

Fungal characteristics and varietal reactions of powdery mildew species on cucurbits in the steppes of Ukraine

Y. Tomason¹ and P.T. Gibson²

¹Department of Selection and Seed Production, Dnepropetrovsk State Agrarian University, Voroshilov 25, 49600 Dnepropetrovsk, Ukraine; e-mail: yantomason75@mail.ru

²Department of Plant, Soil and Agriculture Systems, Southern Illinois University, 62901, Carbondale, IL, USA; e-mail: pgibson@siu.edu

Abstract. Powdery mildew caused by fungal species *Sphaerotheca fuliginea* (Schlect ex. Fr.) Poll. [syn. *Podosphaera xanthii* (sect. *Sphaerotheca*) *xanthii* (Castag.) U. Braun & N. Shish. Comb. nov.] and *Erysiphe cichoracearum* D.C. [syn. *Golovinomyces cichoracearum* (D.C.) Huleta] is the most frequent cucurbit (*Cucurbitaceae*) crop disease in the northern steppes of Ukraine. Species commonly cultivated in Ukraine (melon, watermelon, squash) were colonized by both species. Ash gourd (*Benincasa hispida* Cogn.) and bottle gourd (*Lagenaria siceraria* Standl.) were colonized only by *E. cichoracearum*. Sponge gourd (*Luffa cylindrica* M.J. Roem) and fig-leaved gourd (*Cucurbita ficifolia* L.) were not colonized by either of these fungi. Varieties within species varied from resistant to highly susceptible. Four melon varieties were agronomically elite and had resistance to both species. Those two species were consistently differentiated by the location on the conidia where germination occurred, and by the ratio of conidial length to width. Indicator host species and varieties can provide a tentative identification of which powdery mildew species are present in the field.

Varietal differentials suggested that races 1 & 2 of *S. fuliginea* were present on watermelon, and that race 1 and at least one other race were present on melon.

Key words: powdery mildew, *Sphaerotheca fuliginea*, *Erysiphe cichoracearum*, cucurbit, *Cucurbitaceae*, resistance, melon, *Cucumis melo*, watermelon, *Citrullus lanatus*, races

INTRODUCTION

Powdery mildew is an endemic disease of many crop species worldwide. It is also one of the most damaging diseases affecting melon crops (Alvarez et al., 2000). Six species of powdery mildew fungi have been reported on various hosts from different regions of the world. Among these, *Sphaerotheca fuliginea* (Schlect ex. Fr. Poll. [syn. *Podosphaera xanthii* (sect. *Sphaerotheca*) *xanthii* (Castag.) U. Braun & N. Shish. Comb. nov. (McCreight, 2006), syn. *Sphaerotheca fusca* (Rnakovic, 2003), *Podosphaera fusca* (Perez-Garcia et al., 2006)] and *Erysiphe cichoracearum* D.C. [syn. *Golovinomyces cichoracearum* (D.C.) Huleta] are the most common and widespread (Nayar & More, 1998; McCreight, 2006). *S. fuliginea* is serious in many countries, including Israel, Spain, Greece, Armenia, and Japan (Dutin, 1981; Cohen et al., 2000; Fanourakis et al., 2000; Kuzuya et al., 2000). Both species occur in Germany, France, Hungary, Czech Republic, India, New Zealand and Montenegro (Lebeda & Kriskova, 2000; Rankovic, 2003). Multiple races of both species attack cucurbits in the USA and

elsewhere (McCreight, 2006). Powdery mildew attacks almost all the cucurbitaceous (*Cucurbitaceae*) crops. In addition to both *S. fuliginea* and *E. cichoracearum* occurring on the commonly cultivated cucurbits species (melon, watermelon, squash, pumpkin, etc.), the latter has also been reported on *Momordica balsamina*, *Trichosanthes dioicia*, *Lagenaria vulgaris*, *Coccinia cordifolia*, and *Benincasa hispida* (Nayar & More, 1998).

S. fuliginea and *E. cichoracearum* are distinguished by the size and shape of fungal structures, including asci and conidia (Frolov, 1996; Dutin & Sokolov, 1978; Rankovic, 2003). Ascus structures vary in size and number with the environment and plant host, but the general shape of structures and the number of appendages and ascospores differ enough that the composite picture created by ascus structures are considered reliable in differentiating the two fungal species. However, asci do not develop on all hosts, so conidial observations are much more generally useful. *S. fuliginea* conidia are oval and germinate from a lateral face and produce a broad, clavate germ tube. *E. cichoracearum* conidia are cylindrical and germinate from a corner of terminal and lateral faces. The resulting germ tube is a tapered cylinder (Dutin & Sokolov, 1978). Conidial length/width ratios have been reported to reliably distinguish between the two fungal species (Dutin & Sokolov, 1978; Frolov, 1996; Rankovic, 2003). The race structure of powdery mildew worldwide has been best studied on melon. A total of 28 races of *S. fuliginea* have now been documented, along with two races of *E. cichoracearum* described (Anagnostu et al., 2000; McCreight, 2001, 2006). In watermelon (*Citrullus lanatus* Mansf.), only two races of have been differentiated by varietal reactions to *S. fuliginea* (Nayar & More, 1998).

In Ukraine, powdery mildew was first reported on a cucurbit crop in 1901. In the Kherson area on the coast of the Black Sea, both *S. fuliginea* and *E. cichoracearum* affect cucurbit crops (Frolov, 1996). Although powdery mildew is common on various cucurbits throughout Ukraine, the fungal species involved have not been confirmed for most areas of the country. Therefore, the object of the research reported here was: 1) to identify the species occurring on cucurbits in the northern steppe of Ukraine, 2) to evaluate the varietal reactions of cultivated varieties and genetic accessions of various cucurbit species under local conditions, and 3) to compare the reported host reaction of established race differentials for *S. fuliginea* with their local field response.

MATERIALS AND METHODS

Host reaction to *S. fuliginea* and *E. cichoracearum* was field evaluated each year from 2000 to 2004 in a collection of 200 varieties, populations and genetic accessions of cucurbit crop species (collectively referred to as varieties) (Table 1). Varieties were planted within a one- or two-day period each year, between 1–10 May, and were grown without irrigation. Visual ratings of powdery mildew were assigned, ranging from 0 (immune) to 9 (extremely susceptible). The study involved the subfamily *Cucurbitoidae*, represented by the tribes *Melonthriaca* Endl. (*Cucumis sativus*, *C. melo*), *Benincaseae* Ser. (*Citrullus lanatus*, *Benincasa sp.*, *Lagenaria sp.*, *Luffa sp.*), and *Cucurbitae* Schard. (*Cucurbita ficifolia*, *C. pepo*, *C. moschata* and *C. maxima*). The two locations used were the "Samarsky" farm (48°31'N, 35°25'E) and Novomoskovsk Agrarian College (48°45'N, 35°26'E) of Dnepropetrovsk State Agrarian University.

Table 1. General reaction of each cucurbit species tested against *S. fuliginea* and *E. cichoracearum* in the northern steppe of Ukraine. No pathogen development occurred on any of the 5 varieties of sponge gourd (*Luffa cylindrica* M.J. Roem), or the 2 varieties of fig-leaved gourd (*Cucurbita ficifolia* L.).

Species	Number of varieties	<i>S. fuliginea</i>						<i>E. cichoracearum</i>					
		Stage of fungal development		Host Reaction				Stage of fungal development		Host Reaction			
		Asci	Conidia only	0	1-3	4-5	7-9	Asci	Conidia only	0	1-3	4-5	7-9
Melon (<i>C. melo</i> L.)	100	15	70	15	25	34	26	15	68	17	32	24	29
Watermelon (<i>Citrullus lanatus</i> Mansf.)	40		39	1		39			39*	1		39	
Pumpkin (<i>C. maxima</i>)	15		15			10	5		15			15	
Pumpkin (<i>C. moschata</i>)	10		10			10			10			10	
Summer squash (<i>C. pepo</i>)	10	10				10			10			10	
Cucumber (<i>C. sativus</i> L.)	10		8	2	2	6			8	2	2	6	
Bottle gourd (<i>Lagenaria siceraria</i> Standl.)	4			4					4			4	
Ash gourd (<i>Benincasa hispida</i> Cogn.)	4			4					4			4	

Table 2. Characteristics of ascal structures of powdery mildew fungi on summer squash and melon in the northern steppe of Ukraine. Ranges shown are the extreme values observed among means (n=50) of either of 2 varieties per host species in either location in either year.

Fungus	Ascal structure									
	As-ma*, μ	App (N)	As (N)	As (L), μ	As (W), μ	As(L/W)	Sp (N)	Sp (L), μ	Sp (W), μ	Sp(L/W)
Summer squash										
<i>S. fuliginea</i>	67–97	5–7	1	60–75	47–63	1.0–1.3	6–7	13.6–15.1	12.5–13.7	1.0–1.2
<i>E.cichoracearum</i>	64–121	Numerous	3–17	47–65	27–37	1.4–2.2	2	20.7–24.5	13.5–15.8	1.4–1.8
Melon										
<i>S. fuliginea</i>	57–89	5–8	1	56–73	48–64	1.0–1.5	8–9	14.4–16.0	11.6–13.2	1.1–1.3
<i>E. cichoracearum</i>	74–125	Numerous	2–15	46–61	23–39	1.4–2.6	2	18.9–23.6	15.1–16.5	1.2–1.5

*As-ma – diameter of ascomata; App (N) – number of appendages; As (N) – number of asci per ascal sac; As (L), As(W), As(L/W) – length, width, and length/width ratio of asci; Sp (N) – number of ascospores; Sp (L), Sp (W), Sp (L/W) – length, width, and length/width ratio of ascospores.

Table 3a. Analysis of variance of size of ascomata and asci of 2 species of powdery mildew (*S. fuliginea*, *E. cichoracearum*) on 4 varieties of cucurbits (summer squash, melon) in the northern steppe of Ukraine¹.

Source of Variation	Df	Diameter of ascomata (μ)			Ascal length (μ)			Ascal width (μ)			Ascal L/W		
		MS	F	Den	MS	F	Den ²	MS	F	Den ¹	MS	F	Den
Location (L)	1	4439	6.65	LY	33.6	0.06	LY	15.6	0.04	LY	0.10	4.01*	Err
Year (Y)	1	6320	9.47	LY	287.2	0.49	LY	0.01	0.00	LY	0.37	14.37***	Err
L x Y	1	667.3	9.01**	Err	587.3	35.9***	Err	366.0	28.45***	Err	0.02	0.93	Err
Species (S)	1	11869	25.1	LYS	6007	38.63	LYS	21935	49.45+	LYS	10.63	90.46+	LYS
L x S	1	9377	19.8	LYS	293.4	1.89	LYS	42.9	0.10	LYS	0.10	0.86	LYS
Y x S	1	1000	2.11	LYS	96.8	0.62	LYS	1.3	0.00	LYS	0.15	1.25	LYS
L x Y x S	1	473.5	6.4*	Err	155.5	9.53**	Err	443.6	34.48***	Err	0.12	4.55*	Err
Varieties (V)	3	688.8	1.05	LYV	57.8	0.77	LYV	253.0	3.60	LYV	0.33	2.57	LYV
L x V	3	1115	1.70	LYV	14.1	0.19	LYV	443.7	6.32+	LYV	0.53	4.17	LYV
Y x V	3	322.4	0.49	LYV	192.0	2.55	LYV	262.6	3.74	LYV	0.72	5.67+	LYV
L x Y x V	3	654.5	8.84***	Err	75.3	4.61**	Err	70.2	5.46***	Err	0.13	4.91**	Err
S x V	3	909.3	0.80	LYSV	135.3	0.23	LSV+ YSV	62.4	0.52	LYSV	0.12	0.23	LYSV
L x S x V	3	310.5	0.27	LYSV	393.9	24.1***	Err	173.7	1.44	LYSV	0.52	1.02	LYSV
Y x S x V	3	406.5	0.36	LYSV	323.5	19.8***	Err	167.9	1.40	LYSV	0.13	0.25	LYSV
L x Y x S x V	3	1134	15.32***	Err	30.9	1.89	Err	120.3	9.35***	Err	0.30	19.54***	Err
Error (Err)	136	74.0			16.3		-	12.9	-		0.026		

¹ +, *, **, *** significant at $P = 0.1, 0.05, 0.01, 0.001$, respectively.

² Den signifies F-test denominator, which is dependent on the relevant expected mean squares involved in interactions with that particular source of variation. The actual compound F-test used was $F = (SxV + LxYxSxV)/(LxSxV + YxSxV)$.

Table 3b. Analysis of variance of size of ascospores of 2 species of powdery mildew (*S.fuliginea*, *E. cichoracearum*) on 4 varieties of cucurbits (summer squash, melon) in the northern steppe of Ukraine¹.

Source of Variation	Df	Ascospore length (μ)			Ascospore width (μ)			Ascospore L/W		
		MS	F	Den	MS	F	Den	MS	F	Den
Location (L)	1	0.06	0.03	Err	1.09	0.20	Err	0.01	0.71	Err
Year (Y)	1	1.35	0.71	Err	5.48	1.38	Err	0.03	1.88	Err
L x Y	1	4.73	2.47	Err	3.97	3.82	Err	0.00	0.16	Err
Species (S)	1	2212	380.8*	LS	240.1	60.49+	LYS	3.77	54.03+	SL
L x S	1	5.81	3.04+	Err	0.26	0.06	LYS	0.07	4.81*	Err
Y x S	1	1.29	0.67	Err	2.12	0.53	LYS	0.01	0.35	Err
L x Y x S	1	0.03	0.02	Err	3.97	3.82+	Err	0.03	2.32	Err
Varieties (V)	3	5.33	0.30	LV+ YV	0.57	0.55	Err	0.02	0.11	LV
L x V	3	17.78	9.29**	Err	1.74	1.68	Err	0.15	10.40***	Err
Y x V	3	4.26	2.23+	Err	1.72	1.65	Err	0.00	0.33	Err
L x Y x V	3	1.32	0.69	Err	0.53	0.51	Err	0.01	0.50	Err
S x V	3	12.34	0.53	LSV +YSV	6.58	6.34***	Err	0.25	6.76+	LYSV
L x S x V	3	16.53	8.64***	Err	1.04	1.01	Err	0.13	3.40	LYSV
Y x S x V	3	8.71	4.55**	Err	0.74	0.71	Err	0.06	1.51	LYSV
L x Y x S x V	3	1.15	0.60	Err	2.20	2.12	Err	0.04	2.55+	Err
Error (Err)	136	1.91			1.00			0.013		

¹ +, *, **, *** significant at $P = 0.1, 0.05, 0.01, 0.001$, respectively.

² Den signifies F-test denominator, which is dependent on the relevant expected mean squares involved in interactions with that particular source of variation. The actual compound F-tests used were $F = (SxV + LxYxSxV)/(LxSxV + YxSxV)$ and $F = (V + LxYxV)/(LxV + YxV)$, respectively.

Table 4. Size of conidia of powdery mildew on cucurbit crops in the northern steppe of Ukraine in 2003–2004.¹ Ranges shown are the extreme values observed among means ($n = 50$) in either location in either year.

Species	Variety or collection number ²	Fungi powdery mildew					
		<i>Sphaerotheca fuliginea</i> Poll.			<i>Erysiphe cichoracearum</i> DC.		
		Length of conidia, μ	Width of conidia, μ	Ratio -- length/ width	Length of conidia, μ	Width of conidia, μ	Ratio -- length/ width
	<i>Lipneva</i>	32.9–35.0	22.2–23.2	1.43–1.54	37.5– 40.5	17.3–18.8	2.15–2.30
Melon (<i>Cucumis melo</i> L.)	PMR–45	26.3–26.8	18.–20.5	1.28–1.44	–	–	–
	G–16	–	–	–	35.2–38.5	16.4–18.5	1.98–2.08
	<i>Sicheslav</i>	20.5–22.5	13.8–14.6	1.48–1.59	22.2–25.9	10.5–12.2	1.92–2.32
Watermelon (<i>Citrullus lanatus</i> Mansf.)	Carolina cross#183	–	–	–	–	–	–
	<i>Voronezkii</i>	21.8–22.6	15.4–15.9	1.38–1.47	28.2–29.0	12.9–13.8	2.04–2.28
Cucumber (<i>Cucumis sativus</i> L.)	Feniks 640	–	–	–	–	–	–
Summer squash (<i>Cucurbita pepo</i> var. <i>qiranmonas</i> Duch.)	<i>Zuksha</i>	22.7–23.6	14.2–15.7	1.54–1.63	25.9–27.4	10.9–12.4	2.26–2.42
Pumpkin (<i>Cucurbita maxima</i> L.)	<i>Valok</i>	36.0–37.2	22.1–23.3	1.55–1.66	32.9–34.0	15.4–15.9	2.15–2.26
Pumpkin (<i>Cucurbita moschata</i> L.)	<i>Divo</i>	32.3–34.1	19.9–22.2	1.49–1.66	34.2–35.3	16.0–17.6	1.97–2.23
Bottle gourd (<i>Lagenaria siceraria</i> Standl.)	NDSAU 12	–	–	–	29.2–33.0	14.5–15.8	2.03–2.19
Ash gourd (<i>Benincasa hispida</i> Cogn.)	NDSAU 21	–	–	–	30.5–31.4	13.1–13.7	2.17–2.27

¹ “–” indicates conidia not present.

² varieties included in the analysis of variance in Table 5 are italicized.

Table 5. Analysis of variance of the size of conidia of powdery mildew (*S. fuliginea*, *E. cichoracaerum*) on 6 cucurbits varieties representing 5 species (see Table 4) in the northern steppe of Ukraine¹.

Source of variation	Df	Length		Width		Ratio (L/W)	
		MS	F	MS	F	MS	F
Location (L)	1	0.1	0.02	7.7	2.80	0.05	0.98
Year (Y)	1	5.4	0.82	3.8	1.38	0.13	2.39
L x Y	1	9.0	1.35	0.3	0.11	0.02	0.35
Species (S)	1	459.0	68.94***	1017.2	370.96***	26.10	473.78***
L x S	1	2.4	0.36	3.9	1.43	0.00	0.02
Y x S	1	4.3	0.64	1.2	0.43	0.00	0.08
L x Y x S	1	0.2	0.03	0.5	0.19	0.00	0.00
Varieties (V)	5	1448.3	217.52***	460.0	167.76***	0.13	2.32*
L x V	5	10.8	1.62	1.0	0.36	0.02	0.35
Y x V	5	2.5	0.38	0.9	0.31	0.01	0.16
L x Y x V	5	4.4	0.67	1.4	0.51	0.06	1.17
S x V	5	103.0	15.47***	29.1	10.60	0.11	1.92+
L x S x V	5	3.3	0.50	4.4	1.59	0.07	1.21
Y x S x V	5	0.2	0.03	2.0	0.74	0.03	0.61
L x Y x S x V	5	4.2	0.64	0.5	0.18	0.03	0.61
Error (Err)	192	6.7	–	3.1		0.06	

¹ +, *, **, *** Significant at 0.05, 0.01, 0.001 levels, respectively. Error (Err) was the appropriate F-test denominator in all cases.

Table 6. Field susceptibility ratings (0–9, worst) in the northern steppe of Ukraine for differential genotypes of watermelon and melon compared to their reported response of to races 1–3 of *Sphaerotheca fuliginea* Poll.

Genotype	Reported response to race ¹			Field susceptibility ratings ²				
	1	2	3	S2003	S2004	NM2003	NM2004	Average
Watermelon								
Sugar Baby	S*	S	–	7.3	7.2	7.6	7.8	7.5
Charleston Gray	S	R	–	3.2	3.2	4.0	3.6	3.5
Melon								
Lipneva	S	S	S	8.8	8.8	9.0	9.0	8.9
Top mark	S	S	S	7.2	8.0	8.3	8.0	7.9
PMR–45	R	S	S	4.2	5.0	5.3	6.3	5.2
Amarillo	R	S	S	4.2	5.5	5.5	5.3	5.1
Edisto 47	R	R	S	2.0	2.0	2.2	2.3	2.1

¹ S = susceptible; R = resistant – not identified.

² S = Samarsky, NM = Novamoskovsk.

Samples of powdery mildew were gathered from infected cucurbit plants in 2003–2004 at the time when symptoms developed rapidly, which was mainly from the 10th–20th of August, but encompassed 5–25th August. Infected leaves were collected from multiple replications. Fungal material was scraped from multiple fresh leaves onto glass slides and stored at 5°C. Observations of fungal stage, size of fungal structures, and characteristics of conidial germination were carried out during the one to two days following tissue collection. Conidial shape and germination were examined at 150× magnification. Annually for each host variety, 10 random conidia were measured from each of 5 glass slides, using a screw micrometer (Dutin & Sokolov, 1978; Braun, 1987). Fungal species were identified based on the morphology of fungal bodies and conidia. For observation of conidial germination, glass slides on which conidia rested were placed on wet filter paper in covered Petri dishes at 22°C for 24 h. The Petri dishes were placed on shelves that received daylight through windows. The suitability of fungal measurements (ascal structures and conidia) for differentiating the two fungal species was evaluated using multi-factor ANOVA, with 4–6 varieties, 2 locations, 2 years, and 5 samples per variety per environment. The mean of the values from 10 conidia per slide was considered one sample. Location, year, and sample were considered random-effects factors. Fungal species and host variety were considered fixed-effects factors. Appropriate F-tests were chosen for each source of variation according to the presence or absence of relevant interactions (Snedecor & Cochran, 1976, p. 374).

The host reaction of established differential varieties was evaluated in the field in 2003–2004 and compared with reported response to known races in an attempt to infer the possible racial characteristics of *S. fuliginea* in the field. These differentials, except ‘Lipneva’, are available through the Cucurbits Genetic Cooperative, Cornell Univ., Ithaca, NY and all are available through Vavilov Institute of Plant Production (VIR), St Petersburg, Russia. Lipneva is also available through the first author, or through Umesh Reddy, West Virginia State Univ.

RESULTS AND DISCUSSION

Host reaction. Table 1 shows the distribution of host ratings, averaged over two locations and five years (2000–2004). Comparative ratings were very consistent across environments. No pathogen development occurred on any of the 5 varieties of sponge gourd (*Luffa cylindrica* M.J.Roem), or the 2 varieties of fig-leaved gourd (*Cucurbita ficifolia* L.). Bottle gourd (*Lagenaria siceraria*) and ash gourd (*Benincasa hispida*) were colonized only by *E. cichoracearum*. All other species of cucurbit crops that were investigated were colonized by both species of powdery mildew that are present in this area. The host reaction of melon varieties to each fungal species was almost uniformly distributed across the severity range, with almost one-half showing complete or moderate resistance. There were 12 melon varieties that were immune to both species, and several more that were at least moderately resistant to both. Of the 12 that were immune, four also displayed high yield and quality, and early maturity. These were: Line 22/1 (proprietary line of Dnepropetrovsk State Agrarian University, contact first author), F1 Hybrid Promitei, Tabolinka and Kurume (all available commercially or through VIR). Cucumber also displayed the full range of host response to both species, with two varieties, including Feniks 640 (available commercially or through VIR)

resistant to both fungal species. In contrast, all 40 watermelon varieties except the resistant variety, Carolina Cross #183 (available through the Cucurbits Genetics Cooperative, Cornell Univ., Ithaca, NY), plus all 10 summer squash varieties, and all 10 *C. moschata* pumpkin varieties were intermediate in response to both species. In addition, all varieties tested of *C. maxima* pumpkin, bottle gourd, and ash gourd had intermediate reactions to *E. cichoracearum*. Five varieties of *C. maxima* were highly susceptible to *S. fuliginea*.

Fungal species morphology. Species of powdery mildew are generally identified by the morphology of the fungus (Dutin & Sokolov, 1978). Ascal characteristics were measured for 2003 and 2004, the years of strongest powdery mildew development (Table 2). In this study in the northern steppe of Ukraine, two species were identified – *Sphaerotheca fuliginea* and *Erysiphe cichoracearum*. In local conditions, powdery mildew progresses through all stages of development on summer squash and melon before these crops senesce. In other cucurbit host species, only the conidial stage is reached, if the pathogen develops at all.

Table 2 shows the range encountered for ascal structures among two locations, two years, and two varieties each of summer squash and melon. Thus, these are minimums and maximums from 8 means ($n = 50$) for each of the four combinations of fungal and host species. There is considerable overlap in the range of the two fungal species for size of ascomata and length of asci. On the basis of the means involved, ascal width and length/width ratio and spore dimensions overlapped little, if at all. Rankovic (2003) reported dimensions and ratios very similar to ours for ascal structures for *S. fuliginea* and for *E. cichoracearum* in Montenegro, except that some ascomata were larger (up to 140μ for *E. cichoracearum*). In the ANOVA of our data, species differences showed mean squares at least 150 times greater than sampling error for all ascal measurements (Table 3). However, because of interactions with the environment, species were only significantly different ($P < 0.05$) for ascospore length. Both the overlap of the ranges and the strong effect of interaction with the environment preclude a confident identification of the fungal species based on measurements alone. Appendage and spore numbers were clearly different between the two species, and could be used in combination with ascal and spore measurements (especially length/width ratio) to confidently identify the fungal species involved in the environments tested. These conclusions are in agreement with Rankovic (2003). However, asci did not develop on several of the cucurbit species tested, nor even on a number of relatively susceptible varieties in melon and watermelon. Consequently, the identification of fungal species during plant vegetative growth was more comprehensively carried out on conidial stages.

Germination of conidia in the moist Petri dishes coincided with observations of others (Dutin & Sokolov, 1978; Frolov, 1996; Rankovic, 2003) that *S. fuliginea* sprouted only from a lateral face with the germ tube being clavate in form, and that *E. cichoracearum* sprouted from a surface corner with the germ tube forming a tapered rod. Conidia of *S. fuliginea* had dimensions of $20.5\text{--}37.2 \times 13.8\text{--}23.3 \mu$. The ratio of length to width varied from 1.38–1.66 (Table 4). Conidia of *E. cichoracearum* ranged from $22.2 - 40.5 \times 10.5\text{--}18.8 \mu$, with a length/width ratio of 2.04 up to 2.42. The center of the range of absolute sizes of conidia reported by Frolov (1996) in Kherson (southern Ukraine, on the Black Sea) and by Dutin & Sokolov (1978) in Astrakhan for

these two fungal species was almost identical to ours, but encompassed a narrower range of values. Frolov reported length/width ratios of 1.32–1.76 for *S. fuliginea*, almost identical to our results. Dutin & Sokolov's value of 1.57–1.79 for this species was very similar except that the range did not extend as low as did ours. Frolov's values of 1.91–1.96 for *E. cichoracearum* were noticeably lower than ours, but still clearly distinct from those of *S. fuliginea*. The values of Dutin & Sokolov for *E. cichoracearum* were in general agreement, but encompassed a wider range (1.86–2.45). It is apparent from these ranges that conidial length and width cannot be used for identification of these species, since the species dimensions overlap and change considerably due to the characteristics of the plant host. Both conidial length and width varied strongly by variety and showed fungal species x host variety interactions (Tables 4 & 5). All varieties had length/width ratios consistently below 1.7 for *S. fuliginea* in all environments and consistently above 1.9 for *E. cichoracearum* (Table 4). Varietal differences were small for conidial length/width (barely significant at $P < 0.05$), and the interaction of fungal species x host variety was not significant at $P < 0.05$. It could be argued that variety should be considered a random effect in constructing F-tests when the purpose is to evaluate the use of a particular variable to differentiate between the fungal species regardless of variety. If variety were considered a random variable, the F-test denominator for species would be species x variety. Using this alternate F-test, species differences for length were not significant at $P < 0.05$, while those for width and length/width remained highly significant ($P = 0.002$ and $P < 0.0001$, respectively). Although the ANOVA confirmed that differences for conidial width within a variety were consistent across fungal species and environment, the absolute magnitude varied strongly with variety and overlapped between fungal species. Therefore, only the length/width ratio of conidia reliably differentiated between the fungal species. These results along with those of Frolov (1996), Dutin & Sokolov (1978) and Rankovic (2003) represent widely separated locations with very different environments. There was close correspondence among authors of conidial sizes and length/width ratios, indicating that the length/width ratios should be reliable for distinguishing these two fungal species across multiple cucurbits species in a range of environments.

Fungal species differentiation by host response. As previously mentioned, bottle gourd and ash gourd were moderately susceptible to *E. cichoracearum* but immune to *S. fuliginea*. Melon variety PMR-45 was resistant to *E. cichoracearum*, while variety G-16 was resistant to *S. fuliginea*. Thus, bottle gourd, ash gourd, and melon variety G-16 can be used as field indicators of the presence of *E. cichoracearum*, and melon variety PMR-45 can be used as an indicator of *S. fuliginea* if races other than race 1 are present.

Field response of race differentials. The reported host reactions of standard differentials of watermelon and melon to *S. fuliginea* races (Hosoye et al., 1999; Fanourakis et al., 2000; McCreight, 2001, 2006; Fukino et al., 2004) are shown in Table 6. Race differentials were not available for other host species (Bardin et al., 1997; Sakata et al., 2006). It was not possible to test differential responses to single isolates in a controlled environment. However, field responses of known differentials allow some inferences about the races present. The consistency of response of the differentials in the four environments tested suggests that the race composition of *S. fuliginea* was stable across the locations and years involved. In watermelon, the large

difference between “Sugar Baby” and “Charleston Gray” is consistent with a substantial presence of race 2. In addition, the presence of race 1 is suggested by the failure of “Charleston Gray” to be fully resistant in the field.

In melon, the strong susceptibility of Lipneva and Top Mark compared to the intermediate response of PMR-45 and Amarillo suggests a substantial presence of Race 1. The substantially lower disease rating of Edisto 47 suggests a substantial presence of *S. fuliginea* other than Race 1. The variable response of Edisto 47 to race 2 reported by McCreight (2006) precludes further inferences about which race other than race 1 was present.

CONCLUSIONS

In the steppe of Ukraine, two powdery mildew fungal species were identified on cucurbit crops: *S. fuliginea* and *E. cichoracearum*. Sponge gourd and fig-leaved gourd were not colonized by these species, and ash gourd and bottle gourd were colonized only by *E. cichoracearum*. The common cucurbit species were colonized to varying degrees by both species, although varieties resistant to both species were observed in melon, watermelon and cucumber.

The absolute size of ascus structures was not a reliable criterion for differentiating these two fungal species, but the ratio of length to width of asci and ascospores was. However, the lack of development of asci on many species and varieties limits the usefulness of ascus measurements. The absolute measurements of length and width of conidia also showed overlap between the two fungal species. However, the length/width ratio of conidia and the conidial location of germination consistently differentiated the two species across locations, years, host species, and host varieties within species within the northern steppe of Ukraine. Bottle gourd, ash gourd, and the G-16 variety of melon were susceptible to *E. cichoracearum*, but not to *S. fuliginea*, and can therefore be used as field indicators of *E. cichoracearum*. Melon variety PMR-45 is resistant to *E. cichoracearum* but susceptible to all but race 1 of *S. fuliginea*, and can therefore be used as a field indicator of *S. fuliginea* as long as a race other than race 1 is present. Melon differentials for *S. fuliginea* races showed varying degrees of susceptibility in the field, consistent with the presence of a mixture of pathogen race 1 with some other race. Similarly, the field reaction of watermelon differentials suggested the presence of a mixture of races 1 and 2.

REFERENCES

- Anagnostu, K., Jahn, M. & Perl-Trevis, R. 2000. Inheritance and linkage analysis of resistance to zucchini yellow mosaic virus, watermelon mosaic virus, papaya ringspot virus and powdery mildew in melon. *Euphytica* **116**, 265–270.
- Alvarez, J.M., Gomez-Gullaman, M.L., Toreas, N.A., Canovas, I. & Floris, E. 2000. Virulence differences between two Spanish isolates of *Sphaerotheca fuliginea* race 2 on melon. *Proceedings of Cucurbitaceae 2000. The 7th EUCARPIA Meeting of Cucurbit Genetics and Breeding. Acta Horticulturae* **510**, 67–71.
- Bardin, M., Nicot, P., Normand, P. & Lemaire, J. 1997. Virulence variation and DNA polymorphism in *Sphaerotheca fuliginea*, casual agent of powdery mildew of cucurbits. *European Journal of Plant Pathology* **103**, 545–554.

- Braun, U. 1987. *A monograph of the Erysiphales (powdery mildews)*. Beihet Nova Hedwigia (J. Cramer, Berlin). 700 pp.
- Cohen, Y., Baider, A., Petrov, L., Sheck, L. & Voloisky, V. 2000. Cross-infectivity of *Sphaerotheca fuliginea* to watermelon, melon, and cucumber. *Proceedings of Cucurbitaceae 2000. The 7th EUCARPIA Meeting of Cucurbit Genetics and Breeding. Acta Horticulturae* **510**, 85–88.
- Dutin, K.E. & Sokolov, U.V. 1978. Identification of powdery mildew species on cucurbit crops using conidia. *Breeding and technology: irrigation of melon crops*; pp. 11–14. All Russian Sci. Res. Inst. of Vegetable & Melon Crops, Astrakhan (in Russian).
- Dutin, K.E. 1981. Sources of powdery mildew resistance in melon crops. *Mycology and Phytopathology* **15**, 4, 299–301. All Russian Sci. Res. Inst. of Irrigated Vegetable & Melon Crops, Astrakhan (in Russian).
- Fanourakis, N., Tsekoura, L. & Nanou, E. 2000. Morphological characteristics and powdery mildew resistance of Cucumis melo land races in Greece. *Proceedings of Cucurbitaceae 2000. The 7th EUCARPIA Meeting of Cucurbit Genetics and Breeding. Acta Horticulturae* **510**, 241–246.
- Frolov, V.V. 1996. Species structure of fungi powdery mildew fungi in melon in Kherson. Proceedings of the international conference “Selection and technology: cucurbits production”; pp. 114–117. *Hola Pristania* (Ukrainian Science Research Institute of Southern Vegetable and Melon Crops, Kherson) (in Russian).
- Fukino, N., Kuniyama, M. & Matsumoto, S. 2004. Characterization of recombinant inbred lines derived from cross in melon (*Cucumis melo* L.) “PMAR No 5×Harukei No3”. *Breeding Science* (Japan) **54**, 141–145.
- Hosoya, K., Narisawa, K., Pitrat, M. & Ezura, H. 1999. Race identification in powdery mildew (*Sphaerotheca fuliginea*) on melon (*Cucumis melo*) in Japan. *Plant Breeding* **118**, 259–262.
- Kuzuya, M., Hosoya, K., Masuya, Y.H., Tomita, K. & Ezura, H. 2000. Histological observations of powdery mildew resistance in diploid and haploid melons. *Proceedings of Cucurbitaceae 2000. The 7th EUCARPIA Meeting of Cucurbit Genetics and Breeding. Acta Horticulturae* **510**, 71–77.
- Lebeda, A. & Kriskova, E. 2000. Interactions between morphotypes of *Cucurbita pepo* and obligate biotrophs (*Pseudoperonospora cubensis*, *Erysiphe cichoracearum* and *Sphaerotheca fuliginea*). *Proceedings of Cucurbitaceae 2000. The 7th EUCARPIA Meeting of Cucurbit Genetics and Breeding. Acta Horticulturae* **510**, 219–229.
- McCreight, J.D. 2001. Reactions of melon powdery mildew resistance sources to race 2U.S. In *XXVIth International Horticultural Congress, Acta Horticulturae* **637**, 10.
- McCreight, J.D. 2006. Melon–powdery mildew interactions reveal variation in melon cultigens and *Podosphaera xanthii* races 1 and 2. *J. Amer. Soc. Hort. Sci.* **131**(1), 59–65.
- Nayar, N.M. & More, T.A. 1998. *Cucurbits*. Enfield: Science Publishers, 340 pp.
- Perez-García, A., Mingorance, E., Rivera, M. E., Del Pino, D., Romero, D., Tores, J. A., & De Vicente A. 2006. Long-term Preservation of *Podosphaera fusca* Using Silica Gel. *J. Phytopathology* **154**, 190–192.
- Rankovic, B. 2003. Powdery mildew fungi (order Erysipales) on plants in Montenegro (Chernogoria). *Mycology and Phytopathology* (Russian Acad. of Sci., Leningrad Sci. Publ.) **37**(3), 42–52.
- Sakata, Y., Kubo, N., Morishita, M., Kitadani, K., Sugiyama, M. & Hirai M. 2006. QTL analysis of powdery mildew resistance in cucumber (*Cucumis sativus* L.). *Theor. Appl. Genet.* **112**, 243–250.
- Snedecor, G.W. & Cochran, W.G. 1976. *Statistical Methods* (6th ed.). Oxford & IBH, New Delhi.