Transmission of *Chickpea chlorotic dwarf virus* in Chickpea by the Leafhopper *Orosius albicinctus* (Distant) in Pakistan – Short Communication

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Abstract

Akhtar K.P., Ahmad M., ShahT.M., Atta B.M. (2011): **Transmission of *Chickpea chlorotic dwarf virus* in chickpea by the leafhopper** *Orosius albicinctus* (Distant) **in Pakistan – Short communication**. Plant Protect. Sci., **47**: 1–4.

Chickpea chlorotic dwarf virus (CpCDV, genus *Mastervirus,* family *Geminiviridae*) is the most common viral disease of chickpea in Pakistan. Two aphid [*Aphis craccivora* Koch, *Myzus persicae* (Sulzer)], two leafhopper [*Empoasca devastans* Distant, *Orosius albicinctus* (Distant)] species and an unidentified brown leafhopper were collected in a chickpea field by hand and sweep nets for transmission studies of CpCDV. Transmission results showed that only the leafhopper *O. albicinctus* successfully transmitted the CpCDV from diseased to healthy chickpea plants. The presence of CpCDV in inoculated plants and the vector *O. albicinctus* were confirmed by DAS-ELISA test using specific polyclonal antibodies.

Keywords: chickpea; CpCDV; leafhopper; *Orosius albicinctus*; stunt disease; transmission

The chickpea (*Cicer arietinum* L.) is an important, cool-season grain legume of high nutritive value (Millan *et al*. 2006). More than 50 pathogens including viruses and 54 insect pests have been reported on chickpeas from different parts of the world (Nene 1980; van Rheenen 1991; Singh *et al*. 1994; Kumar *et al.* 2008). Viral diseases often cause significant yield losses (Bos *et al*. 1988; Kumar *et al*., 2008). The chickpea chlorotic dwarf virus (CpCDV, genus *Mastervirus,* family *Geminiviridae*) is commonly found in Pakistan, Iran and Sudan (Horn *et al*. 1995; Makkouk *et al*. 1995, 2001). It has also been documented from India, Egypt, Iraq, Syria, and Yemen (Kumari *et al*. 2006). CpCDV can cause stunting, internode shortening, phloem browning in the collar region and leaf reddening in desi-type while yellowing in kabuli-type chickpea varieties (Nene & Reddy 1987; Nene *et al*. 1991; Horn *et al*. 1993). CpCDV was found to be transmitted by a leafhopper *Orosius orientalis* (Matsumura) in India (Horn *et al*. 1993) and by *O. albicinctus* (Distant) (Cicadellidae: Hemiptera) in Syria (Kumari *et al*., 2004). It nearly caused 100% yield loss of individual plants when infection occurred before flowering and 75–90% losses when infection occurred during flowering (Horn *et al*. 1995). In Pakistan CpCDV is known to have occurred since the 1990s showing 10–40% disease incidence (Makkouk *et al*. 2001; Mughal & Bashir 2007). Despite the prevalence of CpCDV in chickpea in Pakistan since long, its vector was not known. The present study was therefore undertaken to find the vector of CpCDV in Pakistan, which is essential to understand the viral epidemiology and to devise a management strategy for limiting the spread of the disease.

MATERIAL AND METHODS

Different (two aphid and three leafhopper species) species of aphids and leafhoppers were col-

lected by hand and sweep nets from chickpea plants in fields with a high incidence of chickpea stunt disease (CSD) in 2005–2008 at Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan. At first each insect species fed on CpCDV-infected chickpea plants for 3 days for the acquisition of the virus. A batch of ten insects per plant for each insect species was then transferred onto ten 4-week-old healthy chickpea seedlings in isolation cages for an inoculation access period of 5 days. Insects were then killed with Confidor (0.8 ml/l H_2O) and immediately stored at –20°C for ELISA tests. A similar set of chickpea plants was inoculated with insects immediately collected from infected fields. These studies were carried out in a greenhouse where the temperature was maintained between 25°C and 32°C. The exposed chickpea plants were observed for symptom expression and tested for viral infection by DAS-ELISA using the method described by Kumari *et al*. (2006). For the virus detection in insects, a batch of ten insects that had been deep-frozen separately after feeding on test plants was considered as a single unit and the CpCDV presence was detected using the method described by Horn *et al*. (1994). The CpCDV antiserum was provided by Dr. Safaa Kumari, International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria (Kumari *et al.* 2006).

RESULTS AND DISCUSSION

Two aphid (*A. craccivora*, *M. persicae*), two leafhopper (*E. devastans*, *O. albicinctus*) species and an unidentified brown leafhopper were collected in chickpea fields and used as potential vectors for the CpCDV transmission studies. The CpCDV was successfully transmitted from diseased to healthy chickpea plants using the leafhopper *O. albicinctus* (Figure 1). *A. craccivora*, *M. persicae*, *E. devastans* and an unidentified brown leafhopper failed to transmit the virus and to produce CSD symptoms. The rate of the virus transmission was 80% (8 out of the 10 inoculated plants became infected) when chickpea plants were inoculated with *O. albicinctus*, which at first fed on CpCDV-infected chickpea plants for three days (Table 1). When *O. albicinctus* captured in the field were directly released onto healthy chickpea seedlings, 4 out of the 10 inoculated plants (40%) became infected with the virus. CpCDV-transmitted plants showed typical CSD symptoms as reported by other researchers (Nene & Reddy 1987; Nene *et al*. 1991; Horn *et al*. 1993). Out of the 100 plants inoculated with five trapped insects, only 12 plants showed disease symptoms and the *O. albicinctus* collected from these plants showed a strong positive reaction to the antiserum of CpCDV by developing yellow colour and vice versa.

O. albicinctus (thought to be similar to *O. orientalis*) is known as a vector of plant viruses and phytoplasmas. In the present study, CpCDV was successfully transmitted by *O. albicinctus* and these results agree with the previous studies in India by Horn *et al*. (1993) and in Syria by Kumari *et al*. (2004) where the CpCDV in chickpea was reported to be vectored by *O. albicinctus*. However, recently Farzadfar *et al*. (2008) in Iran successfully transmitted the CpCDV using *O. orientalis* to a range of plant species in the *Chenopodiaceae, Fabaceae, Solanaceae* and induced symptoms like

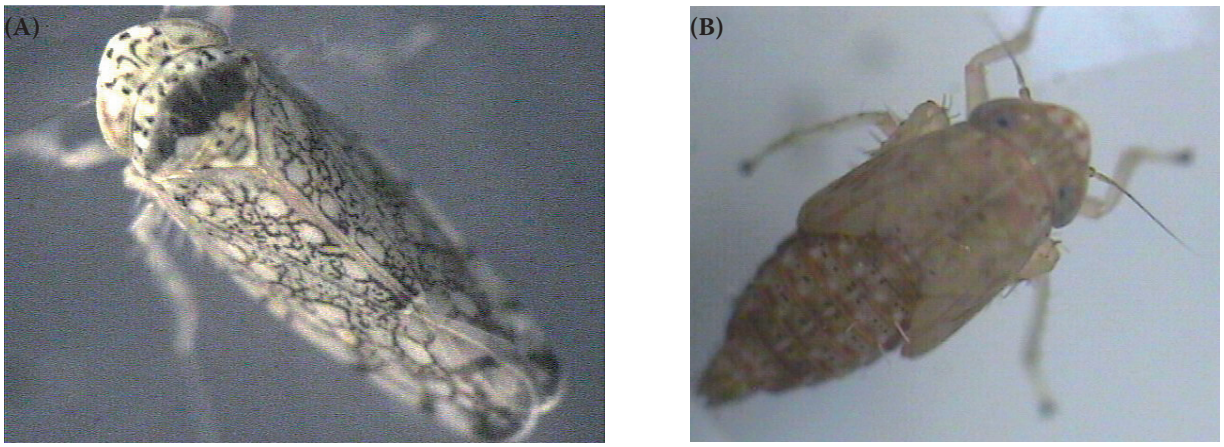

Figure 1. Dorsal view of an adult (A) and a nymph (B) of *Orosius albicinctus*

Table 1. Summary of the insect transmission trials

Insects used for CpCDV transmission		No. of plants used for transmission trials	No. of plants produced disease symptoms and reacted +ve to ELISA	Disease transmission (% age)
Plants exposed to insects after a 3 day AAP on CpCDV infected plants	*A. craccivora*	10	0	0
	M. persicae	10	0	0
	O. albicinctus	10	8	80
	E. devastans	10	0	0
	unidentified brown leafhopper	10	0	0
Plants inoculated with insects immediately after capture from diseased field	*A. craccivora*	10	0	0
	M. persicae	10	0	0
	O. albicinctus	10	4	40
	E. devastans	10	0	0
	unidentified brown leafhopper	10	0	0

AAA = acquisition access period

those reported for CpCDV. *O. albicinctus* was also found to be a natural vector of phytoplasma causing the phyllody disease of chickpea in India and Pakistan (Ghanekar *et al*. 1988; Akhtar *et al*. 2009b) and of sesame in India, Thailand, Upper Volta, Iran, Italy and Pakistan (Schneider *et al*. 1995; Esmailzadeh-Hosseini *et al*. 2007; Sertkaya *et al*. 2007; Akhtar *et al*. 2009a). The present discovery of a natural and experimental vector of CpCDV in Pakistan will provide a valuable lead and experimental tool for the understanding of the disease epidemiology to develop efficient management strategies to minimise yield losses and to control the further spread of the disease.

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