# Effect of Grapevine Fanleaf Virus on the Reproduction and Survival of its Nematode Vector, Xiphinema index Thorne & Allen

SITANATH DAS AND D. J. RASKI<sup>1</sup>

Abstract: Studies on the virus-vector interaction between the grapevine fanleaf virus (GFV) and its nematode vector, Xiphinema index, indicate the virus had no measurable effect on the rate of reproduction of its vector, but significantly influenced survival of the nematodes.

An important aspect of a virus-vector relationship is the biological effect of one on the other, a phenomenon well recognized in many plant viruses. Several instances of such effects, either detrimental or beneficial, have been described for plant viruses and their arthropod vectors. Little information, however, is available on nematode-borne viruses.

Ayala (1) presented experimental evidence to show that the California strain of tobacco rattle virus (CTRV) directly or indirectly increases the rate of reproduction of the vector, Trichodorus allius Jensen. Recently, Roggen (10) reported that grapevine fanleaf virus (GFV) affects the osmoregulatory mechanism of Xiphinema index Thorne & Allen. He also suggested the increase in size of nuclei in the lateral chords, enlargement of the pseudocoelomic cavity and increase in the amount of RNA in the viruliferous nematodes may be the effects of GFV. A virus-like agent has been associated with harmful effects on a nematode which is not a virus vector, Meloidogyne incognita (Kofoid & White) Chitwood (4). In the present study attempts were made to determine the influence of GFV on the rate of reproduction and survival of its vector, X. index.

## MATERIALS AND METHODS

A virus-free population of X. index was collected from Lodi, California and raised on rooted cuttings of fig (Ficus carica L.) and healthy grape (Vitis rupestris Scheele 'St. George'). The nematodes were wet-screened from soil by a combination of Cobb's screening technique (20- and 100-mesh screens) and Baermann funnel extraction. They were introduced into potted soil through small holes around the plants with the help of a small dropper or pipette. After all the nematodes were released, the holes were covered and the pots were watered lightly. Viruliferous nematodes were also recovered and added in the same way.

The source of GFV isolate was the infected clones 1 and 2 of *Vitis vinifera* L. 'French Colombard' (A-5-1) from the old vine collections of Plant Pathology Department, University of California, Davis, maintained by Dr. W. B. Hewitt and Dr. A. C. Goheen.

Methods of virus assay were after Taylor and Hewitt (13) and techniques of keeping nematodes in moist soil were according to Taylor and Raski (12).

## EXPERIMENTS AND RESULTS

EFFECT OF GFV ON REPRODUCTION: A population of virus-free nematodes (about 10,000) including larvae and adult females was allowed to feed on GFV-infected 'French Colombard' rooted cuttings. After 20 days, the nematodes were recovered, their infectivity was tested (adults: 100%, juve-

Received for publication 25 July 1968.

<sup>&</sup>lt;sup>1</sup> Department of Nematology, University of California, Davis, California 95616. Present address of senior author: Department of Mycology & Plant Pathology, University of Agriculture and Technology, Bhubaneswar-3, Orissa, India. Thanks are due to Dr. Wm. B. Hewitt, Dr. A. C. Goheen, and Dr. T. A. Shalla for their helpful suggestions.

Time in months	Viruliferous		Virus-free	
	Recovery/pot <sup>a</sup>	Juveniles/adult <sup>a</sup>	Recovery/pot	Juveniles/adult
0	$325 \pm 23.7$	$1.28 \pm 0.92$	$325 \pm 41.0$	$1.28 \pm 0.92$
1/2	$257 \pm 41.6$	$1.46 \pm 0.36$	$391 \pm 56.6$	$2.67 \pm 0.40$
1	$755 \pm 195.4$	$4.50 \pm 0.95$	$720 \pm 212.7$	$5.05 \pm 1.82$
2	$2.798 \pm 215.0$	$9.30 \pm 1.04$	$2,295 \pm 264.7$	$6.15 \pm 0.21$
3	$3,724 \pm 578.6$	$7.06\pm2.04$	$3,275 \pm 806.4$	$4.16 \pm 0.45$

TABLE 1. Increase in population of viruliferous and virus-free Xiphinema index maintained on figroots.

<sup>a</sup> Mean of three replications and standard error of the mean.

niles: 60%) and finally, they were distributed into pots containing rooted young fig plants in aliquots of  $325 \pm 41$  nematodes per pot. In the same manner another population, which did not have access to any virus source, was first released on roots of healthy 'French Colombard' plants and then transferred into pots with fig as control. After intervals of  $\frac{1}{2}$ , 1, 2, and 3 months the nematodes were extracted separately from the three replicates of the viruliferous series and compared with those of the virus-free series.

Nematode counts are presented in Table 1. The rate of reproduction has been estimated on the basis of juveniles per adult ratio. No statistically significant differences in rate of reproduction between viruliferous and virus-free nematodes were detected.

EFFECT OF GFV ON SURVIVAL: Viruliferous and nonviruliferous nematodes were obtained from diseased and healthy 'French

TABLE 2. Survival of viruliferous and virus-free *Xiphinema index* held in soil in the absence of host plants.

	Average nematode counts per pot		
months	Viruliferous	Virus-free	
0	325	325	
1/2	155	137	
1	154	120	
2	131	86	
3	111	78	

L. S. D. at 5% level = 40.28. L. S. D. at 1% level = 55.48. Colombard' plants, respectively. About 300-350 nematodes, including developmental stages, were introduced into moist sterilized soil in each pot. Every treatment was replicated three times. After  $\frac{1}{2}$ , 1, 2, and 3 month intervals the nematodes were collected and counted. Average counts of surviving nematodes are presented in Table 2.

Statistical treatment of the data by an analysis of variance showed significantly higher percentage of survival of virus-carrying nematodes (significance level 5%). The design of the experiments with only three replicates is a definite limitation in interpret-



FIG. 1. Survival of viruliferous (V<sup>+</sup>) and virusfree (V<sup>-</sup>) Xiphinema index ( $\Diamond$ ,  $\heartsuit$  = adult males and females;  $\theta_{4}$  = fourth stage pre-adults;  $\theta_{2+3}$  = second and third stage juveniles) maintained in fallow soil.

ing data from the studies. However, the significant differences (at 5% level) in rate of survival of viruliferous nematodes suggests there is some effect on infected vectors and further study is encouraged. There was a rapid decline in the number of juveniles after  $\frac{1}{2}$  month, although a few of them were usually encountered even after three months (Fig. 1). It was also observed that the body contents were relatively more depleted and size was smaller toward the end of the experiment in the case of virus-free nematodes.

## DISCUSSION

Retention of infectivity of viruses in their vectors for fairly long periods, 8 months for arabis mosaic virus and GFV (2, 12) and 10 months for tobacco rattle virus (14), points to an intimate interaction between the virus and vector. To what extent the virus affects the nematode physiology is not fully understood. On the basis of available literature on changed physiology of insect vectors, Maramorosch (6) categorizes them as beneficial or harmful effects of virus on vector. Thus, increased rate of reproduction of T. allius on CTRV infected tobacco (1) may have been due to beneficial effect of the virus on the nematode rather than an effect of the diseased host on the vector (7). Raski and Hewitt (8) speculated that failure of adult Longidorus elongatus to transmit tomato black ring virus may have been due to harmful effects of the virus on the nematode. Possible relationships between plant viruses and root-knot nematodes (nonvectors) have also been suggested in the studies of Loewenberg et al. (4). Parallel instances in insect-borne viruses are very common (3, 5, 6, 11). In this investigation, however, no significant correlation could be established between GFV and rate of multiplication of X. index. Roggen (10) observed changes in osmoregulation, increase in size of nuclei in hypodermal chords, and relatively higher content of RNA in GFV-carrying X. index. Evidence here indicates X. index survival during starvation may be significantly higher for viruliferous adults than for virus-free nematodes (Table 2). Maramorosch (5) observed that the leafhopper, Dalbulus maldis, which acquires and retains aster yellow virus in its haemolymph but does not transmit it, survived for longer periods on infected China aster, Callisterus chinensis. Recently, Raski et al. (9) provided evidence that X. index survived 4.5 years in the field and advanced the hypothesis that this might be due to access of the nematodes to virus-infected grape roots remaining from the previous crop. Increased RNA content of infective nematodes may be related to improved survival by affecting the protein metabolism of the nematodes. GFV seems to play an important role in its vector's physiology.

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