RETENTION AND TRANSMISSION OF R*ice yellow mottle virus* (RYMV) BY BEETLE VECTORS IN COTE D'IVOIRE

M.E. ABO[1,3], M.D. ALEGBEJO[2], A.A. SY[3], and Y. SERE

[1]National Cereals Research Institute (NCRI), Badeggi, P.M.B. 8, Bida, Nigeria

[2]Department of Crop Protection, Institute for Agricultural Research Faculty of Agriculture,

Ahmadu Bello University, PMB 1044, Zaria, Nigeria.

[3]West Africa Rice Development Association (WARDA), 01 BP 2551, Bouaké, Côte d'Ivoire.

ABSTRACT

The investigation to determine the ability of some rice beetles to transmit Rice yellow mottle virus (RYMV), genus sobemovirus, was carried out in the screen house in Côte d'Ivoire. In the study visual assessment based on Standard Evaluation System (SES) for rice and Enzyme Linked Immunosorbent Assay (ELISA) test of rice leaves from plants, which the viruliferous insects were placed to feed and transmit the virus, were used to detect infection. The Chrysomelidae (*Trichispa sericea* Guerin, *Chaetocnema pulla* Chapius) and a phytophagus Coccinelid (*Chnootriba similis* Thunberg/*Epilachna similis* Mulsant) transmitted RYMV in the screen house test. *T. sericea* and *C. similis* transmitted it in a semi-persistent manner, while *C. pulla* transmitted it in a persistent manner. *C. similis* that is phytophagous is reported in this study for the first time as a vector of RYMV.

Keywords : rice, yellow mottle, transmission, beetle, Côte d'Ivoire.

RESUME

RETENTION ET TRANSMISSION DU VIRUS DE LA PANACHURE JAUNE DU RIZ (RYMV) PAR LES COLEOPTERES VECTEURS EN COTE D'IVOIRE

*Une étude a été menée dans un insectarium en Côte d'Ivoire en vue de déterminer la capacité de certains coléoptéres du riz à transmettre la panachure jaune du riz (RYMV). Pendant l'étude, l'évaluation visuelle basée sur le système d'évaluation standard (SES) du riz et le test ELISA des feuilles des plants de riz sur lesquelles les pucerons virulifères avaient été placés pour se nourrir et transmettre le virus, ont été utilisés pour détecter l'infection. La chrysomèle (*Trichispa sericea *Guerin,* Chaetocnema pulla (*Chapius*) *et une coccinelle phytophage (*Chnootriba similis *Thunberg/*Epilachana similis *Mulsant) ont transmis la panachure (RYMV) dans le test de l'insectarium.* T. sericea *et* C. similis *l'ont transmise de manière semi-persistante. l'a transmise.* C. similis *qui est phytophage est rapporté pour la première fois dans cette étude comme vecteur du virus de la panachure jaune du riz (RYMV).*

Mots clés : *riz, panachure jaune, transmission, coléoptère, Côte d'Ivoire.*

INTRODUCTION

Rice yellow mottle virus (RYMV) belongs to the sobermovirus group (Seghal, 1981; Hull, 1988). It is a serious constraint to rice production in many countries in the heart of Africa and some adjoining islands (Abo *et al.*, 1998).

Rice is attacked by more than 80 species of insects (Alam *et al.*, 1984). However, a few insects belonging mainly to the family Chrysomelidae have been found to transmit RYMV in East Africa (Bakker, 1971, 1974 and 1975) and Madagascar (Reckhaus and Andriamasintseheno, 1995). These insects include *Trichispa* spp., *Sesselia pussilla*, *Dicladispa* spp., *Chaetocnema* spp and those in the genus near *Apophylia* (Bakker, 1974, 1975). In West Africa *T. sericea* Guerin has been reported to cause considerable damage to young rice crops

in Niger and Mali (Sy, 1994), and the damage is widespread on farmers' farms in Côte d'Ivoire. *Chaetocnema* spp has been suspected to be resplnsible for the spread of RYMV in West Africa (Raymundo *et al.*, 1979 ; Raymundo, 1980 ; Fomba, 1990). Also high populations of some of these insects and their associated damages have been observed on rice fields in the West African sub-region (Akibbo-Betts and Raymundo, 1980 ; Breniere, 1983 ; Alam *et al.*, 1984) and where RYMV incidence is increasing (Sy, 1994). However, their population peaks were not correlated with RYMV incidence (Heinrichs *et al.*, 1995). Furthermore, most of the observations were not substantiated with adequate quantitative data.

The objective of this investigation was to determine the ability of some of the beetles found in large numbers in rice ecologies in Côte d'Ivoire, West Africa to retain and transmit RYMV in screen house test.

MATERIAL AND METHODS

Four types of beetles (*Trichispa sericea* Guerin, *Chaetocnema pulla* Chapius, *Altica* species, and *Chnootriba similis* Thunberg) were collected from rice fields at Mbe/Bouake and at Sakassou, Côte d'Ivoire with a sweep net or aspirator and maintained in entomological cages. However, the adults of *T. sericea* and *Altica* species and *Chnootriba similis* were picked using a combination of sweep net and camel hair paintbrush. The eggs, larvae and nymphs of *T. sericea* were collected from the leaves of rice and *Leersia hexandra* Sw. They were reared in cages in the laboratory at the West Africa Rice Development Association (WARDA) head-quarters, Bouake, Côte d'Ivoire.

The adults of the field collected insects and those reared in the laboratory were picked and starved for 3 hours and then the insects were placed on RYMV infected plants for 3 days to feed and acquire the virus. Thereafter, the insects from each group were picked and placed on a healthy, 14 day old seedling of Bouake 189, the RYMV susceptible check rice variety for feeding transmission test for another period of 3 days. For *C. pulla* the plants on which they fed were agitated and the insects flew and settled on the walls of the cages. Then the plants were removed and another set of healthy plants were replaced in the cages for which the insects returned and settled on them and continued their feeding and possibly the transmission of the virus. The exact number of insects transferred on each seedling and the total seedlings tested for each insect species are indicated in table 1. This number was determined by the total recovery of life insects after the virus acquisition period because mortality of the insects was high on the infected plants. At the end of each test period spraying with insecticide killed the insects after the removal of the test plants. The plants were then kept in the screen house and monitored for the symptoms of RYMV for 3 months.

Table 1 : The transmission and non-transmission of *Rice yellow mottle virus* by some rice beetle insects in screening tests.

Transmission et non-transmission du virus de la panachure jaune du riz par des scarabées du riz en essai de criblage.

Common Name	Scientific Name & Family	Number of Viruliferous Insects/Plant[c]	Number of Test Plants[c]	Source of Insects	Number of Infected Plants/Number of Plants Tested
Hispid Beetle	*Trichispa sericea* Guerin,	10	15	a	5/15(33)
	Chrysomelidae (Hispinae)	10	15	b	7/15(47)
Flea Beetle	*Chaetocnema pulla* Chapius, Chrysomelidae (Halticinae)	10	15	b	8/15(53)
Flea Beetle	*Altica* spp., Chrysomelidae (Halticinae)	10	10	b	0/10(0)
Ladybird beetle	*Chnootriba similis* Thunberg, Coccinelidae	10	10	b	7/15(47)

a : *Laboratory reared insects,* ***b :*** *Field collected insects,* ***c :*** *RYMV susceptible check variety (Bouake 189) Numbers in bracket are percentages.*

To determine the mode of transmission of RYMV by *T. sericea, C. pulla, Chootriba similis* and *Altica* species the adults of the insects were allowed virus acquisition period of 3 days. Thereafter, one insect each was picked and placed on each of the 14 days old seedling of Bouake 189 and at different time regimes of 15, 30, 45, 60 minutes and 1, 2 days. Five seedlings per each time regime were tested. The test plants were then placed in the screen house and monitored for symptoms of RYMV for the period mentioned above.

The retention period of RYMV by the insects was carried out as outlined in the preceding paragraph. However, a single insect was picked and placed on a single healthy young seedling of Bouake 189 on day one. At the end of day one and subsequent days up to 10 days the same insect was picked and placed on the next healthy young plant in another cage.

The method for assessing successful trnsmission of RYMV by each group of insects was based on Standard Evaluation Scale (SES) for rice (IRRI, 1988) and Enzyme Linked Immunosorbent Assay (ELISA) test (Clark and Adams, 1977) on rice leaves. In the ELISA test the working dilution for the antigen was 1:10 while those of the antibody and conjugate was 1:1000. The alkaline phosphatase (ALP) enzyme was conjugated to antiglobulin (Koenig, 1981) and antigen was directly trapped on the micro titre plate and detected by conjugate RYMV antibody introduced after the antigen. The blocking solution contained phosphate buffered saline and 3 % of 99 % fat free milk (Marvel). Each antigenic leaf sample and the control samples were replicated in two wells of ELISA micro titre plate. About 100µ of 0.6mg/ml of 4-nitrophenyl phosphate buffers at pH 9.8 were dispensed into each well of the plate and incubated at 37^{o}C for 30 minutes. Colour change was measured with METERTECH 960 ELISA Microreader after 1 hr. Absorbance values (A 405 nm) were accepted as positive (+) when the reading was greater than twice the mean absorbance of the virus free control sample. Any other value below that was considered as negative (-). For the ability of the insects to transmit RYMV the result was expressed as the number of plants infected to the number tested. Descriptive statistics was employed to present the results.

RESULTS AND DISCUSSION

The results of the ability of the insects to transmit RYMV are presented in table 1. Two chrysomelidae (*T. sericea* and *C. pulla*) and a coccinelidae (*C. similis*) transmitted RYMV efficiently. The *Altica* spp could not transmit RYMV in the screening test. In this test it was observed that the symptoms of RYMV first appeared on the youngest emerging leaves at about 21 days after insect feeding transmission test.

The mode of transmission and retention period of RYMV by three insect identified are presented in Tables 2 and 3. *T. sericea* and *C. similis* retained and transmitted the virus up to 2 days while *C. pulla* retained and transmitted RYMV for 6 days. The adults of *T. sericea, C. pulla* and *C. similis* transmitted RYMV in an efficient manner and are therefore, potential vectors of RYMV. *T. sericea* and *C. pulla* have been reported in Kenya as vectors of RYMV (Bakker, 1974 and 1975). This is a first report of *C. similis*, a phytophagous coccinelid as a vector of RYMV. *C. similis* transmitted RYMV as efficiently as *T. sericea* and *C. pulla.* These three vectors are prevalent throughout Africa (Grist and Lever, 1969 ; Breniere, 1983) and the adjoining island, Madagascar (Bouriquet, 1946).

T. sericea and *C. similis* transmitted it in a semi-persistent manner while *C. pulla* transmitted in a persistent manner according to the classification scheme of Papkova (1989). The above findings conforms with the earlier report in Kenya by Bakker (1974 and 1975) who indicated that *T. sericea* retained and transmitted RYMV for 1 day whereas *C. pulla* did it for 8 days.

Table 2 : Mode of transmission of RYMV by *Trichispa sericea* Guerin, *Chaetocnema pulla* Chapius and *Chnootriba similis* Thunberg.

Mode de transmission du RYMV par Trichispa sericea Guerin, Chaetoenema pulla Chapius et Chnootriba similis Thunberg.

Insect Groups	Test Number	Number of insects per plant[a]	Time regimes/inferctivity					
			Minutes				Days	
			15'	30'	45'	60'	1	2
Prichispa sericea Guerin,	1	1	-	+	+	+	+	-
Chrysomelidae (Hispinae)	2	1	+	+	+	+	+	-
	3	1	+	+	+	+	+	-
Chaetocnema pulla	1	1	+	+	+	+	+	+
Chapius, Chrysomelidae	2	1	+	+	+	+	+	+
(Halticinae)	3	1	+	+	+	+	+	+
Chnootriba similis	1	1	+	+	+	+	+	+
Thunberg, Coccinelidae	2	1	+	+	+	+	+	+
	3	1	+	+	+	+	+	+

[a]Bouake 189, RYMV susceptible check.
+ : Positive test in both visual assessment and ELISA.
+ : Negative test in both visual assessment and ELISA.

Table 3 : Retention period of RYMV by *Trichispa sericea* Guerin, *Chaetocnema pulla* Chapius and *Chnootriba similis* Thurnberg in daily transfer test on healthy seedlings of Bouake 189.

Période de rétention du RYMV par Trichispa sericea Guerin, Chaetocnema pulla et Chnootriba similis Thunberg dans le test de transfert quotidien des plantules saines de Bouaké 189.

Insect group	Test Number	Number of insects per plant[a]	Time regimes/infectivity									
			1	2	3	4	5	6	7	8	9	10
Trichispa sericea Guerin,	1	1	+	-	-	-	-	-	-	-	-	-
	2	1	+	+	-	-	-	-	-	-	-	-
	3	1	+	-	-	-	-	-	-	-	-	-
Chaetocnema pulla Chapuis	1	1	+	+	+	+	+	-	-	-	-	-
	2	1	+	+	+	+	-	-	-	-	-	-
	3	1	-	+	+	+	+	-	-	-	-	-
Chnootriba similes	1	1	+	+	+	-	-	-	-	-	-	-
Thunberg,	2	1	+	+	+	-	-	-	-	-	-	-
	3	1	+	+	+	-	-	-	-	-	-	-

[a]Bouake 189, RYMV susceptible check.
+ : Positive test in both visual assessment and ELISA.
- : Negative test in both visual assessment and ELISA.

CONCLUSION

The adults of *T. Sericea, C. pulla* and *C. similis* transmitted RYMV very efficiently. Thus, these are potential vectors of RYMV. The prolonged retention of RYMV by these insect vectors is a sure way of spreading the virus in the field especially where the susceptible hosts are found and environmental factors are favourable.

ACKNOWLEDGEMENTS

We are grateful to the African Development Bank (AfDB) for a fellowship to carry out this work and to Mr. Olivier Kassoum A. Traore of the Entomology Unit, WARDA for his technical assistance.

REFERENCES

ABO (M. E.), (A. A.) SY and (M. D.) Alegbejo. 1998. *Rice yellow mottle virus* (RYMV) in Africa: evolution, distribution, and economic significance on sustainable rice production and management strategies. Journal of Sustainable Agriculture, 11 (2/3) : 85-111.

AKIBO-BETTS (D. T.) and (S. A.) RAYMUNDO. 1980. Aphids as rice pests in Sierra Leone. International Rice Research Newsletter, 3 (6) : 15-16.

ALAM (M. S.), (V. T.) JOHN and (K) ZAN. 1984. Insect pests and diseases of rice in Africa. In : Rice in Africa. IITA Rice Production Manual, IITA, Ibadan, Nigeria.

ALEGBEJO (M. D.). 1986. Aphid transmission of Pepper veinal mottle virus. Samaru Journal of Agriculture Research 4(1 & 2) : 71-77.

BAKKER (W). 1971. Three new beetle vectors of *Rice yellow mottle virus* in Kenya. Netherlands Journal of Plant Pathology, 77 : 201-206.

BAKKER (W). 1974. characterization and ecological aspects of *Rice yellow mottle virus* in Kenya. Agricultural Research, Doctoral thesis, University of Wageningen, The Netherlands, 152pp.

BAKKER (W). 1975. *Rice yellow mottle virus*. CMI/AAB Descriptions of Plant Viruses. No. 149.

BOURIQUET (G). 1946. Maladies du riz. In : (I) Lechevalier (Ed.) : Les maladies des plantes cultivées a Madagascar. Encycl. Mycol., Paris, 12 : 374-382.

BRENIERE (J). 1983. The principal insect pests of rice in West Africa and their control. West Africa and their control. West African Development Association, Monrovia, Liberia, 87p.

CLARK (M. F.) and (A. N.) ADAMS. 1977. Characteristics of the micro plate method of enzyme linked immunosorbent assay for the detection of plant viruses. Journal of General Virology, 34 : 475-483.

FOMBA (S. N.). 1990. Rice yellow mottle virus on swamp rice in Guinea. International Rice Research Newsletter, 15 : 6.

FULTON (J. P.), (R. C.) GERGERICH and (H. A.) SCOTT. 1987. Beetle transmission of plant viruses. Annual Review of Phytopathology, 25 : 111-123.

GRIST (D. H.) and (R. A. J. W.) LEVER. 1969. Pests of rice. Longmans, London, 3-9 pp.

HARRIS (V. W.). 1937. Annual report of the entomologist ; In : Rep. Dep. Agric. Tanganyika for 1936. Government Printer, Dares Salam. 88-94 p.

HEINRICHS (E. A.), (A. A.) SY, (S. K.) AKATOR and (I) OYEDIRAN. 1995. Seasonal occurrence of *Rice yellow mottle virus* on the WARDA research farm. Paper presented at the 1st International Symposium on RYMV held at WARDA, Mbe/Bouaké, Côte d'Ivoire, 18-22 September 1995.

HULL (R). 1988. The sobemovirus group. In : (I. R.). Koenig, (ed.), 113-146 p. The plant viruses Vol. 3. Polyhedral virus with monopartite RNA genomes Plenum Press, New York.

IRRI (International Rice Research Institute), 1988. The International Rice Testing Program-Standard Evaluation System (IRTP-SES) for rice. Los Banos, Laguna, The Philippines. 3rd Edition, 23 p.

KOENIG (R). 1981. Indirect ELISA methods for the broad specificity detection of plant viruses. Journal of General Virology, 55 : 53-62.

MATTHEWS, (R. E. F.). 1991. «*Plant Virology*». 3rd Edition Academic Press, London, 835pp.

NAULT (L. R.) and (E. D.) AMMAR. 1989. Leafhopper and plant hopper transmission of plant viruses. Annual Review of Entomology, 34 : 503-529.

PAPKOVA (K. V.). 1989. general Phytopathology (In Russian). Agropromizhat, Moscow, 398p.

RAYMUNDO (S. A.). 1980. Diseases of rice in Sierra Leone. Technical Bulletin, No. 1

Rokupr. Rice Research Station, Sierra Leone.

RAYMUNDO (S. A.), (I. W.) BUDDENHAGEN, (S. N.) FOMBA and (D. T.) AKIBBO-BETTS. 1979. Recent advances in the knowledge of rice viruses and resistance to a beetle transmitted mottle of rice in West Africa. In WARDA (Ed.). Rice breeding and varietal Improvement II. West Africa Rice Development Association, September 13-17, 1979

RECKHAUS (P. M.) and (H. E.) ANDRIAMA-SINTSEHENO. 1995. Development of an IPM Strategy to fight RYMV and Constraints to its Implementation in Madagascar. 1st International Symposium on RYMV, 18-22 Sept. 1995, Mbe, WARDA/ADRAO, Bouaké, Côte d'Ivoire.

SEGHAL (O. P.). 1981. Southern bean mosaicvirus group. In (E.) Kurstak, (Ed). Handbook of plant virus infections, comparative diagnosis Elsvier/North Holland Bio-medical press, Amsterdam, 91-121.

SY (A. A.). 1994. The Integrated Pest Management. Task Force (IPM-TF) Monotiring Tour in Mali, November 1994 (Available at WARDA Library).

WALLTERS (H. J.). 1969. Beetle transmission of plant viruses. Advances in Virus Research, 25 : 339-363.