Table 1 3	The number of	nemetodes of	f rice root recovered	from within and	outside rice roots
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		Nematode numbers (Total 15)			
The No. of days after which observation was made following nematode inoculation	Treatments, the number of plates observed at different intervals	Within the roots	Within the medium	In lactophenol washings	
10 days	Carbofuran 1	0	11+4 (enmeshed with roots)	0	
	Phorate 1	6	8	1	
	Phorate 2	2	13	0	
	Control 1	Ō	15	0	
	Control 2	6	8	1	
17 days	Carbofuran 1	0	13	2	
	Carbofuran 2	0	12	3	
	Carbofuran 3	0	10	5	
	Phorate 1	9	5	1	
	Phorate 2	5	9	!	
	Phorate 3	5	9	1	
	Control 1	6	4	5	
	Control 2	5	10	0	
20 days	Carbofuran 1	0	12	3	
	Carbofuran 2	0	12	3	
	Carbofuran 3	0	9	6	
	Carbofuran 4	0	11	4	
	Phorate I	5	5	5	
	Phorate 2	4	9	2	
	Phorate 3	3	10	. 2	
	Control 1	5	7	3	
	Control 2	2	8	5	
	Control 3	7	6	2	
	Control 4	3	2	10	

efficacy of systemic nematicides is checking parasitic nematode penetration of the host roots.

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- 1. Curtis, O. F. and Clark, G. D., An Introduction to Plant Physiology, McGraw Hill Book Company, London, 1949, p. 752.
- 2. Goodey, J. B., Tech. Bull. No. 2, Minst. Agric., London, H.M.S.O., 1957, p. 47.
- 3. Van Gundy, S. D., Nematologica, 1958, 3, 112.
- 4. Krusberg, L. R., Nematologica, 1959, 4, 187.

## EGG ADAPTATION OF INFECTIOUS BURSAL DISEASE VIRUS

A. B. KULKARNI, V. L. PARANIPE, P. D. SARDESHPANDE and S. M. AJINKYA

Department of Microbiology and Pathology, Bombay Veterinary College, Bombay 400012, India.

INFECTIOUS bursal disease (IBD) or Gumboro disease is emerging as a viral disease in India. Recently, during the investigations into a series of Ranikhet disease (RD) outbreaks, in a number of broiler farms located around the city of Bombay, it was observed that there was an association of IBD in all the cases<sup>1,2</sup>. The present work was undertaken to study the behaviour of the local isolate of IBD<sup>2</sup> virus in serial egg passages in chicken embryo. Antiserum for confirmation was obtained from Intervet Laboratories, U.K. and the immune serum was raised in rabbits<sup>1</sup>.

Chicken embryos (ten day-old) were inoculated with 0.2 ml of virus suspension by chorio-allantoic route. The embryos showing death after 48 hr and those still surviving on the sixth day post-inoculation were chilled at 4°C. Embryos were harvested after overnight chilling. Chorio-allantoic membranes (CAMS) and embryos were examined for the presence of lesions, characteristic of infectious bursal disease virus (IBDV). CAMS were washed in phosphate buffer saline (PBS, pH 7.2) and 20% suspension (w/v) was prepared. This suspension was inoculated in another batch of embryos and ten serial passages were carried out in a similar way. To demonstrate the presence of viral entity at every passage level agar gel precipitation test (AGPT) was performed.

Egg infective dose (50%) (EID<sub>50</sub>) of passage ten 1BDV (IBDV/10) was determined<sup>3</sup> and the neutralization test was performed by mixing equal volumes of 10 EID<sub>50</sub>, 100 EID<sub>50</sub> and 1000 EID<sub>50</sub>/0.2 ml of the virus and the heat inactivated (56°C for 30 min) immune and normal serum to confirm the identity of virus. Virusserum mixtures were incubated at 37°C for 1 hr and 0.2 ml was inoculated in embryonating eggs by CAM route.

The embryos showed 100 % and 45 % mortality after 72 hr in passage numbers one and two respectively. However, in the subsequent passages, no mortality was observed and hence the embryos were sacrificed on 6th day post-inoculation. The CAMS of dead and sacrificed embryos showed congestion, edema and thickening. Embryos showed curling after fifth passage. In the subsequent passages both curling and dwarfing were noticed. Thus the CAM route was successfully used for the serial passages of IBDV in chicken embryo as reported earlier<sup>4-6</sup>. The apparent reason for the absence of mortality after second passage is not fully understood. The route of inoculation is important in influencing embryo mortality<sup>5,7</sup>, while the absence of mortality can be attributed to the propagation of the agent in fertile eggs from non-susceptible flock and the selection of wrong embryo materials for sub passages. Failure to cause embryo deaths in subsequent passages in the present study cannot be explained on this basis since: (i) there was no evidence of infection of IBD on the farms from which the eggs were utilized for the propagation of the agent, (ii) other characteristic lesions of IBDV such as congestion and edema of CAM, curling of embryo etc were noticed in the subsequent passages, (iii) CAMS were utilized for the serial passages which were shown to have higher titres than allantoamniotic fluids<sup>7</sup>, (iv) presence of viral entity was demonstrated at every passage level by AGPT and

(v) a titre of  $10^{6.38}$  EID<sub>50</sub>/0.2 ml was obtained at IBDV/10. It is therefore necessary to use specific pathogen-free eggs for such purposes.

In conclusion the findings suggest that the isolate was well adapted to the chicken embryo confirming the earlier reports<sup>4, 5, 8</sup>.

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- 1. Ajinkya, S. M., Survashe, B. D. and Sardeshpande, P. D., Indian Vet. J., 1980, 57, 265.
- 2. Manda, A. V. and Paranjpe, V. L., Curr. Sci., 1979, 49, 221.
- 3. Reed, L. J. and Muench, H., Am. J. Hyg., 1938, 27, 493.
- 4. Wilson, P. H., (1964) Cited by Faragher, J. T., Vet. Bull., 1972, 42, 361.
- 5. Snedekar, C., Wills, F. K. and Moulthrop, J. M., Av. Dis., 1967, 11, 519.
- 6. Jayaramaiah, B. and Mallick, B. B., Indian J. Anim. Hlth., 1974, 13, 99.
- 7. Hitchner, S. B., Poult. Sci., 1970, 49, 511.
- 8. Dorn, P., Kronthaler, O. and Schindler, P. 1968, Cited by Faragher, J. T., Vet. Bull., 1972, 42, 361.

A NEW RECORD OF DIAPHANOSOMA SENEGALENSIS GAUTHIER, 1951 (CLADOCERA, SIDIDAE) FROM MADURAI, SOUTH INDIA.

## K. VENKATARAMAN and S. KRISHNASWAMY,

Department of Environmental Biology, School of Biological Sciences, Madurai Kamaraj University, Madurai 625 021 India.

ALTHOUGH Cladocera are among the commonest micro-crustaceans, a perusal of literature of this group shows that they are poorly known taxonomically throughout India<sup>1-4</sup>. There is no comprehensive systematic study of the species of Cladocera in Tamil Nadu except that of a 'guide to the freshwater organisms<sup>2</sup>, which is a preliminary attempt to identify eight of the common genera occurring in Madurai. The present work was undertaken in 1979 to study the