

7. Highlights

- The four major pests of tea, looper (*Buzura suppressaria*), red slug (*Eterusia magnifica*), tea mosquito bug (*Helopeltis theivora*) and red spider mite (*Oligonychus coffeae*) have a wide occurrence in both organic and conventional plantations of the Darjeeling foothills and their adjoining plains of Terai and the Dooars.
- Greater amount of amylase activity found in the midgut than in the salivary gland of *B. suppressaria* indicated greater digestion of polysaccharides of young leaves in midgut than their break down at the time of ingestion in the oral cavity.
- The presence of protease activity both in salivary gland and midgut of *H.theivora* indicated that this pest can well utilize the protein source of the tea leaf by extra oral and gut digestion.
- The activity of lipase in all the pests in question was much reduced than other two digestive enzymes.
- Of the oxidoreductases, the catalase assay of *B.suppressaria* and *Et.magnifica* was found to be marginally higher in salivary gland homogenate as compared with the midgut homogenate. As this enzyme is involved in inhibiting the action of toxic plant phenolics besides removing the hydrogen peroxide, an active blocking of these oxidants possibly takes place at the salivary gland level followed by that at gut level.

- Catalase present in salivary gland homogenate of *H.theivora* initially showed more activity as compared to that of midgut. Catalase in the saliva has the ability to prevent the formation of plant protective compounds such as quinone.
- In *H.theivora* the peroxidase assay was found to be similar both in the salivary and midgut homogenate, thus indicating the oxidative activity as a defence measure at both levels. In *O.coffeae* the enhanced peroxidase level could possibly minimise the effects of the toxic products that the mites ingest or experience while colonizing the tea leaves.
- In *Et.magnifica* presence of polyphenol-oxidase was detected in both saliva and midgut homogenate, but the enzyme was lacking in *B.suppressaria*. Occurrence of high quantity of polyphenol-oxidase in *O.coffeae* suggested that the mite could easily overcome the plant defenses.
- The enzyme possibly enabled the species to oxidize a wide range of tea phenolic compounds leading to neutralization the defence allelochemicals of the host leaves ingested along with food.
- Among the detoxifying enzymes, a significantly high quantity of the general esterases (EST) in salivary gland homogenate and midgut homogenate of the pesticide-exposed field specimens of *B.suppressaria* and *Et.magnifica* larvae over laboratory-reared unexposed ones possibly indicated a greater esterase-based detoxifying activity in the former ones.

- The EST-2 and EST-3 bands showed intense staining in *B.suppressaria* specimens collected from conventional plantations. Such high intensity may be related to greater pesticide tolerance / resistance of *B.suppressaria* populations.

- Two soluble esterase isozymes, designated as EST-3 and EST-4 due to their prominent presence in the pesticide-exposed larvae of *Et.magnifica* specimens appeared to be related to pesticide detoxification.

- Higher midgut esterase activity was found in *H.theivora* specimens of conventional plantations, possibly endowing the bug with a greater insecticide tolerance (*vis-a-vis* resistance).

- Esterase bands on polyacrylamide gel showed that the pesticide-exposed female *O.coffeae* possessed 3 major co-migrating band whereas the pesticide unexposed female *O.coffeae* possessed only one. Enhance quantity of esterases as well as the additional bands (isozymes) of *O.coffeae* of the conventional plantation were possibly involved in the detoxification of synthetic acaricides and insecticides.

- Significantly high glutathione S-transferase (GST) quantity in field-collected specimens of *H.theivora* as compared to laboratory-reared specimens was noted. The banding pattern also showed a single low-intensity band in laboratory-reared specimen where as field-collected specimens registered a parallel but a high intensity band.

- Acetylcholinesterase (AChE) quantified in the homogenate of cerebral ganglia of four major pests in question showed a significant difference between the laboratory-reared and pesticide-exposed field-collected ones.
- The zymogram of the acetylcholinesterase of *B.suppressaria* and *Et.magnifica* showed a single band formation with a higher intensity in the pesticide-exposed larvae as compared to the laboratory-reared individuals.
- It was also evident that quantities of acetylcholinesterase that bind with the organophosphates and carbamates, was significantly higher in the specimens of *H.theivora* collected from field compared to the laboratory-reared ones. Electrophoregram pattern of acetylcholinesterase also indicated high band-intensity in the pesticide-exposed specimens and low in unexposed (laboratory-reared) ones.
- Both quantitative and qualitative analysis of the homogenate of *O.coffeae* showed that the quantity of acetylcholinesterase in the pesticide-exposed specimen was significantly higher as compared with the laboratory-reared ones.
- In insecticide inhibition test, treatment with field-recommended doses of organophosphate showed partial or no inhibition of esterase gel bands in all the pesticide-exposed field specimen of the above tea pests. Where as in the laboratory-reared specimens all the bands were largely blocked as evident through their disappearance, suggesting the role of esterase-based pesticide detoxification in these species. The uninhibited or over produced of esterase might confer a greater insecticide tolerance / resistance to these pest species.