scribing the relationship between electrophoretic distance and time are currently unreliable[13]. Nevertheless, the relative degree of temporal isolation between species can be appreciated when compared to levels of differentiation between conspecific populations. For example, similarities among geographic samples of continuously distributed North American fishes typically have I = 0.95–0.98[10, 14, 15]. In Lake Malawi, similarities among 3 geographic samples of *Labeotropheus fulleborni* (table 1, B) ranged from I = 0.940 to 0.977. 8 separate comparisons between endemic cichlids exhibited I >0.95, 3 of which had I in excess of 0.980 (table 1, A–D, C–E, F–G). Against the background of intraspecific divergence, most interspecific differences are trivial and suggest very recent genetic continuity. This idea is consistent both with electrophoretic estimates in other cichlid fishes (Kornfield, Ritte, Richler and Wahrman, in preparation) as well as with stratigraphical estimates of divergence (5000 years) for the endemic cichlids of Lake Nabagaboo, Uganda[3].

The high degree of similarity among these fishes makes it imperative that specific status be critically established. Viable hybrids can be artifically produced between some Mbuna[16], but hybrids in the lake are unknown[2]. The 7 Malawi endemics differ in breeding coloration and morphology[17], and most species are ecologically distinguishable[2]. However, morphological and ecological distinctness may be misleading. In New World cichlids, discrete ecomorphotypes may exist sympatrically within a single species of *Cichlasoma*[18]. Though some evidence has cast doubt upon this interpretation[19, 20], recent ethological observations support conspecificity of trophic morphs (Taylor, personal communication).

Morphological polymorphism must be considered for the Lake Malawi cichlids. If any of these endemics were conspecific, allele frequencies at all loci would be the same for morphotypes collected simultaneously at any locality. That is, panmaxia would ensure allelic uniformity among morphs. Allele frequencies, however, differ significantly among species collected sympatrically in Lake Malawi (table 2). This genetic evidence further supports reproductive isolation among these species.

Several recent studies have also reported high similarities among reproductively isolated taxa[10, 14, 21]. The apparent discrepancy between such similarities and those presumably typical of most other species emphasizes that a general timetable for taxonomic differentiation may not be realistic. That is, rates of speciation may differ dramatically among taxonomic groups[22, 23]. Though structural genes provide convenient benchmarks to assess the rates of phyletic evolution, the structural loci are apparently not involved in speciation per se.

Table 2. Allele frequencies from endemic Malawi cichlids collected in sympatry at 2 localities*

Locus	Allele	Locality 1					Locality 2	
		A	B	C	D	E	F	G
PGI-1	1.00	1.0	0.89	1.0	1.0	0.99	0.83	0.99
	1.05		0.11			0.01	0.17	0.01
	n	80	97	90	102	111	48	100
GP-1	1.00	0.69	1.0	1.0	1.0	1.0	1.0	1.0
	1.10	0.31						
	n	26	44	74	80	96	62	96
GP-3	1.00	1.0	1.0	1.0	1.0	1.0	0.79	1.0
	1.05						0.04	
	0.95						0.17	
	n	52	46	74	102	96	56	100
EST-1	0.85	0.23						
	0.90	0.61	0.09	0.21	0.52			
	0.95	0.11	0.55	0.38	0.06	0.01		
	1.00		0.10	0.10	0.40	0.08		
	1.05			0.02		0.86		
	0.00	0.05	0.26	0.29	0.02	0.05		
	n	48	90	97	102	108		
LDH	1.00						0.44	0.64
	1.10						0.56	0.36
	n						64	98

*Locality 1 (14° 02'00" S; 34° 54' E); Locality 2 (14° 17' S; 34° 56' E). Species and loci abbreviations as in table 1.

13 A. C. Wilson, S. S. Carlson and J. J. White, A. Rev. Biochem. *46*, 573 (1977).
14 M. S. Johnson, Copeia *1975*, 662.
15 J. C. Avise and M. H. Smith, Evolution *28*, 42 (1974).
16 P. Loiselle, Buntbarsche Bull. *27*, 9 (1971).
17 E. Trewavas, Ann. Mag. nat. Hist. *16*, 65 (1935).
18 R. D. Sage and R. K. Selander, Proc. natl Acad. Sci. USA *72*, 4669 (1975).
19 K. R. McKaye and N. M. McKaye, Evolution *31*, 647 (1977).
20 A. J. Ribbink, Nature *267*, 243 (1977).
21 H. L. Carson, W. E. Johnson, P. S. Nair and F. M. Sene, Proc. natl Acad. Sci. USA *72*, 4521 (1975).
22 A. C. Wilson, G. L. Bush, S. M. Case and M. C. King, Proc. natl Acad. Sci. USA *72*, 5061 (1975).
23 L. Van Valen, Nature *252*, 298 (1974).

Biological relationship of cucumis virus-3 and powdery mildew fungus in bottle gourd

M. A. Qureshi and C. D. Mayee

Department of Plant Pathology, Marathwada Agricultural University, Parbhani 431 402, M. S. (India),
27 July 1977

Summary. 2 kinds of interrelationship between cucumis virus-3 and powdery mildew fungus in bottle gourd were noted. The fungal conidia transmitted the mosaic virus infection from infected to healthy bottle gourd plants with ease. The transmission relation was found to be external. Powdery mildew symptoms were restricted on mosaic infected leaves and appeared late, while virus symptoms appeared precociously in mildew infected plants.

Viruses and fungi interact in many ways in their host plants. Fungal transmission of plant viruses has been established beyond doubt and fungus vector-virus relationship has been extensively studied[1]. Synergism or antagonism between viruses and non-vector fungal pathogens in plants has also been described by many workers[2–11]. However, the interaction between the vector fungus and the transmitted virus in respect of disease development has not been sufficiently explored. Moreover, the majority of the examples of transmitting fungi are found in soil-borne group where the infection is brought through host roots. The present investigation reveals both the aspects of relationship between the 2 leaf pathogens, namely *Oidium* sp. (powdery mildew) and cucumis virus-3 (mosaic) of bottle gourd (*Lagenaria siceraria* Mol. Standl.).

Materials and methods. Cucumis virus-3 strain M-BGMV was maintained in bottle gourd plants by sap inoculations[12]. The powdery mildew fungus identified by C.M.I., Kew (Surrey, England) as *Oidium* sp. (C.M.I.211351) was maintained by periodical subculturing on virus-free seedlings.

In fungal transmission studies, the mildew conidia from virus-infected as well as healthy leaves were aseptically collected and subjected to uniform dry dusting on injured and uninjured cotyledons. The treated plants were kept for 24 h in humid chambers before transferring them to isolated blocks for observations. In an attempt to know the mode of transmission, conidia harvested from virus-infected and virus-free leaves were treated for 6 h with specific virus antiserum, prepared by the method described earlier[12] and then transferred to test plants.

In a separate experiment, the interaction was studied by arranging various combinations of virus and mildew inoculations as shown in table 2. Each treatment was repeated thrice.

Results. Transmission: Cucumis virus-3 was easily transmitted by the conidia of *Oidium* sp., as is evident from the data in table 1. It is only in cases where the conidia were harvested from mosaic-affected leaves that transmission

Table 1. Transmission of mosaic virus by the conidia of *Oidium* sp.

Treatments	Percent infection	
	Mosaic*	Mildew**
Conidia from virus-infected leaves given transmission feeding on:		
Cotyledons injured	80	90
Cotyledons not injured	60	100
First true leaf injured	60	80
First true leaf not injured	70	90
Conidia from virus-free leaves given transmission feeding on:		
Cotyledon injured	0	100
Cotyledon not injured	0	100
First true leaf injured	0	80
First true leaf not injured	0	90
Antiserum-treated conidia from virus-infected leaves	0	60
Antiserum-treated conidia from virus-free leaves	0	70

* Reaction of virus was systemic. ** Reaction of powdery mildew was localized.

Table 2. Interaction between cucumis virus-3 and powdery mildew fungus in bottle gourd

Treatments	Appearance of symptoms (days)		Severity* of powdery mildew on days after inoculation				
	Mosaic	Mildew	4	6	8	10	12
Virus alone	7	–	–	–	–	–	–
Fungus alone	–	4	+	+++	++++	++++	++++
First virus**, then fungus	7	7	–	–	+	++	++
First fungus**, then virus	5	4	+	++	++	+++	+++
Virus and fungus simultaneously	6	7	–	–	++	+++	+++
By macerating both and inoculating simultaneously	8	–	–	–	–	–	–

* Severity grades: + = 1–25% Leaf area occupied by powdery growth; ++ = 26–50% Leaf area occupied by powdery growth; +++ = 51–75% Leaf area occupied by powdery growth; ++++ = 76–100% Leaf area occupied by powdery growth; ** Second inoculation 8 days after first one.

occurred irrespectively of injury or no injury made prior to transfers. The respective control treatments remained free from virus infection. Mildew developed in all the treatments and percent incidence did not significantly vary between the 2 sources. The period taken for systemic mosaic expression ranged between 15 and 20 days from the time of inoculation, while the localized mildew symptoms developed within 6–7 days. Fungal transmission of virus in external manner was indicated when the antiserum treated conidia acquired from mosaic-affected leaves failed to transmit the virus disease. Antiserum wash, however, did not inhibit powdery mildew infection.

Interaction: Table 2 depicts the results of combined infection on mosaic and mildew development. The powdery mildew was found restricted to green patches of mosaic-infected leaves. The percent leaf area thus occupied by the mildew was comparatively less than on virus-free leaves. The inhibition was further displayed by enhanced incubation period of mildew. Normally, the fungus developed symptoms within 4 days but on virus-infected leaves it was found to take consistently 7 days. In contrast, the mosaic symptoms appeared precociously on plants previously infected with the fungus. In case of simultaneous inoculations, only mildew appearance was delayed. The mildew did not develop in plants inoculated with crude sap prepared from mildew-mosaic-affected leaves.

Discussion. There are extremely few examples of mosaic viruses transmitted by powdery mildew fungi. Yarwood[13] demonstrated that tobacco mosaic virus (TMV) contaminated conidia of *Sphaerotheca lanestris* (powdery mildew) from oak plants when heavily dusted on *Chenopodium quinoa* leaves produced lesions resembling those of local lesions of virus which were followed by systemic infection. Nienhaus[14] extracted strains of TMV from conidia of *S. lanestris* and *Erysiphe graminis*. From the present studies it is evident that the conidia of *Oidium* sp. acquire the virus as an external contaminant. The fact that antiserum-dipped conidia failed to transmit the virus indicated the inactivation of surface-borne virus particles and thus supported the view of external relationship. The virus is mechanically transmissible and hence a slight injury made by the infecting conidium during germination, entrance, appresorium formation or haustorial initiation might have been sufficient to gain virus entry into proper cells. Uninjured leaves prior to conidial transfer also brought successful virus infection, which suggests that the virus particles reach the appropriate site of replication.

1　R. G. Grogan and R. N. Cambell, A. Rev. Phytopath. *4*, 29 (1966).
2　A. C. Gohen and W. C. Schanthrost, Pl. Dis. Reptr *45*,641 (1961).
3　D. F. Bateman, Phytopathology *51*, 574 (1961), abstr.
4　J. H. M. Temink, An ultra structural study of Olpidium brassicae and its transmission of Tobacco Necrosis virus, p. 135. Ed. H. Veenman and N. V. Zonen Wageningen, 1971.
5　K. C. Chadha and S. P. Raychaudhuri, Indian J. agric. Sci. *36*, 133 (1966).
6　K. C. Chadha and S. P. Raychaudhuri, Indian Phytopath. *21*, 171 (1968).
7　K. Mahmood, M. Akram, S. Qamar, A. Naqvi and M. M. Alam, Indian Phytopath. *27*, 627 (1974).
8　K. O. Muller and J. Munro, Anns appl. Biol. *38*, 765 (1951).
9　S. Blumer, L. Stadler and A. Horder, Phytopath. Z. *25*, 39 (1955).
10　S. P. S. Beniwal and R. T. Gudauskas, Phytopathology *64*, 1197 (1974).
11　W. C. Schanthrost, Phytopathology *49*, 562 (1959).
12　M. A. Qureshi, Thesis Agr. Univ. Parbhani (M. S.) India, 1977.
13　C. E. Yarwood, Pl. Dis. Reptr *55*, 342 (1971).
14　F. Nienhaus, Virology *46*, 504 (1971).