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Medium and Embryo Placement Characteristics Affected 5 varieties of Hybrid Kathi Coconut Embryo Germination

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ABSTRACT

Kathi coconut cannot germinate in nature, the embryo rescue technique was used. But the efficiency of seedling production is still low. This research aimed to develop embryo culture for breeding work of the Department of Agriculture. The result showed that medium and embryo placement characteristics affected 5 varieties, NamHom x Kathi (NHK), Malayan Dwarf x Kathi (RDK), Thungkled x Kathi (TKK), West African Tall x Kathi (WAK) and Malayan Yellow Dwarf x Kathi (YDK), of hybrid Kathi coconut embryo germination. In the dark, the germination rate of solid medium ranged from 69.0 to 99.7 percent, while the liquid culture was 50.7 - 72.3 percent, embryo germination in solid medium was better than in liquid mediums. The hybrid coconuts of RDK, TKK, WAK and YDK cultivars where were grown in Murashige and Skoog (MS) solid medium with the addition of 2,4-Dichlorophenoxyacetic acid (2,4-D) 1 mgl^{-1} showed that embryo germination was better than in modified Y3 solid medium, and also MS solid medium with the addition of 2,4-D 1 mgl^{-1} found longer shoots than the modified Y3 solid medium.

Keywords: *embryo placement, germination, hybrid, Kathi Coconut, medium*

INTRODUCTION

The coconut (*Cocos nucifera* L.) is a major Thai economic crop. At present, the major growing areas are still in the south especially in Prachuap Khiri Khan, Chumphon and Surat Thani provinces. in 2008 – 2013, productive area and yield decreased with age and plant conditions, because most of the area is the old coconut plantation. And in 2010, an outbreak of coconut pests and the drought during the relatively dry weather to suitable for the infestation of such insects. As a result, the coconut yield is less. Conduce to a shortage of raw materials, high price per fruit. As a result, farmers have increased demand for good coconut varieties. But the government has insufficient production capacity.

Embryo culture is a technique that has been practiced by breeders for a long time. The key benefit is Helping the embryos of plants that cross-species or cross genus and become sterile to grow into a complete plant. Kathi coconut cannot germinate in nature, the embryo rescue technique was used. But the efficiency of seedling production is still low. Using Kathi coconut embryo rescue technique, the zygotic embryos were successfully cultured in several laboratories. (Ashburner, 1991, Assy-Bah, 1989, Karunaratne et al., 2009, Rillo and Paloma, 1990) In Thailand, Somchai et al. (2008) successfully made Kathi coconut embryo culture and this technique is currently used as a good Kathi coconut production system of the Department of Agriculture. The result showed that medium and embryo placement characteristics affected 5 varieties, NamHom x Kathi (NHK), Malayan Dwarf x Kathi (RDK), Thungkled x Kathi (TKK), West African Tall x Kathi (WAK) and Malayan Yellow Dwarf x Kathi (YDK), of hybrid Kathi coconut embryo germination.

MATERIALS AND METHODS

Embryos of hybrid Kathi coconut 5 varieties, NamHom x Kathi (NHK), Malayan Dwarf x Kathi (RDK), Thungkled x Kathi (TKK), West African Tall x Kathi (WAK) and Malayan Yellow Dwarf x Kathi (YDK) were isolated from 11 months old fruit from Suratthani Seed Research and Development Center Tha Chana District, Suratthani 88170. They were shaken in 70% alcohol for 5 min fallowed by in 15 and 10% Clorox solution for 15 and 10 min and then washing with sterile distilled water 3 times in laminar air flow station.

The experiment has a completely randomized design with 3 treatments, consist of hybrid Kathi coconut 5 varieties' embryo that the fruiting age 11 months and culture medium with Embryo Placement Characteristics; modified Y3 liquid medium (Parinda, 2018) (Figure 1A), modified Y3 solid medium with placed upward (Figure 1B) and Murashige and Skoog (MS) solid medium with the addition of 2,4-Dichlorophenoxyacetic acid (2,4-D) 1 mgl^{-1} (Orathai, 2019) with placed upward. Each embryo was cultured in the dark and taken for 8 weeks. Number of embryo's germination and development were observed and recorded each 2 weeks from 2 months after culturing.

Eight weeks later when shoots started growing, they were sub-cultured in modified Y3 solid medium and transferring to the light under the illumination of cool-white, fluorescent tubes of about 37 µmol m^{-2} s^{-1} for 16 h/day photoperiod, 25+2 ^{o}C about for 8 weeks. Number of shoot and plantlet's development were observed and recorded each 2 weeks from 2 months.

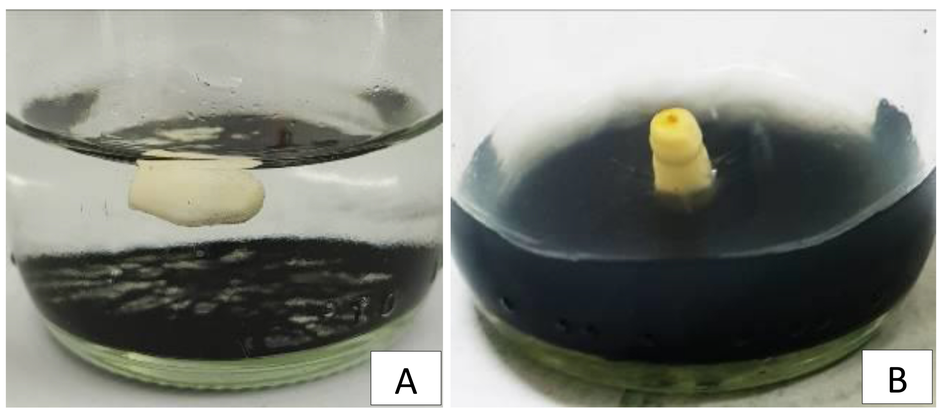

Figure 1 Embryo in liquid medium (A) and in solid medium with placed upward (B)

RESULTS

Table 1 Embryo germination percent of five varieties hybrid Kathi coconut after 8 weeks of culturing in medium with embryo placement characteristics in the dark.

Treatment	Embryo germination in the dark (percent)				
	NHK	RDK[1/]	TKK[1/]	WAK	YDK
modified Y3 liquid medium	51.0	53.3 b	50.7 b	60.0	72.3
modified Y3 solid medium with placed upward	86.7	74.0 ab	86.7 a	69.0	74.0
MS solid medium with 2,4-D 1 mgl^{-1} with placed upward	80.0	86.7 a	99.7 a	93.3	82.3
C.V. (%)	44.8	20.0	18.5	29.5	22.7

[1/] The averages in the same column that follow with the same letter were not statistical difference at 95% confidence level by DMRT

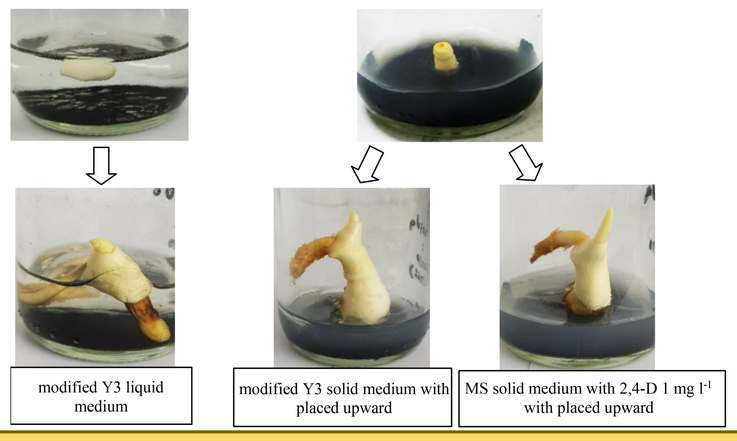

Figure 2 Embryo germination of hybrid Kathi coconut after 8 weeks of culturing in medium with embryo placement characteristics in the dark.

Table 2 Embryo development percent of five varieties hybrid Kathi coconut, from were cultured medium with embryo placement characteristics in the dark, after 8 weeks of sub-culturing in modified Y3 solid medium and transferring to the light.

Treatment	Embryo development in the light (percent)				
	NHK[1/]	RDK	TKK[1/]	WAK[1/]	YDK
modified Y3 liquid medium	28.7 b	45.0	35.7 b	53.3 b	45.3
modified Y3 solid medium with placed upward	80.0 a	62.7	86.7 a	69.0 ab	62.7
MS solid medium with 2,4-D 1 mgl^{-1} with placed upward	73.3 a	86.7	99.7 a	93.3 a	82.3
C.V. (%)	35.5	34.8	27.8	27.5	30.6

[1/] The averages in the same column that follow with the same letter were not statistical difference at 95% confidence level by DMRT

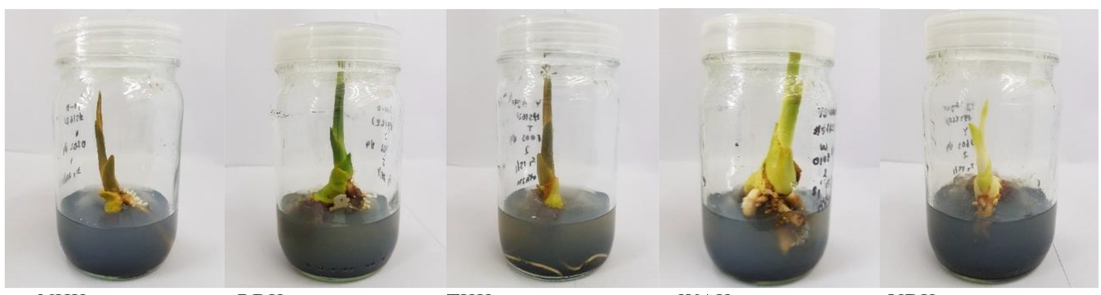

Figure 3 Embryo development percent of five varieties hybrid Kathi coconut after 8 weeks of sub-culturing in modified Y3 solid medium and transferring to the light.

DISCUSSION

To study the effect of Medium and Embryo Placement Characteristics on the percent germination in the dark, found that 5 varieties hybrid Kathi coconut embryo germination in solid medium was better than in liquid mediums (Table 1). After culturing 8 weeks in the dark, when they were sub-cultured in modified Y3 solid medium and transferring to the light, it was found that percent of embryos development from were cultured in the both solid mediums are better than from were cultured in liquid medium (Table 2). In accordance with Pech Y Ake et al. (2004) studied to enhanced aerobic respiration improves In Vitro coconut embryo germination and culture. Germination of Malayan Green Dwarf (MGD) coconut embryos was tested in liquid and solid medium. It was found that the percentage of germination increased when the embryo was fed on solid medium, especially when the embryo with the micropyle side is placed upward. Causing exposure to the air inside the bottle and embryo proliferation is inhibited when the ambient atmosphere is replaced by N_2 or when anaerobic respiration inhibitors are added to medium. The result showed that embryo proliferation requires aerobic respiration and germination in an upward position will result in better seedling development.

CONCLUSIONS

Medium and embryo placement characteristics affected 5 varieties of hybrid Kathi coconut embryo Germination, it was found that the embryos were cultured in solid medium in the dark showed the best embryo germination and development to plantlet.

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