Journal of Plant Pathology (2019) 101:481–489
https://doi.org/10.1007/s42161-018-00221-2

ORIGINAL ARTICLE

Prevalence and aetiology of *Phytophthora* fruit and stem rot of solanaceous and cucurbitaceous crops in the Pampas region of Argentina

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Published online: 4 January 2019

Abstract
In Argentina, agriculture and horticulture industries account for 7% of the country's employment. Moreover, this country is considered one of the major organic agriculture producers. Horticulture production is widespread all along the country, but the Pampas region is the most vital section because it provides the greatest economic wealth and houses 80% of the population of Argentina. The genus *Phytophthora* includes remarkably destructive pathogens to an extensive diversity of plant species, especially solanaceous and cucurbitaceous crops. *Phytophthora* diseases can affect roots, stems and fruits. Due to the increasing economic importance of this disease in Argentina, effective and feasible management strategies need to be established. The current study aims at describing *Phytophthora* species associated with rots in solanaceous and cucurbitaceous crops in the North-east of Buenos Aires. Field symptoms comprise the aerial parts of the plants in association with the humid conditions that prevail in that region. *Phytophthora capsici* was determined as the main pathogen causing stem and fruit rot of globe squash, eggplant, tomato and pepper. The prevalence was higher in globe squash, followed by eggplant, compared with the other hosts probably because of the structure of the crop and the susceptibility of the commercial cultivars used in the region. All *P. capsici* isolates obtained belonged to the A1 mating type and were sensitive to metalaxyl. The isolates of *P. capsici* tested were pathogenic to all the inoculated fruits, regardless of the host of origin.

Keywords *Phytophthora* · Horticulture · Argentina

Introduction

In Argentina, agriculture and horticulture industries provide over half the foreign exchange and 7% of the country's employment (Ministerio de Agroindustria 2017). Moreover, this country was recently ranked as one of the major organic agriculture producers along with Australia and China, with an area of 4 million hectares (National Centre of Organic Farmers 2017). Horticulture is widespread all along the country, from the Andes to the west, lowland plains in the north, the central Pampas, and the southern region of Patagonia. The Pampas in central Argentina are the largest loessic plain in the southern hemisphere (Soil Survey Staff 1999), and are characterised by a temperate and humid climate and represent the most fertile and densely populated part of the country. The Pampas region is perhaps the most vital section of the country, as it provides the greatest economic wealth and houses 80% of the population of Argentina. In this region, the province of Buenos Aires is the main horticultural production center (Mercado Central de Buenos Aires 2017). Farmers in the province of Buenos Aires, where our project was centered, produce a diversity of crops, such as solanaceous, cucurbitaceous, asteraceous, cruciferous, chenopodiaceous and amarilidaceous species in small acreage (1 to 10 ha). The solanaceous and cucurbitaceous families comprise most of the horticultural crops of economic importance in

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the region; among them: squash, tomato, pepper and eggplant (Mercado Central de Buenos Aires 2017).

The genus *Phytophthora* includes pathogens like *P. palmivora, P. cinnamomi, P. ramorum, P. nicotianae, P. sojae, P. infestans* and *P. capsici* that are remarkably destructive to a wide range of plant species, causing damages to both agricultural and natural ecosystems (Hansen et al. 2012). Some of these *Phytophthora* species, i.e. *P. sojae* and *P. infestans,* have a narrow host range, whereas others, like *P. cinnamomi*, *P. nicotianae* and *P. capsici* have a wide host range (Lamour 2013).

P. capsici is distributed worldwide (Erwin and Ribeiro 1996) and can be a significant limiting factor to horticultural production (Hausbeck and Lamour 2004). It attacks all cucurbits, pepper, tomato and eggplant, and, recently, snap and lima beans (Lamour et al. 2012). Management of *P. capsici* is challenging and is limited by the long term survival of oospores in the soil (Lamour and Hausbeck 2003) by the occurrence of fungicide-resistant populations (Lamour and Hausbeck 2000; Granke et al. 2012) and the lack of commercially acceptable resistant cultivars (Lee et al. 2001; Gevens et al. 2006; Foster and Hausbeck 2010). Since resistant cultivars may perform well in the presence of isolates from a certain region, but not with isolates from another region, breeders need to test their lines and cultivar with isolates from diverse geographic regions within a single location, as it was demonstrated by Foster and Hausbeck (2010).

The variation in virulence among *P. capsici* populations from different host plants has been extensively studied. In Georgia, one the most important horticulture regions of the USA, Yin et al. (2012) assessed the aggressiveness on pepper of a series of 49 *P. capsici* isolates obtained from roots, stems and fruits of solanaceous and cucurbitaceous crops and identified 10 pathotypes. Glosier et al. (2008) and Sun et al. (2008) obtained similar results in China. Additionally, Lee et al. (2001) had previously observed a significant cultivar-isolate interaction, indicating that host specialisation was present in some isolates. Furthermore, in a study carried out on pepper, it was stated that *Phytophthora* root rot, stem and foliar blight must be considered three different disease syndromes, each requiring a single and different gene for the expression of resistance (Sy and Bosland 2005). As symptoms vary considerably according to the host, plant part infected and environmental conditions (Lamour 2013), the behaviour of pathogen populations varies as well. For example, in dry areas, like south-western USA and southern France, infection on tomato and bell or chilli pepper is generally on the roots and crown, while in areas where rainfall is more common all parts of the plant are infected, including fruits. The disease, whose symptoms include root, stem, leaf and fruit rots, is favoured by poor drainage during the wet season (Lamour 2013). Disease progress is very quick under favourable conditions.

Early detection of the disease is often difficult for farmers and local extension workers. Although the disease was first described in Argentina in cultivated peppers (Lindquist 1932), not much was known about the causal agent. The very few studies conducted since then were in the middle-west region of the country (Gobena et al. 2012). The disease was never reported in detail and the pathogen was not conclusively determined. Therefore we decided to determine its prevalence, defined as the proportion of fields affected by the disease. Prevalence is strongly associated with incidence, as it depends on the incidence and pathogen survival. Nevertheless the prevalence is the first approach to understand the magnitude of the problem.

Due to the increasing economic importance of this disease in Argentina, effective and feasible management strategies need to be established. As a starting point of a larger project focused on an integrated management control strategy, the current study aims at describing *Phytophthora* species associated with rots in solanaceous and curbitaceous crops in the North-east of Buenos Aires. The specific objectives of the study were: (i) to analyse the prevalence of the pathogen in a small but important horticultural production region; (ii) to identify taxonomically isolates of this pathogen in this area using classical and molecular methods; and (iii) to perform pathogenicity tests using selected isolates.

Materials and methods

Survey and sampling strategy A survey for the prevalence of *Phytophthora* spp. in globe squash (*Cucurbita maxima* cv. 'Zapallito') eggplant (*Solanum melongena*), tomato (*Solanum lycopersicum*) and pepper (*Capsicum annuum*) was conducted from January to April 2010 and January to April 2011. Three growing regions in the Northeast of Buenos Aires were selected: the localities of Luján, General Rodríguez and Exaltación de La Cruz. This area belongs to the Pampas Region and is located between 34°19′14.05"S, 59°11′58.87"W and 34°41′33.75"S, 58°54′4.25"W. It is characterised by a humid temperate climate, with a historical average precipitation of above 1000 mm/year. In Luján eight farms were sampled, in General Rodriguez 17 and in Exaltación de La Cruz 12. Because of the complexity of the production systems in these horticulture region, a wide diversity of species is grown in small farms (5 ha in average) on lots ranging from 0.01 to 0.2 ha, the lots instead of the farms were used as single sample units in this survey. Along the two years of survey, a total of 112 lots were sampled randomly, 68 in the first year and 44 in the second one collecting infected tissues from fruits, stems, shoots or seedlings. The disease was identified based on the available literature relative to diseases of pepper, tomato, eggplant and cucurbitaceous crops (Pernezny et al. 2003; Jones et al. 1991; Gevens et al. 2008; Zitter et al.

2004; Erwin and Ribeiro 1996). Each year, the percentage of lots with the presence of *Phytophthora* spp. was recorded out of the total number of sampled lots to obtain the prevalence of the pathogen during the survey.

Pathogen isolation and phenotypic characterisation Isolates of *P. capsici* were collected from symptomatic tissues of globe squash, eggplant, tomato and pepper from the commercial production fields of the Northeast of Buenos Aires (Table 1).

Plant tissues were thoroughly washed under running tap water to remove soil residues, and then disinfected with a sodium hypochlorite solution at 1% by immersion for two min, rinsed with distilled water, and dried with filter paper. Small pieces from lesion margins were first plated individually onto potato dextrose agar (PDA, Oxoid), following manufacturers' instructions without the addition of any antibiotic or fungicide and incubated for two to three days at 24 °C in the dark. Hyphal tips of putative *Phytophthora* colonies were transferred to a 33%-V8 agar (V8A), supplemented with $CaCO_3$ in order to adjust the pH to 7, and incubated to establish an axenic culture for storage at 15 °C. All the 311 *Phytophthora* isolates obtained were examined for morphological characteristics, but 46 isolates were arbitrarily selected for specific measurements. Isolates were observed under a microscope, and *P. capsici* was initially identified based on morphological characteristics (Stamps et al. 1990; Erwin and Ribeiro 1996). The length, breadth and pedicel length of 60 arbitrarily chosen sporangia were determined under a microscope.

Mating type of *Phytophthora* isolates was determined by paring with known A1 (CBS 111.334) and A2 (CBS 370.72) reference isolates of *P. capsici* obtained from the Fungal Biodiversity Centre of Utrecht (Netherlands). Sexual structures were produced by crossing each isolate with known A1 and A2 *P. capsici* isolates. Each isolate was grown on V8A, incubated at 24 °C in the dark and examined microscopically after one week to verify heterothallism and to take morphological measurements. All observations were made using an light microscope (Carl Zeiss KF 2 ICS, Germany). The morphology of antheridia and the size of 60 arbitrarily chosen oogonia of each isolate were recorded. For these determinations a camera was adapted to the microscope and the TSView programme was used. Unpaired test isolates were used as negative controls. Pairing between the opposite mating types of tester isolates was used as a positive control.

Metalaxyl sensitivity was determined by placing a 5-mm-diameter agar plug from a one-week-old culture on the center of a Petri dish with V8A amended with 100 ppm of metalaxyl (at 35% *v*/v, suspended in distilled water and added to cooled agar). Controls for every isolate consisted of V8A plates cultured as previously mentioned. Plates were incubated at 24 °C for seven days and colony diameters were measured. Growth of an isolate in an amended medium compared with that in an unamended medium was classified as sensitive (S, < 30% of the control), intermediately sensitive (I, 30 to 90% of the control) and resistant (R, > 90% of the control) (Gevens et al. 2006).

Molecular identification of *Phytophthora* isolates The preliminary taxonomic assignation of these isolates to *P. capsici* based on phenotype characteristics was confirmed by molecular analyses using a nuclear DNA region sequence including the Internal Transcribed Spacer (ITS) 1, 5.8S rRNA gene, ITS2 and a 28S rRNA gene portion.

For DNA extraction, cultures from storage were transferred to fresh V8A broth and grown in the dark at 24 °C for seven days. Mycelium of each isolate was harvested and freeze-dried. High molecular weight DNA was extracted with the protocol of Ristaino et al. (1998) based on the CTAB method. The DNA was solubilised in 25 µl TE and stored at −20 °C for subsequent use.

Amplification of the target gene region was performed in an Eppendorf Master Cyclerep gradient S. Universal primers ITS4 (5'-TCCTCCGCTTATTGATATGC-3′) and ITS5 (5'-GGAAGTAAAAAGTCGTAACAAGG-3′) (White et al. 1990) were used. PCR were carried out in 50 µl volumes

Table 1 Host prevalence for the two years of sampling (2010–2011)

Sample units / Host		Year	*Phytophthora* spp.		Total of sample units by host
			Absence (% absence)	Presence (% presence)	
Host	Globe squash	2010	7 (30%)	16 (70%)	23
		2011	6 (38%)	10 (62%)	16
	Eggplant	2010	10 (44%)	13 (56%)	23
		2011	8 (67%)	4 (33%)	12
	Tomato	2010	13 (76%)	4 (24%)	26
		2011	9 (75%)	3 (25%)	12
	Pepper	2010	3 (60%)	2 (40%)	5
		2011	1 (25%)	3 (75%)	4
Total			57	55	112

containing 10 ng of DNA, 1× PCR buffer (Promega, USA), 0.125 mM of dNTPs mix (Genbiotech, Argentina), 0.4 μM of each primer (Genbiotech, Argentina). Sterile distilled water, and *Taq* DNA polymerase (0.25 U, Promega, USA) were added. PCR products were electrophoresed on 1% agarose gels to verify the amplification of a single fragment of the appropriate length. Amplicons were purified using a commercial kit (Wizard SV Clean-Up System Promega, USA) following manufacturer's instructions. Purified PCR products were resuspended in 10 μl of water and sequenced bidirectionally using primers ITS4 and ITS5, at the Genomics Unit of INTA (National Institute of Agriculture Technology, Buenos Aires, Argentina). Each sequence obtained was compared with sequence data from GenBank by using BLAST-n (Altschul et al. 1990), in order to determine whether they corresponded to *P. capsici* or to other *Phytophthora* species. Multiple sequence alignment was carried out with ClustalW (default parameters) as implemented in Mega 5.0 (Tamura et al. 2011).

Pathogenicity tests Pathogenicity and aggressivity tests were carried out with 14 isolates of *P. capsici* obtained from six globe squashes (cv. 'Angelo'), four eggplants (cv. 'Classic'), two tomatoes (cv. 'Elpida') and two peppers (cv. 'Almuden') provided by local farmers. Three replicates were done for each isolate with each fruit (each replicate consisted of a different fruit). Fruits were detached from the plants at the mature stage immediately before the experiment to avoid the long-term storage conditions of the market. The fruits were selected according to their size and weight to secure standardised conditions (tomato and globe squash: 100 g; eggplant: 200 g; pepper: 150 g; approximately), were first washed in tap water, submerged in a 1% sodium hypochlorite solution for two min, then rinsed with sterile distilled water and dried in a laminar flow hood. Each fruit was labelled. Two delimited regions in opposite faces were selected, inoculated by placing a 5-mm mycelial agar plug that was covered with a 10-mm paper tape. Inoculated fruits were placed in large plastic trays filled with wet filter paper, which were wrapped in nylon bags and incubated for up to seven days in the laboratory. Three replications were done for every isolate. Controls consisted of sterile V8A plugs placed on the fruits. Disease aggressivity was rated after seven days of incubation by measuring two perpendicular diameters of the lesions. Analysis of variance (ANOVA) was conducted to determine the effect of the isolate in every host. The aggressivity of the isolates was compared after seven days post inoculation using Tuckey's and Bonferroni's tests in SPSS v.21.

Tests were carried out with 14 *P. capsici* isolates obtained from six globe squashes, four eggplants, two tomatoes and two peppers. With another experiment, the pathogenicity of selected isolates was tested on 30-day-old seedlings of globe squash, eggplant, tomato and pepper of the above-mentioned cultivars. Three *P. capsici* isolates (obtained from acorn squash, *Cucurbita pepo* cv. 'Medullosa', Pc12; globe squash, Pc13 and eggplant, Pc14) were each inoculated on seedlings using two different methods: (i) millet seed medium (MSM) according to Quesada-Ocampo and Hausbeck (2010); (ii) placing a 5-mm-diameter mycelial agar plugs directly on the crown tissue (V8 AM), both prepared in pots with four replicates each. Four additional control pots were inoculated with sterile mixture of millet seed and other four with sterile V8 agar plugs. These experiments were conducted twice. The pots were irrigated daily and incubated under natural conditions. For both experiments, *Phytophthora* crown and root rot were evaluated every three days post inoculation up to three weeks when plants died, using the following scale: 0 (healthy), 1 (minor wilting), 2 (moderate wilting), 3 (severe wilting) and 4 (plant death) (French 2004).

Results

Disease survey and prevalence study Observation of disease development was as described by Erwin and Ribeiro (1996). The most common symptoms caused by *Phytophthora* on globe squash, eggplant, tomato and peppers were fruit rots consisting of round dark brown areas which, in the case of tomatoes, assumed the appearance of concentric rings (Fig. 1a, b, c, d, e, f, g and h). The rot appeared at any stage of fruit maturity. In particular, more than one lesion was observed on eggplant fruits that remained attached to the plant and mummified. Stem and shoot lesions turned dark brown to black and occurred at any height (Fig. 1b, d and g). Shoot rot was only observed in eggplant (Fig. 1f).

Phytophthora spp. was found in globe squash, eggplant, tomato and pepper. Among the 112 lots sampled in the two years, almost 50% (55 sample units) were affected by *Phytophthora* spp., 47% of which were grown with globe squash; 31% with eggplant; 13% with tomato and only 9% with pepper. The number of samples collected in the survey was the same as the number of lots grown with these species in the area.

When disease prevalence was compared in both years of sampling a negative tendency was observed for globe squash (70% in 2010 and 62% in 2011) and eggplant (56% in 2010 and 33% in 2011). The percentage recorded for tomato was similar in both years (24% in 2010 and 25% in 2011), whereas in pepper a positive tendency (40% in 2010 and 75% in 2011) was observed. The low number of samples collected in the latter case should be taken into account (Table 1). There was a difference regarding the number of sample units between both years (68 records in 2010 and 44 records in 2011) which was partially due to the farmers' moving from one place to another, a predominant characteristic of this horticultural region. Most of them rent the land to produce in unstable conditions. In this sense, we could not sample the same lot or farm both years.

A total of 311 isolates were obtained from diseased fruits, stems, shoots and seedlings of all horticultural crops and examined for morphological characteristics. Three species of *Phytophthora*: *P. capsici*, *P. nicotianae* and *P. drechsleri* were identified based on morphological characteristics. *P. capsici* was the most abundant (233 isolates), followed by *P. nicotianae* (61) and *P. drechsleri* (17). In eggplant the three *Phytophthora* species were identified; two in tomato: *P. nicotianae* and *P. capsici,* and in pepper: *P. capsici* and *P. drechsleri. P. capsici* was the only species identified in globe squash. Other pathogens were also recorded: *Alternaria* spp. in fruits, *Phomopsis* spp., *Colletotrichum* spp. and *Phoma* spp. in fruits and stems; *Verticillium* spp. and *Fusarium* spp. in stems.

Phenotypic characterisation Detailed examination of 46 *P. capsici* colonies out of 311 arbitrarily selected revealed that they were stellate, petaloid and radial on V8A (Fig. 2a). Morphological characteristics included torulose hyphae (Fig. 2b) and sympodial sporangiosphores (Fig. 2c) that were umbrellate in a few isolates (Fig. 2d). Sporangial shapes ranged from obpyriform, limoniform, obovoid to ovoid (Fig. 2e and f). Sporangia were wide (14 μm × 41 μm) and long (23 μm to 58 μm), with a L/B ratio of 1.1–2.0 (Table 2), caducous with pedicels up to 70 μm long; papillate, rarely with two apices and tapered at the bases. No chlamydospores were observed in any of the isolates. Mating tests showed that the isolates were heterothallic, forming amphigynous antheridia and oogonia (Fig. 2g).

Mating type analysis with a series of 71 *Phytophthora* isolates (60 of *P. capsici*, eight of *P. nicotianae*, three of *P. drechsleri*) revealed that those of *P. capsici* and *P. drechsleri* belonged to the A1 mating type; produced oogonia only when plates of each isolate included the reference isolate: A2 MT (CBS 370.72), while *P. nicotianae* presented both mating type in a ratio 7A1/1A2. The same series subsequently analysed for metalaxyl revealed that its addition to the medium significantly reduced growth rates (> 90%) in all the *Phytophthora* isolates tested, regardless of the host of origin of the isolates studied: globe, squash, eggplant, tomato or pepper, which were classified as sensitive.

Molecular identification All *Phytophthora* isolates identified morphologically were verified by PCR and sequence analysis of the ITS region (see Materials and Methods) which yielded amplification products, ranging from 750 to 850 bp for all isolates. BLAST analyses confirmed with high score values and 98–99% identity (comparing with different sequences from GenBank) that 46 isolates were *P. capsici*, eight were *P. nicotianae* and three were *P. drechsleri*, in accordance with morphological characterisation. Subsequently, the multiple alignment analysis using the MEGA program allowed the identification of 505 conserved sites and 158 variable sites, most of which were localised in the ITS2 segment.

Pathogenicity test At the second day post inoculation, 100% of the 14 *P. capsici* isolates evaluated produced infection on the fruits of every host, while controls remained healthy. There were differences in aggresivity among the isolates inoculated in each host ($p < 0.05$) seven days post inoculation.

The three *P. capsici* isolates inoculated in seedlings with both methods were pathogenic to all the hosts, except for eggplant where no symptoms were expressed in the four weeks of trial. The percentage of mortality observed after

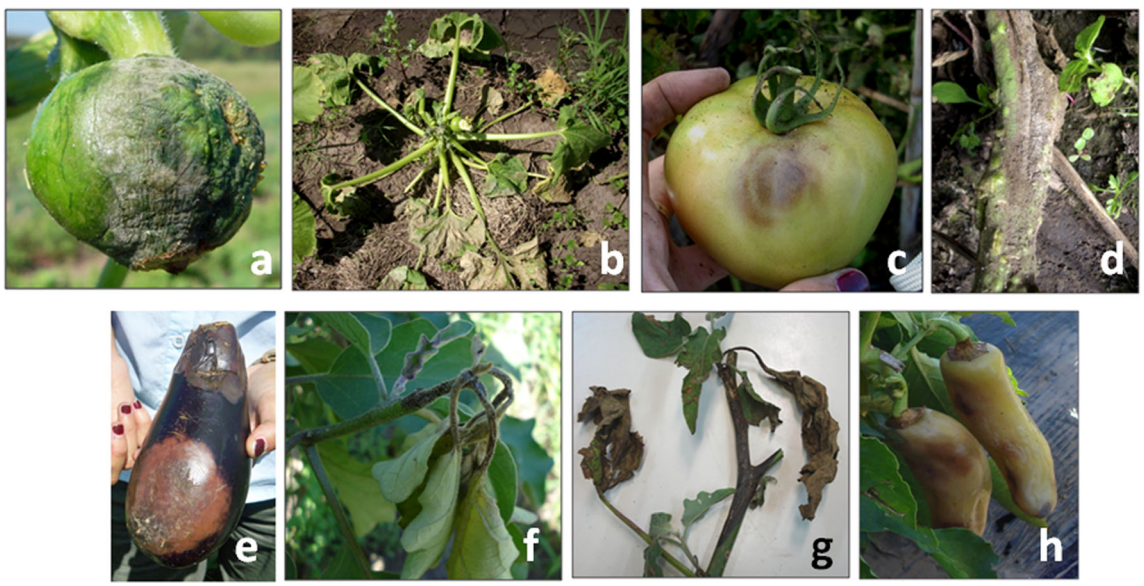

Fig. 1 Symptoms of *Phytophthora* fruit, stem and shoot rot of globe squash, tomato, eggplant and pepper in Buenos Aires: (**a**) aqueous lesion on fruit of globe squash; (**b**) plant of globe squash blight; (**c**) fruit lesion of tomato fruit; (**d**) stem lesion of tomato; (**e**) fruit lesions on eggplant; (**f**) shoot rot of eggplant; (**g**) stem lesion of eggplant; and (**h**) fruit rot of pepper

inoculation of host plants with a selection of *P. capsici* isolates under the V8 AM method ranged from 0% to 100%. The reaction of plants varied with the inoculated isolate. The isolate from acorn squash (Pc12) was the only one that produced mortality in globe squash and the greatest wilting severity in tomato and pepper (0.75 and 0.50, respectively) (Table 3). The MSM method was not consistent among the four replicates so these results were discarded.

Discussion

Since the first identification of *P. capsici* in pepper in Buenos Aires (Lindquist 1932), many reports have addressed diverse pathosystems in a vast area of Argentina (Nome et al. 2017). However, this is the first report from an area systematically surveyed for prevalence. The present results provide evidence that *Phytophthora* fruit and stem rot is widely distributed in Buenos Aires, wherever solanaceous and cucurbitaceous crops are grown. The identification of *P. capsici* in tomato, eggplant and globe squash is the first report for these hosts in the Pampas region.

The prevalence was higher for globe squash compared with the other hosts and followed by eggplant; probably in association to the structure of the crop (the whole plant is in straight contact with the ground compared to the solanaceous crops and that may help soilborne pathogens to infect the plant and the susceptibility of the commercial cultivars used in the

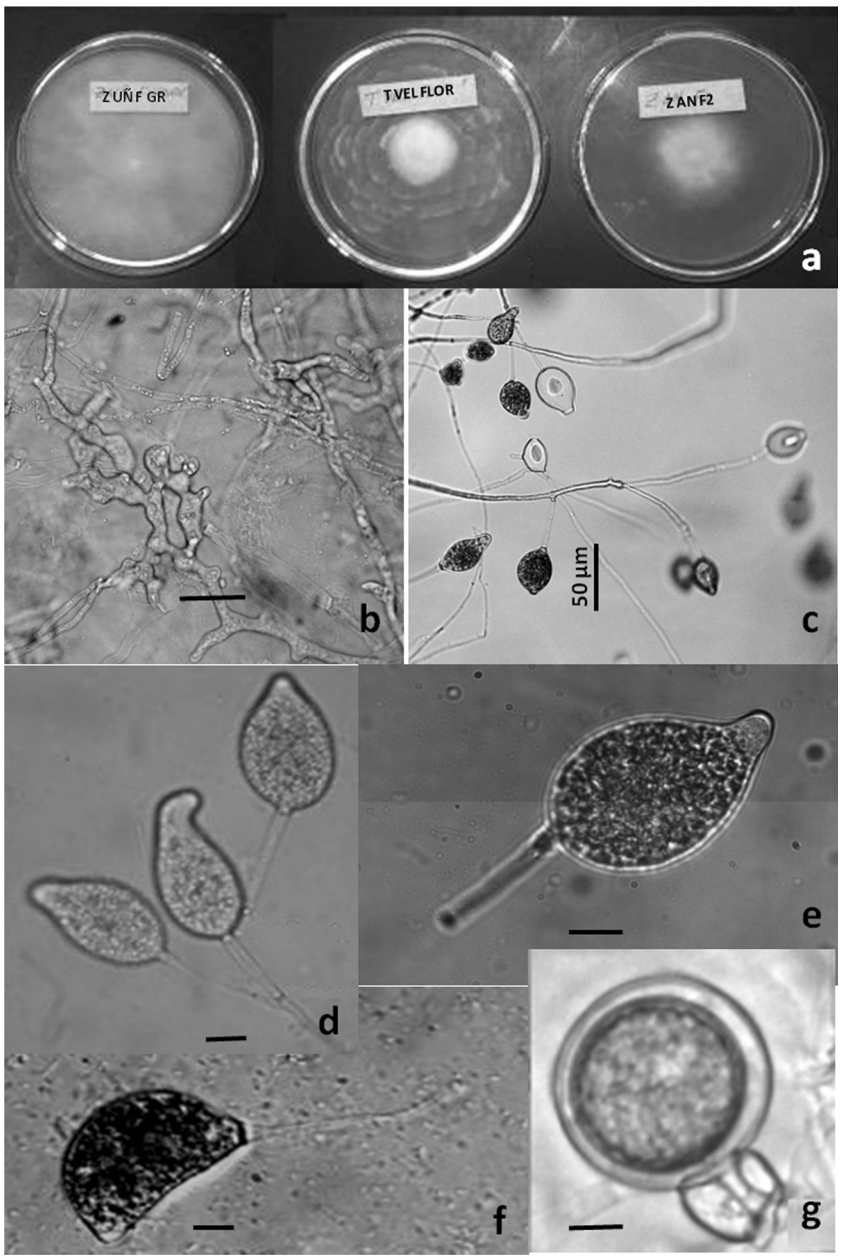

Fig. 2 Morphology of *P. capsici* isolates from globe squash, eggplant, tomato and pepper: (**a**) colonies on V8A, the initial in the label refers to the crop of origin, Z for globe squash or "Zapallito" in Spanish and T for tomato; (**b**) torulose hyphae; (**c**) simpodial sporangiophore; (**d**) umbrellate sporangiophore; (**e**) obovoid pedicelled sporangium; (**f**) sporangium with long pedicel; and (**g**) amphigynous antheridium and oogonium. Scale bar = 10 µm, except where otherwise stated

region. On the other hand, the lowest prevalence recorded for tomato along the two years is in line with the availability of certain cultivars tolerant to *Phytophthora* grown in the region.

P. capsici was the predominant species in all the sampled hosts, except for tomato, and the only species identified in globe squash. In the case of tomato, the predominant species was *P. nicotianae*. These results are consistent with the pathogenicity reports for solanaceous and cucurbitaceous crops worldwide. For instance, *P. capsici* was reported as the main pathogen of globe squash by Zitter et al. (2004) and Koike et al. (2007). On the other hand, four species, i.e. *P. capsici*, *P. nicotianae*, *P. drechsleri* and *P. cryptogea* along with others are reported in eggplant (Erwin and Ribeiro 1996; Gevens et al. 2008) and in tomato, as well as *P. infestans* (Jones et al. 1991; Koike et al. 2007), whereas only *P. capsici* and *P. nicotianae* were reported from pepper (Pernezny et al. 2003; Koike et al. 2007).

Field symptoms comprised the aerial parts of the plants likely due to the humid conditions prevailing in the Pampas, whereas in western Argentina, a region characterised by an arid climate, Gobena et al. (2012) observed symptoms in the roots of peppers. Likewise, Lamour (2013) has shown the influence of climatic conditions on the symptoms expressed on chilli pepper (*Capsicum annuum*) crop, reporting that under the semi-arid and arid conditions of the South-western USA, below-ground symptoms are more common. However, symptoms on leaves, stems and fruit are found in production systems using sprinkler irrigation or following summer monsoonal rain. This is due to the dispersal of soil inoculum and increased soil moisture during overhead irrigation and rain events. Thus, in irrigated commercial fields, affected plants are generally found in row-bound patterns. This behaviour depends also on the dispersal mechanisms of the pathogen (Ristaino and Johnston 1999).

Testing of the North-east Buenos Aires *P. capsici* isolates for mating type and metalaxyl sensitivity revealed only the A1 mating type and metalaxyl sensitiveness in all isolates just like in a previous study on peppers from the western region of Argentina carried out by Gobena et al. (2012). These authors pointed out that the limited genetic variation found in pathogen populations on pepper in the Central-west of Argentina could allow disease resistance remaining viable for extended periods. However, there is still insufficient information on the pathogen diversification in Argentina, considering the diversity of climates where *P. capsici* is present. Studies are needed to test and evaluate different disease management practices to control the pathogen. Currently, the management of the stem and fruit rot is carried out without much knowledge of the epidemiology of the causal organism. Methalaxyl is used in different commercial forms along with other fungicides for general control, without a precise knowledge of the disease to be controlled. In this sense, instead of breeding for

Table 3 Mortality percentage and wilting intensity observed on globe squash, eggplant, tomato and pepper cultivars three weeks postinoculation with three isolates of *P. capsici* (Pc12, Pc13 and Pc14) with the V8 AM method

Fruit – isolate inoculated	Mortality percentage	Wilting intensity[a]
Eggplant - Pc12	0	0
Eggplant - Pc13	0	0
Eggplant - Pc14	0	0
Globe squash - Pc12	100	4
Globe squash - Pc13	0	0.75 (0–3)
Globe squash - Pc14	0	0.75 (0–3)
Tomato - Pc12	0	0.75 (0–3)
Tomato - Pc13	0	0.25 (0–1)
Tomato - Pc14	0	0.25 (0–1)
Pepper - Pc12	0	0.5 (0–2)
Pepper - Pc13	0	0.25 (0–1)
Pepper - Pc14	0	0.25 (0–1)
Control	0	0

[a] Average of 4 repetitions and extreme values between parentheses: intensity scale between 0 (health plant) and 4 (dead plant)

Table 2 Morphological characteristics of 46 isolates of *Phytophthora capsici* isolated from globe squash, eggplant, tomato and pepper

Number of isolates/host	Breadth (μm)		Sporangium length (μm)		L/B ratio		Pedicel length (μm)		Oogonium diameter (μm)	
N	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Globe Squash										
23	25.3	3.7	39.0	6.8	1.6	0.3	35.7	21.2	30.9	4.2
Eggplant										
12	29.5	6.1	44.3	14.3	1.6	0.4	43.3	21.1	31.6	2.0
Tomato										
8	23.4	3.0	31.9	4.3	1.4	0.2	33.1	21.9	31.7	3.6
Pepper										
3	28.6	4.4	47.7	9.7	1.7	0.3	44.1	25.1	32.4	3.1

Values are presented as means, with the Standard Deviation (SD)

resistance to one disease, breeders must now screen all the interactions plant-pathogen in breeding programmes (Foster and Hausbeck 2010).

The isolates of *P. capsici* obtained from globe squash, eggplant, tomato and pepper were pathogenic to all inoculated fruits. The isolates inoculated to seedlings had a different performance; no symptoms were expressed by eggplant seedlings.

Based on these preliminary results, eggplant appears to be a potential source of resistance for root and crown rot. Fruit rot was the primary symptom observed in the fields under study for all the hosts. Other reports have indicated that fruits are more susceptible to *P. capsici* than roots and crowns of pepper (Foster and Hausbeck 2010). Gevens et al. (2008) have also reported that fruit rot is the primary symptom caused by *P. capsici* in eggplant. The differences in virulence among the 14 isolates obtained from the four hosts in the region are consistent with previous results highlighting the range of virulence of *P. capsici* isolates from different hosts and geographical locations (Granke et al. 2012; Foster et al. 2013).

Pathogenicity trials and field observations showed no differences in the symptoms expressed on fruits of each of the four host plants sampled. These observations along with the lack of correlation of virulence with the origin of the isolate are markers of the variability present in the pathogen population (Tamietti and Valentino 2001). The lack of pathogenic specialisation in *P. capsici* has also been confirmed by Isakeit (2007) with isolates from pumpkin that were inoculated to chilli pepper in Texas, by Glosier et al. (2008) in California and by Yin et al. (2012) in Georgia.

The information presented here sheds light on the idea that securing information from diverse geographic regions within a single state is a necessity, considering the wide range of virulence among the isolates of this narrow geographical area under study and the reports worldwide (Glosier et al. 2008; Quesada-Ocampo and Hausbeck 2010; Quesada-Ocampo et al. 2011; Foster et al. 2013). Further research is also warranted to test the virulence of these *P. capsici* isolates to other alternative hosts that will provide more information for understanding the epidemiology the pathogen.

Acknowledgements This research was supported by Conicet (www.conicet.gob.ar) and the National University of Luján (UNLu). We thank Claudia Ojeda (Central laboratory, UNLu) for her valuable contribution in the isolates processing and maintaining for long term survival.

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