseased plant to healthy plants. So, we were identified stem bulging as a disease. Therefore, it was decided to carryout studies to correctly identify the causal factors and develop control measures for stem bulging of passion fruit in low country wet zone area.



Fig.2: Green bark split as a result of bulging



Fig.3: Longitudinal Section of an affected stem

II. MATERIALS AND METHODS

A survey was conducted in stem bulging reported areas of Ratnapura and Kaluthara districts to find out the causal factors of passion stem bulging. Soil samples were collected from disease infected and disease free locations

and analyzed for soil pH, major nutrients and some micro nutrients.

The field trail - 1 was conducted to observe the disease transmission of stem bulging from infected plants to healthy plants. In here, 100% infected field was selected for the experiment. 60 healthy plants were grown adjacent to the diseased plants and 60 healthy plants were grown in disease free field. Disease incidence level was recorded at two months after planting.

Microscopic observations were made in bulged tissues to detect fungal infection. Affected tissues were microscopically inspected and it was observed fungal mycelium and two types of morphologically different conidia. Pieces of bulged tissues were kept on Potato Dextrose Agar (PDA) medium and incubated 7 days under room temperature. The mycelia development and conidia development on PDA was observed. Then single spore inoculants of fungi isolates were made. Ten isolates were collected and each isolate also wound inoculated into healthy passion fruit vines but and observed for development of bulging symptoms on inoculated stems to confirm Koch’s postulates. Also, Passion stem bulging samples were taken and dispatched to the CABI for further diagnosis of causal organism.

The field trail - 2 was conducted at Ratnapura District (Agro ecological zone WL1a) to develop control measures for stem bulging. The experiment was laid out in a Randomize Complete Block Design and five treatments were used with 3 replicates. The treatments were as follows.

Treatment	Description
T1	Foliar spraying of insecticide - Thiamethoxam25 WG at the rate of 3g/10 l, two months after planting and spraying was continued 3 times in 2 weeks intervals
T2	Foliar spraying of fungicide - Thiophanate methyl 70 WP at the rate of 6g/ 10 l, two months after planting and spraying was continued 3 times in 2 weeks intervals
T3	Alternative foliar spraying of Thiamethoxam 25 WG and Thiophanate methyl 70 WP, two months after planting and spraying was continued 3 times in 2 weeks intervals
T4	Alternative foliar spraying of Thiamethoxam, Thiophanate methyl and Albert solution (liquid fertilizer) 5g/10 l, two months after planting and spraying was continued 3 times in 2 weeks intervals
T5	Control

III. RESULTS AND DISCUSSION

3.1 Analysis of soil samples collected from passion stem bulging infected fields

Table 1: Analysis of soil samples collected from passion stem bulging infected fields

Factor	Results
pH	4 – 5 Acidic
N,P	Normal
K	Higher than normal(800-1000 ppm)
Electrical conductivity	Very high
Micronutrients (Fe, Cu, Mn, Zn)	Normal

Note: Soil samples collected from 10 locations of healthy and disease infected passion fruit cultivation were used for analysis.

Results revealed that soils of those areas were acidic and pH of soil samples was varied 4 - 5. K level is higher than normal level. N and P levels are within normal range and no Fe, Cu, Mn, Zn deficiencies observed in the tested soil samples.

3.2 Transmission of stem bulging from infected plants to healthy plants

Table 2: Passion Stem bulging incidence of newly planted passion fruit plants which were planted in adjacent to the diseased field and new field

Treatment	Percent disease incidence after 2 months of field planting
T1- Healthy plants growing adjacent to the diseased plants	33.3
T-2 Healthy plants growing in new land	1.7

Stem bulging symptoms were first observed in T1 treatments within 5 weeks after field planting and later symptoms progressively developed and 33 % plants showed stem bulging symptoms. However, only one plant showed mild symptoms on stem of T2 treatment during the period. Results numerated that, stem bulging can be transmitted from infected plant to healthy plants.

3.3 Microscopic observations and characters of isolation of *Fusarium* collected from bulged stem samples of passion fruit.

Colony colour on PDA was white, and then turns into brown when older. Reverse colony colour on PDA was light brown and then turns into brown when older. There were two types of conidia. Macro conidia were abundant,

strait and pointed at ends 3-6 cells. Larger micro conidia were few, cylindrical, 1-2 cells and comparatively smaller. Isolates of fungi were identified as *Fusarium spp* by comparison of their colony characters and morphological features of conidia with published data [1]. Further, it was noticed that re-inoculation of purified *Fusarium* isolates failed to development of bulging symptoms of passion fruit vines. However, it was identified by culturing test that *Fusarium* fungi remained viable in tissues of inoculated sites of vines, indicating that the *Fusarium* fungi survives in stem tissues of the passion fruit vines without showing bulging symptoms.

3.4 The field trial based on fungicide and insecticide

Table 3: Passion Stem bulging incidence five months after field planting

Treatments	Mean Passion Stem Bulging incidence per vine at 5 months after planting
T1	11.1
T2	4.2
T3	13.2
T4	9.5
T5	13.7

Lowest disease incidence was observed in Thiophanate methyl 6g/ 10 l i.e. T-2 treated plots of field trials indicating *Fusarium* species may be the causal factor of the problem. Thiophanate methyl is an approved EU fungicide used widely to control fungal diseases on crops. A systemic fungicide is effective with protective and curative activity against a broad spectrum of diseases in fruits, vegetables and other crops [2]. However, Thiophanate methyl is highly effective on *Fusarium sp.* [3]. Foliar spraying of insecticide application, alternative foliar spraying of insecticide and fungicide and alternative foliar spraying of insecticide, fungicide and Albert solution were failed in controlling stem bulging in the field conditions. However, according to the CABI report No: IMI 504215 & IMI 504216, the isolation made from internal tissues and two fungal cultures were subjected for molecular identification and *Gibberella fujikuroi* and *Fusarium sp.* were identified as associate fungi of stem bulging of passion fruit[4].

The fungus, *Gibberella fujikuroi* produces plant growth hormones called gibberellins. Some strains of the fungus *Gibberella fujikuroi* (perfect stage of *Fusarium moniliforme*) are the industrial source of gibberellic acid [5]. These strains infect rice and cause the disease known as bakanae in Japan. Bakanae disease of rice is caused through infection by this fungus and in rice this causes an exaggerated growth response and the plants to become

extremely tall with a pale spindly appearance [6]. Other hosts are also infected by this fungus although we are unable to find any references concerning passion fruit. The fungus was only isolated from within the bulging areas and therefore we suspect the fungus colonization of the tissues is a localized infection.

IV. CONCLUSION

The isolation made from internal tissues of passion fruit and two fungal cultures were subjected for molecular identification and *Gibberella fujikuroi* and *Fusarium* sp. were identified as associated fungi of stem bulging of passion fruits. Also, these results are compatible with field trails' data. However, Thiophanate methyl 70 WG 6g/ 10 l can be recommended as an effective fungicide to minimize passion fruit stem bulging.

V. ACKNOWLEDGEMENT

Grateful acknowledgements are made to Sri Lanka Council for Agriculture Research Policy for the financial assistance provided for this study.

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