

Abstract

Tea, cherished worldwide as a beverage of choice, holds a dominant position in India's agricultural landscape, representing 75% of the country's tea production. The North East region of India holds immense importance, making a substantial contribution of approximately 75% to the overall output. However, despite its paramount importance, tea production faces numerous challenges, chief among them being pest attacks. These attacks pose a significant threat, often leading to substantial losses that can range from 30% to 50%. Among the array of pests, the Red Spider Mite (RSM) poses a significant threat, characterized by a short life cycle and prolific reproductive capacity. In North East India, the economic threshold level for RSM is identified at 5 mites per leaf. The persistent use

of conventional pesticides, coupled with the mite's rapid life cycle, has fostered considerable pesticide tolerance, leading to recurrent and challenging resurgences, thereby impacting the Indian tea trade.

The tea plant is susceptible to diverse pests, including tea mosquito bugs, red slugs, red spider mites, thrips, jassids, etc. Among these, red spider mites and tea mosquito bugs stand out as predominant pests in the region, causing substantial crop losses in the tea industry annually. The association of *Oligonychus coffeae* with tea dates back to the inception of tea production in Assam, North East India, in 1868. While Nietner initially discovered *O. coffeae* on coffee plants (*Coffea arabica*) in Sri Lanka in 1861, it was Wood-Mason who, in 1884, designated it as a new species,

Tetranychus bioculatus. However, Pritchard and Baker's 1955 studied on the family Tetranychidae prompted a reevaluation, leading to the recognition of *T. bioculatus* as a synonym of the coffee mite, eventually placing it under the name *Oligonychus*.

According to early data, the red spider mite, *Oligonychus coffeae* Nietner, (Acarina: Tetranychidae), is one of the most significant pests of Indian tea, and causing up to 35–40% of the crop to be lost. Red spider mites and tea mosquito bugs are seasonal pests. The former occurs during the summer months and the latter during rainy winter. RSM is a polyphagous pest that feeds on coffee, rubber, indigo, grape, cashew nut, citrus, mango, *Camellia*, camphor, mulberry, oil palm, and many other tropical plants. It normally infests the upper surface of mature tea leaves, feeds along the midrib and veins, and gradually

spreads to the entire surface of the leaf, thereby changing the color of the leaf to ruddy bronze. Larvae, nymphs, and adult mites wreak havoc on the tea plant by feeding on the sap of the leaves and sporadically on the petioles. In severe infestation, it damages the younger and older leaves and ultimately leads to defoliation and debilitation of the tea bush causing a crop loss of 14–18%. In South India, the economic threshold level (ETL) of RSM in tea is reportedly 4 mites per leaf. and 2–3 mites/cm² in North East India. However, ETLs vary from area to region and even from field to field due to crop phenology, the cost of pesticides and manpower, weather patterns, etc.

The RSM lives under the cover of a silken web spun on the leaf surface as protection against inclement weather. Leaf temperature and light penetration within tea bushes also influence mite

distribution; *O. coffeae* prefers the middle zone of the bush (30 cm below the plucking surface) because of optimum temperatures associated with plant shading. The temperature in the upper zone of the tea plant may reach 40–45 °C yet shading can bring down the temperature of the middle tier of a bush to ambient levels of 30–32 °C.

Tea plantations use synthetic pesticides to manage pests, but researchers need to explore plants' biopesticidal properties for environmental problems. Native plant materials like oils, extracts, and secondary metabolites protect against pests and illnesses. Plant-based preparations, including powders, solvent extracts, essential oils, and insecticidal properties, are also used as biopesticides.

Botanicals play a vital role in organic tea production by effectively controlling pests such as red spider

mites and keeping their population below the Economic Threshold Level (ETL). Consistent use of plant extracts from the same species mitigates herbivore responses and minimizes the risk of pest resistance development.

Numerous studies from previous literature affirm that plant materials are abundantly accessible within local regions. The utilization of crude or raw plant extracts, containing a diverse amalgamation of bioactive components, emerges as a straightforward and economically feasible solution. This approach, exemplified by the utilization of aqueous extracts derived from various indigenous plants in India, underscores the practicality and cost-effectiveness of harnessing natural resources for various applications. Plant based remedies for the red spider mite have included *Clerodendron infortunatum* L., *Acorus calamus* L.,

Aegle marmelos (L.) Corrêa, *Xanthium strumarium* L., *Terminalia chebula* Retz., *Duranta repens* L., and neem kernel. The selection of plants should not limit the available plants to guarantee a wide range of protectants, and ongoing research into more potential plants is encouraged. It is significantly more important to carry out multiple bioassays when evaluating botanical pesticides.

So objective of our study is the survey and documentation and characterization of plants used in preparing bioformulation for managing the pest. In different tea estates of tarai and dooars region and tea research institute start to introduce some cultural practices in combating tea pest using some locally available weed plants. With such information in together, we chose ten weed plants based on the results of the survey data. Then collect those plant and prepare a

bioformulation. We also prepared individual plant aqueous extracts to know which plant is more effective than a formulation and found *Clerodendron infortunatum* L., *Artemisia vulgaris* L., and *Justicia adhatoda* L. confer better results than other plants when applied to a red spider mite in a laboratory condition.

Next, we formulate bioformulations using locally sourced plants, drawing upon both existing literature and the traditional knowledge of tea garden inhabitants. Initially, to evaluate the effectiveness of these bioformulations, greenhouse experiments were conducted, administering them at different dosages to infected samples.

Following the acquisition of promising outcomes, we advanced to field trial experiments carried out in two distinct tea estates, situated in both the Terai and Dooars regions. As part of our ongoing exploration into

the composition of the bioformulation, we embarked on comprehensive GC-MS analysis, revealing the presence of specific compounds within its formulation.

To gain deeper insights into the potential impact on insect pests, particularly the two-spotted red spider mite (*Tetranychus urticae*), we selected relevant proteins from the PDB database. Employing molecular docking studies, we substantiated that the presence of these identified compounds in the bioformulation could be a contributing factor to the mortality of the insect pest. Additionally, our findings are in line with earlier literature studies that underscore the pesticidal activity linked to these compounds.

In the metagenomic approach applied to the bioformulation, an intricate exploration of the microbial flora within the bioformulation was

unveiled. Furthermore, the reverse ecology study lends support to the notion that there exists a robust interaction among the bacteria, influencing their impact on the pest population.

Upon inflicting damage to the pest, the ensuing generation of reactive oxygen species (ROS) triggers the activation of antioxidant enzymes. Our investigation involved the collection of leaves from both infected plants and those treated with the bioformulation, alongside a positive control. Subsequent analyses, including various enzymatic and non-enzymatic tests, revealed that our bioformulation contributes to the reduction and mitigation of biotic stress.

Moreover, our bioformulation demonstrated a positive influence on the activity of Plant Growth-Promoting activity by inducing

changes in soil physiochemical composition. This, in turn, resulted in improvements in plant height, leaf count, and flushes, showcasing the

multifaceted benefits conferred by our bioformulation.

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