



Influence of Specific Gravity Grading Using Floatation Technique on seed and Seedling Quality Characteristics in Amaranthus CV. PLR 1

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Study was undergone to evaluate the efficacy of floatation grading on the seeds of Amaranthus Cv. PLR 1 using organic solvents in Department of Seed Science and Technology, TNAU, Coimbatore during March 2023. The experiment was laid out in Completely Randomized Design with four

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replications in each five treatments. We considered four organic solvents viz., Acetone, Methanol, Dichloromethane and Petroleum Ether along with water and observed the seed recovery percentage, germination percentage, root length, shoot length and vigour index of all the treatments. The results showed that sinker fraction regardless of organic solvent and water performed better than floater fraction. The grading ability of Dichloromethane was greater among the assessed organic solvents. The sinkers seed recovery percent of dichloromethane (93%) was higher than sinkers recovered by water (63%). Also, the sinkers of dichloromethane recorded higher germination percent(77%) and vigour index(304) than the sinkers of water whose germination percent is 55% and vigour index is 272. This specific gravity grading using dichloromethane can be adopted to segregate well filled and ill filled seeds in a seed lot of Amaranthus. This method of floatation grading is a good substitute for sieve grading which requires skilled labour and availability of specific size sieve to properly segregate seeds. So, this can be used as an alternate method for grading Amaranthus seeds.

Keywords: *Amaranthus*; *dichloromethane*; *germination*; *grading*; *sinkers*; *specific gravity*.

1. INTRODUCTION

Amaranthus which belongs to the family Amaranthaceae is a genus of 50 species of herbaceous annuals found in tropical and subtropical regions worldwide. Generally, members of this genus are weeds possess rapid vegetative growth and produce plentiful small seeds with good-quality protein. *Amaranthus* is an underutilized inexpensive leafy vegetable which are rich sources of vitamins, proteins, dietary minerals and bioactive compounds with potential health benefits. Dietary fiber is abundant in leafy foods. According to the Indian Council of Medical Research, an adult man needs 300 grams of vegetables each day which includes 100 grams of roots and tubers, 125 grams of leafy vegetables and 75 grams of other vegetables. Vegetables are necessary to human health as it is the repository of nutrition. Improved seed production is the first step to boost availability of organic vegetables. As a labor-intensive crop, it requires considerable skill and attention. Improved varieties and better management practices help reduce production costs and increase yield through seed production technology. A crucial ingredient in the cultivation of vegetables is good seed. Homogenization of seed lot is the ultimate goal of seed processing. Physical characteristics are the main focus of homogenization since they are the most readily adaptable. *Amaranthus* is propagated through seeds which are tiny. Due to its indeterminate growth habit, seeds are continuously produced on branches, which generate immature, smaller seeds from late formed inflorescence. Such immature or empty seeds must be eliminated from good ones in seed processing. The seeds of the *Amaranthus* pass through the smallest sieve size of BSS 22 x 22 (British Standard Sieve), so

size grading could be a tedious one. Hence, attempts were made in *Amaranthus* Cv. PLR 1 to grade the seeds by density using organic solvents and were evaluated for the seed and seedling quality characters.

Density grading is a technique where seeds are separated based on their weight using solvents or equipments like specific gravity separator. According to Jacqueline and Ramaswamy [1], high-density brinjal seeds produced higher germination and vigor indices. The difference in weight of seed is due to the storage reserves and its mobilisation occurring during germination and early stages of seed development determine the physiological parameters of seedling [2].

In the context of *Jatropha curcas* seeds, large seeds displayed higher germination rates and biomass production compared to medium and small seeds [3]. Ramandane and Ponnuswamy [4] opined that high and medium density seeds obtained after grading with specific gravity separator had good germination and vigour than low density seeds.

2. MATERIALS AND METHODS

2.1 Materials

The experiment was conducted at Department of Seed Science and Technology, Agricultural College and Research Institute, Coimbatore. Fresh seeds of *Amaranthus polygonoides* Cv. PLR 1 were obtained from Vegetables Research Station, TNAU, Palur formed the base material for the study. The organic solvents used in this experiment were procured from Department of Seed Science and Technology

and Department of Environmental Sciences, TNAU, Coimbatore.

2.2 Methods

The seeds were subjected to floatation grading using organic solvents and water by dropping 1 gram of seeds in 50 ml of solvents. This method works based on the principle of specific gravity of liquids to separate seeds based on its density. The organic solvents used were acetone, methanol, dichloromethane and petroleum ether. Seeds were stirred well and allowed to stand for one to two min. The well filled seeds get separated as sinkers (sink in the column of solution/water) and the ill filled tend to separate as floaters (seeds that float in the column of solution/water). Both the grades were then collected separately and dried to its original moisture content. The respective grades were evaluated for recovery percentage by using the following formula

$$\text{Sinkers}(\%) = \frac{\text{Weight of sinkers}}{\text{Total weight of seeds}} \times 100$$

$$\text{Floaters}(\%) = \frac{\text{Number of floaters}}{\text{Total weight of seeds}} \times 100$$

Then, each of the fractions were sown in quadruplicate in a paper medium using inclined plate method and allowed to germinate in a germination room maintained at 25 ± 2 °C and 85 per cent relative humidity and illuminated with fluorescent light. At 8th day, seed and seedling quality characters viz., germination (%) [5], root and shoot length [6], drymatter production (mg 10 seedlings⁻¹), vigour index [7] were recorded. Vigour Index was computed using the following formula;

$$\text{Vigour Index} = \text{Germination}(\%) \times \text{Dry matter production (mg/10 seedlings)}$$

Specific gravity of the considered organic solvent was estimated using the below mentioned formula [8].

$$\text{Specific Gravity} = \frac{\text{weight of sample in bottle} - \text{weight of empty bottle}}{\text{weight of water in bottle} - \text{weight of empty bottle}}$$

The experiment was conducted with Completely Randomized Design (CRD). The recorded observations were statistically analysed by adopting the procedure described by Gomez and Gomez [9] for evaluating the treatment significance ($P = 0.05$). Percentage values were transformed to arc-sine values prior to statistical analysis.

3. RESULTS AND DISCUSSION

The seed weight evaluated for both fractions after dropping 1 gram of seed in 50 ml of solvent (Fig. 1) have shown that sinker seed were heavier in weight than floater seeds in both water and dichloromethane (Data was depicted in Table 1). This is in accordance with Nascimento [10] where pea 1000 seed weight and purity decreased with decreasing density.

The recovery of seeds was higher in dichloromethane followed by water. Dichloromethane was able to recover 93% of seeds as sinkers with 77 % germination. This could be obtained by rejecting 7% of seeds which floated on the surface with 24% germination, whereas water able to recover 63% of seeds as sinkers with 55% germination by rejecting 37% of seeds which floated on the surface with 53% germination. This correlates with the findings of Sivakumar et al., [11] where *Casuarina equisetifolia* seed lots were upgraded using petroleum ether exhibited efficient separation of *Casuarina* seeds as witnessed by higher germination in sinker fraction (90%) than in floater fraction (4%). This implies that the germination of seeds increased with increase in weight of the seeds recovered in each fraction (Table 1).

In rest of the other solvents seeds just got settled down. This aids us to look into the solvents' specific gravity using specific gravity bottle (Fig. 2). The specific gravity of universal solvent i.e; water is 1 and dichloromethane is 1.27. The considered remaining organic solvents viz., acetone, methanol and petroleum ether's specific gravity are 0.73, 0.77 and 0.64 respectively (Table 2).

From this we can conclude that Amaranthus seeds got separated as sinkers and floaters in solvents whose specific gravity is greater than or equal to 1. In dichloromethane, ill filled seeds got separated as floaters precisely when compared with water where both well filled and ill filled found to float on the surface for first few minutes due to their lighter weight but thereafter the seed sink to the bottom based on the adsorption principle because of which the recovery of seeds were lower in water than dichloromethane. Several authors have recorded their experience on relationship between specific gravity of seed and seedling vigour [12,13].

Though the seeds retained as sinkers by water recorded higher root length (cm), shoot length (cm) and DMP (mg 10 seedlings⁻¹) as 5.47, 3.81 and 4.95 respectively than dichloromethane sinkers. The vigour index of dichloromethane sinkers were higher than sinkers of water which endorse the mixture of ill filled and well filled seeds in sinker fraction of water which lead to lower germination percentage. Highly significant differences were observed between sinkers and floaters of water and dichloromethane for the evaluated parameters viz., seed recovery, sinkers and floaters seed weight, germination and vigour index as depicted in Table 3.

Similar results were found in upgrading petunia seeds by Natarajan and Srimathi [14] where

sinkers found in acetone performed better than water. Results are in conformity with Selvaraju and Selvaraj [15] where marigold seed lot was said to be improved by adopting density grading by organic solvent.

The result of the present study indicated that among the four organic solvents used for grading the amaranthus seeds, dichloromethane stood efficient in segregating the ill filled seeds and well filled seeds as sinkers and floaters. Apart from dichloromethane solvent, sinkers and floaters fraction were found in water also. The seeds got settled down in remaining organic solvents completely which indicate that the solvents' specific gravity is not sufficient to separate as fractions.



Fig. 1. Floatation grading in Amaranthus Cv. PLR 1 seeds using organic solvents

Source-picture taken during my research work, 2023 in TNAU

Table 1. Effect of seed grading on seed weight, seed recovery percentage of Amaranthus Cv. PLR 1

Treatment	Seed weight (g)		Seed recovery (%)	
	Sinkers	Floaters	Sinkers	Floaters
T ₁	0.63	0.37	63(52.54)	37(37.47)
T ₂	1	0	100(89.71)	0(2.866)
T ₃	1	0	100(89.71)	0(2.866)
T ₄	0.93	0.07	93(74.66)	7(15.34)
T ₅	1	0	100(89.71)	0(2.866)
Mean	0.91	0.09	91.2	8.8
SEd	0.019	0.003	1.352	0.180
CD(P=0.05)	0.041	0.007	2.881	0.383

T₁-Water, T₂-Acetone, T₃- methanol, T₄-dichloromethane, T₅- Petroleum Ether

Numbers in parentheses indicate Arc-sine values

Table 2. Specific gravity of solvents used

Solvents	Specific gravity
Water	1.00
Acetone	0.73
Methanol	0.77
Dichloromethane	1.27
Petroleum Ether	0.64

Table 3. Effect of sinker and floater seeds on physiological parameters of Amaranthus Cv. PLR 1

Treatment	Germination%		Root length (cm)		Shoot length (cm)		Dry matter production (mg 10 seedlings ⁻¹)		Vigour index	
	Sinkers	Floaters	Sinkers	Floaters	Sinkers	Floaters	Sinkers	Floaters	Sinkers	Floaters
T ₁	55(47.87)	53(46.72)	5.47	3.78	3.81	2.25	4.95	4.37	272	231
T ₂	70(56.79)	0(2.866)	4.00	0	2.44	0	2.52	0	176	0
T ₃	62(51.94)	0(2.866)	4.15	0	3.22	0	3.08	0	191	0
T ₄	77(61.34)	24(29.33)	4.82	3.29	2.86	1.98	3.95	0.57	304	14
T ₅	68(55.55)	0(2.866)	3.31	0	3.07	0	2.9	0	197	0
Mean	66.4	15.4	4.35	1.42	3.08	0.846	3.48	0.988	228	49
SEd	0.992	0.403	0.097	0.050	0.071	0.031	0.082	0.040	5.446	2.135
CD(P=0.05)	2.114	0.858	0.207	0.106	0.151	0.067	0.176	0.085	11.601	4.551

*T₁-Water, T₂-Acetone, T₃- methanol, T₄-dichloromethane, T₅- Petroleum Ether
Numbers in parentheses indicate arc-sine values*



Fig. 2. Specific gravity bottle

Source- picture taken during my research work, 2023 in TNAU

4. CONCLUSION

Hence considering both the recovery and quality of seed, grading using dichloromethane could be adopted. The principle behind the separation might be the relationship between the seed density and the buoyancy it exhibit. Thus the study expressed that grading of Amaranthus seed with dichloromethane established proper stratification of well filled and ill filled seeds.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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