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#### SHORT COMMUNICATION

# THE EFFECT OF NATURAL SUBSTANCES ON THE RADICLE DEVELOPMENT OF CERTAIN PLANT SEEDS DURING THE EARLY GERMINATION STAGE

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#### ABSTRACT

Successful seed germination is crucial in many plants. Natural substances and chemicals have been used as growth enhancement tools. In the meantime, natural substances are considered environmentally friendly and cost-effective. Hence, the present study was focused on the effects of natural substances such as red rice, white rice washed water, orange coconut, green coconut, and matured coconut water on radicle development of black gram, pumpkin, rice, and maize. Twenty-five healthy sterilized seeds (85% germination rate) of the above plants were pre-soaked in 30 ml of the natural suspension for 24 hours separately at room temperature and repeated twice. Then seeds were transferred to the sterilized petri dish with moistened filter paper and mean values of length of the radicle were measured. Data were subjected to one-way analysis of variance (ANOVA) (P<0.05) followed by Tukey test. Natural substances have not affected radicle development of the tested seeds, except pumpkin, and maize after 24hours whereas in the control radicle length of these two species was 5.48 and 0.68mm, respectively. However, interestingly after 48hours, radicle formation of most tested seeds was highly accelerated by red rice and white rice washed water, except for black gram. Hence, orange coconut, green coconut, and matured coconut water have showed an inhibitory effect on root length development, except for maize at 48 hours. On the other hand, among the selected natural substances red rice and white rice washed water revealed better effects on radicle development. So, they can be used as an environmentally friendly method to stimulate the radicle development.

Keywords: Radicle development, natural substances, coconut water, rice water

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#### **1. INTRODUCTION**

In many plants, successful seed germination is a crucial step in the environment. Various methods can be used to enhance seed germination, such as chemical treatments  $(H_2SO_4, KNO_3)$  and growth regulators [1]. In the meantime, natural substances are considered better for the environment and cost-effective to accelerate germination.

Sprouts get their nutrients from stored food rather than from the environment during the germination process. However, in order for the germination process to take place properly, a good growing environment is needed to promote the mobilization of stored food.

Coconut water is a natural source of growth regulators, also known as phytohormones. It's used in micropropagation as a growth regulator [2]. Sugar and minerals are the primary components of coconut water, Auxin (Indole Acetic Acid – IAA), Abscisic acid (ABA), gibberellins, and zeatin (cytokines) are phytohormones found in coconut water in varying quantities [3]. The sugar content in the young coconut water is 3.23 %, in half old it is 5.73 % and in the old (nine months old) it is 5.19 %. [4], according to the auxin-cytokinin hypothesis, cytokinins, together with auxins, play an important role in plant morphogenesis by controlling the formation of roots and shoots and moderating their relative growth [5] and Gibberellic acid is a crucial chemical molecule in the process of seed germination. In addition to stimulating the formation of amylase, gibberellins also stimulate the synthesis of ribonuclease, protease, cellulase and peroxidase [6]. The presence of gibberellin, cytokinin and auxin in coconut water medium allows the translocation of glucose and amides from the storage area to the growth area, cell division and cell enlargement and the next it will be a process of cell differentiation. Consequently, seeds that had been able to germinate [3].

However, the chemical composition of coconuts varies depending on their stage of development, variety, and cultivation techniques [2]. Therefore, Different forms of coconuts have different phytohormone content.

The phytohormone content of different coconut varieties is thought to have a different influence on seed germination [2]. In addition, Coconut water is also used in plant cuttings to speed up root initiation and growth [7].

Washing rice water contains up to 7% protein, 30% crude fiber, 15% free amino acids, 25% calcium (Ca), 47% total phosphorus (P), 47% iron (Fe), 11% zinc (Zn), 41% potassium (K), 59% thiamine, 26% riboflavin, and 60% niacin [8]. Hence, rice wash water is high in basic plant nutrients such as N, P, and K, but it is also high in sugars, mostly Amidon, as well as other carbohydrates. Moreover, wash rice water has enriched with a high content of starch. It is beneficial to seed germination [8]. As a result, rather than discarding washed rice water, it can be used as an ingredient for enhancing seed radicle development.

Hence, the present study was conducted to screen the effect of selected natural substances on the radicle development of *Oryza sativa* (Rice), *Cucurbita maxima* (Pumpkin), *Vigna mungo* (Black gram), and *Zea mays* (Maize), which are most extensively cultivated crop in Sri Lanka, where these natural substances are low cost, eco-friendly and easily available.

# 2. MATERIALS AND METHODS

## 2.1 Collection of plant seeds and natural substances

Plant seed, *Oryza sativa* (Rice), *Cucurbita maxima* (Pumpkin), *Vigna mungo* (Black gram), and *Zea mays* (Maize) were collected from Agriculture farm, Eastern University, Sri Lanka. Five different natural substances such as red raw rice washed water (100g rice was soaked in 250ml distilled water for 30 minutes), white raw rice washed water (100g rice was soaked in 250ml distilled water for 30 minutes), green king coconut water, orange king coconut water, and matured coconut water also collected. Red and White raw rice was collected from the Department of Agriculture, Vantharumoolai and different coconuts were collected from Agriculture farm of the Eastern University, Sri Lanka.

## 2.2 Experiment site and design

A lab experiment was conducted at the Department of Botany, Faculty of Science Eastern University, Sri Lanka. The trial was laid out in a Completely Randomized Design (CRD) with three replicates.

## 2.3 Seed germination study

Twenty-five surface-sterilized healthy seeds (85% germination rate) of black gram, pumpkin, rice, and maize plants were pre-soaked in 30 ml of different natural suspension (red rice washed water-T1, white rice washed water-T2, orange coconut water -T3, green coconut water-T4, and matured coconut water-T5) for 24 hours, separately. Whereas seeds soaked in sterilized distilled water used as the control. Before transfer the seed into the Petri dishes, the seeds were fully washed with distilled water. Then seeds were transferred in filter paper which was moistened with distilled water in a 9 cm diameter Petri dish, (25 seeds/ petri dish) for radicle development studies. Radicle length was measured by using a ruler in different intervals, such as 24 and 48 hours. Data were statistically analyzed using analysis of variance (ANOVA) (P<0.05), and mean separation was done using the Tukey test.

## **3 RESULTS AND DISCUSSION**

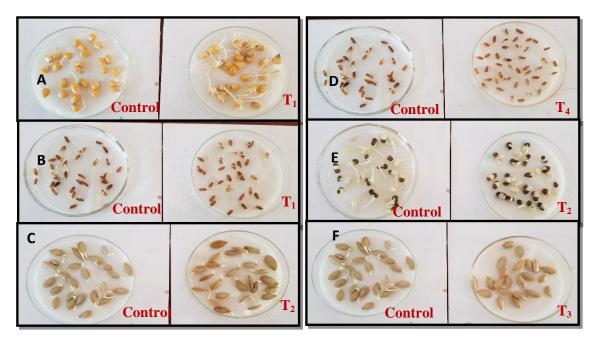
The Effect of radicle development depends on the type of natural substances. The treatments were significantly influenced in selected seed radical development except Maize. (Rice P=0.000, Pumpkin P=0.000, Black gram P=0.000 and Maize P=0.680 and P=0.250in Day 1 &2). Natural substances showed different degrees of radicle development that leads to germination in different plant seeds. The effect of locally available selected natural substances on radicle formation is shown in Table 1.

**Table 1:** The mean radicle length (mm) in seeds of study species with different natural substances in Day 1 and Day 2. Differences among the treatments given by results of ANOVA, Turkey analysis. In columns, different letters indicate significance at the given probability (P<) at 0.05 level. T<sub>1</sub>- Red rice washed water, T<sub>2</sub>- White rice washed water, T<sub>3</sub>- Orange coconut water, T<sub>4</sub>- Green coconut water, T<sub>5</sub>- Matured coconut water.

Seed Type	Treatment	Average length of the radicle±SE(mm) in day1	Average length of the radicle±SE(mm) in day2
Seed Type	T1	0.96±0.9 <sup>b</sup>	22.28±3.6 <sup>a</sup>
	T2	0.36±0.5b °	17.32±2.9 <sup>b</sup>
Rice	T3	$0^{\circ}$	$6.88\pm2^{c}$
	T4	0.12±0.3 <sup>c</sup>	$9.72\pm9^{c}$
	T5	$0.96 \pm 0.2^{b}$	$8.76 \pm 4.6^{\circ}$
	Control	$2.4\pm1.5^{a}$	$14.5\pm5.8^{b}$
	Control	P < 0.05	110_010
	T1	$8.28{\pm}1.5^{a}$	43.12±3.6 <sup>a</sup>
	T2	$6.36 \pm 2.3^{b}$	37.56±5.3 <sup>b</sup>
Pumpkin	Т3	$0^{c}$	$0^{c}$
	T4	$0^{c}$	$0^d$
	T5	$0^{c}$	$2.8{\pm}2^{d}$
	Control	$5.48{\pm}1.8^{ m b}$ P < 0.05	23.8±10.6 <sup>d</sup>
	T1	8.76±3.4 <sup>c</sup>	26.32±9.9 <sup>b</sup>
	T1 T2	$8.56\pm4.2^{\circ}$	$10.12\pm6.6^{\circ}$
Black gram	T2 T3	$12.24 \pm 4.6^{b}$	$27.36\pm8.9^{b}$
	T3 T4	$12.68 \pm 4.8^{b}$	$36.4\pm8.7^{a}$
	T5	$12.44\pm2.8^{b}$	22.76±5.8 <sup>b</sup>
	Control	$12.44\pm2.5^{a}$ 16.04±2.5 <sup>a</sup> P <0.05	24.48±11 <sup>b</sup>
	T1	1.28±1.1ª	7.68±8 <sup>a</sup>
	Τ2	$0.84{\pm}0.6^{a}$	$4.64 \pm 3.6^{a}$
Maize	Т3	$0.8{\pm}0.7^{\mathrm{a}}$	$4.4{\pm}3.2^{a}$
	T4	$0.44{\pm}0.3^{a}$	$7.48{\pm}7^{a}$
	T5	$0.68{\pm}0.5^{a}$	$8.44{\pm}6.44^{a}$
	Control	$0.68{\pm}0.4^{a}$	$3.96 \pm 6^{a}$
		p > 0.05	

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In the formation of radicle development of rice and black gram, none of the treatments were effective in 24 hours, but  $T_1$  and  $T_2$  were effective in the radicle development of the *Oryza* seeds in 48h, and treatments  $T_1$ ,  $T_3$ , and T4 were effective in black gram in 48 hours. In pumpkin treatments,  $T_1$  and  $T_2$  were effective on both days. In Maize, except for  $T_4$ ,  $T_5$  other treatments were effective on day 1 compared to control.



**Figure 1:** Effect of different treatments compared to the control treatment on radicle development of maize (A), Rice (B & D), Pumpkin (C & F) and Black gram (E).

Moreover, all the treatments were effective in enhancing radicle development after 2 days of treatments (Table 1). This may be because; the growth regulators, such as auxin, gibberellins, cytokines, and abscisic acid, present in coconut water have an enhancing effect on radicle development [1]. As an example, it has been reported that Gibberellins induce germination by activating the enzymes that remodel food reserves [9].

In 48h, black gram showed a higher root length on different varieties of coconut water such as orange coconut water and green coconut water, and the radicle lengths were 27.4 and 36.4mm, respectively, whereas the control seeds had a radicle length of 24.5mm. Setyaningsih [10] mentioned that red palm seeds treated with coconut water germinated within 14 days, whereas without coconut water, seeds took > 14 days to germinate, and also, Wulung coconut water showed a better effect on Charismas palm seedlings germination [2]. Among the studied types of coconut water, green coconut water showed better results than other types. A previous study also revealed that coconut with different ages and different varieties had different effects on the germination of cucumber seeds [3]. The treatments T3, T4, and T5 have an inhibitory effect on tested seeds of rice and pumpkin.

Radicle development of rice, maize, and pumpkin were induced by red raw and white raw rice washed water. Among these seeds, pumpkin seeds were germinated more effectively. It was observed 8.3mm and 43.1mm for  $T_1$  and  $T_2$  revealed 6.4mm and 37.6mm of radicle at 24h and 48h, respectively. In contrast, control showed less radicle development than rice washes water, which was 5.5mm (24h) and 23.8mm(48h).

The different treatments were significantly influenced in selected radicle development except in Maize. Among different treatments in radicle development, the control showed a significant effect on rice and black gram on day 1. The treatment  $T_3$ ,  $T_4$ , and  $T_5$  have an inhibitory impact on most tested seeds. On the other hand, among the selected natural substances  $T_1$  and  $T_2$  revealed better effects on seed germination. There were no effects of natural substances on the radicle development of the tested seeds except pumpkin and maize seeds treated with  $T_1$  and  $T_2$  in 24 hours. However, more interestingly, after 48 hours (Figures 1), radicle formation of most tested seeds was highly accelerated by  $T_1$  and  $T_2$ , except for black gram treated with  $T_2$ .

#### CONCLUSION

In this study, the radicle development of seeds of most of tested species was not enhanced by orange, green, and matured coconut water. On the other hand, among the selected natural substances red and white rice water had better effects on radicle development of the seeds of studied species. Thus, Red and white rice water can be used as eco-friendly and low-cost methods to induce seed radicle development.

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