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Short Communication

OCCURANCE OF BUD NECROSIS DISEASE IN MUNG BEAN IN HAMBANTHOTA DISTRICT

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INTRODUCTION

The mung bean or green gram (*Vigna radiata* L.) plays a vital role in the protein deficient rice – based diets of the rural and urban population in Sri Lanka. It can be used as a green manure crop, combined cash and a soil improvement crop with residues incorporated in to soil after pods have been harvested. In Sri Lanka, it is mainly cultivated in the Dry zone regions, where about 80% during *Maha* season under rainfed upland crop and the rest in *Yala* season paddy fields with supplement any irrigation. Mung bean is a short term warm season crop, popular as an intercrop as well as a mixed crop. Due to short age, its ability to grow under low moisture and low fertility condition, it is a popular as a third season crop.

Mung bean suffers from several diseases, especially Cercospora leaf spot (*Cercospora canescens, C. cruenta*), Powdery mildew (*Erysiphe polygony*), root disease complex (*Pythium* spp, *Rhizoctonia solani, Fusarium* spp.) and the reinform (*Rotylenchulus reniform*) and root knot (*Meloidogyne* spp.) nematode. Moreover, mung bean harbours different viruses namely, alfalfa mosaic virus (AMV), bean common mosaic virus (BCMV), cucumber mosaic virus (CMV), leaf crinkle virus (LCrV), leaf curl virus (LCV), mosaic mottle virus (MMV), black gram mottle virus (BGMV) and mung bean yellow mosaic virus (MYMV) (Bashir *et al.* 1991; Aftab *et al.*, 1993; Malik, 1991).

In Sri Lanka, recorded viruses in mungbean cultivation are MYMV, cowpea mosaic virus, BCMV, southern bean mosaic virus, LCV, soybean mosaic virus, LCrV and tobacco ring spot virus (Shivanathan, 1980). In 2012, an unknown disease condition of mung bean was reported during inter seasonal cultivation from Hambanthota area. The recorded initial symptoms were leaf mottling, yellowing, mosaic and necrosis of the terminal bud at around three weeks after planting. Afterwards growth reduction was observed. Infected plants usually bear very low flowers and pods resulting very low yield. Moreover, high population of thrips were also observed in the infected fields. Even though farmers applied different chemicals to manage this situation, it has not been successful. The causal agent for these conditions was unknown and proper management techniques could not be practised. Therefore, this study was carried out to identify the causal agent/s of this problem.

MATERIALS AND METHODS

Mung bean plants showing bud necrosis symptoms (Figure 1) and soil samples from the infected fields in Hambanthota district were collected. Seed samples used for planting were also collected from the farmers in affected areas and tested for seed borne viral infection. For that collected seeds were planted in earthen pots, kept inside an insect proof net house and observed for any symptom development. Those seedlings at cotyledon stage and symptomatic immature leaves of collected plants were tested for several viruses by Enzyme Linked Immunosorbant Assay (ELISA) using commercially available antiserum. Tested viruses were Cucumber mosaic virus (CMV), Poty group virus, Cowpea mosaic virus (CoMV), Southern bean mosaic virus (SBMV), Tomato spotted wilt virus (TSWV) and Ground nut bud necrosis virus (GNBN).

Symptomatic leaf samples of collected mung bean plants were checked for the infection of endophytic pathogens. In this regards, those leaf samples were washed with running tap water for few minutes and cut into 1 cm size pieces. Then those leaf cuttings were sterilized with 75% ethanol for few minutes at three times and then washed with sterilized distilled water. Sterilized cuttings were air dried and placed in Potato Dextrose Agar (PDA) and Nutrient

Agar (NA) plates separately and incubated three days at 28 ^{0}C. Collected soil samples were checked for soil borne pathogens by using serial dilution technique and incubating in PDA and NA medium containing plates. Pathogenecity of isolated fungi and bacteria was done by standard steps of the Koch's postulation.

Figure: 1 Symptoms of mungbean bug necrosis disease.

Mechanical inoculation was carried out to confirm the viral infection. The immature leaves with initial yellowing symptoms of collected plants were macerated in a pestle and mortars by adding 0.1M phosphate buffer, pH 7.0, containing 1% 2-mercatoethanol. The resulting pulp was strained through a muslin cloth. Carborandom powder (400 mesh) was dusted on to first and second trifoliate leaves of the test plant (Cowpea) for making microscopic punctures in epidermis. Then, the inoculation was made by rubbing the surface of leaves with cotton swab dipped in extract (inoculum) gently, uni-directionally. Excess inoculums were washed with water using wash bottle and the inoculated plants were kept in an insect proof net house for symptom development.

RESULTS AND DISCUSSIONS

In the isolation of soil borne pathogens, creamy white colour bacterial colonies were observed. Through gram staining and KOH test, it was confirmed as gram negative bacteria. But in the pathogenicity test, it did not observe any effect of this gram negative bacteria to mung bean plants. In leaf cultured PDA plates, fungi *Colletotrichum* spp, *Curvularia* spp and *Phythium* spp were observed. It also not developed any bud necrosis disease symptoms in mung bean plants in pathogenicity test. Symptomatic leaf samples and collected seeds were tested for five suspected viruses and poty group virus using commercially available antisera (Table 1).

Table: 1. ELISA test results for the tested viruses.

Sample	ELISA values of tested viruses					
	CMV	TSWV	SBMV	CoMV	Poty group	GNBNV
Mung bean leaves -1	-	-	-	-	-	+
Mung bean leaves-2	-	-	-	-	-	+
Mung bean leaves -3	-	-	-	-	-	+
Mung bean leaves-4	-	-	-	-	-	+
Mung bean leaves-5	-	-	-	-	-	+
Mung bean seeds -1	-	-	-	-	-	-
Mung bean seeds -2	-	-	-	-	-	-
Positive control	0.396	0.807	0.579	1.317	0.739	0.968

Note: +=positive; -=negative.

CMV is one of the most abandoned viruses in the world, which has the widest host range including the family Fabaceae. In 1971, Bashir *et al.*, reported that mung bean is a host for CMV. But mung bean bud necrosis diseased samples negatively reacted with monoclonal antiserum for CMV. SBMV, belongs to sobemovirus group, is a seed borne virus, transmitted by beetles. Even though, mung bean act as a host for SBMV, this bud necrosis symptomatic samples gave negative results with SBMV antibodies.

Mung bean also a host of CoMV, a type member of the comovirus group, transmitted by beetles, seed borne in cowpea. Tested mung bean bud necrosis samples showed negative reaction with CoMV antibodies. Mungbean harbours some viruses belongs to poty group. But tested samples gave negative reaction with poty group antiserum.

TSWV and GNBV both are Tospo virus, which are becoming an emerging problem in the agriculture sector in many countries including Sri Lanka (Mandal *et al.*, 2012). Both are transmitted by thrips. Vidana Gamage *et al.* (2014) has confirmed the presence of TSWV in tomato and ground nut in Sri Lanka. In this study, mung bean bud necrosis symptomatic samples showed negative reaction with TSWV antiserum and positive reaction with GNBV antiserum. GNBV is characterized by leaf mottling and necrotic streaks on veins, shortened internodes and necrosis of terminal buds. It is a severe problem in ground nut cultivation in Sri Lanka and mung bean is a host plant.

Symptomatic mung bean bud necrosis leaves mechanically inoculated cowpea (*Vigna anguculata*), which is the diagnostic host plant for Tospo virus, showed both localized as well as systemic infections on cowpea leaves. This observation proved that bud necrosis diseases in mung bean is due to tospo virus. Seeds planted in an insect protected house did not show any virus symptoms throughout the observation period as well as all the ELISA tests gave negative results including GNBV test.

CONCLUSIONS

Causal agent of mung bean bud necrosis problem in inter season mung bean cultivation in Hambanthota district was identified as Ground nut bud necrosis virus. Further, it was not a seed borne virus. This is the first record of ground nut bud necrosis virus in mung bean in Sri Lanka. As it affects the yield severely, it is important to find a suitable management strategy. It is also important to carry out host range studies for ground nut bud necrosis disease in order to find suitable management strategies.

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