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Evolution of Genetic Groups of whiteflies *Bemisia tabaci*

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Introduction

Bemisia tabaci is a member of the family Aleyrodidae under the Homoptera suborder of Hemiptera. The distribution of the insect varies from warm to hot climates between 30°N and S of the equator and is considered amongst the world's worst 100 invasive insects (Chaubey *et al.*, 2015). The nymph and adult of this highly polyphagous pest insect suck sap from phloem tissues of the plant and causes damage by attacking a wide range of more than 1000 plant species. The yield reduction caused by the pest often goes more than 50 per cent due to the combined effect of feeding and vectoring plant pathogenic viruses. The populations of whiteflies with varied geographical distribution, virus transmission efficiency, host specificity and insecticide resistance, would have varied genetic constitutions where mere morphological distinctiveness would not be sufficient to distinguish them. The recent studies categorized B. *tabaci* as a cryptic species complex. In this article, we tried to include the development of the cryptic species concept of whitefly.

The pest status of *B.tabaci*

B. tabaci as a pest was first reported in tobacco plants in Greece in 1889 and described as *Aleyrodes tabaci* by Gennadius. Quaintance (1900) described another species of whitefly, *Aleyrodes inconspicua*, in the USA which was later moved to a new genus, *Bemisia*. As a result of a series of reports of whiteflies from different countries on a variety of host plants, synonymization took place and the species '*tabaci*' has been placed in the genus *Bemisia* (Takahashi,1992).

A broadening of the host range and host shift in was also taken place during this period. Studies revealed that *B. tabaci* emerged from weeds such as *Convolvulus arvensis* and *Brassica* spp. in the early days of a year, and spread to cotton, cucumber and Hibiscus as the temperature increases. The population declines in October and adults revisit *Brassica* spp., *Solanum* spp. and other weed plants. By that time, reports of *B. tabaci* incidence was

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observed from 315 host plants from different countries including Sudan, Egypt, Taiwan, USA and Israel. Further, the status of the pest has grown as a highly polyphagous pest with preferred hosts from the families, Malvaceae, Solanaceae, Cucurbitaceae, Leguminosae and Verbenaceae. Presently, the distribution of *B. tabaci* extends to more than 60 countries (De Barro *et al*., 2011).

In India, the incidence of *B. tabaci* was first recorded in 1905. In the 1920s and 1930s, the pest had an outbreak in cotton ecosystem in some parts of Rajasthan and Punjab. Later it has spread to other crops *viz*., brinjal, tomato, okra, tobacco and many ornamental plants across the country. With whitefly, the virus diseases associated also gained momentum. There exists a considerable difference in the mode of transmission of the virus. *Begomovirus*, *Crinivirus*, *Ipomovirus*, *Carlavirus* and *Torradovirus* are the genus transmitted by *B. tabaci*. The genus *Begomovirus* is responsible for Cotton leaf curl virus (CLCV), a major disease of cotton, which is more prevalent in Asia and Africa, and Tomato yellow leaf curl virus (TYLCV) that harms tomato crops worldwide. Besides, some other important viruses other than the genus *Begomovirus*, transmitted by *B. tabaci* are Cucurbit yellow stunting disorder virus (CYSDV; *Crinivirus*), Tomato chlorosis virus (ToCV; *Crinivirus)*, Cucumber vein yellowing virus (CVYV; *Ipomovirus*) and Tomato torrado virus (ToTV; *Torradovirus*) (Horowitz *et al*., 2020). Sriganganagar in Rajasthan and nearby districts of Punjab faced an outbreak of the disease, CLCV in 1994 and then the epidemic has reached across cotton-growing areas. Thereafter, outbreaks were reported in 2004-2005, 2009-10, 2013-14, 2015-16 crop seasons causing complete devastation of cotton crops in North India (Datta *et al*., 2017).

The cryptic species status and genetic group

After the description of whitefly species from tobacco as *Aleyrodes tabaci* by Gennadius in 1889, the species has undergone numerous synonymization and finally ended up in the name *Bemisia tabaci* with 18 other previously described whitefly species (Perring, 2001). The host range of the pest has widened enormously owing to its high polyphagy nature and, thereafter researchers confirmed *B. tabaci* as morphologically indistinguishable species or cryptic species, which exhibit typical biological, physiological, and genetic variation. Thorough knowledge of the genetic relationship among individuals can delineate the species

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group boundaries and precisely place individuals to populations or groups (Dinsdale *et al.*, 2010).

The discovery of morphologically indistinguishable populations in the 1950s confirmed the existence of biotypes or host races in *B. tabaci.* When the host range of the pest widened, researchers recorded them based on the host plant and called them *host races*. Such two races, *Sida* race and *Jatropha* race are reported from Puerto Rico. A Brazilian cultivar of cassava which never had a history of colonization of *B. tabaci,* were readily colonized by the pest when introduced to Africa. Subsequently, two different host races were also reported from the Ivory coast; *Cassava biotype* colonizing cassava and eggplant, and *okra biotype* colonizing plants other than cassava. Meanwhile, it is been suggested that the host races can be called biotypes. Esterase based markers were used to distinguish these populations, called *biotypes* (Perring *et al.,* 2001).

Biotype concept had gained popularity among whitefly researchers from the invasion event of *B* biotype to the Southern United States in the late 1980s via the trade of ornamentals. The host utilization and esterase profile of the *B biotype* was different from the indigenous one, *A biotype*, and also found the existence of reproductive incompatibility between these biotypes. Later, allozyme profile and RAPD-PCR was used successfully to differentiate A and B biotypes. Furthermore, genetic differences can be identified through polymerase chain reaction (PCR) using mitochondrial cytochrome oxidase 1 (*mtCO1*) and ribosomal internal transcribed spacer (ITS)1 primer. The analysis of *mt COI* sequence could place the populations in well-defined clades or lineages with strong geographical evidence on a continental scale (Frohlich *et al.*, 1999). Multiple detection methods helped in designating *B. tabaci* populations into different biotypes and among them, the two extremely invasive biotypes, B and Q which have a native range extending through the Middle East into Asia Minor and Mediterranean Basin through to Egypt and are named as Middle East Asia Minor (MEAM) and Mediterranean (MED) respectively.

The continued research in this aspect concluded that considerable genetic diversity exists in *B. tabaci* populations and they represent a cryptic species complex (Dinsdale *et al.*, 2010). Phylogenetic analysis of ITS and *mtCOI* sequence data has become one of the strongest pieces of evidence for delineating the *B. tabaci* populations into genetic groups. De Barro *et al.* (2005) used ITS data and confirmed six major genetic groups and published

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global phylogeny of *B. tabaci*. The six groups were Asia, Bali, Australia, sub-Saharan Africa, Mediterranean/Asia Minor/Africa and the New World. There is increasing evidence that COI sequences are constructive in delineating species by matching the species boundaries. Boykin *et al.* (2007) analyzed 366 *mtCOI* sequences and concluded the presence of 12 genetic groups worldwide, Mediterranean/Asia Minor/Africa, Mediterranean, Indian Ocean, Sub-Saharan Africa (silver leaf), Asia I, Australia, China, Asia II, Italy, New World, sub-Saharan Africa non-silver leafing, and Uganda sweet potato. Later, Dinsdale *et al.* (2010) analysed 549 sequences and the pair-wise comparison of the clades represented two distinct breaks at 3.5% and 11%. A break in the divergence >11% yielded 11 distinct genetic groups Asia 1, Australia/Indonesia, Australia, China, Asia II, Asia II India, Italy, sub-Saharan Africa, Uganda, New World and Africa/ Middle East/ Asia Minor and >3.5 % yielded 24 distinct genetic groups namely, Asia I, Australia/ Indonesia, Australia, China 1 and 2, Asia II 1–8, Italy, sub-Saharan Africa 1–4, Uganda, New World, MED, MEAM 1 and 2, and Indian Ocean. This revealed the presence of 11 well defined high-level groups containing 24 morphologically indistinguishable *B. tabaci* species globally (De Barro *et al.*, 2011). Further, reconstruction of the phylogenetic tree using 383 unique *mtCOI* haplotypes that cover complete diversity of *B. tabaci* known. A new genetic group of *B. tabaci* was reported from Tamil Nadu that diverged 6.2 % from Asia I sequence and was named Asia I India (Chowda Reddy *et al.*, 2012). Phylogenetic rebuilding of *B. tabaci* revealed that the lineage of ancestral members of the species complex came from Sub Saharan Africa followed by the New World.

Later studies revealed the existence of many genetic groups across the world. New genetic group Japan- 2 was reported by Lee *et al.* (2013). The phylogenetic tree reconstructed by Firdaus *et al.* (2013) based on increasing the divergence threshold for species boundaries up to 4% from 3.5%, added seven new groups, Asia IV, Asia II 11, Asia II 12, Japan 1, Japan 2, Africa and Sub-Saharan Africa 5 and finalized 36 genetic groups globally. Roopa *et al.* (2015) confirmed the presence of a new genetic group, in Karnataka (India), MEAM-K, which shows 92.6 % genetic relatedness with MEAM-1. Two new putative cryptic species, China 5 and Asia V were identified in China from 571 sequences with 738 bp length (Hu *et al.*, 2018). A recent study on phylogenetic analysis of *B. tabaci* by Kanakala and Ghanim (2019) analysed 4253 mtCOI sequences from 82 countries and identified two new genetic groups, Asia II 13 and Spain I. The reclassification of the phylogeny of *B. tabaci* based on

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3.5 and 4% divergence revealed 44 distinct genetic groups. The 44 genetic groups of *B. tabaci* existing worldwide are Asia I, Asia I-India, Asia II 1-13, Asia III, Asia IV, China 1-5, Japan1, Japan 2, MEAM 1, MEAM 2, MEAM K, MED, Australia, Australia/Indonesia, Africa, Spain I, Ru, Italy1, Italy 2, Sub Saharan Africa 1-5, Indian Ocean, New World1, New World 2, Uganda are the 44 genetic groups. China has the largest diversity of *B. tabaci* genetic groups (16) followed by India. The genetic groups present in India are Asia I, Asia I-India, Asia II 1, Asia II 5, Asia II 7, Asia II 8, Asia II 11, Asia II 13, MEAM K, China 3, MEAM1 (Ellango *et al.*, 2015; Roopa *et al.*, 2015; Kanakala and Ghanim, 2019). Recently Tamilnayagan *et al.* (2019) reported the presence of Asia II 6 genetic group in the Ash gourd field of Tamil Nadu and also this study reported the co-existence of Asia I and Asia II 6 genetic groups in the same field.

Conclusion

This article mainly focuses on the development of pest status, the cryptic species staus and the delineation of *B.tabaci* populations across the world into different genetic groups. This could help in better understanding of the invasions of *B. tabaci* species complex across the world. The patterns of spread and diversity of *B.tabaci* will offer valuable insights into considerabley differing biologically attributed aspects such as virus transmission efficiency, insecticide resistance development *etc*.

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