P: ISSN No. 0976-8602 RNI No.UPENG/2012/4262 VOL.-8, ISSUE-2 (Part-1) April 2019

E: ISSN No. 2349-9443 Asian Resonance

Mechanical Transmission of Cucumber Mosaic Virus in *Cucurbita pepo* (Pumpkin) in Doon Valley

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Abstract

Field survey was conducted during 2018 to document the occurrence and distribution of virus infecting cucurbits in Doon valley of Uttarakhand. Member of cucurbitaceae family are very susceptible to viral infection. During the field study the plants infected with virus and those showing symptoms were assessed. Virus like CMV, WMV cause severe destruction to these crops. These viruses are transmitted mechanically as well as non-persistently by aphids. Leaf sample from various virus infected pumpkin of different varieties of *C.pepo* were included in earlier growing season (July- Oct.) from different locations of Dehradun. Infectious nature of disease, biological characterization and identification of isolated virus were performed by mechanical inoculation in different varieties of *C.pepo* by rubbing crude sap on healthy leaf of pumpkin seedling under field condition. Most common symptoms of mosaic disease transmitted mechanically shown mosaic symptom like vein bending, chlorosis, curling, vine decline, mottling and blistering. This study would provide the information regarding the occurrence, symptomatology, characterization and transmission of CMV, WMV and its retaining infectivity period in Doon valley of Uttarakhand.

Keywords: *Cucumber mosaic virus(CMV), Watermelon mosaic virus(WMV), Cucurbita pepo, Cultivars*

Introduction

Cucurbita pepo (Pumpkin) is native to central America. This crop is grown globally and is extensively grown in warm and semi tropic regions of world. It is commercially important crop of Dehradun after other three cultivated cucurbits viz. watermelon, pumpkins, cucumber. These vegetable crops are effective fulfill the necessity of local people. The vegetable is highly nutritious and a source of vitamins, minerals and fatty acids. There are many elements that leads to decrease in its quality and quantity. Disease causing viruses is one of the most limiting element that leads to low the production of these vegetables.Major viruses that infect *Cucurbita pepo* are Cucumber Mosaic Virus (CMV), Watermelon mosaic virus (WMV), Cucumber Green Mottle Mosaic Virus(CGMMV). Symptoms produced by these virus may vary from mild to severe and cause significant loss to the crops. These viruses are specify by symptoms viz, Hypertrophy, Vein bending, leaf deformation , yellowing of leaf, reduced leaf size and stunting of plant. CMV is mainly transmitted by mechanical inoculation of the crude sap of the plant. *Aphis gossypii* was considered to cause 100% transmission efficiency.The emerging isolate of WMV-Tr obtained from naturally infected watermelon showing mosaic symptoms .

Objectives of Study

The aim of this work were to determine the identification and confirmation of CMV on the basis of biophysical properties Longevity 'in vitro' (LIV).

Review of Literature

CMV was first reported as a casual agent in cucumber plant (Doolittle 1916). Presently about 60 viruses reported that infect the member of cucurbitaceous family and every year new viruses were also described on same host (Romay et al., 2014). Efficient aphid vectors are also responsible for transmission of CMV in persistent and non- persistent manner. *Aphis gossypii* and *Myzus persicae* are very common species for the transmission of different isolates of virus (Tian et al., 2012).The isolated pathogen transmitted by aphid was confirmed by the methods of biophysical properties, insect transmission and host range. (Chandankar et

al., 2013).The isolates of WMV were characterized on the basis of host range study that produce mosaic symptoms on *C. pepo* and other cucurbit member.(Santosha et; al 2018). Longevity 'invitro' in mild and severe isolates of bottleguard mosaic shows the reduction in percentage infection with increase in temperature (Koranga 2018). The biological properties of the virus is similar to those reported for the WMV isolates from different part of the world.The isolate was efficiently transmitted by *M. persicae* (Sulzer) with an average transmission rate of 95% when aphids were used on each test plant of *C.pepo* cultivar (Kamberoglu et al.,2015).Seeds act as primary source of transmission for Cucumber mosaic virus in peeper. The report suggest that virus may transmitted through infected gamete during fertilization in reproductive tissue like embryo and seed coat (Ali et al., 2010).Various method of diagnosing the virus at advance stage of disease is characterized on basis of biological, serological nature, host range and its transmission through vectors(Raj et al.,2017).The results suggest that increase in temperature of Dehradun may involve change in the infection rate incidence or behavior of CMV.

Material and Methodology

The present research work was carried out at D.B.S PG College Dehradun Department of Botany.

Pathogenicity Assay

During study period samples of leaf infected with virus were collected from the field area in Dehradun. These samples were collected from naturally infected cucurbit plant showing symptoms viz:- Leaf distortion, vein clearing, chlorosis, hypertrophy, vein bending. After collection infected leaves were subjected to lab for their recognition. Inside insect proof glass house stock culture of virus isolate were conserved to prevent them from contamination and insect visit. Seedling of test plant were grown in polybags while seedlings of stock culture were raised in plastic pots of 25cm height. Experiments of sprouted seedling were done in summer (June-July).

Inoculum

From the infected leaves of *Cucurbita pepo* inocula of different viruses isolates were taken. This sap or inocula is used for experiments. Inocula were prepared by plucking infected leaves from plant and with the help of mortar and pestle crush the leaves. The crude sap of infected leaves were extracted by pressing /compressing the pulp through muslin cloth or filter. Sap was poured into watchglass and mix it with silicon carbide. Gently rub the forefinger dipped in crude sap to made inoculation done. After inoculation the leaves of test plant were sprayed with water to remove excess of inoculum and to prevent the test plant from damage/ injury destruction to tissues. Healthy young seedlings at cotyledon stage were selected forinoculation. The filtrate or infective sap was applied on whole foliage gently.

Physical Properties

Biophysical properties of Cucumber mosaic virus in *Cucurbita pepo* were determined by performing experiments in laboratory and Longevity in vitro were directed by standard method givenby(Noordam1973).Trails of experiment was conducted to establish the infectivity of sap.

Longevity 'in vitro'

Two tails of experiment was organized to check the infectivity of crude sap. Longevity in vitro is the time in hours, days, weeks to determine the infectivity of virus that is present in sap kept at room temperature and to check the infectivity rate at which the virus is inactivated. Crude sap is taken viz:- Room temperature.After fixed period of time inoculate the test plant with extracted sap and then kept them in glass house and observe the symptoms periodically.

Results and Discussion

Mechanical Transmission of Virus

The present study reveals the natural occurrence of virus in *C.pepo* under field condition in Dehradun .Sap inoculation in *C.pepo* shows transmission of virus. In the present report the inoculated sap was shown to be transmitted mechanically with the crude sap that was taken in dark at room temperature at an interval of 8hr, 16hr, 24hr, 36hr, 48hr, 60hr, 72hr, 84hr, 96hr, 108hr. It is reported from the results that there is decline in infectivity rate and percent transmission with time period. Variation of time period is taken under consideration. The inoculated plant show same symptoms as found on the grown plant in the field. Data presented in Table1. clears that CMV under investigation retain infectivity for a period of 4 days with 40% transmission rate under room temperature 30ºC-32ºC.

Table1: Longevity 'in vitro' (LIV) of CMV in *C.pepo*

Sr. No.	Testing of samples after extraction	No. of plant inoculated	Percent transmission
1	Control	10	100%
2	8hr	10	80%
3	16hr	10	70%
4	24hr	10	70%
5	36hr	10	60%
6	48hr	10	60%
7	60hr	10	50%
8	72hr	10	40%
9	84hr	10	0%
10	96hr	10	0%
11	108hr	10	0%

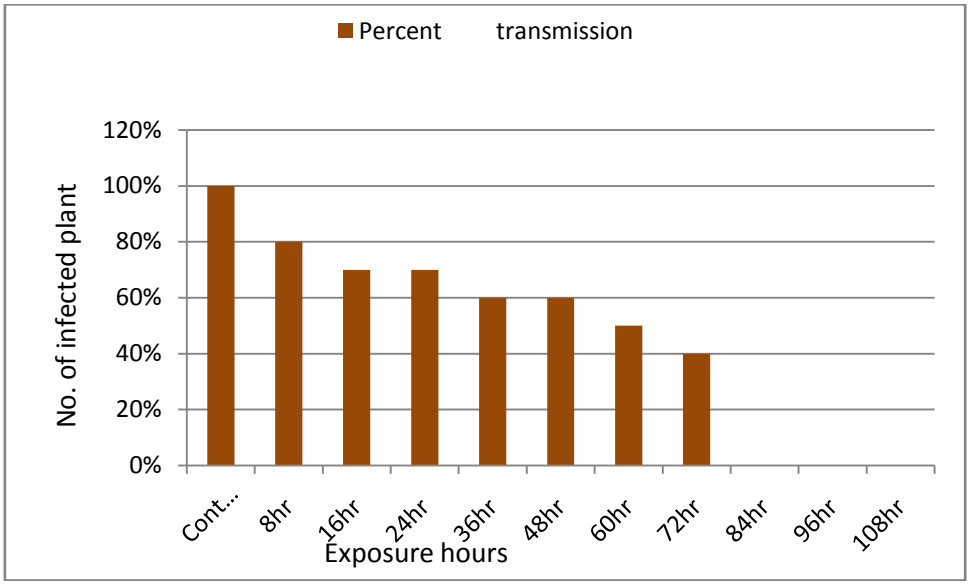

Fig.1 Plot show the percent of transmission of Cucumber Mosaic Virus in *Cucurbita pepo* with interval after extraction

Acknowledgement

The author wish to thanks Dr. Nirmala Koranga Senior Assistant Professor Department of Botany D.B.S (P G) College Dehradun for all their kind assistance. The author also wish to thank the gardener in the surveyed study area for the sanction to access their garden for the collection of samples.

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