## J. Appl. Environ. Biol. Sci., 5(4)87-93, 2015 © 2015, TextRoad Publication

ISSN: 2090-4274
Journal of Applied Environmental
and Biological Sciences
www.textroad.com

## Agrobacterium rhizogenes Mediated Hairy Root Induction in Justicia gendarussa Burm.f

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Received: December 23, 2014 Accepted: February 15, 2015

## **ABSTRACT**

Five strains of *Agrobacterium rhizogenes* (LB510, LB509, YMB072001, A4T, and ATCC 15834) were tested to determine their effects on the induction of hairy root on leaf explants of *Justicia gendarussa* Burm. f.. Leaf explants were sterilized, and then infected within liquid MS medium + sucrose containing *A. rhizogenes* OD600 = 0.1, for 20 minutes while being shaken gently by hand. Teh explant was co-cultivated on Murashige and Skoog (MS) solid medium at 25°C for 48 hours. After 48 hours of co- cultivation, explants were transferred to MS solid medium with the addition of 250 mg/L cefotaxim, and repeated 1-2 times. Observations were carried out during 6 weeks to observe the specified parameters. Data were analized by Kruskal-Wallis Test and followed by Mann-Whitney Test. All strains of *A. rhizogenes* were used in this study were able to produce hairy root. The treatment of LB510 and YMB072001 strain was the best strain in the hairy root induction of *J. gendarussa* Burm. f.. The infection frequency of LB510 strain was highest (100 %), longest hairy root (2.23 cm), and highest of hairy root wet weight (0.0243 g). The treatment of YMB072001 strain was the highest number of hairy roots (3.3) per explant and the highest number of hairy roots dry weight (0.0098 g).

**KEYWORDS**: Agrobacterium rhizogenes, hairy root, Justicia gendarussa Burm. f.

## INTRODUCTION

Gandarusa (*Justicia gendarussa* Burm. f.) is an Indonesian medicinal plant, Acanthaceae family. Gandarussa is used for migraine headaches, fever, hemiplegi, paralysis of facial muscles, swelling, ear pain, inflammation, bronchitis, dyspepsia, eye diseases, bleeding, muscle pain, antirheumatic, antinociception, antihepatotoksic and anti malaria [1, 2]. Based on ethnomidicine study, gandarusa (local name) is used for male birth control drug in the Irian Jaya, Indonesia [3]. Prajogo et al. [4] proved the male antifertility gandarusa efficacious clinically. According Prajoga et al. [5] gandarusa is also useful as antiviral.

Utilizations of gandarusa in the industrialized world are faced with a problem its availability in nature. Gandarusa has not been cultivated and grow wildly. Gandarusa cultivation is a relatively long time traditionally. The level and quality of the desired active compounds are not as expected. Levels of 0.9% of the total flavonoid extraction, while gendarusin A 0.03% [6]. Modern biotechnology interventions are needed to address this problems, one of which is the hairy root culture.

Hairy root culture is the hairy root induction techniques by genetic transformation using *Agrobacterium rhizogenes*. Excess hairy root culture is capacity in the production of secondary metabolites similar to or greater than the parent plant [7]. Trigonelline production in *Trigonella foenum-graecum* hairy root cultures produces three to five times that of plant origin [8]. In industry, the hairy root culture is much more effective than cell culture because of its genetic stability. Culture of the hairy roots can also produce recombinant proteins for potential pharmaceutical industry [9].

A. rhizogenes is a genus of gram-negative soil bacterium, is responsible for the formation of hairy root at the site of infection. These bacteria can transfer the T-DNA, which is in Ri (root inducing) plasmid, the size of few hundred kb, from bacteria to plant cells [10]. Many factors affect the success of the hairy root culture. That are strains of bacteria [11,12,13,14,15], the type of plants [11,13,15,16], parts of plants [11,13,15,16,17], and culture medium [7,11,15,18,].

The study of the efficiency of various strains of *A. rhizogenes* on induction on hairy root of gandarusa plants still exist. This study aims to find the right kind of strain for the optimization of the hairy root induction of gandarusa leaf.