Patterns of antigenic variation in whitefly-transmitted geminiviruses

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Geminiviruses cause many of the world's most economically important virus diseases of crops in tropical and sub-tropical countries. Some of the viruses are transmitted by whiteflies (*Bemisia tabaci*) but others have leafhopper vectors. The most important diseases caused by the whitefly-transmitted viruses are cassava mosaic, tomato leaf curl and yellow leaf curl, tobacco leaf curl, yellow mosaics of many legume species, and leaf curl or mosaic of cucurbits, pepper, okra and cotton. In cassava alone, the value of crop losses in Africa caused by mosaic is thought to exceed £200 million annually. We have studied many of these whitefly-transmitted viruses, and have prepared a series of diagnostic reagents of wide applicability.

Of these reagents, polyclonal antisera and monoclonal antibodies (MAbs) have proved to be invaluable and have been used extensively in ELISA. Early work[1] in which polyclonal antisera were used in immunosorbent electron microscopy instead of ELISA showed that serological relationships between different whitefly-transmitted geminiviruses are common whereas leafhopper-transmitted geminiviruses are not serologically related to the whitefly-transmitted viruses, and most are not related to one another. The production
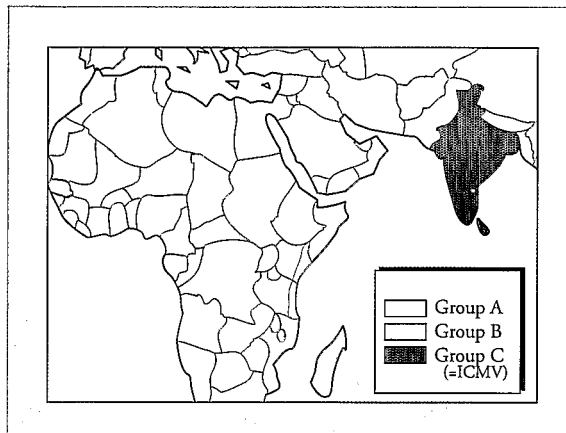
of panels of MAbs to particles of the whitefly-transmitted African cassava mosaic (ACMV) and Indian cassava mosaic (ICMV) geminiviruses has enabled us to investigate these relationships in more detail, and to assess the extent of variation among isolates of the same virus.

Initial tests, in which crude extracts from mosaic-affected cassava leaves from plants sent from a range of countries were tested for reactivity with 17 ACMV MAbs, showed unexpectedly that some of the virus isolates were not closely related to others. They fell into three categories: Group A, reacting with at least 14 of the MAbs; Group B, reacting with 4 to 9; and Group C (=ICMV), reacting with only 2 or 3[2]. Tests with MAbs to ICMV have confirmed this grouping. Group C isolates reacted with almost all these MAbs, Group A with about half of them and Group B with only one. Moreover, the groups can also be distinguished in other ways, such as ability to infect and to multiply at different temperatures in *Nicotiana* spp., and the extent of the nucleotide sequence similarity of the smaller of two genomic DNA species. Thus groupings based on different criteria coincide.

When the geographical origin of isolates of the three groups is mapped (Fig. 1), it can be seen that each group of isolates occurs in a different region. Group A isolates occur in many countries of West Africa, and in South Africa, Uganda, western Kenya and western Tanzania. In contrast, Group B isolates occur in coastal Kenya, coastal Tanzania, Malawi and Malagasy, and Group C isolates occur in India and Sri Lanka. This pattern of distribution, and the apparent absence of similar virus isolates in South America, led us to suggest that all cassava was geminivirus-free when originally brought from the Americas across the Atlantic Ocean by Portuguese colonists and traders in the 16th to 18th centuries, that after introduction to locations in West Africa, East Africa and India it became infected by geminivirus isolates which already occurred in these regions in other plant species, and that the virus isolates then endemic in West Africa differed from those in East Africa and the Indian subcontinent[3]. Group A isolates would have spread with cassava cultivation across much of Africa from west to east. If this hypothesis is correct, one might expect to find cassava-infecting geminiviruses in wild plants in the three regions. Indeed, an isolate from a wild euphorbiaceous species from Malagasy has proved to be antigenically indistinguishable from Group B isolates of ACMV, although there is now no way of knowing whether the plant was infected by inoculum from other such wild plants or from cassava.

Leaf curl and yellow leaf curl of tomato occur in many tropical regions ranging from Central America to Australia and cause serious crop losses in many countries. Some of the ACMV and ICMV MAbs can be used to detect these viruses but, as with cassava mosaic, isolates from different geographical regions share different combinations of epitopes with ACMV and ICMV. From their epitope profiles, the tomato viruses examined to date can be assigned to four main groups with different geographical distributions (Fig. 2).
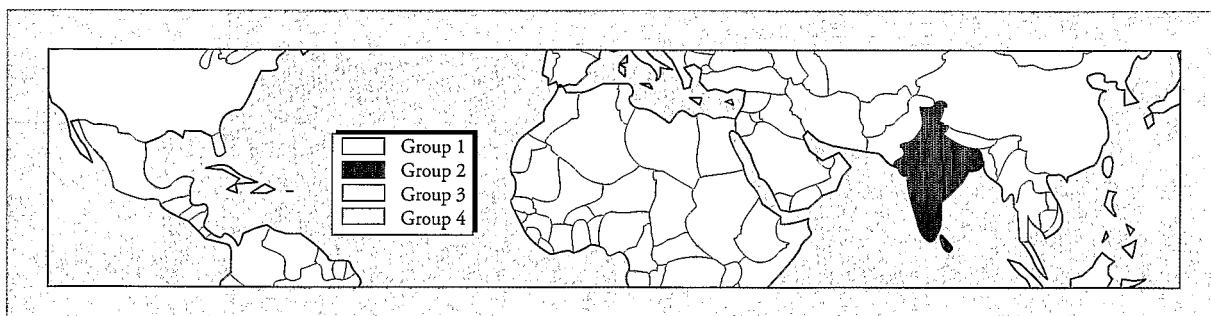
Group 1 occurs in Mediterranean countries, Senegal, Egypt and Yemen, Group 2 occurs in India, Group 3 in Thailand and Group 4 in Mexico, USA (Florida) and, probably, the West Indies.

Geminiviruses also occur widely in okra (*Abelmoschus esculentus*). However, isolates of okra leaf curl virus (OLCV) from West Africa have very different epitope profiles from those of bhendi (okra) yellow vein mosaic virus (BYVMV) from India. Indeed, OLCV shares many epitopes with ACMV (Group A isolates) whereas BYVMV has an epitope profile very like that of ICMV. Tests of several other geminiviruses from different host species in India[4] for reactivity with the full range of ACMV and ICMV MAbs show that although they differ in epitope profile, many of them have a general resemblance to ICMV in their patterns of reaction. Similarly, geminivirus isolates from the southern United States and Central America have a general similarity in epitope profile notwithstanding their different host ranges.

Summarising these results, we reach the remarkable conclusion that geminiviruses causing the same disease in cassava or tomato in different continents have different epitope profiles, whereas distinct geminiviruses which have non-overlapping host ranges but occur in the same geographical area show a general similarity in epitope profile. Within the geminivirus group, evolution seems to have proceeded differently in different continents, either by parallel or convergent changes in different progenitor viruses occurring in the same region, or by adaptation of a different progenitor geminivirus occurring in each region to a variety of host species.

All whitefly-transmitted geminiviruses for which vectors are known are transmitted by the same species, *Bemisia tabaci*. Moreover, the vector specificity of different members of the geminivirus group seems to be determined by the specificity of their coat protein[1,5]. This raises the question of whether *Bemisia tabaci* may occur as different biotypes in different geographical regions, with each biotype acting to select virus isolates with a coat protein that has structural features particularly suited for transmission by that biotype. If this situation exists, both the differences in epitope profile of viruses causing the same disease in different regions, and the epitope similarity of different viruses occurring in the same region, could readily be explained.

B. tabaci contains esterase enzymes which can be separated by electrophoresis in polyacrylamide gels[6]. We have adapted this approach for analysis of the patterns of esterases occurring in single whiteflies. Such analyses show that although the esterase patterns of *B. tabaci* from one plant species in one country are very similar, substantial differences exist in the patterns given by *B. tabaci* from the same host in four different regions: India, Malawi, Ivory Coast and United States. Further work is needed to establish whether geminiviruses are preferentially transmitted by the *B. tabaci* biotype from their source region and to exploit further the unique opportunity to study the nature and causes of evolutionary change in this important group of plant viruses.

References

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