African Cereal Streak, a New Disease of Cereals in East Africa

D. E. Harder and W. Bakker

Plant Pathologist, University of Manitoba-Canadian International Development Agency, Plant Breeding Station, Njoro, Kenya (present address is c/o Canada Agriculture Research Station, 25 Dafoe Road, Winnipeg, R3T. 2M9, Manitoba, Canada); and Plant Virologist, National Agricultural Laboratories, Nairobi, Kenya (present address is Laboratory of Virology, State Agricultural University, Wageningen, The Netherlands).

Published with the approval of the Chief Research Officer, Ministry of Agriculture, Kenya, and L. H. Shebeski,

Dean, Faculty of Agriculture, University of Manitoba and project director, C.I.D.A., P.B.S., Njoro.

Mr. R. E. Allen, presently with U.N.D.P., Mogadishu, Somalia, initially assisted with insect collections and identified some of the aphid species. Leafhopper specimens were identified at the Commonwealth Institute of Entomology.

Accepted for publication 14 May 1973.

ABSTRACT

A new disease of small-grain cereals in East Africa is reported. The disease is caused by a spherical virus, which is 24 nm in diam, limited to the phloem, induces phloem necrosis, and is transmitted by the delphacid planthopper, *Toya catilina*.

The proposed name of the disease is "African cereal streak", and its causal pathogen African cereal streak virus (ACSV).

ACSV has been successfully transmitted in the greenhouse and growth chamber to wheat, oats, barley, rye, triticale, rice, Eragrostis chalcantha, E. tenuifolia, Chloris pycnothrix, Aristida adoensis, and Harpachne schimperi. Attempts to transmit it to maize and linseed were unsuccessful.

The main symptoms are faint chlorotic streaks originating at the base of the leaf, elongating to form broad bands of alternate yellow and green streaks. Young infected plants remain severely stunted and die prematurely. The inflorescence becomes yellow, and awned types in particular become distorted. Seed yield is almost completely suppressed.

The possible relationships of ACSV to other viruses affecting the Gramineae are discussed, as well as some observations with epidemiological implications.

Phytopathology 63:1407-1411

Wheat and other cereals have been grown with varying degrees of success in Kenya since the early part of the present century (8). Wheat is the major small-grain cereal cultivated, and the main limiting factors in production have been the rust diseases. Consequently, little attention has been focused on other disease problems. One such problem is the occurrence of an unfamiliar disease with symptoms generally corresponding to those of the yellows type. During the period 1969-1972 the disease at times reached epidemic proportions in plots at the Plant Breeding Station, Njoro, and has seriously affected commercial fields of wheat and barley. No disease of this description has been previously recorded in Kenya.

The present paper is a preliminary description of this disease: symptoms, identification of the pathogen, transmission, some hosts that are affected, and geographical distribution are discussed.

MATERIALS AND METHODS.—Transmission.—Mechanical.—Wheat tissue of various ages collected from plants with typical symptoms was ground in a mortar and pestle in distilled water or in 0.01 M phosphate buffer, pH 7.0. The juice was filtered through cheesecloth and finger-rubbed on leaves of test hosts which had been dusted with a fine grade of Carborundum prior to inoculation. Sterilized soil was used for planting all test material. Test plants used were seedlings of the wheat cultivars 'Florence Aurore' (FA) and 'Tobari 66', the barley cultivar 'Union', Nicotiana tabacum, N. rustica, Gomphrena globosa, Datura stramonium, and Chenopodium amaranticolor.

Transmission.—Seed.—Young wheat plants in the field showing clear symptoms were tagged for later identification. After ripening, heads were collected and seed was obtained where possible. The seeds were planted in sterilized soil and placed on greenhouse benches.

Transmission. - Soil. - Soil was collected from around

the roots of diseased plants growing in the field, placed in pots, and used for planting the wheat cultivar FA. Also, part of the above soil was placed around the roots of FA seedlings which had been previously planted in sterilized soil.

Transmission.—Aphid.—Five aphid species, Schizaphis graminum (Rond.), Macrosiphum avenae (F.), Rhopalosiphum rufiabdominalis (Sasaki), R. padi (L.), and R. maidis (Fitch) were used separately in transmission tests. The aphids were collected from diseased plants in the field, and transferred to healthy plants of wheat (FA and Tobari 66), barley (Union), and oat ('Rodney') seedlings growing in sterilized soil and caged to prevent the entry of other insects. In a second test, aphids were allowed to feed and multiply on diseased plants which previously had been transplanted from the field into large pots, screened, and placed on greenhouse benches. Fifteen days later, the aphids were transferred to healthy, screened FA and Tobari 66 seedlings growing in sterilized soil.

Transmission.—Leafhopper.—A number of leafhopper species were tested in essentially the same manner as were the aphids. The leafhoppers were collected in the field, and were either allowed to feed directly on healthy seedlings of the wheat cultivars FA or Tobari 66, or were held on diseased plants for 14 days prior to transfer to healthy plants. All leafhoppers were allowed a 5-day transmission-feeding period. A total of about 50 randomly selected adult male or female leafhoppers of each species were tested.

Electron and light microscopy.—Pieces of tissue approximately 1×2 mm were cut from leaves, stems, and glumes of wheat plants showing characteristic symptoms. The tissue pieces were fixed overnight at 4 C in 4% glutaraldehyde in sym-collidine buffer (Polysciences, Inc., Warrington, Pennsylvania), pH 7.1, washed six

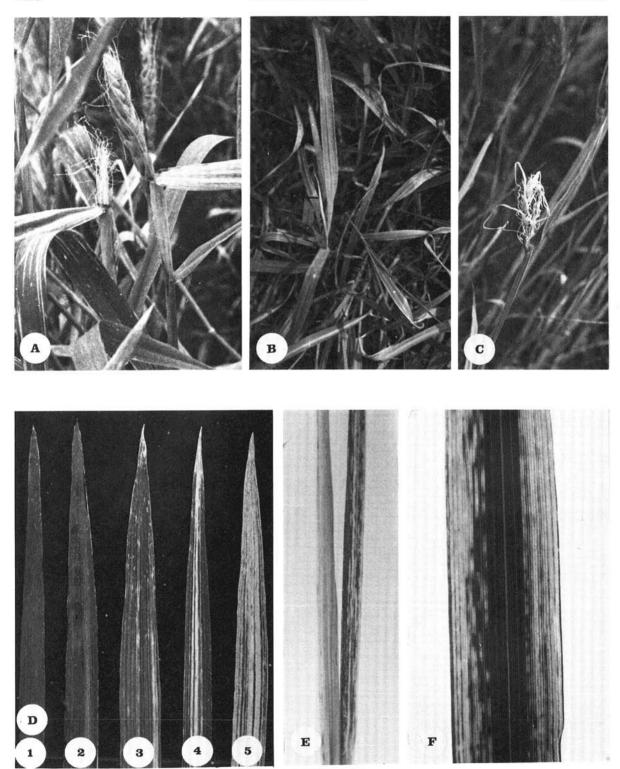


Fig. 1. Symptoms of African cereal streak on wheat, oats, barley, and rice. 1A) Streaking of leaves and awn distortion in wheat; 1B) "Shoestring" leaf formation in oats; 1C) Head and awn distortion in barley; 1D) Progression of symptoms in wheat leaves: 1D1 healthy, 1D2-1D5 increasing severity of symptoms with succeeding stages of infection; 1E) Symptoms on third and fourth leaves of rice; 1F) Broken nature of streaks in early stage of oat leaf infection.

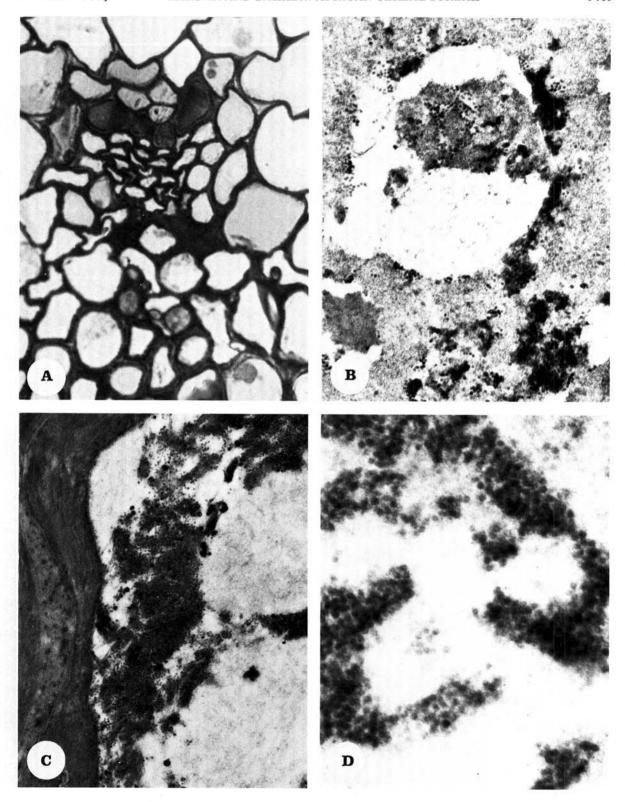


Fig. 2. Internal symptoms of African cereal streak in wheat. 2A) Phloem necrosis in outer vascular bundle of stem of nearly mature plant; 2B-C) Electron micrographs of two leaf vascular phloem cells with spherical particles. $2B \times 30,000, 2C \times 19,800; D$) Enlarged area of cell in $2C \times 81,000$.

times in buffer over a 6-hr period, then post-fixed overnight at 4 C in 1% osmium tetroxide in sym-collidine buffer, pH 7.1. After fixation, the tissue was dehydrated in an ethanol series, transferred to propylene oxide, and embedded in Spurr's Low Viscosity Embedding Medium (Polysciences, Inc.). Ultrathin sections were taken up on uncoated 200- or 300-mesh grids, stained 60 min with uranyl acetate and 20 min with lead citrate, vapor-coated with carbon, and examined in a Hitachi HU-12 electron microscope. Magnification was calibrated using carbon replicas with 2,160 lines/mm.

For light microscopy, larger 3-micron sections were cut from the plastic blocks with an LKB Pyramitome, flattened by heating slightly in 30% aqueous acetone, and stained with toluidine blue in 1% sodium borate.

RESULTS.—Transmission.—Repeated attempts to transmit the virus by mechanical means, with seed from infected plants, with soil from the roots of infected plants, and by the five aphid species, S. graminum, M. avenae, R. rufiabdominalis, R. padi, and R. maidis, failed.

Of the leafhoppers tested, only one, the delphacid *Toya* catilina (Fennah) regularly transmitted the disease. In subsequent tests, *T. catilina* nymphs were hatched from eggs laid on caged plants in the greenhouse, and used successfully as vectors. Field-collected females were also transferred individually to healthy seedlings, and where no symptoms appeared after 50 days, any resulting progeny were retained and increased for additional tests.

Symptoms.—The disease can be identified on wheat, oats, barley, rye, and triticale (see host range), with only minor variations in symptoms. The first symptom is the appearance of faint, somewhat broken chlorotic streaks (Fig. 1D2), typically beginning near the base of the leaf (wheat, Fig. 1A), then extending upwards. The broken nature of the young streaks is clearly defined on oat leaves (Fig. 1F). Pronounced, alternate yellow and green streaks then develop along the entire leaf blade, and it is at this stage that the disease is easily identified. In later stages the leaves become almost completely yellow (Fig. 1D5). Newly developing infected leaves tend to acquire a "shoestring" habit (oats, Fig. 1B), and soon die. Rice ('Sindano') was also infected in transmission experiments, and symptoms were similar to those on other cereals, although the streaks were more broken (Fig. 1E), resulting in a somewhat mottled appearance.

If plants are infected as young seedlings, the entire plant becomes chlorotic, remains severely stunted, and dies prematurely. In later-infected plants the heads emerge but are conspicuously yellow, and often become distorted, particularly if awned (wheat, Fig. 1A; barley, Fig. 1C). Through either infertility or abortion, seed yield is almost completely suppressed. Infected plants become soft and flaccid, being somewhat velvety to the touch.

Electron and light microscopy.—The objective of electron microscopy was to ascertain the presence and nature of any possible pathogen. Some cells in the phloem of all diseased plants examined contained numerous spherical particles (Fig. 2B-C). The particles are approximately 24 nm in diam, with a slightly electron-lucent center (Fig. 2D). No comparable particles were found outside the phloem or in the phloem of healthy plants.

A common feature in diseased plants was phloem necrosis, as seen in the outer bundle from the stem of a nearly mature wheat plant (Fig. 2A).

Host range.—In preliminary host range studies, virus obtained from a single wheat plant was successfully transmitted to oat (Rodney), barley (Union), rye ('Petkuser'), triticale, and rice (Sindano) plants, with the pattern of symptom development similar in each case. Attempts at transmission to maize and linseed were unsuccessful.

Plants of several grass species near Njoro, including Chloris gayana (Kunth.), C. pycnothrix (Trin.), Eragrostis chalcantha (Trin.) [syn. E. racemosa (Thunb.) Steud.], and E. tenuifolia (Hochst.), show yellowing and some streaking among otherwise healthy stands. The vector, T. catilina, was readily found in stands of native grasses. In subsequent transmission tests, using infected wheat as virus source, symptoms developed on C. pycnothrix, E. chalcantha, and E. tenuifolia. Additional grasses successfully infected were Harpachne schimperi (Hochst.) and Aristida adoensis (Hochst.).

Geographical distribution.—The disease has been observed in all wheat growing areas of Kenya, but is of most frequent occurrence at lower altitudes. Plots of all cereals at Njoro (2,300 m altitude) are generally heavily infected. Heavy infection in commercial fields is sporadic, although infection levels of about 40% were seen in single fields of wheat at Rongai (altitude about 2,100 m), and at Lanet (altitude about 2,000 m), and 25% in barley at Njoro. Infected plants are found less frequently in the Mau Narok, Molo, and Timau wheat growing areas, which are in the 2,500 to 2,900 m altitude range. However, occasional infection levels of up to 15% were found in experimental plots at Molo. At Njoro, there appears to be a relation between increased levels of infection and high temperatures.

A disease syndrome similar to that described was also found in some of the wheat growing areas of Ethiopia. Although the identity of ACS in Ethiopia could not be verified, the symptomatic features were strikingly similar, and the vector of ACS, *T. catilina*, was found at Awassa. The highest infection levels were found at Awassa, which at 1,650 m altitude was the lowest area visited. In other areas visited, Bako, Kulumsa, Holetta, and Debra Zeit, the disease was only sporadic in occurrence.

DISCUSSION.—The principle diagnostic features of the disease described are: 1) symptoms—broadly streaked leaves, yellowing, awn and head distortion, stunting, flaccidness, and lack of seed formation; 2) transmission by *Toya catilina*; 3) presence of apparent spherical virus particles, 24 nm in diam, limited to the phloem; 4) phloem necrosis; and 5) hosts affected—presently the small-grain cereals wheat, oats, barley, rye, triticale, and rice have been found susceptible.

Of the above features, particle morphology, size, and distribution in the host are similar to those of barley yellow dwarf virus (BYDV) (2, 7), but the disease is differentiated from BYDV by symptoms and transmission. The symptoms and transmission are somewhat similar to those of wheat striate mosaic virus (WSMV). However, the virus causing North American wheat striate mosaic is a bacilliform particle (4), and Lindsten (5) found no phloem necrosis associated with European wheat striate mosaic. In comparison with other virus diseases of Gramineae in the tropics or in Kenya (1, 3, 6, 9, 11, 12), similarity can be ruled out by one or more

features, or there is insufficient information to draw clear relationships. Slykhuis (10) reported a disease of wheat in Egypt with comparable features, and it is possible that it is similar to the disease reported here.

Preliminary epidemiological observations indicate that spread of the disease is facilitated by high temperatures, as it is most widespread at lower altitudes, and it appears to be more serious at Njoro in warm periods. The natural reservoir of the virus needs to be determined, although grasses with similar symptoms can be found, and the vector is easily found in stands of native grasses. The grass species to which the virus has been successfully transmitted are common in the Njoro area. It is suggested that grasses may play an important epidemiological role. The introduced, cultivated cereals serve as alternative and highly susceptible hosts.

The above considerations indicate that the disease in question has not been previously recorded, and that this is the first published description. Because no similar disease has been reported from outside the African continent, and the common cultivated cereals are congenial hosts, the name proposed for the disease is African cereal streak (ACS), and for the causal virus African cereal streak virus (ACSV).

LITERATURE CITED

 BAKKER, W. 1970. Rice yellow mottle, a mechanically transmissable virus disease of rice in Kenya. Neth. J. Plant Pathol. 76:53-63.

- JENSEN, S. G. 1969. Occurrence of virus particles in phloem tissue of BYDV-infected barley. Virology 38:83-91.
- KULKARNI, H. Y. 1969. Transmission of the pathogen of molasses dwarf disease by Maloxades farinosus. Phytopathology 59:1783-1786.
- LEE, P. E. 1968. Partial purification of wheat striate mosaic virus and fine structural studies of the virus. Virology 34:583-589.
- LINDSTEN, K. 1961. Studies on virus diseases in Sweden. II. On virus diseases transmitted by the leafhopper, Calligypona pellucida (F.). Ann. Royal Agric. College of Sweden 17:199-271.
- MARTYN, E. B. (ed.). 1968. Plant Virus Names. An annotated list of names and synonyms of plant viruses and diseases (including Supplement No. 1). Phytopathological Paper No. 9, Commonwealth Mycological Institute, 204 pp.
- PALIWAL, Y. C., & R. C. SINHA. 1970. On the mechanism of persistence and distribution of barley yellow dwarf virus in an aphid vector. Virology 42:668-680.
- PINTO, F. F., & E. A. HURD. 1970. Seventy years with wheat in Kenya. East Afr. Agric. Forest. J. Vol. 36 -Special Issue. 24 p.
- ROBINSON, R. A. 1960. Notes on Kenya Agriculture VIII: Important plant diseases. Revised 1962. East Afr. Agric. J. 25:1-16.
- SLYKHUIS, J. T. 1962. An international survey for virus diseases of grasses. FAO Plant Prot. Bull. 10:1-16.
- SLYKHUIS, J. T. 1967. Virus diseases of cereals. Rev. Appl. Mycol. 46:401-429.
- STOREY, H. H., & A. P. D. MC LEAN. 1930. The transmission of streak disease between maize, sugar cane, and wild grasses. Ann. Appl. Biol. 17:691-719.