

5.0 Conclusion

In the attempts to establish a reliable and optimum transformation system for *Cryptocoryne willisii* via *Agrobacterium tumefaciens* strain LBA4404 harbouring the binary vector pCAMBIA1304, factors influencing *Agrobacterium*-mediated transformation i.e. infection time and co-cultivation period were investigated. Six mins of infection followed by one day of co-cultivation period was found to give higher transformation efficiency among all combinations of parameters tested. Expression level of GUS reporter gene was determined using fluorometric assay. The highest GUS enzyme activity was found to be 582.09 ± 84.30 pmol 4MU/ mg/ min. PCR analysis revealed the presence of the reporter gene in regenerated plantlets of *C. willisii*. The integration frequency of *mgfp5* gene in the plant genome was found to be acceptable through Southern Blot analysis with two copies *mgfp5* genes integrated in the sample. In conclusion, the optimized system for *Agrobacterium*-mediated transformation of *C. willisii* could be applied for further transformation studies of other fresh water plants. Nonetheless, further study is required in order to generate more stable transformans.