

Host dynamics and molecular characterization of neo tropical invasive Bondar's Nesting Whitefly (BNW), *Paraleyrodes bondari* Peracchi (Hemiptera: Aleyrodidae) in Andhra Pradesh

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ABSTRACT: The Bondar's Nesting Whitefly (BNW), *Paraleyrodes bondari* Peracchi is a new exotic pest of coconut since December, 2018 in India. It has spread across the nation since its first report and occurring in several horticultural crops especially in the East Godavari District of Andhra Pradesh. The host dynamics of BNW were studied and its incidence, intensity and severity on different host plants were also recorded. The host plants observed were Coconut, Oilpalm, Banana, Cinnamom, False rubber, Mango, Jackfruit, Guava, Temple pod and Hibiscus etc. The incidence and intensity were observed to be highest in Coconut and Oilpalm, while least was recorded in Hibiscus plant. The identity of BNWwas further confirmed through amplification of mt. COI gene obtained for the isolate which was sequenced and submitted to NCBI- GenBank (Acc. No. ON739183).

Keywords: *Paraleyrodes bondari*, BNW, Andhra Pradesh, host dynamics, neotropical, invasive, molecular characterization.

INTRODUCTION

Whiteflies (Hemiptera: Aleyrodidae) are plantfeeding, polyphagous sucking pests of global significance because of their function as vectors of plant diseases as well as inducing secondary deposits of sooty moulds on leaf surfaces by honeydew production, thus disturbing photosynthetic activity (Dickey et al. 2015). They superficially resemble tiny flies, with more than 1550 species described worldwide (Ouvrard and Martin, 2018). Among the insect pests, globally over the past 25 years, exotic whiteflies invaded several countries causing direct losses in agriculture, horticulture and forestry. Such reported invasive whiteflies in India are the spiralling whitefly, Aleurodicus dispersus Russell, which is known to breed on 320 host plants belonging to 225 genera and 73 families (Sundararaj and Pushpa, 2012); solanum whitefly, Aleurothrixus trachoides (Back) reported to breed on 24 plant species including medicinal plants (Sundararaj et al. 2018) and rugose spiraling whitefly (RSW), Aleurodicus rugioperculatus Martin that invaded in 2016 (Shanaset al. 2016; Sundararaj and Selvaraj, 2017; Chalapathi Rao et al. 2018 and Sushmitha et al. 2020) and found breeding on more than 20 host plants including coconut. In December 2018, Central Plantation Crops Research Institute (CPCRI) recorded two exotic whitefly species, *P. bondari* Peracchi and *P. minei* Iaccarino on coconut palms of Kerala and issued a pest alert (CPCRI, 2019).First incidence of the neotropical invasive Bondar's Nesting Whitefly (BNW), *P. bondari* Peracchi (Hemiptera: Aleyrodidae) in India was reported on coconut palms from Kerala (Chandrika *et al.* 2018). Very recently, it was observed that BNW has been invaded into different host plants of Andhra Pradesh. The occurrence of this pest on different host plants was confirmed during the survey in East Godavari District of Andhra Pradesh. The host range of BNW was discussed in this article.

MATERIALS AND METHODS

Systemic and continuous surveys were conducted to identify different host plants of BNW and percentage of BNW incidence and intensity on different host plants were worked out. The severity of BNW was assessed based on the following criteria, i.e. low: 0-10 live adult nests /leaf; moderate: 11-20 live adult nests/leaf; severe: >20 live adult nests /leaf.

Occurrence of Bondar's Nesting Whitefly in Andhra Pradesh state

In Andhra Pradesh, Bondar's Nesting Whitefly (BNW)

Common Name	Scientific Name	Order	Family	Distribution	Whiteflies intermingled on the infested leaf	Incidence (%)	Intensity per leaf (%)	Severity
Coconut	Cocos nucifera	Arecales	Arecaceae	A.P, Kerala and Tamil Nadu	Aleurodicus rugioperculetus Martin, P.minei laccarino	59	32	Medium to high
Oilpalm*	Elaeis guineensis	Arecales	Arecaceae	A.P	Aleurodicus rugioperculetus Martin	45	27	Low to medium
Banana*	Musa paradisiaca	Zingiberales	Musaceae	A.P, Kerala	Aleurodicus rugioperculetus Martin	27	16	Low to medium
Cinnamom*	Cinnamomum verum	Laurales	Lauraceae	A.P	Aleurodicus rugioperculetus Martin	23	14	Low to medium
False Rubber	Ficus elastic	Rosales	Moraceae	A.P		26	18	Low to medium
Mango*	Mangifera indica	Sapnidales	Anacardiaceae	A.P	Aleurodicus rugioperculetus Martin	24	12	Low to medium
Jackfruit	Artocarpus heterophyllus	Rosales	Moraceae	A.P	Aleurodicus rugioperculetus Martin	18	14	Low to medium
Guava	Psidium guajava	Myrtales	Myrtaceae	A.P	Aleurodicus disperses Russell, Aleurodicus rugioperculetus Martin	19	28	Low to medium
Temple pod*	Cassia fistula	Fabales	Fabaceae	A.P		12	15	Low
Hibiscus*	Hibiscus rosa-sinensis	Malvales	Malvaceae	A.P	Aleurodicus rugioperculetus Martin	16	6	Low to Medium

Table 1. New host plants of *Phondari* Peracchi in Andhra Pradesh

104

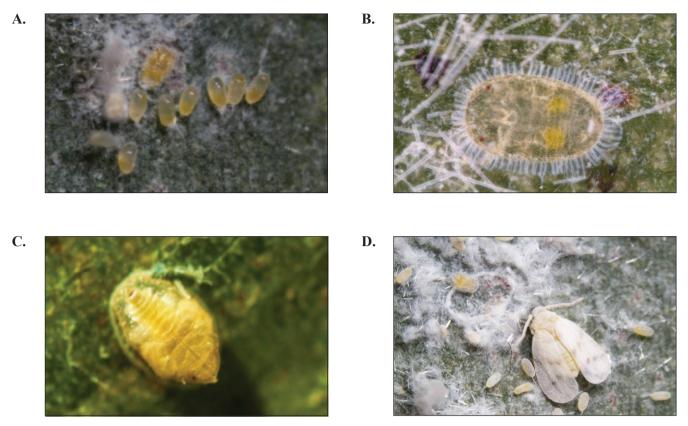


Fig 1. Life stages of BNW: A) Stalked Eggs and CrawlersB) Nymphs C) Pupae D) Adults

was observed on coconut plantations firstly in villages of East Godavari district. Based on typical characteristics of BNW, samples were collected from different locations and brought to the Biocontrol Research Laboratory, Coconut Research Station, Ambajipeta for morphological identification which was confirmed by Principal Scientist Dr. N. B. V. Chalapathi Rao as Bondar's Nesting Whitefly based on characteristics described by Perrachi. The incidence of BNW was confirmed after the survey in East Godavari District of Andhra Pradesh state. The pest was detected in all the coconut growing villages present in the vicinity of Konaseema region of East Godavari District. During the survey, this pest was also observed on different host plants and their samples were collected for further identification.

A total of 10 different hosts (Table 1) were identified during survey. Among them, coconut is already identified as host plant of BNW in Andhra Pradesh, while host plants like oilpalm, Guava, Banana and some ornamental plants etc. are firstly recorded as preferred host of BNW in India. As higher economic importance of plantation crops mainly in Konaseema region of Andhra Pradesh state, the concern about occurrence of BNW has raised the risk of becoming major pest.

However, this perilous neotropical invasive Bondar's Nesting Whitefly (BNW) pest was firstly reported from

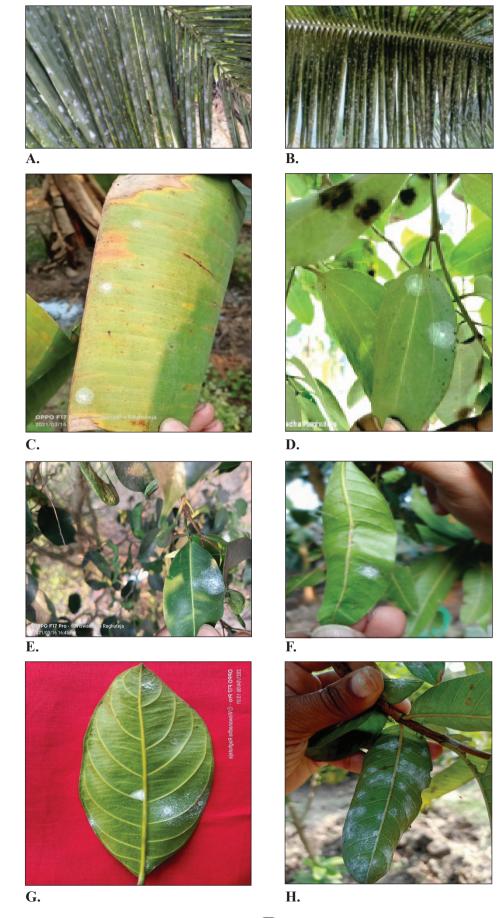
coconut (*Cocos nucifera* L.) palms at Kayamkulam, Kerala, Peninsular India during December, 2018 (Joseph rajkumar *et al.*, 2019). Vidya *et al.* (2019) reported the incidence of BNW on Custard Apple, Jackruit, Capsicum, Cinnamom, Mango, Banana, Guava, Teak, *Macaranga peltata, Brideliaretusa, Morinda citrifolia, Leucanea leucocephala* in Andaman and Nicobar Islands.

Biology

Adults lay stalked eggs in clusters around the nest and mobile crawlers are seen active in the colony. The crawlers settle down on finding a suitable feeding point.

Nymphs are creamy yellow, transparent, oval shaped, absolutely flat up to 0.9 mm. The margin of the nymph is covered with a band of white wax and short setae are observed all around. On the dorsal side of the final-instar nymph, there are two shiny fibre glass like strands on the anterior side, eight on the posterior side and four setae (two longer and two shorter) located on the middle region. As the nymphs advance to the fourth-instar, there is excessive production of shiny fibreglass-like strands (Josephraj kumar *et al.* 2019).

Adults are powdery white, about 0.95 mm long, with two conspicuous oblique grey bands across the forewings together forming a **typical "X" pattern**.

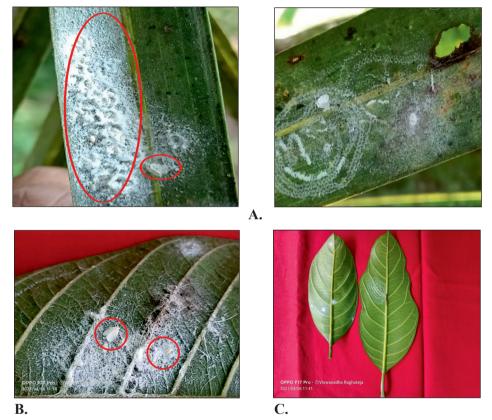


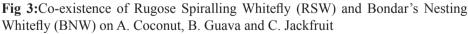
Pest Management in Horticultural Ecosystems Vol. 28, No.2 pp 103-109 (2022)

106



Fig 2: Occurence of Bondar's Nesting Whitefly, *P. bondari*in different hosts A. Coconut, B. Oilpalm, C. Banana, D. Cinnamom, E. False Rubber, F. Mango, G. Jackfruit, H. Guava, I. Templepod and J. Hibiscus





Severity of Bondar's Nesting Whitefly

The invasive pest *P.bondari* Perrachi (Hemiptera: Aleyrodidae), commonly known as the Bondar's Nesting Whitefly (BNW) was firstly observed in the 10 different host plants after reported in coconut from Konaseema region of Andhra Pradesh. The characteristics of BNW were shown same as observed in the coconut palm which is usually confined to the abaxial surface of palm leaflets shying away from the sunlight on account of its photosensitiveness. Adults and nymphs produce honeydew leading to deposits of sooty mould which affects the photosynthetic efficiency. The feeding damage by BNW has not become as intense as that of RSW with minimum honey dew and sooty mould deposits recorded so far on palm leaflets.

The following stages (Fig. 2 and 3) were observed during the survey for the confirmed report of this invasive pest in all these hosts.

Molecular Characterization of exotic Bondar's Nesting Whitefly (BNW)

Genomic DNA was isolated from individual adults

Accession Number	Species	Location	Per cent Identity (%)	Reference
MW488198.1	P. bondari	Karnataka, India	100.00	Shivaji et al. 2020
MW041899.1	P. bondari	Karnataka, India	100.00	Shivaji et al. 2020
MK343480.1	P. bondari	Kerala, India	100.00	Josephrajkumar <i>et al.</i> 2019
KP032215.1	P. bondari	Florida	100.00	Dickey et al. 2015
MZ026894.1	P. bondari	Tamil Nadu, India	100.00	Ramasubramanian <i>et al.</i> 2021

Table 2. List of nucleotide sequences of BNW retrieved from GenBank

of *P. bondari* collected from Horticultural Research Station (HRS), Ambajipeta using HiPurATM. Insect DNA Purification Kit (Hi-Media, India) following the manufacturer's instruction. The extracted DNA was eluted in 200 μ l elution buffer. The quality of extracted DNA was tested using the spectrophotometer and 1.2 per cent agarose gel electrophoresis and stored at -20°C for further use. The DNA samples of RSW were subjected to PCR analysis for confirmation using mitochondrial COI gene specific primers, LCO1490 (5'-GGTCAACAAATCATAAAGATATTGG-3') and HC02198 (5'- TAAACTTCAGGGTGACCAAAAAAT CA-3') (Dickey *et al.* 2015 and Josephraj Kumar *et al.* 2020).

Amplification was carried out in a 20 µl reaction volume which contained 2 µl of 10X Buffer [10 mMTris-HCl (pH 9.0), 50 mMKCl, 1.5 mM MgCl, 0.01% gelatin], 0.8 µl dNTPs (0.25 mM each), 3 µl of the forward and reverse primer's (2µM each), 0.3 µl of 3U Taq DNA polymerase (Genei Laboratories Pvt. Ltd., India) and 35 ng DNA. The PCR cycling condition consisting of an initial denaturation at 94°C for 2 min followed at 35 cycles of denaturation at 94°C for 30 sec, annealing at 50 °C for 1 min and extension at 72 °C for 1 min, with a final extension at 72 °C for 10 min. After amplification, the PCR products were fractioned on 1.2 per cent agarose gel stained with ethidium bromide in 1X TBE buffer by electrophoresis. The expected amplicon size was found to be 700 bp in all the samples. The DNA was eluted from gel and sequenced by Sanger's di-deoxy chain termination method (Barcode Biosciences Pvt. Ltd.).

The nucleotides were aligned and edited using Bio edit software. The sequences were deposited in NCBI and accession number was obtained (Acc. No. ON739183).

Based on the nucleotide sequence analysis of *P. bondari* isolates shared 100 per cent similarity with previouslyreported BNW isolate coconut cytochrome c oxidase subunit I (COX 1) gene, partial cds m (Acc. No. MW488198.1), isolate BNWKa1 (Acc. No. MW041899.1), isolate WFM 2 (Acc. No. MK343480.1), isolate BNW_856-1 (Acc. No. KP032215.1), isolate SBITR-Pb01 (Acc. No. MZ026894.1) (Table 2).`

Omongo *et al.* (2018) confirmed the identity of BNW, *P. bondari* adults by sampling from the affected Cassava field of Uganda and subjecting partial mt. COI gene sequences (632 bp) to NCBI BLAST. The sequences matched 100 per cent nucleotide identity (haplotype Acc. No. MH 178372) and 100 per cent nucleotide identity with bondar's nesting whitefly, *P. bondari*, Peracchi (Acc. No. KP032215). Josephraj Kumar *et al.* (2019) determined the identity of bondar's nesting whitefly (BNW) by confirming the partial mt. COI gene sequences (675 bp) (Acc. No. MK343480) shared 100 per cent nucleotide identity with bondar's nesting whitefly (Acc. No. KP 032215.1 *P. bondari* isolate BNW 856–1).

As the invasive pest, *P. bondari* has been already reported in coconut with causing significant damage in Konaseema region of Andhra Pradesh state. But recently it has been seen that this pest invaded into the different ten host plants. After the morphological and molecular identification of this pest, this is the first confirmed report of *P. bondari* on Oilpalm, False rubber, Temple pod, Hibiscus in India and first time observed in Mango, Banana, Cinnamom and Guava in Andhra Pradesh state. This evident the alarming situation to the growers of these crops as this pest may spread to other growing areas of Andhra Pradesh state.

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