7. CONCLUSION

On compiling the entire sets of study, it may be concluded that during post-harvest preservation, aggregation of macromolecules inside xylem lumen and proliferation of microbes causes obstacle in normal conducting pathway, resulting in accumulation of ROS causing disintegration of membrane consequence to senescence, as manifested by yellowing of the leaves preserved with distilled water. Beside this, at post-harvest stage mulberry leaves exhibited gradual decrease in chlorophyll, protein, carbohydrate, and other primary essential metabolite content. Accumulated free radicals resulted in oxidative damage including lipid peroxidation and thus imposing changes in rearing parameters in the current study. Nanosilver and silver nitrate solution as preservative showed efficiency in inhibiting microbial proliferation and ROS generation, thus extending post-harvest shelf life. Leaves preserved in nanosilver and silver nitrate solution showed two line of defensive strategy; first the activation and up-regulation of enzymatic antioxidants (SOD, CAT, GPOX, GSR, APX, GST) and non-enzymatic antioxidant (glutathione, ascorbic acid and carotenoids) activities; and secondly enrichment in the production of secondary metabolites involved in scavenging activity of free radicals. These metabolic alterations allow the leaves to withstand and extend shelf life at post-harvest stage. Leaves preserved in nanosilver solution exhibits greater retention of photosynthetic pigments and other essential primary metabolites, probably due to greater capability to scavenge generated ROS both by enzymatic and non-enzymatic pathways. Further experimental analysis evident that 6 ppm nanosilver solution was more effective than equivalent silver nitrate concentration as indicated by less number of partial and complete vascular blockages and comparatively better shelf life. The metabolic defensive activities were also found to be more in leaves preserved in nanosilver solution than silver nitrate solution. Transcriptome analysis identified significant retention of photosynthetic and chloroplast proteins in nanosilver preserved leaves responsible for green texture and maintaining shelf life through proper regulation of photosynthetic machineries. Shelf life extension in nanosilver preserved leaves was achieved probably through enhanced expression of proteins involved in providing defence by scavenging toxic substances including ROS. Probably blockage of xylem vessels inhibits the conducting pathways and leakage of storage carbohydrates through cut end caused rapid wilting associated

senescence through accumulation of ROS in distilled water preserved leaves. However enhanced expression of different transporters and immune-modulators were observed in distilled water preserved leaves but their collective summation also failed to prolong shelf life by 7 days.

Moreover, feeding leaves to silkworm larvae preserved in nanosilver solution showed better growth and cocoon parameters than that observed after feeding silver nitrate preserved leaves, indicating the absence of any adverse toxic effects. Thus, at post-harvest stage, preserving mulberry leaves using 6 ppm nanosilver solution will help to overcome the problem of larval feeding during rainy season. Thus it may be concluded that green synthesized (phytosynthesized) silver nanoparticles that was characterized as crystalline and spherical in nature with average particle size of 14 nm, bears the potentiality to extent the shelf life of harvested mulberry leaves at a least effective concentration of 6 ppm. Present findings may regain the interest of landless farmers towards this broad field of application as they can go through the process of larval rearing for a period of 7 days by purchasing or collecting the leaves once in seven days.