ORIGINAL PAPER

Relative incidence of cucurbit viruses and relationship with bio-meteorological variables

Elizabeth Alicia Pozzi[1] · Cecilia Bruno[1,2] · Cecilia Elizabeth Luciani[1,3] · Marcos Giovani Celli[1] · Vilma Cecilia Conci[1,3] · María Cecilia Perotto[1,3]

Abstract

Environmental heterogeneity can shape the plant-virus relationship, furthering the appearance of new diseases in crops, or altering disease incidence and severity. In this work, we studied the virus association with environmental heterogeneity taking into account bio-meteorological variables and virus distribution. In Argentina, three potyviruses, *Watermelon mosaic virus* (WMV), *Zucchini yellow mosaic virus* (ZYMV), and *Papaya ringspot virus* (PRSV), and a cucumovirus, *Cucumber mosaic virus* (CMV), are the most common viruses infecting cucurbits. We identified the bio-meteorological variables that had the greatest impact on disease incidence. All the crops were infected with more than one virus in all the studied environments. Molecular and serological analyses showed the highest relative incidence for WMV (46%), the lowest for CMV (9%), and intermediate values for PRSV and ZYMV (20% and 24%, respectively). Infected plants had a random distribution. Viruses and bio-meteorological variables were highly correlated, with high support (Pearson's r = 0.96, $P < 0.001$). Temperature variables were negatively correlated with the three potyviruses. CMV was positively correlated with temperature. Relative humidity was positively correlated with potyviruses, but was not correlated with CMV. Our research field data and analyses may help to predict the outbreak of viral diseases in cucurbits crops.

Keywords Bio-meteorological variables · Viral disease · Plant-virus interactions

Introduction

Understanding the relationship between plant-viral diseases and environmental heterogeneity is important to prevent the emergence of new viruses as well as the increase of existing virusesincidence and severity. Environmental heterogeneity, defined by biotic and abiotic factors, affects infection patterns, resulting in changes in demographic processes in the virus population (Archie et al. 2009; Biek and Real 2010; Meentemeyer et al. 2012; Fraile et al. 2017).

The family *Cucurbitaceae* includes many important food species, such as watermelon, melon, pumpkin, squash, butter-nut and zucchini (Bisognin 2002). Viral diseases, especially those spread by aphids, are considered the main limiting factors for cucurbits. In Argentina, viral diseases produce yield losses and unprofitable harvests in some years (Perotto et al. 2016). This situation was also reported in other parts of the world (Abou-Jawdah et al. 2000; Coutts et al. 2011). In Argentina, cucurbits are grown from seeds of national origin in 75% of the plantings. So far there are no virus-tolerant/resistant cultivars (Della Gaspera et al. 2013), and seed exchange among producers is very common.

Potyviruses, including *Watermelon mosaic virus* (WMV), *Papaya ringspot virus* (PRSV) and *Zucchini yellow mosaic virus* (ZYMV), and the *Cucumovirus*, *Cucumber mosaic virus* (CMV), are the most common and prevalent virus affecting cucurbits in Argentina (Feldman and Gracia 1992; Gracia and Feldman 1986; Nome et al. 1974; Gracia 2000; Perotto et al. 2016). Moreover, these viruses are present in most cucurbit-

✉ María Cecilia Perotto
perotto.cecilia@inta.gob.ar; mcecilia.perotto@gmail.com

1 Unidad de Fitopatología y Modelización Agrícola (UFyMA), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Córdoba, Argentina

2 Estadística y Biometría, Facultad de Ciencias Agropecuarias, Universidad Nacional de Córdoba, Ing. Agr. Félix Aldo Marrone 746. Ciudad Universitaria, Córdoba, Argentina

3 Instituto Nacional de Tecnología Agropecuaria (INTA), Av. 11 de septiembre 4755 (X5020ICA), Córdoba, Argentina

growing areas worldwide; all of them are transmitted by aphids in a non-persistent manner and cause severe epidemics. Numerous epidemiological studies have been conducted to determine the distribution of these diseases (Alonso-Prados et al. 2003; Ali et al. 2012; Ibaba et al. 2015; Lecoq and Katis 2014; Moreira et al. 2014).

In this work we studied the cucurbit-virus association with environmental heterogeneity, using bio-meteorological variables and virus distribution. We identified the bio-meteorological variables that had the greatest impact on disease incidence. The results of the statistical analyses highlight the importance of considering the numerous bio-meteorological variables involved in the development of viral diseases.

Materials and methods

Study areas

The study was conducted at sites in the main commercial cucurbit areas in Argentina (Fig. 1). We used a Köppen climate updated classification to describe the ecosystems of the study area (Kottek et al. 2006; Reisen 2010). We considered 4 climate types: hot arid climate (BSh), cold arid climate (BWk), temperate dry winter hot summer (Cwa) and temperate without a dry season hot summer (Cfa) (Kottek et al. 2006).

Samples were collected during spring-summer (September–March) 2010–2016. An average crop cycle (from planting to harvest) was 120 days. We combined climate types with crop cycles and obtained 14 environments. The bio-meteorological variables for each environment were recorded throughout the crop cycle. The following variables were included: Minimum Temperature (Tmmin), Average Temperature (Tmmd), Maximum Temperature (Tmmax), Relative Humidity (RH), and Accumulated Precipitation (PPacum). We got the information from INTA Water and Climate Institute (Table 1).

Data collection

Samples were collected from all of the 14 environments. The size of each sampled plot for each environment was different. According to the size of the plot, following a diagonal line, 10 to 50 samples were taken; in total 491 samples of squash and melon leaves were collected. We identified the samples (with and without symptoms) and processed them in laboratory.

Identification of viruses and estimation of relative incidence and relative dispersion index

The presence of each virus was determined by a double antibody sandwich (DAS)-ELISA using commercial antisera specific for the detection of WMV, ZYMV, PRSV and CMV (BIOREBA SRL Latin America, Mar del Plata, Argentina), following the manufacturers' instructions. In addition, molecular analyses were performed to confirm serological results. To this end, specific primers were designed (Table 2) that allowed the amplification of genomic fragments of the expected sizes by reverse transcription - polymerase chain reaction (RT-PCR). 323 samples were positive (at least to one virus); The relative incidence (RI) was estimated for each virus and each environment using those 323 positive samples.

$$RI\ (\%) = \frac{n}{N} \times 100,$$

where n is the number of positives samples for each virus and N is the total number of positive samples.

The Relative Dispersion Index (RDI) was estimated to study the dispersion of viruses in each environment, based

Fig. 1 Geographical location of the sampled environments (black dots) in the main commercial cucurbit areas. Location of the studied climate types: hot arid climate (BSh), cold arid climate (BWk), temperate dry winter hot summer (Cwa), and temperate without dry season hot summer (Cfa)
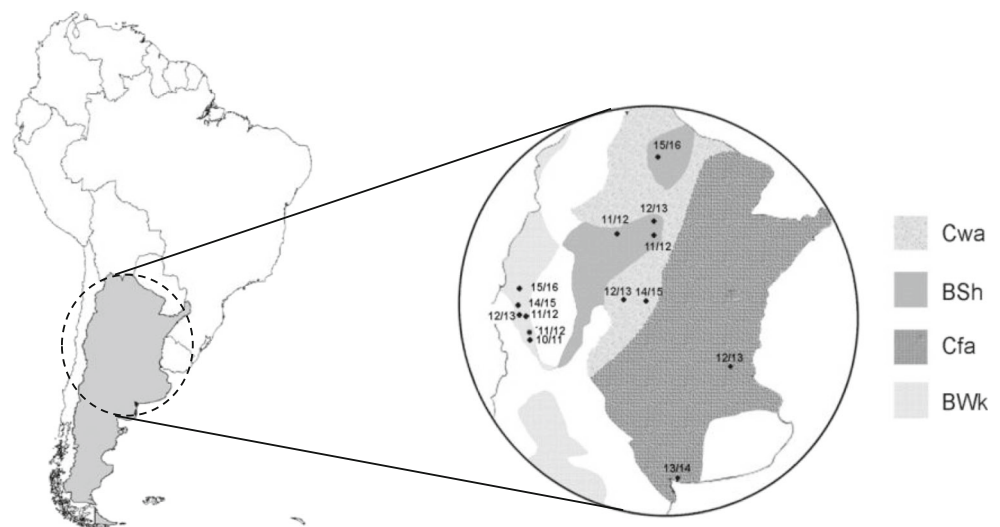
Table 1 Average values of meteorological variables measured during of the crop cycle (120 days) in 14 environments from 2010 to 2016

Environments		Bio-meteorological variables				
Climatic type	Crop cycle	T_{mmax}	T_{mmd}	T_{mmin}	PP_{acum}	RH
Cfa	12/13	28.6	22.7	16.8	477.1	59.3
Cfa	13/14	30.8	23.0	15.1	143.4	49.6
Cwa	12/13	28.6	21.6	15.5	373.2	63.2
Cwa	14/15	28.6	22.1	15.6	767.1	71.1
BWk	10/11	28.9	20.6	12.2	105.3	44.9
BWk	14/15	29.6	21.2	12.8	148.8	54.0
BWk	11/12	34.0	26.5	19.0	69.0	43.8
BWk	12/13	32.9	25.6	18.3	87.5	47.3
BWk'	14/15	32.7	25.7	18.7	35.9	48.2
BWk	15/16	32.3	25.6	18.8	51.8	54.7
BSh	11/12	33.9	26.6	19.3	412.8	62.8
BSh'	11/12	32.2	25.6	19.1	287.4	62.8
BSh	12/13	34.4	27.2	20.0	143.8	61.8
BSh	15/16	31.4	25.7	20.0	523.8	69.3

Tmmin Average Minimum Temperature, *Tmmd* Mean Average Temperature, *Tmmax* Average Maximum Temperature, *HR* Average Relative Humidity, *PPacum* Accumulated Precipitation produced during the crop cycle in the evaluated environments

on the concept of Moreira et al. (2014). The RDI estimates dispersion by comparing the variance of the observed incidence relative to the expected variance. RID values equal to 1.0 indicate random distribution, RID values >1.0 indicate aggregation, and those lower than 1.0 indicate regularity.

Statistical analysis

Samples of melon and squash were analyzed for the presence of studied viruses using a Multiple Correspondence Analysis (MCA) of categorical data.

A frequency analysis of each virus was performed for cucurbit crops in each environment.

A Principal Component Analysis (PCA) was performed to study the variability of environments and to correlate the bio-meteorological variables with relative incidence of each virus. Furthermore, the biplot allows the visualisation of the behaviour of the environments, the virus relative incidence and the bio-meteorological variables at the same time (Gabriel 1971).

A Canonical Correlations Analysis (CCA) was performed to analyze the relationship between the virus and the bio-meteorological variables by calculating unequal combinations and determine the correlation between the two sets of variables.

Finally, an algorithm with low prediction error Regression Trees (CART algorithm) was used to identify the bio-meteorological variables that had the greatest impact on disease presence. The thresholds of bio-meteorological variables that favored the increase of the presence of each virus were also established. Each virus was studied separately.

All the statistical analyses were performed with InfoStat software (Di Rienzo et al. 2018).

Results

All four viruses WMV, ZYMV, PRSV and CMV in the collected samples were found. For subsequent analyses, the data sets of melon and squash were combine. This was possible because the MCA showed that the presence of these viruses was not dependent on the host (data not shown).

Molecular and serological analysis showed that WMV had the highest RI (46%), CMV had the lowest RI (9%), and PRSV and ZYMV had intermediate RI values (20% and 24%, respectively) (Fig. 2).

All the crops were infected with more than one virus in all the studied environments. WMV was present in all the analysed environments with an incidence higher than 50%, except for BSh'11/12. ZYMV and PRSV were detected in 11 of 14 environments, with incidence ranging from 12.5 to 100%. CMV was in 7 of 14 environments, all of them being arid climates (BSh and BWk). The four viruses together were

Table 2 Primers used for the detection of WMV, PRSV, ZYMV and CMV

Primer	Sequence	Length	Tm (°C)	Reference
WCPr	GCACTAGTCGACCCGAAATGCTAACTG	950	61	Mohammed et al. (2014)
WCPd	GACCTAGTGAYGGKTGCTGTGAATCAG			
PRSVCP326	TCGTGCCACTCAATCACAAT	475	57	Mohammed et al. (2012)
PRSVCP800	GTTACTGACACTGCCGTCCA			
ZYMVCP117	TGTCACGAAGGACAAGGATG	549	57	This study
ZYMVCP665	TCMGGAGTTTTGGAATTGAC			
CMV5CP	CTCGAATTCGGATCCGCTTCTCCGCGAG	870		Anonymous
CMV3CP	GGCGAATTCGAGCTCGCCGTTAAGCTGGATGG AC			(1998)

WMV Watermelon mosaic virus, *PRSV* Papaya ringspot virus, *ZYMV* Zucchini yellow mosaic virus, *CMV* Cucumber mosaic virus
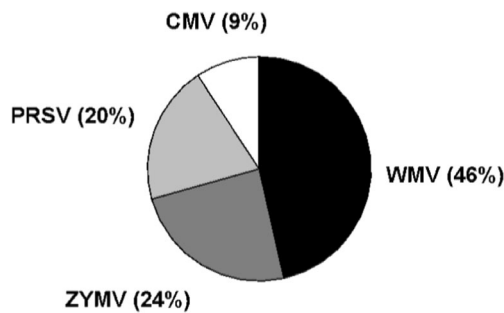
Fig. 2 Average relative incidence of the viruses *Watermelon mosaic virus* (WMV), *Zucchini yellow mosaic virus* (ZYMV), *Papaya ringspot virus* (PRSV) and *Cucumber mosaic virus* (CMV)

in six environments, all of them with arid climates (BSh and BWk) (Fig. 3).

RDI was estimated considering only infected plants. The distribution of the infected plants was random because the RDI was equal to one.. This meant that the observed incidence was the same as expected.

The PCA biplot (Fig. 4) shows that 68.7% of the total variability was explained by the first two principal components (PCs). The first PC differentiates BSh´11/12, BWk11/12, BWk12/13, BWk14/15 and BWk15/16 environments from Cfa12/13, Cwa12/13, Cfa13/14 and Cwa14/15 environments. A high positive correlation was observed between CMV and temperature variables (Tmmd, Tmmin and Tmmax). ZYMV and PRSV showed a high correlation with PPacum and RH. There was also a negative correlation between potyviruses (WMV, ZYMV and PRSV) and temperature variables.

The CCA scatter plot showed a high correlation between viruses and bio-meteorological variables (Fig. 5), with high support (Pearson's r = 0.96, $P < 0.001$).

Temperature variables were negatively correlated with the three potyviruses, with statistical significance only for WMV ($p = 0.0083$).

CMV was positively correlated with temperatures. Relative humidity was positively correlated with potyviruses but was not correlated with CMV (data no shown).

The CART algorithms showed that Tmmd was the variable with most weight to explain WMV presence. At Tmmd lower than or equal to 25.6 °C, there was 0.93 probability of WMV presence in cucurbits crops; at Tmmd above 25.6 °C, the probability decreased to 0.74 (Fig. 6 up-left).

Tmmin was the most important variable to explain CMV presence. At Tmmin lower or equal to 18.9 °C, a very low presence was expected (0.05), whereas at Tmmin above 18.9 °C, probability of virus occurrence was 0.76 (Fig. 6 up-right).

For PRSV and ZYMV, RH was the variable that best predicted virus presence. For RH values lower than or equal to 58.3%, a 26% presence of ZYMV was expected, whereas for RH values higher than 58.3%, a presence of 82% was expected (Fig. 6 down-right).

For PRSV, RH lower than or equal to 61.5% and Tmmax lower than or equal to 33.5 °C, the probability of PRSV presence was 6%. In contrast, for higher RH values (61.5%), probability of virus presence was 99% (Fig. 6 down-left).

Discussion

Environmental heterogeneity can shape the plant-virus relationship, furthering the appearance of new diseases in crops, or altering their incidence and severity. The environment can modulate the plant-virus interactions, without inducing genetic changes in the virus or the host (Fraile et al. 2017).

We conducted this study in different environments during 6 years; we identified the bio-meteorological variables involved in the relationship between environmental heterogeneity and virus incidence. All viruses studied in this work are non-persistent and transmitted by aphids. These viruses can be
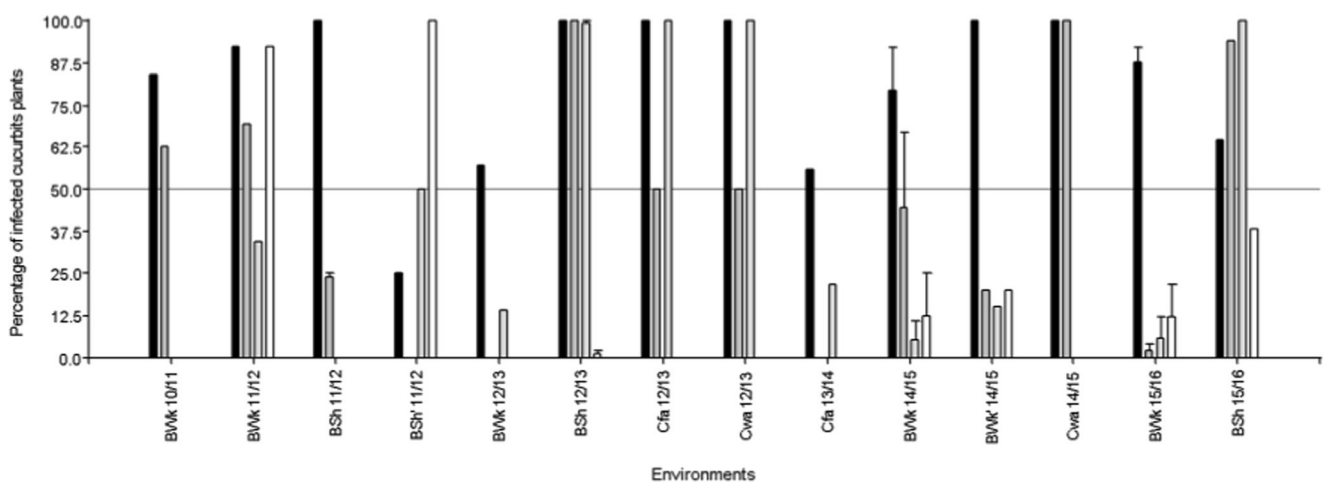

Fig. 3 Percentage of cucurbit plants infected with *Watermelon mosaic virus* (WMV) (black), *Zucchini yellow mosaic virus* (ZYMV) (gray), *Papaya ringspot virus* (PRSV) (light gray) and *Cucumber mosaic virus* (CMV) (white) viruses, in the different environments (14)
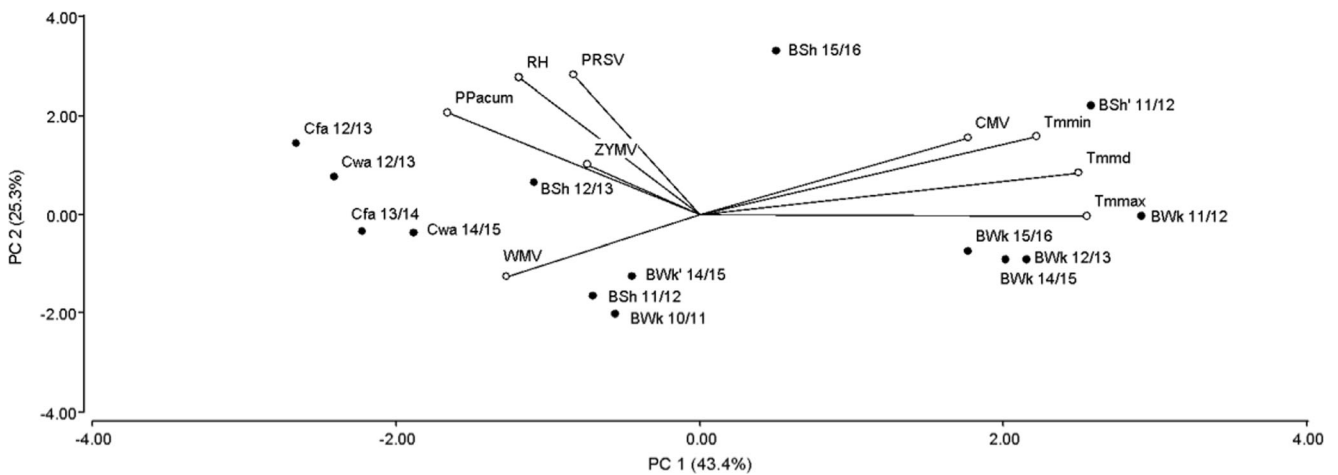
Fig. 4 Principal Component Analysis considering bio-meteorological variables (of the 120 days of the crop cycle), the viruses present (*Watermelon mosaic virus* (WMV), *Zucchini yellow mosaic virus* (ZYMV), *Papaya ringspot virus* (PRSV), and *Cucumber mosaic virus* (CMV), and the different environments

transmitted by several aphid species, but with different efficiency (Lecoq and Desbiez 2012; Francki et al. 1979; Gal-On 2007; Alonso-Prados et al. 2003). In this work, we inferred the relationships between the viruses and environments with highly reliable results. Other works have mentioned the same relationships (Fraile et al. 2017; Kone et al. 2017; Roossinck and García Arenal 2015).

The combination of melon and squash data resulted in a higher total number of samples and provided more powerful results of the statistical analysis. WMV, PRSV, ZYMV and CMV are the most important viruses reported in cucurbits crops worldwide (Alonso-Prados et al. 2003; Ali et al. 2012; Bananej and Vahdat 2008; Lecoq and Desbiez 2012; Vučurović et al. 2012). These viruses cause serious damage that may lead to 100% yield loss and 95% decrease in the value of marketable production (Massumi et al. 2007; Lecoq and Desbiez 2012; Desbiez et al. 2009; Köklü and Yilmaz 2006; Luis-Arteaga et al. 1998; Lecoq et al. 1998; Vučurović et al. 2012). Argentina is not an exception, as WMV, PRSV, ZYMV and CMV were detected in all climates and environments over the 6 years of cucurbit monitoring.

WMV was the most common virus present in cucurbit crops in Argentina, both in arid and temperate climates. A high presence of this virus is expected even in unfavourable environments. CMV was the virus with lowest incidence, in agreement with results reported for South Africa (Ibaba et al. 2015). Some virus species (CMV and WMV) are more influenced by temperature variables, whereas others are influenced by variables of precipitation and relative humidity (PRSV and ZYMV).

Fig. 5 Scatter plot for canonical variables that represent the virus presence against canonical variable of bio-meteorological variables. Polynomial of order 1
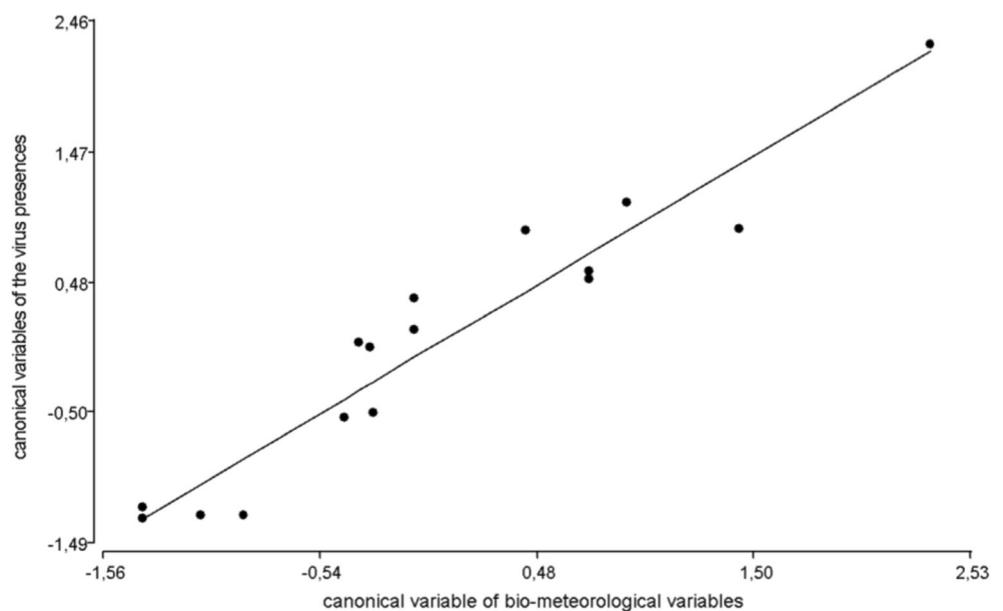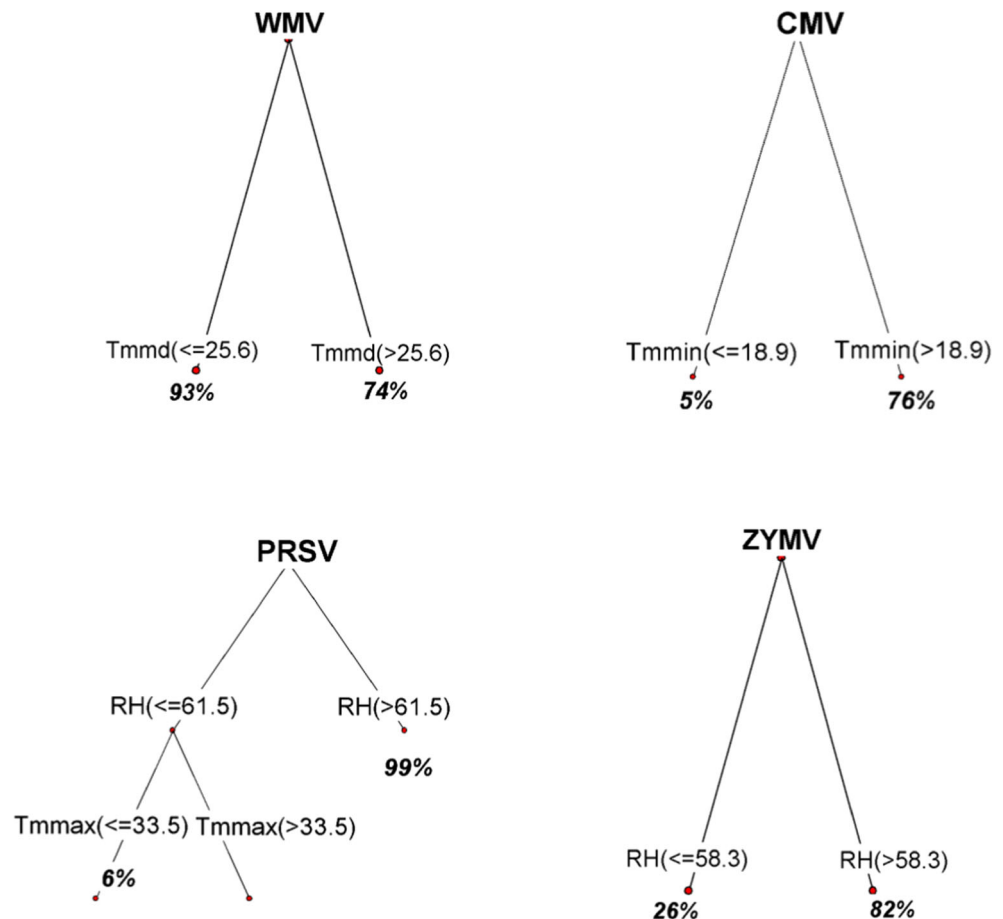
Fig. 6 Regression trees for each virus and climatic conditions, showing percentages of incidence under predisposing climatic conditions. *Watermelon mosaic virus* (WMV), up-left, *Cucumber mosaic virus* (CMV) up-right, *Papaya ringspot virus* (PRSV) down-left and *Zucchini yellow mosaic virus* (ZYMV) down-right

Furthermore, our results show that all the environments had more than one virus, independant of the environment heterogeneity. There are about 130 genera of curcubits: however only four are of commercial importance: squash, watermelon, melon and cucumber. In Argentina, the main cucurbit crop is squash. *Cucurbita moschata* type (70%) and *Cucurbita maxima* (30%) are grown over 27,000 ha. .

The low virus genetic diversity in the all monitored environments and the presence of more than one virus suggest a low or no genotype-environment interaction. We clearly observed a high prevalence of viral diseases in all environments. The diversity and species composition of plant communities can have significant effects on virus prevalence in some productive systems (Power et al. 2011).

Viral diseases were detected in all environments but in different proportions. This difference (between arid and temperate climates) in virus proportion could be due to the different climatic conditions among environments, which also affect virus transmission, replication, incidence, and severity, and the biology of the host and/or vectors (Hull 2009; Fajinmi 2011).

The spatial pattern of viral disease provides an insight into the pathogen dynamics. A random distribution can be attributed to low biodiversity and lack of improved virus resistant/tolerant cultivars.

High temperatures during the 120 days of the crop cycle favoured CMV infection; similarly, Alonso-Prados et al. (2003) reported that CMV incidence was positively correlated with mean maximum and average temperatures. In addition, recent studies have found that high temperature results in symptoms and increases CMV titers (del Toro et al. 2017). WMV was negatively correlated with temperatures, unlike previous results (Alonso-Prados et al. 2003). This disagreement could be attributed to the different growing seasons when the studies were conducted. These results confirm that temperature is an environmental variable that affects the interaction of compatible hosts with different RNA viruses in different ways (del Toro et al. 2015).

The information provided by CART is very useful as a reference parameter. It can predict disease incidence based on prevailing environmental conditions during the crop cycle. However, it is also possible that under favourable conditions, this algorithm cannot predict the virus presence; in this case, the environment should be considered as a risk environment.

All the future analyses performed in other environments will contribute to a better understanding of plant-virus interactions with the environment. The field data obtained for this study and the analyses performed may help to predict the outbreak of viral diseases in cucurbit crops.

Compliance with ethical standards

Conflict of interest Authors declare that they have no conflict of interest.

References

Abou-Jawdah Y, Sobh H, El-Zammar S, Fayyad A, Lecoq H (2000) Incidence and management of virus diseases of cucurbits in Lebanon. Crop Prot 19:217–224

Ali A, Mohammad O, Khattab A, Science B (2012) Distribution of viruses infecting cucurbit crops and isolation of potential new virus-like sequences from weeds in Oklahoma. Plant Dis 96:243–248

Alonso-Prados JL, Luis-Arteaga M, Alvarez JM, Moriones E, Batlle A, Laviña A, García-Arenal F, Fraile A (2003) Epidemics of aphid-transmitted viruses in melon crops in Spain. Eur J Plant Pathol 109(2):129–138

Archie EA, Luikart G, Ezenwa VO (2009) Infecting epidemiology with genetics: a new frontier in disease ecology. Trends Ecol Evol 24:21–30

Bananej K, Vahdat A (2008) Identification, distribution and incidence of viruses in field-grown cucurbit crops of Iran. Phytopathol Mediterr 47:247–257

Biek R, Real LA (2010) The landscape genetics of infectious disease emergence and spread. Mol Ecol 19:3515–3531

Bisognin DA (2002) Origin and evolution of cultivated cucurbits. Ciência Rural 32(4):715–723

Coutts BA, Kehoe MA, Jones RAC (2011) Minimising losses caused by Zucchini yellow mosaic virus in vegetable cucurbit crops in tropical, sub-tropical and Mediterranean environments through cultural methods and host resistance. Virus Res 159:141–160

del Toro FJ, Aguilar E, Hernández-Walias FJ, Tenllado F, Chung BN, Canto T (2015) High temperature, high ambient CO_2 affect the interactions between three positive-sense RNA viruses and a compatible host differentially, but not their silencing suppression efficiencies. PLoS One 10(8):e0136062

del Toro FJ, Rakhshandehroo F, Larruy B, Aguilar E, Tenllado F, Canto T (2017) Effects of simultaneously elevated temperature and CO_2 levels on *Nicotiana benthamiana* and its infection by different positive-sense RNA viruses are cumulative and virus type-specific. Virology 511:184–192

Della Gaspera P, Rodríguez RA, Elisei VR, Ayastuy ME, Khier M, Miglierina A M, Delhey R (2013) Manual del cultivo del zapallo anquito (*Cucurbita moschata* Duch) (No. 635.62). Instituto Nacional de Tecnología Agropecuaria (Argentina)

Desbiez C, Joannon B, Wipf-Scheibel C, Chandeysson C, Lecoq H (2009) Emergence of new strains of watermelon mosaic virus in South-Eastern France: evidence for limited spread but rapid local population shift. Virus Res 141:201–208

Di Rienzo JA, Casanoves F, Balzarini MG, Gonzalez L, Tablada M, Robledo YC (2018) InfoStat versión 2018. Grupo InfoStat, FCA, Universidad Nacional de Córdoba, Argentina URL http://www.infostat.com.ar

Fajinmi AA (2011) Agro-ecological incidence and severity of *Pepper veinal mottle virus*, genus *Potyvirus*, family Potyviridae, on

cultivated pepper (*Capsicum annuum* L.) in Nigeria. Arch Phytopathol Plant Protect 44(4):307–319

Feldman JM, Gracia O (1992) Un nuevo virus de cucurbitáceas en Argentina: el virus de la mancha anillada de la papaya. Revista de Investigaciones Agropecuarias 23:63–67

Fraile A, McLeish MJ, Pagán I, González-Jara P, Piñero D, García-Arenal F (2017) Environmental heterogeneity and the evolution of plant-virus interactions: viruses in wild pepper populations. Virus Res 241:68–76

Francki RIB, Mossop DW, Hatta T (1979) Description of plant viruses. No. 213. CMI/AAB

Gabriel KR (1971) The biplot graphic display of matrices with application to principal component analysis. Biometrika 58(3):453–467

Gal-On A (2007) Zucchini yellow mosaic virus: insect transmission and pathogenicity-the tails of two proteins. Mol Plant Pathol 8(2):139–150

Gracia O (2000) First report of zucchini yellow mosaic virus in Argentina. Plant Dis 84:371

Gracia O, Feldman JM (1986) Virus identificados en cultivos de cucurbitáceas. IDIA 445-448:1–6

Hull R (2009) Comparative plant virology, 2nd edn. Academic press, Norwich

Ibaba JD, Laing MD, Gubba A (2015) Incidence and phylogeny of viruses infecting cucurbit crops in KwaZulu-Natal, Republic of South Africa. Crop Prot 75:46–54

Köklü G, Yilmaz Ö (2006) Occurrence of cucurbit viruses on field-grown melon and watermelon in the Thrace region of Turkey. Phytoprotection 87:123–130

Kone N, Asare-Bediako E, Koita O, Kone D, Winter S (2017) Seasonal and spatial variation in the prevalence of viral diseases and associated aphid-borne viruses in cucurbits in cote d'Ivoire. Ann Agric Sci 62(2):227–234

Kottek M, Grieser J, Beck C, Rudolf B, Rubel F (2006) World map of the Köppen-Geiger climate classification updated. Meteorol Z 15:259–263

Lecoq H, Desbiez C (2012) Viruses of cucurbit crops in the Mediterranean region: an ever-changing picture. Adv Virus Res 84:67–126

Lecoq H, Katis N (2014) Cucurbit virus control. In advances in virus research control of plant virus diseases, pp. 255–296. Eds. G. Loebenstein and N. Katis. San Diego, USA- London UK: Academic Press is an imprint of Elsevier

Lecoq H, Wisler G, Pitrat M (1998) Cucurbit viruses: the classics and the emerging. In: McCreight JD (ed) Cucurbitaceae '98: evaluation and enhancement of cucurbit germplasm. ASHS, Alexandria, pp 126–142

Luis-Arteaga M, Alvarez JM, Alonso-Prados JL, Bernal JJ, García-Arenal F, Laviña A, Batlle A, Moriones E (1998) Occurrence, distribution, and relative incidence of mosaic viruses infecting field-grown melon in Spain. Plant Dis 82:979–982

Massumi H, Samei A, Pour AH, Shaabanian M, Rahimian H (2007) Occurrence, distribution, and relative incidence of seven viruses infecting greenhouse-grown cucurbits in Iran. Plant Dis 91(2):159–163

Meentemeyer RK, Haas SE, Vaclavik T (2012) Landscape epidemiology of emerging infectious diseases in natural and human-altered ecosystems. Annu Rev Phytopathol 50:379–402

Mohammed H, Manglli A, Zicca S, El Hussein A, Mohamed M, Tomassoli L (2012) First report of *Papaya ringspot virus* in pumpkin in Sudan. New Dis Rep 26:26

Mohammed HS, Zicca S, Manglli A, Mohamed ME, El Siddig MA, Tomassoli L, El Hussein AA (2014) Identification and phylogenetic analysis of common pumpkin viruses in Sudan. J Plant Pathol 96(1):77–84

Moreira AS, Filho AB, Rezende JAM (2014) Comparative epidemiology of three virus diseases on zucchini squash. J Phytopathol 163:386–394

Nome SF, March GJ, Giorda LM (1974) Disminución de la productividad de plantas de zapallito de tronco (*Cucurbita máxima* Duch var. zapallito (Carr. Millán) infectadas con el virus del mosaico de la sandía, raza 2 (*Watermelon Mosaic Virus*-2). IDIA, 321–324

Perotto MC, Celli MG, Pozzi EA, Luciani CE, Conci VC (2016) Occurrence and characterization of a severe isolate of *Watermelon mosaic virus* from Argentina. Eur J Plant Pathol 146:213–218

Power AG, Borer ET, Hosseini P, Mitchell CE, Seabloom EW (2011) The community ecology of barley/cereal yellow dwarf viruses in Western US grasslands. Virus Res 159(2):95–100

Reisen WK (2010) Landscape epidemiology of vector-borne diseases. Annu Rev Entomol 55:461–483

Roossinck MJ, García Arenal F (2015) Ecosystem simplification, biodiversity loss and plant virus emergence. Curr Opin Virol 10:56–62

Vučurović A, Bulajić A, Stanković I, Ristić D, Berenji J, Jović J, Krstić B (2012) Non-persistently aphid-borne viruses infecting pumpkin and squash in Serbia and partial characterization of *Zucchini yellow mosaic virus* isolates. Eur J Plant Pathol 133(4):935–947