SUMMARY

Eleven samples were randomly collected from cruciferous plants with green or yellow mosaic symptoms in different areas of the Sichuan province (southwest China). The virus recovered from all samples was recognized as a possible potyvirus by electron microscope observations (negative staining and ultrathin sections) and identified as Turnip mosaic virus (TuMV) by RT-PCR. The coat protein (CP) gene of these isolates was cloned and sequenced. Sequence comparisons showed that these isolates shared 97-100% identity among themselves at the nucleotide level, whereas the identity level with TuMV strains isolated from other parts of China was 89-99.2%. Phylogenetic analysis of the CP sequences revealed that Sichuan isolates clustered in the world-B group. The results also showed that TuMV is a common pathogen of various cruciferous crops in the Sichuan province.

Key words: TuMV, coat protein gene, sequence analysis, cruciferous crops.

In recent years, virus infections have emerged as one of the limiting factors to cruciferous crop production in China, where Turnip mosaic virus (TuMV, genus Potyvirus, family Potyviridae) is considered a most important viral pathogen, for it causes great economic losses every year (Cai et al., 2005; Li et al., 2005; Tian et al., 2007; Pallett et al., 2008).

So far, TuMV has been reported from many Chinese provinces, such as Shangdong, Hebei (Song et al., 2005; Liu et al., 2006; Tian et al., 2007), Zhejiang, Hunan, Niaoning and Guangxi (Chen et al., 2002; Tan et al., 2004), but very few records exist from Sichuan, which is one of the most important agricultural areas of the country. A survey conducted in Sichuan in 2006 and 2007 showed that cruciferous crops had a very high incidence of viral infections. This led to the collection of eleven samples from plants showing mosaic or mottling symptoms in different geographic and ecological areas of the province.

Figure 1. A. Negatively stained virus particles. Bar = 250nm. B. Cylindrical inclusions (arrowheads) in the cytoplasm of a parenchyma cells. Bar = 200 nm.
When the sequenced Sichuan TuMV isolates were compared with one another and with those from other parts of China, the results showed that, although some molecular divergence existed, sequence identity at the nucleotide level was higher among them (97 to 100%) than with isolates from other districts. Minor differences were found, as a function of host and area of collection. Thus, four *Brassica juncea* var. *foliosa* isolates (WJ-Q, PX-Q1, PX-Q2, PX-Q3) collected from different regions had nucleotide sequence identities of 97.7% to 100% with the sequenced Sichuan TuMV isolates.
confirming Sanchez et al. (2003) report that TuMV isolates from hosts in the genus Raphanus are less variable than those from plants in the genus Brassica.

The TuMV Sichuan isolates shared 90.7%-99.2% CP nucleotide sequence identity with those from southwest China (Guangxi, Yunnan), 89.1%-99.2% with those from the eastern China (Anhui, Shandong, Zhejiang) and 96.5%-98.3% with those from central China (Hu-nan). A relatively lower sequence identity (89.4%-91.2%) was found with isolates from northern China (Hebei, Niaoning, Beijing). Interestingly, Sichuan isolates shared 97.5%-100% identity with those originating from southwest China, including CHSC2, YJBC, CHZC1, HB12, HJ1S, TAGL and TABC, suggesting that the viral CP gene is largely conserved also under different environments and host selection pressure.

To investigate the phylogenetic relationship of the newly obtained TuMV isolates among themselves and with isolates from other parts of China, a tree was constructed with the CP sequences of 75 Chinese TuMV isolates (Fig. 2) and six isolates from other countries. The tree comprised four major clades, consistent with the previously reported TuMV genetic evolution clusters (Ohshima et al., 2002; Tan et al., 2004; Ali et al., 2004), i.e. basal-B, basal-BR, Asian-BR, and world-B. No Chinese isolates were present in basal-B group, consistent with previous analysis of TuMV Asian isolates (Ohshima et al., 2002; Tomimura et al., 2003, 2004; Tan et al., 2004). A single Brassica napus isolate (TAYC) from eastern China was in the basal-BR cluster (Tian et al., 2007), whereas Chinese isolates were much more numerous in the world-B group, including all those from Sichuan. Four of these (WJ-Q, PZ-LB, WJ-LB, PZ-BC) grouped with isolate CHSC2 originating from Chengdu, the capital city of the Sichuan province. Seven isolates grouped into another sub-clade showing a closer phylogenetic relationship with isolates from Shandong or Zhejiang than with the above mentioned four Sichuan isolates.

In China, viral isolates of the Asian-BR group originated mostly from Raphanus while representatives of world-B group came from a wide range of brassica hosts. Phylogenetic tree showed that clustering of the various isolates was not entirely related with their geographic origin. This is in line with the notion that TuMV isolate variability is complex and is unlikely to depend on a single factor, i.e. host plants, environmental conditions, or geographic distribution.

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