New Strain of Okra Mosaic Virus in Nigeria

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ABSTRACT

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The agent causing yellow vein and mosaic diseases of *Sida linifolia, Abelmoschus esculentus*, and *Hibiscus rosa-sinensis* was identified as a new strain of okra mosaic virus (NIN-OKMV). All three isolates of NIN-OKMV had a dilution end point between 10^{-3} and 10^{-4}, thermal inactivation point between 55 and 60 C, and longevity in vitro between 8 and 10 days. The isolates differed from previously described OKMV strains in that they incited local and systemic chlorotic lesions in *Chenopodium quinoa* and *C. amaranticolor* and were infectious to *Solanum melongena, Sesamum indicum*, and *Crotalaria* spp. In double-diffusion tests, all three isolates reacted without spur formation to an antiserum to a Nigerian strain of OKMV.

Okra mosaic virus (OKMV) is confined to West Africa, where it was first isolated from okra (*Abelmoschus esculentus* (L.) Moench) growing in the Ivory Coast (6). This type strain is now designated IC-OKMV (1). The existence of another strain of OKMV in West Africa was first suspected by Lana et al (8,9), who reported that the properties of a Nigerian isolate of OKMV (N-OKMV) differed from those of IC-OKMV. This finding was confirmed by Bozarth et al (1).

Two strains of IC-OKMV commonly occur in the Ivory Coast (2,3). Only one strain of N-OKMV is known, although the possible existence of a second strain was recently suggested by Lana (7).

In 1979, I obtained several virus isolates from okra, hibiscus (*Hibiscus rosa-sinensis*), and sida (*Sida linitolia*) that on diagnostic species incited symptoms similar to those caused by OKMV. Serological tests confirmed their identity, but there were differences in physical properties, host range, and symptomatology between the isolates and IC-OKMV and N-OKMV. This paper describes the host range and properties of these isolates.

MATERIALS AND METHODS

Virus cultures and maintenance. The three isolates of OKMV used in this study were from hibiscus in Igbo-Ukwu, from sida growing in an abandoned cassava

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plot at Adani, and from an okra plant on the University of Nigeria vegetable farm at Nsukka. All three isolates were given three single-lesion serial passages in *Chenopodium quinoa* and thereafter increased and maintained in *Solanum melongena* L. 'Lokoja.'

Host range and properties. With the exception of hibiscus, all test plants were started from seeds in steam-sterilized soil. All plants were kept in an insectproof screenhouse. At least four plants of each species were inoculated, and each test included appropriate controls. Inoculum was prepared by grinding young *S. melongena* leaves with symptoms in a mortar containing 0.05 M potassium phosphate buffer, pH 8.0 (2 ml/g of tissue). Inoculum was applied with cheesecloth pads to Carborundum-dusted leaves that were rinsed with tap water immediately after inoculation. Plants were observed for symptoms for at least 6 wk before back-inoculations were made to *Sesamum indicum* L. or *S. melongena* plants.

Thermal inactivation point, longevity in vitro, and dilution end point were determined by the methods of Ross (10) using *Sesamum indicum* as the test plant.

Serological tests were done in double-diffusion plates of 0.6% agarose, 0.1% sodium azide, and 0.85% sodium chloride. Expressed sap (diluted with distilled water) from infected *S. melongena* plants was the source of the test antigen.

Transmission tests were done with the tobacco whitefly *Bemisia tabaci* Genn. and the beetle *Podagrica uniforma* Jac., which were the predominant insects on infected hibiscus and okra, respectively. Nonviruliferous whiteflies were reared on tobacco (*Nicotiana tabacum* L. 'Xanthi'), starved for 2 hr, given an acquisition access on infected leaves of *S. melongena* for 24 hr, and then transferred to healthy *S. melongena* for inoculation access of 24 hr. Apparently nonviruliferous beetles (*P. uniforma*) collected from healthy cowpea (*Vigna unguiculata* (L.) Walp.) were indexed for OKMV on eggplant and cowpea plants, starved for 2–4 hr, and then transferred to infected okra (two- to four-leaf stage) for acquisition access of 24 hr. Thereafter the insects were allowed to feed on healthy okra for 12–24 hr.

RESULTS

In general, the reactions to inoculation with the three isolates were similar to those with IC-OKMV or N-OKMV in *Abelmoschus esculentus, Arachis hypogaea* L., *Cucumis sativus* L. 'Improved Long Green' and 'Supermarket,' *Citrullus vulgaris* Schrad 'Sugar Baby,' *Gossypium hirsutum, Hibiscus esculentus* L., *Malva* spp., *Sida cordifolia* L., *Sida rhombifolia, Urena lobata* L., and several local cultivars of *Vigna unguiculata*. The tendency for symptoms in cotton, hibiscus, okra, and *U. lobata* to disappear as the plant grew older (4) was confirmed. Furthermore, the three isolates incited local and systemic chlorotic lesions in *Chenopodium amaranticolor* L. and *C. quinoa*. Both IC-OKMV and N-OKMV caused systemic symptoms in *C. quinoa* but were not transmissible to *C. amaranticolor*.

The three isolates infected *S. melongena* 'Lokoja' and 'New York,' *S. aethiopicum, Sesamum indicum*, and *Crotalaria* spp., which are nonhosts of IC-OKMV and N-OKMV (4,8). In addition, the three isolates infected *Solanum nigrum*, a reported host of IC-OKMV (4). In all these hosts, the three isolates incited chlorotic local lesions on inoculated leaves followed by systemic chlorotic veinbanding, which often resulted in bright yellow chlorosis (Fig. 1). Reciprocal transfers between these infected host plants always produced symptoms indistinguishable from those caused by each of the original isolates. Transfers from infected *Solanum melongena, S. aethiopicum, S. nigrum, Sesamum indicum*, and *Crotalaria* spp. to okra or sesame plants produced typical symptoms of OKMV.

The isolates did not infect *Amaranthus hybridus* L.; *Capsicum frutescens* L. 'Tabasco' and *C. annuum* L. 'California Wonder' and 'Nsukka Yellow'; *Glycine max* (L.) Merr. 'Jupiter'; *Gomphrena globosa* L.; *Lycopersicon esculentum* Mill. 'Ife No. 1' and 'Roma'; *Nicotiana glutinosa* L., *N. rustica* L., and *N. tabacum* 'Xanthi,' 'NC 95,' 'Burley 21,' and 'Havana 425'; *Physalis floridana* Rybd.; *Pisum sativum* L.; and *Pueraria phaseoloides*.

In crude sap from systemically infected leaves of *S. melongena*, the three isolates had a thermal inactivation point between 55 and 60 C, a dilution end point between 10^{-3} and 10^{-4}, and longevity in vitro between 8 and 10 days.

The three isolates in crude sap from *S. melongena* reacted with antiserum to N-OKMV (a gift from R. F. Bozarth). The precipitin lines of the three isolates fused completely without spur formation. No reaction was obtained when crude sap from healthy *S. melongena* was tested against N-OKMV antiserum. Antiserum to N-OKMV reacted positively with crude sap from infected *S. melongena, Sesamum indicum*, and *Crotalaria* spp.

In three different tests using 10 insects per test plant, none of the isolates was transmitted to okra by whiteflies from

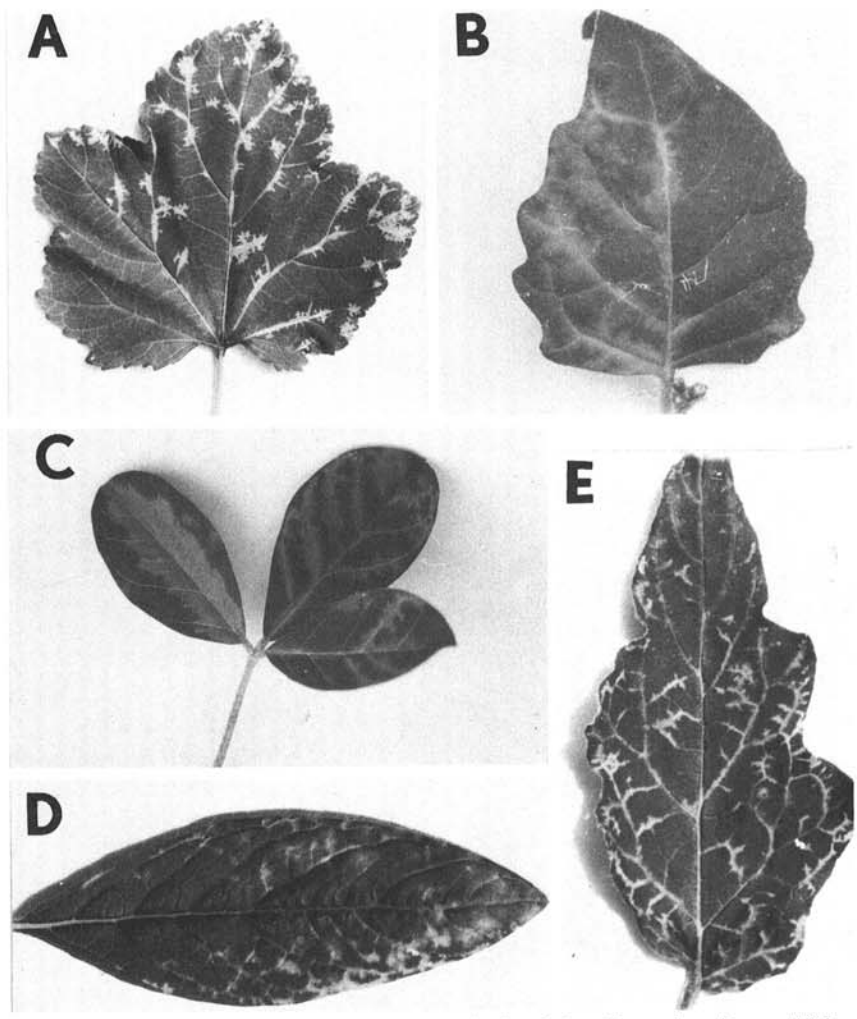

Fig. 1. Leaf symptoms incited by a strain of okra mosaic virus isolated from okra, sida, and hibiscus in Nigeria: Yellow veinbanding in **(A)** *Abelmoschus esculentus* 'Local,' **(B)** *Solanum nigrum*, **(C)** *Crotalaria* spp., **(D)** *Sesamum indicum*, and **(E)** *Solanum melongena* 'Lokoja.'

their respective original hosts. However, all three isolates were transmitted from their original respective hosts to okra by beetles. Also, eggplants and cowpea plants exposed to beetles not fed on OKMV-infected plants remained healthy.

DISCUSSION

Host range, symptomatology, and beetle transmission indicated that the yellow vein and mosaic diseases of hibiscus, okra, and sida observed in scattered areas of Anambra State, Nigeria, were caused by OKMV. Serological tests confirmed the identity of the three isolates with OKMV.

Although host ranges of the three isolates were similar to those of IC-OKMV and N-OKMV, there were differences in host range, symptomatology, and properties in crude sap (2–5,8). They also differed in properties from the virus that Lana (7) suspected to be a strain of OKMV. Because with minor exceptions the properties of the three isolates were similar, I considered them to be the same virus and designated them as Nsukka isolates of a Nigerian strain of OKMV (NIN-OKMV). Until parallel host range, cross-protection, and reciprocal serological tests are done with the type strain of N-OKMV, the exact relationship among OKMV strains remains unknown.

The data showing that NIN-OKMV was infectious to *S. melongena*, *Sesamum indicum*, and *Crotalaria* spp. extend the experimental host range of OKMV and confirm its economic importance. The observation that *Hibiscus rosa-sinensis*, a common ornamental in gardens in Nigeria, and *Sida linifolia*, a common weed in and around cultivated crops, were natural hosts of OKMV suggests that these plant species may serve as reservoirs of the virus.

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