Record of Nomuraea rileyi (Farlow) Samson (Hypocreales: Clavicipitaceae) in Hypocala sp. (Lepidoptera: Erebidae) from the Mangroves of Maharashtra, India

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Abstract
Hypocala sp. the defoliating insect causes regular infestation on Avicennia marina (Forssk.) Vierh. in the mangroves of Mumbai region particularly in Airoli creek of Navi Mumbai areas. This pest is prevalent during the post monsoon period in Airoli and nearby mangroves of Maharashtra State. The assessment intensity level of infestation revealed that moderate to severe infestation was recorded during the year 2018. An entomopathogenic fungus, Nomuraea rileyi (Farlow) Samson strain isolated from the field collected cadavers of Hypocala sp. was evaluated for its efficacy and effectiveness against the larvae of Hypocala sp. in laboratory condition. The native strain of N. rileyi showed highest level of larval mortality of 96 % and 100% in the concentration of 2.4× 10^3 and 2.4 × 10^8 Spores/ml respectively in the fifth day after treatment.

Keywords: Avicennia marina, Hypocala sp., defoliator, intensity, prevalent, infestation

1. Introduction
Avicennia marina (Forssk.) Vierh. is the most common mangrove species in Maharashtra [1]. This is found regularly infested by the defoliating insect pests during the post monsoon period. This pest is reported for the first time in the mangroves of Mumbai region particularly in Airoli creek of Navi Mumbai areas. Pest surveys conducted to assess the infestation caused in the mangrove species in Airoli and Vashi creek revealed the severe infestation of Hypocala sp. on A. marina and A. officinalis during the year 2018 and 2019. The larva feed on entire part of leaf and cause complete defoliation in nurseries, young plantations and in natural mangroves. This major pest was recorded on A. marina and A. officinalis during July to September with high intensity of infestation. In the past H. rostrata was recorded as pest on Ban Oak Quercus leucotrichophora in the western Himalaya [2]. Different species of the genus Hypocala viz. H. aspersa, H. australiae, H.biarcuata, H. deflorata, H. moorei, H. rostrata, H. subsatura and H. violacea were reported from Tamil Nadu part of the Western Ghats consists of the Nilgiri Biosphere Reserve, Kodaikanal, and Palani Hills [3]. The genus Hypocala was recharacterised and provided key to the species of this genus viz: H. subsatura, H. rostrata and H. deflorata [4]. The observation during the pest surveys resulted in detection of a species of an entomopathogenic fungus Nomuraea rileyi (Farlow) Samson for the first time on Hypocala sp. The infected larvae were found died and attached on the shoots and leaves. Earlier studies reported that N. rileyi was very much effective on the defoliating pests of agricultural crops in particular the insect pest Spodoptera litura [5]. Therefore, an attempt was made to assess the potential of this native entomopathogenic fungus for the control of Hypocala sp. in laboratory condition.

2. Materials and Methods
2.1 Study area
Regular insect pest surveys were conducted in mangrove areas of Airoli ( N 19° 14´ 76.5” E 072° 98´ 43.9”) and Ghansoli mangrove plantations (N 19° 11´ 50.9” E 072° 99´ 17.3”) during the year 2018 and 2019 to record the defoliating pests. The intensity of infestation was assessed based on the level of incidence of the insect pest and percentage of the damage/extent of damage caused. Larvae collected from the field were reared in laboratory condition and its life cycle was studied.
2.2 Pathogenicity Test
The fungal isolates of *N. rileyi* was grown on Sabouraud Dextrose Agar medium (SDA) at room temperature 25-28 °C for 8–10 days were harvested and crude extract were prepared (stock solution) with distilled water. The concentration of spores in the final suspension was determined by haemocytometry [6]. Four different concentrations of spore suspensions, 2.4 \times 10^{10}, 2.4 \times 10^8, 2.4 \times 10^6 and 2.4 \times 10^4 Spores/ml were prepared and pathogenicity test was conducted in laboratory condition on the targeted defoliating pest. Healthy larvae reared in the laboratory were first surface sterilized with 1-5% sodium hypochlorite and sprayed with the fungal inocula along with the feeding material (leaves of *A. marina*). The freshly prepared spore suspensions were sprayed evenly on the III instar larvae. Four treatments, namely T1 –T4, corresponding to 2.4 \times 10^{10}, 2.4 \times 10^8, 2.4 \times 10^6 and 2.4 \times 10^4 Spores/ml, were applied. The sprayed larvae and the feeding materials were air dried on filter paper. The larvae sprayed with inoculum were released into 100 ml plastic/glass containers. A control (T5) sprayed with 0.1% Tween 80 in sterile distilled water was also maintained. Each treatment was replicated five times with 10 larvae in each replicate. Observations were made every 24 h. Mortality was assessed up to 5 days when the highest concentration yielded 100 percent mortality or maximum percentage of mortality. The infected larvae were collected and maintained in Petri dishes lined with moist blotting paper for sporulation of the fungal pathogen.

3. Results and Discussion
*Hypocala* sp. was observed as a regular and major pest on *A. marina* and *A. officinalis*. The dark brownish adult moths are having 35 to 40mm wing span. Antennae ciliated in male. Thorax and abdomen scaled smoothly. Forewings with slightly arched costa towards rectangular apex. Larva greenish in colour having five instar stages before pupation. The matured larva ranges from 35 to 40 mm in length. The larval period was about 17 to 18 days. Pupation takes place on the leaf folded. The average duration required to complete the life cycle ranged from 35 to 37 days. The larva feed on entire part of leaf and cause complete defoliation in nurseries, young plantations and in natural mangroves (Fig 1 A & B). Severe intensity of infestation was recorded during the period from July to September ranged from 30 to 65% (Graph 1). In the mangroves allowing the population of pest uncontrolled will favour the pest to build up its population many folds and damage the mangroves severely.

Graph 1: Intensity of *Hypocala* sp. on *A. marina* and *A. officinalis*

Pathogenicity of *N. rileyi* tested on the targeted insect *Hypocala* sp. at four different concentrations 2.4 \times 10^{10}, 2.4 \times 10^8, 2.4 \times 10^6 and 2.4 \times 10^4 Spores/ml were found effective for control of the pest. However efficacy of the fungus in terms of percent mortality was found to vary with different concentrations of the conidial suspension of the fungus in lab condition. The concentrations 2.4 \times 10^{10} Spores/ml resulting 100 percent larval mortality in the fifth day on the third instar larvae and 2.4 \times 10^8 Spores/ml resulted 96 percent mortality. Where as 2.4 \times 10^6 and 2.4 \times 10^4 Spores/ml caused 84 and 72 percent mortality respectively (Fig 1 C & D). Earlier reports confirming the result of this fungus causing infection on caterpillar pests of cabbage, clover, soybean [7,8]. It was reported on groundnut, castor, potato, cotton etc. [9-12]. Entomopathogenic fungi were used as a biocontrol agent worldwide for controlling different insect pests [13]. They affect the insects by penetrating in to the body make use of extracellular cuticle-hydrolyzing enzymes of proteases, lipases and chitinases [14]. The native EPF, *Beauveria bassiana* was evaluated against *H. vitessoides* in laboratory condition and spore concentrations of 2.4 \times 10^{10}, 2.4 \times 10^8 and 2.4 \times 10^6 spores/ml were found effective [15]. The influence of three cultivars of *Sorghum bicolor* (L.) on the activity of the fungus *M. anisoplae* on the stem borer *Chilo partellus* was investigated in the laboratory and field [16]. *B. bassiana* was effective in controlling the major pest of tomato *Helicoverpa armigera* [17]. It was also reported to cause 88.68 percent mortality of *Dicladispa armigera* on rice and also found effective against *Spilarctia oblique* [18,19].

Fig 1: *Hypocala* sp. infestation on *Avicennia marina* (A) *Hypocala* sp. larva on *A. marina* (B) Pupa and adult moth of *Hypocala* sp. (C) *N. rileyi* infected larva of *Hypocala* sp. (D) Conidiospores of *N. rileyi*
4. Conclusion
Increase in pest problems has forced indiscriminate use of pesticides causing resurgence of many insect pests. Impact of various pesticides causing damage to the environment and development of resistance in insects forced to search the alternative, and the solution is use of biopesticides. In this study, the pathogenicity tested against the targeted insect pest *Hypocala* sp. with the native entomopathogenic fungus *N. rileyi* proved that the fungal pathogen was effective. As this pathogen reported causing infection to *Bombyx mori*, the silk moth, therefore care should be taken to use this fungus in the field where the sericulture farms are nearby. Therefore further investigations are required to confirm any strain variations and the host specificity of infection.

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6. References