

Research Article

Genetic Transformation of *Metroxylon sagu* (Rottb.) Cultures via *Agrobacterium*-Mediated and Particle Bombardment

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Sago palm (*Metroxylon sagu*) is a perennial plant native to Southeast Asia and exploited mainly for the starch content in its trunk. Genetic improvement of sago palm is extremely slow when compared to other annual starch crops. Urgent attention is needed to improve the sago palm planting material and can be achieved through nonconventional methods. We have previously developed a tissue culture method for sago palm, which is used to provide the planting materials and to develop a genetic transformation procedure. Here, we report the genetic transformation of sago embryonic callus derived from suspension culture using *Agrobacterium tumefaciens* and gene gun systems. The transformed embryoids cells were selected against Basta (concentration 10 to 30 mg/L). Evidence of foreign genes integration and function of the *bar* and *gus* genes were verified via gene specific PCR amplification, gus staining, and dot blot analysis. This study showed that the embryogenic callus was the most suitable material for transformation as compared to the fine callus, embryoid stage, and initiated shoots. The gene gun transformation showed higher transformation efficiency than the ones transformed using *Agrobacterium* when targets were bombarded once or twice using 280 psi of helium pressure at 6 to 8 cm distance.

1. Introduction

Sago palm (*Metroxylon sagu*) is one of the most important plants contributing to the local economy and grown commercially for starch and/or conversion to animal food or fuel in Malaysia, Indonesia, Philippines, and Papua New Guinea. Sago palm research has been under focus because of the increasing need to explore nontraditional sources of food and fuel. Sago palm has a long life cycle, with an average of 15 years. Due to the long flowering time of sago palm and low seed germination rate, there is no report of breeding programs for sago palm [1], thus requiring alternative means of propagation for sago palm. Successful micropropagation of sago palm leaf tissues via direct shoot formation has been reported by several researchers [2–4]. The development of sago palm tissue culture technique serves as a basis by which genetic transformation can be conducted.

Two of the most common methods for plant genetic transformations are the *Agrobacterium*-mediated and the direct particle bombardment method. *Agrobacterium tumefaciens* is a soil borne, gram-negative bacterium that has a unique characteristic to transfer part of its genome to infect, transform, and parasitize plants. It has the ability to penetrate into cells at a wound site, actively transferring and integrating stably its genetic materials into the plant chromosomes [5]. The transformation mechanism works well with dicotyledonous plants; however, monocotyledonous plants are recalcitrant towards gene transfer using *Agrobacterium*. It is now possible to transform monocots using *Agrobacterium*, namely, via tissue culture; nevertheless various factors needed to be considered that contribute towards a successful genetic transformation. Among the factors involved included the genotype of plants, types and age of tissues used, the *Agrobacterium* strains and binary vectors used, and the various tissue