Seedling Vigor Testing as Affected by Two Methods of Seed Extraction of Tomato (*Lycopersicon esculentum* L.)

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ABSTRACT

This study was conducted to test and compare the seedling vigor performance of dwarf green and dwarf white varieties of tomato as affected by two methods of seed extraction such as fermentation and hydrochloric acid in terms of germination percentage, germination rate, days to seedlings emergence, days to 50% development of true leaves, root length, stem length and shoot length. It was carried using Complete Randomized Design (CRD) in a 2x2 factorial experiment with four treatment combinations with three replications. Dwarf Green variety was affected by seed method extraction with HCl treatment had ideal conditions for seed and seedling quality performance and vigor of tomato such as germination percentage and rate, number of days to seedling emergence, number of days to fifty percent development of true leaves, root, stem and shoot length. Dwarf White performed inferior than Dwarf Green in terms of the parameters investigated.

Keywords - Seed extraction methods, seedling vigor, germination rate

INTRODUCTION

Tomato (*Lycopersicon esculentum L.*) is one of the most widely cultivated vegetable crops not only in the Philippines but also throughout the world. It is usually used as one of the most popular salad vegetables rich in iron and it is an excellent source of vitamin A and C. The tomato fruit is a primary ingredient in the preparation of soup, sauce, catsup, paste, pickles, juice and many other products. It is also an important cash crop for smallholders and medium-scale commercial farmers. Many of the tomato growers face some challenges in the production such as the lack of open-pollinated varieties (non-hybrid), which can tolerate the strict environmental condition, the discontinuous production, and supply of seeds of these varieties that possess a good quality.

For several years, many Filipino farmers still have a problem regarding where to procure good quality tomato seeds to plant in their farms. Although advances in technology and better seed scientists and technologists, government and private seed growers, the problem has been lessened somewhat, for the small scale problems, the increasing cost of seeds and the difficulty in getting seed subsidies from the government has made the procuring of seeds to be a problem for them.

With the application of new innovation and knowledge in seed science and technology, farmers can now procure their own seeds including vegetable fruit seeds like tomato through seed processing. In this process, quality plays great importance. The quality of tomato seeds is affected by seed extraction method. Seed extraction is the process of separating seeds from the fruit and removing the pulp and gelatinous coating surrounding on it which can delay and prevent seed germination.

The seed as planting material plays a very pivotal role in the development of agriculture. One of the major limiting factors in the success and sustainability in crop production is the unavailability or lack of a continuous supply of quality seeds.

Supplying high quality seeds is the basic requirement that contributes greatly to success in any crop production. Because of the importance of tomato to humans and agriculture, large quantities of high quality tomato seeds are required worldwide.

Most of the Maranao farmers in Lanao del Sur do not bother to produce their own seeds. The probable reasons are lack of knowledge, skills, and information on how to produce and maintain seeds of good quality. They rely and are dependent mainly on seeds from agro-industrial supplies produced by commercial seed growers, suppliers and companies like Eastwest, Monsanto, Kaneko, etc. for their planting material. As a result, they hesitate to continue the activity; others even leave crop farming and shift to other crop production ventures due to its being expensive and the unavailability of good quality seeds. Unknown to these people, seed production ventures in the area can be of good help to solve the increasing price of seeds due to high demand and the knowledge and skills needed in seed production is not actually sophisticated.

OBJECTIVES OF THE STUDY

This study was conducted to determine the seedling vigor of two varieties of tomato using two methods of seed extraction based on germination percentage, germination rate, number of days to seedling emergence, number of days to 50% development of true leaves, root, stem and shoot length.

METHODOLOGY

The study on the different seed extraction methods of two varieties of tomato requires evaluation to test its seed vigor.

Seed Extraction Preparations (Treatments)

The study involved the two techniques of seed extraction on two varieties of tomato, namely the Dwarf Green and Dwarf White. The first technique used was fermentation in which the two varieties were extracted. The preparation was started by weighing three kilograms of tomato fruits that were taken from 12 kilograms each of Dwarf Green and Dwarf White tomato variety. The selection of sample fruits was based on the criteria that the fruit should be fully ripe, and uniform in size, and robust. The selected fruits were cut in half through the middle and the seeds were squeezed out and scraped into a small plastic container. The seeds with gelatinous coating were strained to remove the juicy part. The seeds were placed back in the container and fill with water the same as the volume of seeds extracted. The seed extract was fermented for two days at room temperature. Stirring was done three times a day to hasten the separation of seeds from the pulp. Two days after, the seeds were poured into the strainer and rinsed with tap water to remove any remaining flesh from the tomato fruit. Extracted seeds were placed in four layers of clean bond paper and allowed to dry at room temperature for three days and five hours through sun-drying.

The second technique was the used of hydrochloric acid in which another three kilograms of fully ripe, uniformly sized and healthy tomato fruits from each variety were squeezed, scraped and strained. The tomato seeds with gel mass were soaked in water in a glass jar and added with 5% hydrochloric acid. Stirring the seed-gel mass was continuously done for thirty minutes to remove and dissolve the gelatinous coating. The soaked seeds were rinsed and placed in four layers of bond paper and loosen any clumps of seeds. The seeds were properly dried by air-drying at normal room temperature for a period of three days, followed by sun-drying for five hours only (8:00am-11:00am in the morning and 3:00pm-5:00pm in the afternoon. It was recommended to avoid sun-drying in between 12 to 3 pm, since the rays emitted from the sun and the heat may affect the seed viability. During drying, even distribution and a mix of seeds in the drying lot was necessary, and the seeds were also stirred or turned (up-side-down) to avoid clogging. It was observed by some researchers that clogging during seed drying will result in improper drying, fungal infection and poor vigor. After four days of drying, thirty seeds were placed in each prepared seed box of the seed lot for seedling vigor testing. Watering of seed lot was done every morning and afternoon or whenever necessary with 50 ml plain water used for each seedling box of the seed lot.

The concentration of 5% HCl from 250 mL in a volume of HCl was determined using the calculation on ratio and proportion that is:

Variety Used

The two local and open-pollinated varieties (OPV) of tomato such as Dwarf Green and Dwarf White were used in the study. These varieties are being cultivated by local farmers since it is adaptable in the area and has market potential in the local market.

Experimental Design

The study was a 2x2 factorial experiment. The testing for seedling vigor, the experimental design is complete randomized design (CRD) factorial with four treatment combinations and replicated three times.

Variety	Days to harvest (DAT*)	Productive period (mos)	Yield (T/ha)	Color	Fruits Size	Shape	Reaction to Pest and Diseases
Dwarf Green	60	2	25-32	Red orange	Medium large	Oblong	Resistant
Dwarf White	63	2	25-30	Pink orange	Medium	Oblong	Moderately resistant

Table 1. Characteristics of Dwarf Green and Dwarf White Tomato

*DAT= day after transplanting

Treatments

The treatments were as follows: Factor A (Seed Extraction Methods) M_1 = Manual (Fermentation with water) M_2 = Chemical (HCl) Factor B (Varieties) V_1 = Dwarf Green V_2 = Dwarf White Treatment Combinations: $T_1 = M_1V_1$ $T_2 = M_1V_2$ $T_3 = M_2V_1$ $T_4 = M_2V_2$

Randomization

Randomization was done through drawing of lots in all the twelve seed lots using uniform-size pieces of paper just big enough to allow labeling and rolling. The layout was done following the step—by—step procedure in CRD factorial experiment since it was conducted in a controlled environment.

T1R3	T3R1	T2R2	T2R3
T1R1	T4R1	T4R2	T4R3
T3R3	T2R1	T4R2	T3R1

Figure 1. Experimental Layout in CRD

Legend:

Seed lot Dimension	= 34 inches (Length)
	= 33 inches (Width)
Size of Wooden Stick	= 1X2 inches (Thickness)
Bottom Surface	= Plastic Mesh Wire covered with tissue paper

Research Site

This study was conducted at the Plant Science Experimental Area, College of Agriculture, Mindanao State University Main Campus, Marawi City, Lanao del Sur from January to June 2016.

Data Collected

Seedling Vigor Testing

The seedling vigor of tomato varieties was evaluated in this study for each seed box of the seed lot with 30 seedlings but only 20 seedlings were used as data plants and these include the following:

1. Germination Percentage (%). Percent germination was obtained when the length of radicle measured 2mm long. This was done by counting the number of seeds germinated each seed lot divided by the number of seeds sown and multiplied by 100. This was calculated using the following formula:

Germination % = $\frac{\text{Number of seed germinated } x \ 100}{\text{Number of seeds used}}$

- 2. Germination Rate (day). The germination rate of seeds was taken by counting the number of days the seed germinated to at least 50% to the total seeds used each plot.
- **3. Number of Days to Seedling Emergence (day).** This was done by counting the number of days the seedlings developed its plumule and radicle to at least 50% and measured 2cm in length. Each seed lot was counted.
- 4. Number of Days to 50% Development of True Leaves (day). The development of tomato leaves dictates its further growth and

development since these are responsible for the photosynthetic activity of the plant. After sowing, the number of days to 50% development of true leaves was counted from each seed lot.

- **5. Root Length (cm).** The root length was taken at fourteen days of emergency and this was measured from the base of the root up to its tip using a ruler. There were twenty (20) seedlings that were randomly taken as data plants from each seed lot.
- **6. Stem length (cm).** This was taken by measuring the stem of the twenty (20) data plants which were randomly taken seedlings from the seed lot. The stem length was measured from the base up to its tip using a ruler (cm) after fourteen days.
- 7. Shoot Length (cm). The shoot length of seedlings was measured from the base up to the tip of the highest leaf of each (20) randomly taken seedling using a ruler (cm).

Statistical Tools and Analysis

Collected data were analyzed using Statistical software Statistical Tool for Agricultural Research (STAR) by the International Rice Research Institute (IRRI) (STAR v 2.0.1, 2014). One-way Analysis of Variance (ANOVA) was used to determine the differences. Further significant results will be compared using Tukey All Pairwise Tukey Comparison.

RESULTS AND DISCUSSION

Germination Rate and Number of days to Germination

Figure 1 shows the germination rate and a number of days to germination of two varieties of tomato as affected by two methods of seed extraction. For the germination rate, all treatments had greater than 80% germination that is above the recommended germination percentage. Dwarf Green variety using HCl extraction method had the highest percentage germination rate. It was followed by Dwarf White using HCl extraction and comparable to Dwarf Green using the manual extraction method. Dwarf White using manual extraction had the lowest germination rate.

None of the extraction methods was detrimental to percentage germination of seeds and it was greater than 80% for all treatments (Nemati, Nazdar, Azizi, & Arouiee, 2010). Natural fermentation gave lower germination than acid treatment (Silva, Koch, & Moore, 1982). The highest seed quality, as defined by percentage germination and seed recovery observed for acid extraction method and lowest was recorded in the fermentation method.

In terms of the number of days to germination, Dwarf White either extracted using manual or HCl extraction method germinated faster based on the result of the ANOVA. The same trend was observed to Dwarf Green, either extracted using manual or HCl extraction germinated later.

The result of this study correlates to the findings of Singh et al. (1985) where he found out that the seed germination rate is affected by cultivar and methods of seed extraction. Moreover, the advantages of acid treatment are an efficient breakdown of the gelatinous coating and quick cleaning, eradication of bacterial canker, inactivation of tobacco mosaic virus and producing bright looking seed coat (Ritchi, 1971; Silva, Koch, & Moore, 1982; George, 1985, Demit and Samit, 2001).





Number of Days to Seedling Emergence and Days 50% Development of True Leaves

Figure 2 shows the number of days to seedling emergence and number of days to 50% development of true leaves of two varieties of tomato as affected by two methods of seed extraction. The data on the average number of days to seedling emergence of two varieties of tomato as affected by two methods of seed

extraction is presented in Table 4. Regardless of the method of extraction used, Dwarf Green showed earlier seedling emergence than Dwarf White.

In terms of the number of Dwarf Green had less number of days to 50% development of true leaves regardless of the method of seed extraction. The Dwarf White had later days before 50% true leaves develop compare to Dwarf Green based on ANOVA.

Valdes and Gray (1998) reported that the cultivar had a significant effect on some seed quality traits. The effect of temperature of fermentation and duration of fermentation on all traits was significant so that increasing duration and temperature during the fermentation drastically reduces the seed germination and other seed quality traits including seedling emergence height and the number of leaves/seedling.



Figure 2. Number of days to seedling emergence and number of days to 50% development of true leaves of two varieties of tomato as affected by two methods of seed extraction

Root, Stem, and Shoot Length

Figure 3 shows the root, stem and shoot length of two varieties of tomato as affected by two methods of seed extraction. The longest root length was observed from Dwarf Green extracted using the manual method. It was followed by Dwarf White extracted using HCl extraction. Dwarf Green using manual extraction and HCl extraction respectively followed.

According to Yaday et al. (2004) soaking tomato seeds in hydrochloric acid, which is diluted for 200 times, for 3 hours can eliminate tobacco mosaic virus. Desai (2004) pointed out that producing bright looking seed coat are important

features of acid extraction method, but it can be deteriorative on seed quality if the application period and concentration are not appropriate.

For the stem and shoot length, the Dwarf Green was affected by the method of extraction, using HCl extraction method had higher stem length than manual fermentation method. In the case of Dwarf Green, regardless of the method of extraction, it had shorter stem length observed.

This result confirmed to the finding of Miller (1999) that tomato seeds are susceptible to a number of seed-borne diseases. However, he pointed out that chemical soak treatment provides the best protection for tomato seeds. Soaking the tomato seeds in 5% HCl for 5 to 10 hours as treatments can inactivate an internal number of seed-borne diseases.



Figure 3. Root, stem and shoot length of two varieties of tomato as affected by two methods of seed extraction.

CONCLUSIONS

On the basis of the above results, it can be concluded that Hydrochloric acid (HCl) extraction method had created ideal conditions for seed and seedling quality performance and vigor of tomato such as germination percentage and rate, number of days to seedling emergence, number of days to fifty percent development of true leaves, root, stem and shoot length for Dwarf Green tomato variety.

It is further concluded that Dwarf Green tomato variety had better seedling performance in terms of seed viability and seedling vigor than Dwarf White variety when extracted using Hydrochloric acid.

TRANSLATIONAL RESEARCH

It is highly recommended that the used of other varieties to be subjected to different methods of extraction be used. From this, we can determine whether the method of seed extraction is varietal-specific.

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