

RESEARCH ARTICLE

Impact of *Cauliflower mosaic virus* Infection on Morphological Parameters of Radish Plants

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ABSTRACT

Brassicaceae are among the most economically important crops in the world. Radish (*Raphanus sativus* L.) belongs to the family Brassicaceae, and is an economically important root crop grown worldwide. Brassica vegetables are highly susceptible to viral diseases. *Cauliflower mosaic virus* (CaMV), the type member of the genus *Caulimovirus*, is one of the most severe and destructive viral disease that causes serious damage and economic losses across the members of the Brassicaceae family. The virus systemically infects host plants and produces severe symptoms. Infection with virus reduces radish plant vigour and subsequent root size. Trials were conducted in climate chamber and controlled greenhouse to assess the effect of CaMV infection on growth and yield attributes of radish. Plant-root height (7.6-7.5%), leaf length (19.8%), leaf width (23.4%), stalk length (21.1%), plant-root fresh mass (32.2-8.1%), and plant-root dry mass (38.1-9.5%) were significantly lower in infected plants after eight weeks of virus infection as compared to healthy plants. It has been determined the negative effect of CaMV infection in growth, yield and the morphological attributes of radish in the current study.

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Introduction

Radish (*Raphanus sativus* L.) is one of the well-known root crops that belong to the Brassicaceae family (Poudel and Shrestha, 2018) and has a wide range of nutritious and represents a significant part of dishes (Thiruvengadam et al., 2016). World production of radish roots is estimated at seven million tons per year, about 2% of the total world production of vegetables (Kopta and Pokluda, 2013). However, the productivity of radish is heavily affected by virus diseases including *Radish mosaic virus* (RaMV), *Turnip mosaic virus* (TuMV), *Beet western yellows virus* (BWYV), and *Cauliflower mosaic virus* (CaMV) (Coutts and Jones, 2000; Latham et al., 2003).

Cauliflower mosaic virus (CaMV) is one of the top 10 viruses in plant pathology (Scholthof et al., 2011). CaMV has a double-stranded DNA genome and is the type species of the genus *Caulimovirus* in the family Caulimoviridae (King et al., 2012). CaMV is transmitted by sap inoculation, and in nature by aphids at least 25 species in a semi-persistent manner (Yasaka et al., 2014). CaMV causes severe symptoms in vegetable brassicas and CaMV reduces the yield and quality of brassica crops worldwide (Hunter et al., 2002).

The virus systemically infects young host plants and produces severe symptoms including leaf mosaic, mottling, and vein banding, reduced growth, developmental abnormalities and stunting (Farzadfar et al., 2007; Kalischuk et al., 2015). Saunders et al. (1990) have shown that a broad

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range of *Brassica* species exhibiting a variety of susceptibilities to CaMV infection. Infection with virus reduces radish plant vigour and subsequent root size (Guo et al., 2005).

CaMV was also determined in Brassica vegetables in Turkey (Erkan et al., 1990; Tuzlali and Korkmaz, 2014; Sevik, 2016). The radish is an important root vegetable crop in Samsun, Turkey. This study was carried out under controlled environmental conditions at the Experimental field of Faculty of Agriculture, Ondokuz Mayıs University, Samsun, Turkey in order to determine the effect of CaMV on some morphological parameters of radish.

Materials and Methods

Seeds

Seed materials of *R. sativus* (Brassicaceae family)-*Cherry Belle* (standard, early radish variety) were obtained from commercial sources in Samsun, Turkey. Plants were grown under controlled conditions at 18-24 °C, with supplementary lighting, when necessary, to achieve a 16 h photoperiod.

Inoculation

CaMV isolate was used isolated from cabbage plants in a previous study conducted in Samsun province (Sevik, 2016). The original host of this isolate was tested for the other main brassicaceae viruses. The virus was propagated by inoculating to radish seedlings in a climate room (16 hours light at 24 °C and 8 hours dark at 18 °C). The fresh leaves of infected radish were used as source of virus inoculum in the experiments. Radish plants were mechanically inoculated at the two-four leaf stage with CaMV using celite as an abrasive. Leaves of infected plants which displayed CaMV symptoms were triturated in 0.1 M sodium phosphate buffer pH 7.0, and sap was applied to the leaf surface of radish seedlings. The inoculated plants were kept in the greenhouse and observed for eight weeks. Four replicates (each replicate consisting of 20 plants) were conducted for CaMV and the results were analyzed in the study.

Serological Diagnosis

Detection and presence of CaMV was performed by a double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) using a commercial kit in radish seedlings two weeks after CaMV inoculation. The tests were performed according to the protocol recommended by the producer (Bioreba, Switzerland). Samples were ground in phosphate buffered saline (PBS), pH 7.4 and all conjugates were diluted in PBS-T buffer containing 0.1% Tween 20 (w/v 1:5). Each sample was tested in two duplicates. The absorbance was measured at 405 nm using microplate reader (Tecan Spectra, Salzburg, Austria). The samples were considered as infected when the absorbance was at least three times higher than the mean A405 for negative control (Farzadfar et al., 2007).

Morphological Parameters

The plants were harvested at 60 days post-inoculation (p.i.). The different growth and yield parameters such as plant height (cm), root height (cm), leaf length (cm), leaf width

(cm), stalk length (cm), plant fresh mass (g), plant dry mass (g), root fresh mass (g), root dry mass (g), leaf and root water contents (%) were measured in the plants.

Morpho-agronomic Evaluations

The plant leaf and root height were measured with a millimetric ruler. The weight of biomass and roots of the plants were individually measured on a 0.001 g precision digital balance (Kern). For dry weight measurement, the biomass and roots were dried for 72 hours at 70 °C in a drying oven and weighed again on a precision balance. Leaf water content was calculated via biomass and dry mass values after drying of the leaf sample in the drying oven (Wang et al., 2011).

Statistical analyses were performed using an SPSS software package (Version 21.0, IBM Corp., NY). Plant-root height, leaf length-width, stalk length, plant-root fresh mass and plant-root dry mass values were subjected to variance analysis with SPSS statistical package program.

Results and Discussion

The CaMV-inoculated plants were kept in the greenhouse and observed for eight weeks. Mosaic and local lesions symptoms were observed in radish seedlings two weeks after CaMV inoculation. Vein banding, rugosity, and stunting (Figure 1) were observed on *R. sativus* in the following weeks. The symptoms observed in the current study agreed with those of previous reports (Farzadfar et al., 2007). The leaves were analyzed by DAS-ELISA and all inoculated plants were positive for CaMV.



Figure 1. Vein banding and rugosity symptoms on radish leaves infected with CaMV.

The plants were harvested at 60 days post-inoculation (p.i.). Plant height, root height, leaf length, leaf width, stalk length, plant fresh mass, plant dry mass, root fresh mass, root dry mass, leaf and root water contents in radish plants were examined after CaMV infection was confirmed.

It has been determined that CaMV infection has negative impact on radish morphological parameters in the present study. In radish CaMV-infected, plant height (7.6%), root height (7.5%), leaf length (19.8%), leaf width (23.4%), stalk length (21.1%), plant fresh mass (32.2%), plant dry mass (38.1%), root fresh mass (8.1%), root dry mass (9.5%), leaf (2.9%) and root (0.7%) water contents were reduced when compared with healthy control plants 60 dpi (days post inoculation). There were significant differences ($p < 0.05$) found between the averages.

Plant infection by viruses causes physiological disorders responsible for plant diseases of economic and agronomic

significance in many crops (Nicaise, 2014). Radish is cultivated worldwide and is of agronomic importance. CaMV reduces the yield and quality of brassica crops worldwide (Hunter et al., 2002).

The reduction in some growth and yield parameters in CaMV-infected plants were recorded in the current study. CaMV incidence can easily exceed 70%, and subsequent yields may be reduced up to 20-50% (Sutic et al., 1999). Similarly, screen house experiments conducted in Kenya showed that inoculation of cabbage seedlings with CaMV reduced the number and weight of marketable harvested heads (Spence et al., 2007).

Conclusion

Plant height, root height, leaf length, leaf width, stalk length, plant fresh mass, plant dry mass, root fresh mass, root dry mass, leaf and root water contents of radish plants showed a decreasing with CaMV infection. It has been determined the negative effect of CaMV infection in radish in the current study. These findings may help to understand effective management practices to reduce the negative effect of CaMV in radish. There is need to prevent early infection and loss of crop. CaMV infections can be minimized by several protective measures.

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