MJSIR
Mountain Journal of Science and Interdisciplinary Research
PRINT ISSN: 2619-7855
ONLINE ISSN: 2651-7744
January-June 2023 • 83 (1) : 7-18

Performance of Seed-Primed Ricebean (*Vigna umbellata* L.) Using Organic-Based Priming Materials and Priming Duration

Hector C. Gayomba*, Belinda A. Tad-awan, Hilda V. Palaes, and Juniel S. Busiguit

College of Agriculture, Benguet State University
Author email address: h.gayomba@bsu.edu.ph

ARTICLE INFO
Date Received: 03-04-2021
Date Last Revised: 08-12-2022
Date Accepted: 02-15-2023

KEYWORDS
organic-based priming materials
seed priming
priming duration
ricebean

Abstract

Ricebean is valued for its nutritional content and as a legume, it provides soil health benefits which is an essential element in farming systems. Long storage of its seeds affects the seed quality which lessens the production as well as the income of the grower. For these reasons, seed priming is practiced to enhance seed performance for better crop stand. Thus, this study was conducted from January to June 2019 at Benguet State University (BSU) experiment area, La Trinidad, Benguet to determine the best organic-based priming materials for ricebean production; the best soaking duration for ricebean seeds; the interaction effect of organic-based priming material and soaking duration of seeds on the growth and yield of ricebean; and the profitability of ricebean primed at different duration using organic-based materials. Ricebean seeds primed with garlic extract has the earliest emergence and number of normal seedlings, heaviest seeds, and total seed yield. Among the soaking duration, 6 hours has the earliest emergence; highest percent emergence; the greatest number of normal seedlings and seeds per pod; and the heaviest 200 seed-weight and total seed yield. The growth and yield of the ricebean plants were not considerably influenced by the interactions of the priming duration and organic-based priming materials. Garlic extract priming material and six hours of priming showed the highest return on cash expenses.

Introduction

In the Philippines, leguminous crops are well-acknowledged as an inexpensive source of protein. Ricebean (*Vigna umbellata* L.), compared to other legumes of the vigna family has higher nutrient quality with 20.78% protein, 59.96% carbohydrates, and high bio-availability of 59.8% calcium (Bepary, 2016). The crop is commonly grown for food, fodder, and intercropped with cereals in the upland tropics. It is cheaply grown for decades in the highlands of the Philippines (Tad-awan et al., 2005).

Long-stored seeds have low ability to germinate and produce a normal seedling, are less metabolically active, and have poor establishment of seedlings even in favorable conditions. It is known to be seed physiological malfunctioning and physical deterioration (Silva et al., 2018). This condition lessens the production as well as the income of growers.

Seed priming is a promising method to improve the quality of seeds which is likely to increase the production of crops. It is an effective technology to enhance rapid and uniform

emergence and to achieve high vigor leading to better establishment and yield (Farooq et al., 2007). Depending on the plant specimen, seed morphology, and physiology, different priming treatments can be applied triggering the 'pre-germinative metabolism" of the seeds. This physiological process takes place during early seed imbibition and includes the seed repair response such as the antioxidant mechanism, ensuring proper germination and seedling development.

This study was conducted to (1) determine the best organic-based priming materials in ricebean production, (2) identify the best priming duration for ricebean seeds, (3) to determine the interaction effect of organic-based priming material and priming duration of seeds on the growth and yield of ricebean; and (4) to assess the profitability of ricebean primed with organic-based materials at different priming durations.

Materials and Methods

Experimental Design and Field Preparations

The study was laid out following Split-Plot Design. An area of 375m² was thoroughly prepared and then divided into 75 plots measuring 1mx5m each. BSU growers' compost was applied at the rate of five kilograms per 5m² one week before planting. The main plot is the soaking Duration (S) (S_1= Unsoaked (Control), S_2=2 hours, S_3=4 hours, S_4= 6 hours, and S_5= 8 hours) while the sub-plot is the priming materials (O) (O_1= Water (control), O_2= Garlic extract, O_3= Horsetail Tea, O_4= Oriental Herbal Nutrient (OHN), and O_5= Wood vinegar).

Preparation of Priming Materials

Garlic extract was prepared by peeling the composite garlic bulbs before extracting juice. A domestic juicer was used to extract the juice. The extracted garlic juice was diluted with water at a ratio of 1:2 (Islam et al., 2001).

The horsetail tea was prepared by boiling 125 grams of air-dried horsetail plants in 4 liters of water (Koeppe et al., 1971) for 20-30 minutes (Naidu & Paroha, 2008). The tea was cooled and used as a priming material.

Oriental herbal nutrient (OHN) was prepared following the process published by South Asia Rural reconstruction (SARRA) in 2015. One kilogram of ginger (or turmeric) and garlic were chopped and placed in a separate clean glass jar. An equal amount (weight) of brown sugar or molasses was added to each glass jar and was then covered with Muslim paper or clean cloth. After 5 to 7 days, vodka or any liquor with 40% proof was added until the glass jar was full. The mixtures were stirred every morning for 14 days. For the preparation of the priming solution, the two OHN (garlic and ginger) were mixed following a ratio of 1:1 and were diluted with water to a ratio of 1:1000.

For the wood vinegar, the solution was diluted with water to a 1: 100 ratio (Jothityangkoon et al., 2007).

Seed Priming and Planting

Seven-month stored seeds of ricebean were used for the study. The seeds were primed with different organic-based materials at two hours, 4 hours, 6 hours, and 8 hours then it was sown at a distance of 30 x 30 between hills and rows. Three seeds were sown per hill.

Data Collection

Growth and Development

Percent of Emergence. Recorded by counting the seeds germinated after 50% of the seeds sown per plot have fully emerged. It was computed by dividing the number of seeds germinated by the number of seeds sown multiplied by 100.

Days to Emergence. Recorded by counting the days from sowing to the time when at least 50% of the plant emerges.

Number of Normal and Abnormal Seedlings. Gathered per plot by counting the number of normal seedlings or those with normal leaves and roots and abnormal seedlings or those that exhibit curled leaves, thick stems (beyond normal), and are etiolated.

Seedling Vigor. Computed using the formula of Copeland (1976) adding the normal seedling divided by the added days of germination multiplied by 100.

Plant Survival. Obtained by counting the number of seedlings that survived divided by the number of seeds sown multiplied by 100.

Yield and Yield Component

Number of Seeds Per Pod. Gathered by counting the seeds of 10 randomly chosen ripe pods.

Seed Weight (g). Two hundred sample seeds at 14% moisture content were selected randomly and weighed using sensitive weighing scale.

Total Seed Yield Per Plot (kg). Gathered by weighing the seed yield per plot.

Return on Cash Expenses (ROCE). Computed by dividing the net profit over the cost of production multiplied by 100.

Data Analysis

Using SAS software, all quantitative data were statistically analyzed using the analysis of variance (ANOVA) for split-plot design with three replications. The significance of differences among treatment means were tested using the Least Significant Differences (LSD) at 5% level of significance.

Results and Discussion

Percent of Emergence

Soaking duration has significantly affected the percent of emergence of the ricebean seeds (Table 1). The percentage of emergence is higher on primed seeds compared to unprimed seeds. This result implies that seed priming primarily boosted the germination processes in breaking seed dormancy, hydrolysis of inhibitors, and enzyme activation as acknowledged by (Ajouri et al., 2004). Priming induces quantitative changes in biochemical and physiological activities promoting faster germination (Sung & Chang, 1993) and Karssen et al. (1989). Two, four, and six hours of seed priming resulted in faster emergence indicating its significance over other soaking duration used. Arif et al. (2008) also presented that soybean seeds soaked for six hours were superior in terms of percent emergence. These results suggest that proper soaking duration can ensure high percent emergence and prolonged soaking lowers enzymatic activity that may inhibit germination.

Table 1

Percent Emergence and Days to Emergence of Ricebean Seeds Primed with Organic-Based Materials at Different Soaking Durations

TREATMENT	EMERGENCE (%)	DAYS TO EMERGENCE
Soaking Duration (S)		
Control (Unsoaked)	86.77[b]	17.67[b]
2 hours	94.28a	14.33[a]
4 hours	93.89[a]	16.73[ab]
6 hours	94.22[a]	14.07[a]
8 hours	91.89[ab]	16.33[ab]
Organic-based Priming Material		
Water (control)	91.08	17.00[b]
Garlic extract	93.00	14.73[a]
Horsetail tea	92.85	15.40[ab]
OHN	92.52	16.20[ab]
Wood vinegar	92.26	15.80[ab]
N x S	ns	ns
CV_a	1.18	14.64
CV_b	1.00	12.18

It was observed that the percent emergence did not vary with the use of different organic-based priming materials (Table 1). However, seeds primed with garlic extract recorded 93% emergence followed by seeds treated with horsetail tea (92.85%). The lowest percent of emergence was noted on seeds primed with water having 91.08%. This result suggests that without any effect of the priming materials, the percent emergence of ricebean seeds can be improved through the soaking duration. Arif et al. (2003) and Arif et al. (2005) came up with the same result in mungbean and maize.

There was no significant interaction noted between soaking duration and organic-based priming materials for percent emergence.

Days to Emergence

Days to emergence of the seeds were significantly affected by seed priming (Table 1). Seeds soaked for 2 hours and 6 hours were the earliest to emerge but comparable to that of seeds soaked for 4 and 8 hours. The unsoaked seeds were the latest to emerge. Farooq et al. (2007) stated that seed priming is attributed to faster seedling emergence as the seeds' pre-germinative metabolic activities were completed and the seeds were prepared for radicle protrusion before planting. This result may be due to the invigorating effects of priming on the early stages of the germination process through cell division intervention in the germinating seeds as Hassanpouraghdam et al. (2009) stated. Ricebean seeds that were soaked for two and six hours emerged faster compared to other treatments which are the same in sorghum, millet, cotton, beans, and maize as reported by Murungu et al. (2003), Harris (1996), and Harris et al. (1999). It was also reported that seed priming effectively improved better seedling emergence in barley and chickpea (Ghassemi-Golezani et al., 2008).

The use of organic-based priming materials resulted in significant differences in the number of days to the emergence of the seeds (Table 1). Seeds primed with garlic extract were the earliest to emerge with 14 days average followed by horsetail tea (15 days). Seeds primed with water were the latest (17 days) while seeds primed with garlic extract were the earliest to emerge indicating its efficiency over other priming materials used. Ali et al. (2019) made the same observation in eggplant where garlic extract significantly shortened the days to seedling emergence.

Soaking duration and organic-based priming materials have no significant interaction recorded in terms of days to the emergence of ricebean seeds.

Number of Normal Seedlings

Seed priming effectively increased the number of normal seedlings compared to the unsoaked (Table 2). Seeds that were soaked for 8 hours produced 78 normal seedlings comparable with those that were soaked for 4 and 6 hours which produced 77. Un-primed seeds recorded 65 normal seedlings. Priming effectively increased the number of normal seedlings indicating the improved performance of the ricebean through the activated chemical processes inside the seed as indicated by Sivritepe and Sivritepe (2016) and Singh et al. (2015). Seeds soaked for 8 hours had the highest number of normal seedlings. This finding is in support of Nawaz et al. (2013) who stated that normal seedlings were substantially related to seed imbibition wherein the more diffused the seeds are, the more normal the seedlings are, and this is only done by longer soaking duration.

As presented in Table 2, seeds primed with garlic extract significantly produced the highest number of normal seedlings (76). However, it is comparable to OHN, wood vinegar, and horsetail tea which produced 75, and 73 normal seedlings respectively. Garlic extract, horsetail tea, OHN, and wood vinegar produced the highest number of normal seedlings establishing significance over hydro-priming. This may be due to the various compounds, such as vitamins, flavonoids, phenolic compounds and enzymes in the priming materials that act as growth promoting factors during seed germination resulting in normal seedlings (Hayat et al., 2018)

No significant interactions were recorded between soaking duration and organic-based priming materials on the normal seedlings of ricebeans.

Number of Abnormal Seedlings

Primed ricebean seeds have a significantly lower number of abnormal seedlings compared to the non-primed seeds (Table 2). Seeds soaked

Table 2

Number of Normal and Abnormal Seedlings of Ricebean Seeds Primed with Organic-Based Materials at Different Soaking Durations

TREATMENT	NUMBER OF SEEDLINGS	
	NORMAL	ABNORMAL
Soaking duration (S)		
Control (Unsoaked)	64.93^{c}	13.40^{d}
Two hours	74.27^{b}	10.60^{c}
Four hours	77.53^{ab}	7.03^{b}
Six hours	77.20^{ab}	7.60^{b}
Eight Hours	77.87^{a}	4.93^{a}
Organic-based Priming Material (O)		
Water (control)	72.47^{b}	8.93^{ab}
Garlic Extract	75.60^{a}	8.26^{a}
Horsetail tea	73.27^{ab}	10.53^{b}
OHN	75.40^{ab}	7.93^{a}
Wood vinegar	75.07^{ab}	7.93^{a}
N x S	ns	ns
CV^{a}	2.85	44.30
CV^{b}	5.56	23.22

for eight hours noted 5 abnormal seedlings followed by two hours with 11 abnormal seedlings. Seeds that were not soaked recorded the highest (13) number of abnormal seedlings. The number of abnormal seedlings was lowered indicating improvement in crop establishment. This result conforms to Jafar et al. (2012) who stated the same finding on wheat. The enhanced percent emergence of the seeds may have caused the decreased number of abnormal seedlings.

The number of abnormal seedlings was considerably affected by the organic-based priming materials used as presented in Table 2. Seeds soaked with wood vinegar and OHN have the least number of abnormal seedlings (8) which are comparable to those seeds soaked with water and seeds not treated (8 and 9). Lesser abnormal seedlings recorded from wood vinegar, OHN, and garlic extract were probably due to the organic acids, phenolic, alkone, alcohol, and ester compounds with an acetic acid component of wood vinegar (Mu et al., 2003) that enhanced the growth of the roots and stems of the seedlings, OHN activated the seedling growth and increased plant robustness (Chang et al., 2014) lowering the number of abnormal seedlings. Seeds soaked in horsetail tea had the greatest abnormal seedlings due to their phytochemical component (Siddiqui & Al-Whaibi, 2013) that induced the seedling to develop curled leaves.

No interaction effects of soaking duration and organic-based primers were observed on the number of abnormal seedlings of ricebean.

Seedling Vigor

The seedling vigor of the ricebean seeds was not affected significantly by the priming duration (Table 3). However, two, four, six, and eight hours were rated 4 in terms of seedling vigor while unsoaked seeds were rated 3. The seedling vigor of the ricebean seeds was not significantly affected by the priming duration. As Ventura et al. (2012) stated that enhanced seed vigor is dependent on the DNA repair mechanisms activated during seed imbibition, this result may imply that the DNA repair mechanism activated during seed imbibition did not vary significantly.

Table 3

Seedling Vigor and Plant Survival of Ricebean Seeds Primed with Organic-Based Materials at Different Soaking Durations

TREATMENT	NUMBER OF SEEDLINGS	
	NORMAL	ABNORMAL
Soaking Duration (S)		
Control (Unsoaked)	3.14	63.33[b]
2 hours	4.31	74.53[a]
4 hours	4.40	72.73[a]
6 hours	4.16	76.73[a]
8 Hours	4.26	73.93[a]
Organic-based Priming Material (O)		
Water (control)	3.95	70.67
Garlic Extract	4.14	73.27
Horsetail tea	4.20	72.20
OHN	4.19	71.73
Wood vinegar	4.09	73.40
N x S	ns	ns
CV[a]	6.44	12.66
CV[b]	10.03	8.20

Likewise, no significant differences were observed in the seedling vigor of ricebean as affected by organic-based priming materials as presented in Table 3. The result presents that horsetail tea recorded the most vigorous seedlings (4.2) while priming with water resulted in 3.95 seed vigor. This result implies that hydro-priming can produce seedlings that are as vigorous as the seedlings produced by other priming materials. Ghassemi-Golezani et al. (2008) also reported that lentil seeds primed with water resulted in high seedling vigor.

The treatment interaction between soaking duration and organic-based priming materials did not significantly affect the seedling vigor of ricebean seeds.

Plant Survival

The plant survival of ricebean was significantly affected by the soaking duration (Table 3). The ricebean seeds that were soaked at different duration had higher plant survival than those seeds that were not soaked. The same result was observed in the number of days to seedling emergence and the number of normal seedlings. These results imply the faster establishment of seedlings from primed seeds.

Organic-based priming materials affect the survival of the ricebean plants (Table 3). The plant survival ranged from 70.67% to 73.27%. This result suggests that without any effect of the priming materials, plant survival of ricebean seeds can be increased through soaking duration.

There was no significant interaction between the soaking duration and organic-based materials on the number of plant survivals of ricebean.

Number of Seeds Per Pod

The result shows that seed priming effectively increased the number of seeds per pod of ricebean seeds as shown in Table 4. Six hours and 4 hours of soaking recorded 16 seeds per pod which is the highest among soaking duration. The unsoaked seeds and seeds soaked for 2 and 8 hours had the same results which is 10 seeds per pod. The result shows that seed priming effectively increased the number of seeds per

Table 4

Number of Seeds Per Pod and 200 Seed Weight (g) of Ricebean Seeds Primed with Organic-Based Materials at Different Soaking Durations

TREATMENT	SEEDS PER POD	200 SEED WEIGHT (g)
Soaking duration (S)		
Control (Unsoaked)	10.00[b]	25.20[ab]
2 hours	10.00[b]	25.20[ab]
4 hours	16.00[a]	24.47[b]
6 hours	16.00[a]	25.93[a]
8 hours	10.00[b]	24.00[c]
Organic-based Priming Material (O)		
Water (control)	12.00	24.73[ab]
Garlic Extract	13.00	25.47[a]
Horsetail tea	13.00	24.47[b]
OHN	12.00	25.34[ab]
Wood vinegar	13.00	25.20[ab]
N x S	ns	ns
CV[a]	6.44	12.66
CV[b]	10.03	8.20

pod of ricebean seeds. Priming seeds probably improved not only their vegetative characteristics but also their reproductive characters. Basra et al. (2003) reported that priming enhances the number of grains per pod of different field crops tested. An increased by 19% in the number of seeds per pod was also reported by Aboutalebian and Mohagheghi (2015) in lentil seeds.

The number of seeds per pod was not significantly affected by organic-based materials (Table 4). Regardless of the soaking duration, priming seeds with organic-based materials produced 2-3 more seeds than non-primed seeds and OHN. This means that hydro-priming can result in more seeds per pod. The same finding was reported by Agawane and Parhe (2015) in soybean.

The different interaction combinations of soaking duration and organic-based priming materials on the number of seeds per pod of ricebean were not significant.

Two Hundred Seed Weight

Seed priming showed a significant effect on the 200 seed weight of ricebean (Table 4). Ricebean seeds soaked for 6 hours produced the heaviest among the soaking duration and 8 hours of soaking recorded the lightest seed (24g). It shows that seed priming increased the seed weight at a certain level of priming duration, which was also observed by Mehri (2015). In his study, heavier seed weight was produced by 18 hours priming duration than 24 hours priming duration of soybean seeds.

Organic-based priming materials positively affected the seed weight of the ricebean. Table 4 shows that garlic extracts had the heaviest seed (25.47g) comparable to wood vinegar (25.2g), OHN (25.34g), and water (24.73g). Singh et al. (2015) reported that allicin found in garlic extract at low concentration improves seed performance.

No significant difference among treatment interactions between the soaking duration and organic-based priming materials was observed on the 200 seed weight of the ricebean plant.

Total Seed Yield (kg/5m²)

Seed priming has significantly increased

the total seed yield of ricebean (Figure 1). Seed priming regardless of duration (2,4,6,8 hours) significantly increased the total seed yield as compared to the unprimed seeds that gave the lowest yield of 0.65 kgs/5m². This result conforms to Rao and Singh (1997) and Basra et al. (2003) who stated that seed priming increases grain yield. In relation to the result, Harris et al. (2000) explained that the increase in yield may be because primed seeds emerge faster and more uniformly leading to a wide range of yield-associated benefits. Harris et al. (1999) further informed that primed seeds have higher yields than non-primed crops.

Further, the total seed yield among treatments was affected significantly by the organic-based materials used (Figure 1). The highest total seed yield was recorded in garlic extract (1kg) comparable with seeds primed with horsetail tea (0.93kg). On the other hand, OHN and Wood vinegar yielded 0.85kg and 0.86kg accordingly. Generally, seeds treated with garlic extract and horsetail tea showed high total seed yield signifying its advantage over other priming materials. This result may be correlated to faster seed emergence, a higher number of normal seedlings, and heavier seed weight, which were recorded under these priming materials.

Total seed yield was not significantly affected by the treatment interactions between soaking duration and organic-based priming materials.

Return on Cash Expenses (ROCE)

Positive returns on cash expenses were recorded in the five soaking durations used. Two hours and 6 hours of soaking had the highest percentage in terms of return on cash expenses while the unsoaked (control) has the lowest ROCE (Figure 2).

In terms of organic-based priming materials, garlic extract had the highest percentage of return on cash expenses. It was 0.24% higher than that of OHN (Figure 2).

Conclusion

Priming duration using organic-based materials significantly affected ricebean production. Seeds primed with garlic extract were the earliest to emerge. It has the highest number of normal seedlings and the least number of abnormal seedlings. As the result, garlic extract gave the heaviest seeds and the highest total seed yield. On the other hand, ricebean seeds soaked for

Figure 1

Total Seed Yield (kg/5m²) of Ricebean Primed with Organic-Based Materials at Different Soaking Durations

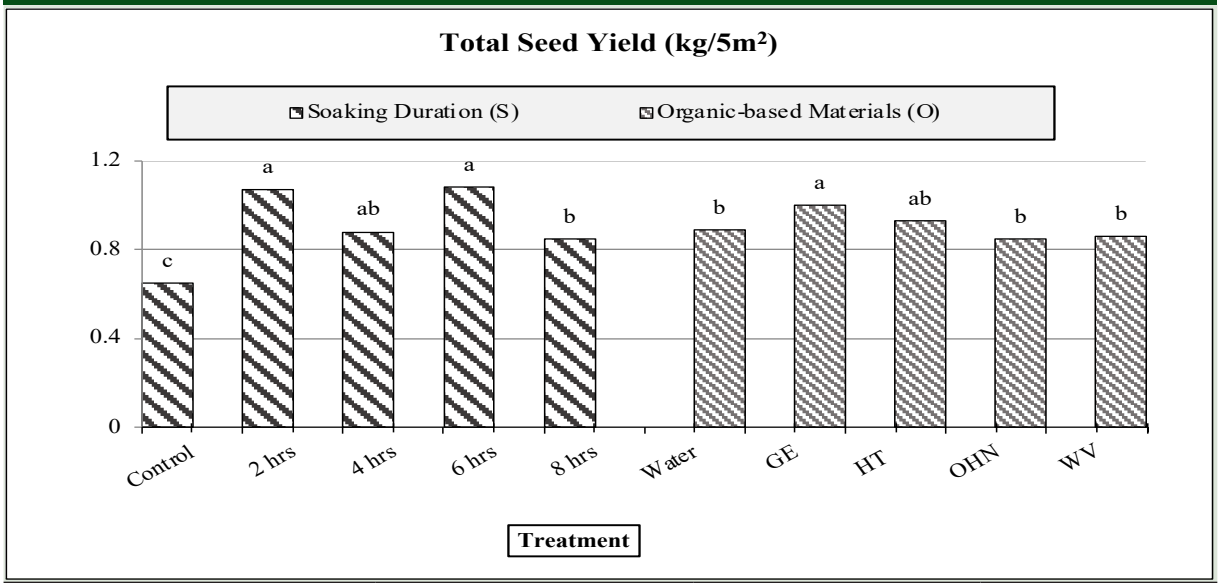
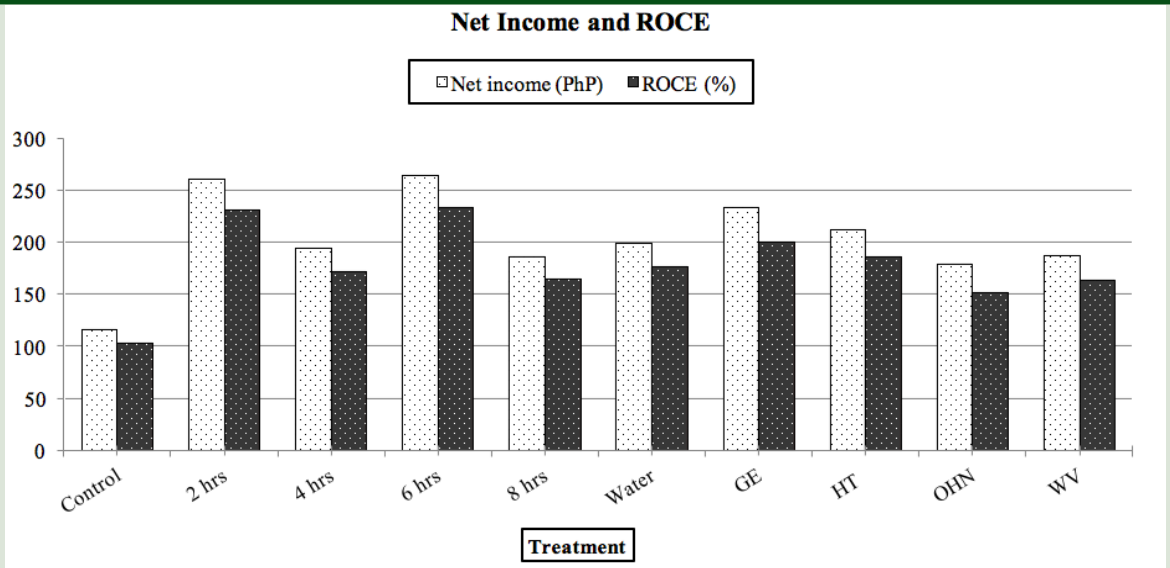


Figure 2

Return on Cash Expense (ROCE) of Ricebean Primed with Organic-Based Materials at Different Soaking Durations



6 hours resulted in early seedling emergence, high percent emergence, a high number of normal seedlings, and a low number of abnormal seedlings. Also, priming for 6 hours gave the greatest number of seeds per pod, heaviest seed, and highest total seed yield. The growth and yield parameter of ricebean plants was not significantly affected by the treatment combinations between priming duration and organic-based priming materials used although the different treatment combinations between soaking duration and organic-based priming material have shown a positive return on cash expense. Soaking ricebean seeds for 6 hours using garlic extract was shown to be the most profitable among the treatment combinations due to high seed yield and profit relative to production cost.

Acknowledgment

We thank the Commission on Higher Education-Cordillera Administrative Region (CHED-CAR) for funding the project. Appreciation is also owed to Benguet State University – La Trinidad for being a counterpart to the project.

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