NOTAS CIENTÍFICAS

Prevalence of *Lettuce mosaic virus* - common strain on three lettuce producing areas from São Paulo State

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RESUMO

Firmino, A.C.; Krause-Sakate, R.; Pavan, M.A.; Silva, N.; Hanai, S.M.; Anbo, R.H.; Nietzsche, T, Le Gall, O. Prevalência da estirpe comum de *Lettuce mosaic virus* em três regiões produtoras de alface do estado de São Paulo. ***Summa Phytopathologica,*** v.34, n.2, p.161-163, 2008

O LMV ocorre em todo o mundo e é considerado um dos patógenos mais importantes para a cultura da alface. De acordo com a habilidade em contornar os genes de resistência *mo1*^{1} e *mo1*^{2} encontrados em alface, os isolados de LMV podem ser dividos em dois sub-grupos: LMV-Most, capazes de contornar a resistência propiciada por estes genes e de serem transmitidos pela semente nestas cutivares, e LMV-Common, que não são capazes de causar sintomas nestes cultivares, além de serem transmitidos pela semente somente em cultivares suscetíveis. Para avaliar a ocorrência destes dois tipos de isolados de LMV foram coletadas, durante 2002-2005, amostras de alface com sintomas de mosaico em áreas de produção de alface comercial das regiões de Campinas, Mogi das Cruzes e Bauru no estado de São Paulo. O RNA total foi utilizado para detecção por RT-PCR utilizando-se oligonucleotídeos universais para LMV que amplificam a porção N-terminal variável da capa protéica, localizada no terminal 3´do genoma. As amostras positivas foram analisadas por um segundo primer que amplifica um fragmento da região central (CI-VPg) do genoma viral. Um total de 1362 amostras foram avaliadas, tendo sido detectado o LMV em 504 amostras (37,29%). O LMV-Common prevaleceu em variedades suscetíveis (77,3%). O LMV-Most foi encontrado frequentemente associado a variedades portadoras do gene de tolerância *mo1*^{1}. Apesar da existência dos LMV-Most capazes de contornar a resistência em alface, estes não predominam em nossa condições.

Palavras-chave adicionais: *Potyvirus*, LMV, RT-PCR, detecção

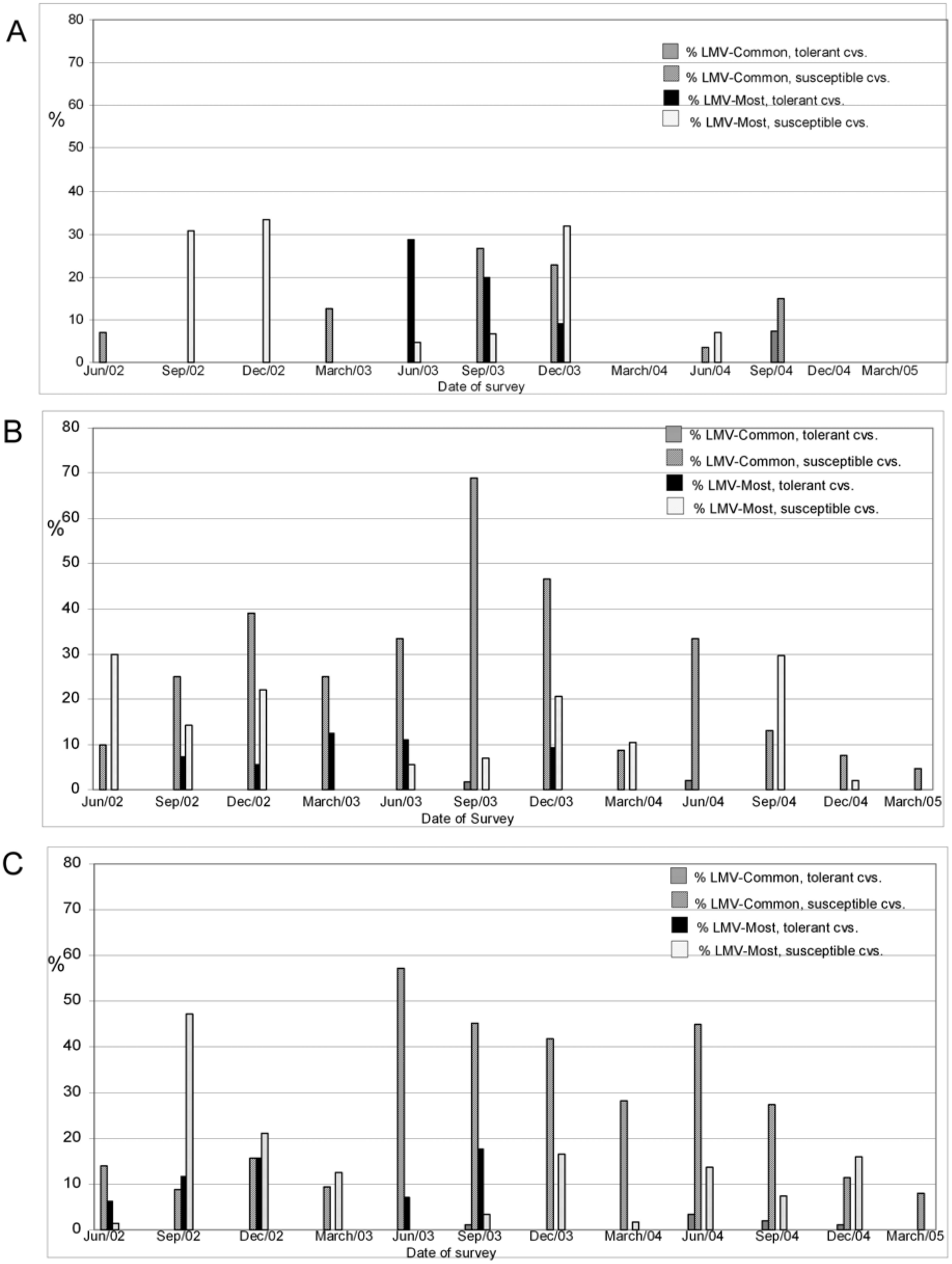
ABSTRACT

Firmino, A.C.; Krause-Sakate, R.; Pavan, M.A.; Silva, N.; Hanai, S.M.; Anbo, R.H.; Nietzsche, T.; Le Gall, O. Prevalence of *Lettuce mosaic virus* - common strain on three lettuce producing areas from São Paulo State. ***Summa Phytopathologica,*** v.34, n.2, p.161-163, 2008

LMV is one of the most important pathogens of lettuce worldwide. Based on their ability to overcome the resistance genes *mo1*^{1} and *mo1*^{2} in lettuce, isolates can be divided in two types: LMV-Most, which can infect and are seed-borne in cultivars containing the *mo1* gene and LMV-Common, which do not cause symptoms on these cultivars and are seed transmitted only in susceptible cultivars. To evaluate the occurrence of these two types of LMV isolates, a survey was carried out during 2002-2005 in three lettuce production areas from São Paulo State. Total RNA was used for the diagnosis of LMV isolates by RT-PCR using universal primers for the variable N-terminus of the capsid protein, in the 3' end of the genome. Positives samples were analyzed by a second RT-PCR using specifics primers for LMV-Most isolates designed to amplify a fragment from the central region (CI-VPg) of the genome. A total of 1362 samples showing mosaic symptoms were collected and 504 (37.29 %) were positives for LMV. On susceptible lettuce cultivars, LMV-Common was prevalent (77.3%). LMV-Most was found frequently associated with tolerant (*mo1*^{1}) lettuce cultivars. Susceptible cultivars correspond today for most of the area of lettuce production. So, despite the ability of LMV-Most isolates to overcome the resistance provided by the recessive *mo1*^{1} gene, they are not prevalent in the conditions of São Paulo State.

Additional keywords: *Potyvirus*, LMV, RT-PCR, detection, resistance breaking

Figure 1: Percentage of LMV-Most and LMV-Common collected in (A) Bauru, (B) Mogi das Cruzes, and (C) Campinas.



Lettuce mosaic virus (LMV) is one of the most important pathogens of lettuce (*Lactuca sativa*) worldwide. LMV belongs to the genus *Potyvirus* within the family *Potyviridae*. The control of lettuce mosaic relies on prophylactic measures such as the elimination of contaminated commercial seed lots and on genetic resistance. Two alleles of the recessive gene *mo1* (*mo1*¹, formerly named *g*, and *mo1*², formerly named *mo*) were introgressed into different lettuce cultivars, conferring either tolerance (systemic virus accumulation but no symptoms) or resistance (no systemic virus accumulation), depending on the virus isolate considered (2). The combined use of genetic resistance and seed sanitary control generally results in proper control of lettuce mosaic in the field. In the last decade however, LMV isolates capable of overcoming the resistance afforded by *mo1* have been described in various parts of the world (8) including Brazil (9). While generally resistance-breaking isolates are not seed-borne, these newly observed LMV isolates combine resistance breaking and efficient seed transmission in resistant hosts (2;8). In a study of the genetic diversity within LMV isolates collected on a worldwide scale (6), such isolates clustered separately, suggesting a monophyletic origin of this group of isolates for which the name LMV-Most (for *mo1*-breaking, seed transmitted) was proposed. Similarly, the name LMV-Common was proposed for another monophyletic group corresponding to the seed-borne isolates that are unable to causes symptoms on *mo1* plants.

Because of their ability to spread in seed lots even in the presence of the two available resistance genes, Most-type isolates are an increasing threat to lettuce production worldwide. A specific test based on RT-PCR was developed by Peypelut et al. (7) for the detection of LMV-Most isolates and was used in this work with the aim to study the occurrence of these isolates in São Paulo lettuce fields. During June 2002 to March 2005, lettuce samples from plants showing mosaic symptoms were collected from the most important lettuce growing areas from São Paulo State (Bauru, Campinas and Mogi das Cruzes). Total RNAs were prepared from infected plants as described by Bertheau et al (1). The broad-spectrum primer pairs 08894p-09171m (11;16) and the Most specific primer pairs 5930p-6544 described in Peypelut *et al*. (7) were used for RT-PCR.

LMV was found all year long in the different regions (Figure 1 A,B,C). During the three years, a total of 1362 lettuce samples were analyzed by RT-PCR and LMV was found on 504 (37.3%) samples. From these samples, 440 plants infected by LMV correspond to susceptible cultivars which are more frequently cultivated in São Paulo State. On susceptible cultivars, LMV-Common was found on 340 samples (77.3%) and LMV-Most on 100 samples (22.7%), indicating that LMV-Most is not prevalent on cultivars lacking resistance genes (Figure 1).

On tolerant commercial cultivars like Gizele (crisphead), Elisa (butterhead) and Vanda (butterhead) carrying the LMV recessive *mo1*^{1} resistance gene, LMV positive samples were not found frequently, but from the positives (64 samples), 86.8% were infected by isolates belonging to the LMV-Most subgroup, and 13.2% by LMV-Common. LMV-Common multiplies on tolerant cultivars but does not normally induce symptoms. The symptoms observed on these plants could be induced by a second virus, explaining the detection of LMV-Common on these cultivars. The negative samples (854) were tested for the presence of Lettuce mottle virus (LeMoV) as described by Jadão et al (3) . Only 94 plants (11.0%) were infected by LeMoV.

The first occurrence of LMV-Most isolates in lettuce fields of São Paulo State was verified in 1992 (9). Their appearance is related either to the previous occurrence of virulent strains that remained undetected until the release of resistant cultivars, or to the recent evolution of common strains into virulent ones once the resistant cultivars were introduced. In our work, LMV-Most were frequently associated with tolerant lettuce cultivars like with the exception of the Bauru region and September 2002 were high occurrence of LMV-Most was also observed on susceptible cultivars (Figure 1A and 1C).

It has been suggested that because of the small size of the genome of plant viruses, their ability to overcome the selection pressure exerted by a resistance gene may have deleterious side effects on the fitness of the virus [Vanderplank (1984) cited by Jenner et al., (5)]. Some examples support this idea: *Turnip mosaic virus* (TuMV) isolate UK 1 belongs to pathotype I and has the ability to overcome the *TuRB01* gene on *Brassica napus,* but are less fit compared to wild type isolates (5).

Here we present indirect evidence that the virulent strain of LMV (LMV-Most) is less fit than the avirulent strain (LMV-Common) in our conditions, where little selection pressure is exerted by the presence of the *mo1*^{1} gene in the natural environment. The study of the aggressiveness of LMV-AF-199 (Most) and LMV-AF198 (Common) on susceptible cultivars reveals that the Most strain reduces drastically the fresh weight, leaf area and chlorophyll content, of the White Boston (susceptible) and Elisa (*mo1*^{1}) cultivars (3). This could be a constraint for the permanence of Most strains in the fields.

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