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Glucose content, viability and vigor of four cucumber seed lots

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Abstract This study assessed the viability, vigor, and soluble glucose content of four cucumber seed lots. The results revealed significant variations among seed lots, with Vanesa 2, Vanesa 3, and Vanesa 4 exhibiting higher vigor index, germination speed, and germination percentage compared to Venus 1. While a strong positive correlation was observed between glucose content and vigor, it was not statistically significant, indicating the potential involvement of other physiological factors. These findings suggested that glucose may serve as a biochemical indicator of seed quality.

Keywords: Alpha - amylase, Enzymatic activity, Cucurbitaceae, Germination, Seed quality

Introduction

Cucumber (*Cucumis sativus* L.) is an annual crop belonging to the Cucurbitaceae family, extensively cultivated for its edible fruit (Mallick, 2022). It is believed to have originated in Asia, where it has been cultivated as a food source for over 3000 years (Chomicki *et al.,* 2020). Cucumber is a rich source of essential nutrients, including minerals, thiamine, niacin, and vitamin C (Uthpala *et al.,* 2020). Cucumber is considered beneficial for individuals with jaundice and other medical conditions (Hossain *et al.,* 2022), and it plays a role in alleviating constipation (Chakraborty dan Rayalu, 2021). Moreover, as a low-calorie food, cucumber is associated with weight management and the regulation of blood sugar levels (Bartimaeus *et al.,* 2016). The cucumber seeds contain oils that are thought to support brain development and maintain overall bodily health (Gupta *et al.,* 2021). The cucumber seeds are also commonly incorporated into Ayurvedic medicinal preparations (Wahid *et al.,* 2021).

High seed quality is essential for ensuring rapid and uniform seedling emergence, which is a critical factor for succesful stand establishment and consistent plant growth and development (Demir and Mavi, 2008). Seed

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deterioration refers to the decline in seed viability and vigor due to exposure to adverse environmental conditions (Kapoor *et al.,* 2010). The rate of seed deterioration is significantly influenced by environmental factors, including temperature, relative humidity, and seed moisture content, as well as biological factors (Jyoti and Malik, 2013). Prolonged storage periods contribute to an increase in seed moisture content which is the most influential factor affecting seed longevity (Ghassemi-Golezani *et al.,* 2010). Seed stored with high moisture content exhibit increased respiration rates which contributes to a reduction in seed vigor and viability (Jyoti and Malik, 2013).

Germination is characterized by elevated enzymatic activity that facilitates the breakdown of stored reserves such as amylases, phytases, proteases, and lipases (Guzmán-Ortiz *et al.,* 2019). Among the various amylases, alpha – amylase plays a crucial role during germination, as it is specifically involved in the hydrolysis of starch, leading to the subsequent release of soluble sugars (Bewley *et al.,* 2013). Alpha – amylase activity increases during seed germination and associate with starch degradation (Padilha *et al.,* 2024). The starch degradation by alpha – amylase enhances seedling development and performance in the field (Heloisa *et al.,* 2015; Wang *et al.,* 2016; Mohamed Zeid, *et al.* 2019). High seed vigor exhibits increased alpha – amylase activity that was associated with high soluble glucose during seed germination (Padilha *et al.,* 2024). This research aimed to assess soluble glucose content, viability and vigor of four cucumber seed lots.

Materials and methods

The research was conducted in Seed Testing and Storage Laboratory and Post-harvest Laboratory, Department of Agronomy and Horticulture, Faculty of Agriculture, Bogor Agricultural University (IPB University), Bogor - Indonesia. The research was conducted using a single factor – Completely Randomized Design (CRD) with three replications. The factor was the seed lots, which comprised four different lots i.e., Venus 1, Venesa 2, Venesa 3, and Venesa 4.

Germination test

Cucumber seed lots were collected from seed suppliers in the seed commercial market surrounding Bogor Agricultural University (IPB University) Campus, Dramaga, Bogor – Indonesia. 50 seeds were germinated in between – paper method with moistened filter paper and incubated in an eco-germinator (25 – 28 ^{o}C) for 8 days. Evaluations were conducted at 4 and 8 days after sowing and the results were expressed as the percentage of normal seedlings (ISTA, 2022).

Vigor index (%) was calculated as the total number of normal seedlings at 4 days after planting divided by the total number of germinated seeds. Germination (%) was calculated as the total number of normal seedlings at 4 and 8 days after planting divided by the total number of germinated seeds. Speed of germination (% etmal^{-1}) was assessed daily from the start of germination until the final count was obtained. It was calculated using the following formula:

$$\text{SG (\%NS/etmal)} = \sum_0^t \left(\frac{\%NS}{etmal}\right)$$

SG was speed of germination (%NS etmal^{-1}), %NS was percentage of normal germination at each observation time, etmal was observation time every 24 hours, and t was observation time (Gundala *et al.,* 2023).

Glucose content assay

A total of 10.6 g of 3,5-dinitrosalicylic acid and 19.8 g of NaOH were dissolved in 1416 ml of water. Subsequently, 306 g of Na-K tartrate was added to the solution, followed by the addition of 7.6 ml of phenol that had been dissolved at 50°C, along with Na-metabisulfite. All components were mixed thoroughly to ensure the homogeneity of the solution. For testing, 3 ml of the DNS reagent was titrated with 0.1 N HCl using phenolphthalein as an indicator. The amount of HCl required should be between 5-6 ml; if the volume of HCl used is less than this range, NaOH should be added at a rate of 2 g for every 0.1 ml deficiency of 0.1 N HCl.

The standard glucose solution was prepared by gradually diluting (10, 20, 30, 40, and 50 µl) in phosphate buffer to a total volume of 1 ml. Subsequently, 3 ml of DNS reagent was added to each solution. Each solution was then vortexed and heated in boiling water for 5 minutes. After cooling, each solution was diluted twice and vortexed again to ensure homogeneity. The absorbance was measured using a spectrophotometer at a wavelength of 550 nm, and the linear equation was obtained as the standard curve. The obtained standard curve is expressed as $Y = 0.0039x - 0.0868$, with a coefficient of determination (R^2) = 0.999. This curve was derived from glucose concentrations of 25, 75, 100, and 125 ppm, corresponding to absorbance values of 0.013, 0.198, 0.295, and 0.401, respectively.

The procedure for glucose determination began with the seeds being moistened for 24 hours. One gram of seeds was weighed for each replicate, resulting in a total of eight samples (4 lots x 2 replicates). The seeds were then combined with 4 ml of extraction buffer solution (phosphate buffer) and ground using a mortar. The resulting mixture was transferred to 2 ml microtubes and

centrifuged at 11,200 rpm for 5 minutes to separate the supernatant. The supernatant from each sample was then divided into two microtubes (supernatants 1 and 2), yielding a total of 16 microtubes. The supernatants were centrifuged again for 5 minutes to obtain a clear supernatant.

Sixteen test tubes were prepared, with the first eight tubes receiving the first supernatant from each sample, followed by the addition of 2 ml of 0.1% starch solution. The second supernatant from each sample was transferred to the remaining eight tubes, to which 2 ml of distilled water was added. The reaction mixtures were incubated for one hour with periodic vortexing. Subsequently, the 16 reaction tubes were incubated in boiling water for 20 minutes to accelerate the reaction, until the samples became clear; if clarity was not achieved, centrifugation was performed. Three ml of DNS reagent was added to each of the new 16 test tubes, followed by 1 ml of the sample. The samples were then placed in boiling water for 5 minutes, resulting in a color change to reddish-orange, and were cooled under running water. Finally, the absorbance of the test samples was measured at a wavelength of 550 nm using a spectrophotometer.

The absorbance values obtained were used to calculate glucose concentrations by substituting the absorbance values as X in the linear regression equation derived from the standard curve. The formula was $X = ((((Y + 0.0868)/0.0039)*fp)/bs)$. X was glucose concentration (ppm g^{-1}), Y was absorbance value of the sample, fp was dilution factor (12), and bs was sample weight (mg).

Results

The vigor index, speed of germination, germination percentage, and glucose content of four cucumber seed lots are presented in Table 1. The result showed that there was not significant differed in vigor index, speed of germination, and germination percentage among lots Vanesa 2, Vanesa 3, and Vanesa 4. The vigor index ranged from 63.3 to 76.0%, speed of germination was about 24.22 to 28.42 % $etmal^{-1}$, and germination percentage ranged from 79.3 – 90.7%. In contrast, lot Venus 1 was significantly lower vigor index (18,7%), speed of germination (11,35% $etmal^{-1}$), and germination percentage (58,7%).

Table 1. Vigor index, speed of germination, germination percentage, and glucose content of four cucumber seed lots

Seed lots	Viability and vigor			Soluble glucose content (ppm g^{-1})
	Vigor index	Speed of germination	Germination percentage	
Venus1	18.7[b1/]	11.35[b]	58.7[b]	1.521[b]
Vanesa 2	76.0[a]	28.42[a]	90.7[a]	2.164[ab]
Vanesa 3	63.3[a]	26.46[a]	86.7[a]	2.739[a]
Vanesa 4	64.7[a]	24.22[a]	79.3[a]	2.235[ab]

1/: Numbers followed by same letter in the same column did not significant based on duncan's test $\alpha = 0.05$.

Table 2. Correlation between soluble glucose content and vigor index, speed of germination, and germination percentage

	Vigor index	Speed of germination	Germination percentage
Soluble glucose content	0.76[ns1/]	0.83[ns]	0.82[ns]

1/: ns = Not significant

In terms of glucose content, Lot Vanesa 3 had highest concentration (2.739 ppm g^{-1}), although this didn't different to Vanesa 2 (2.164 ppm g^{-1}) and Vanesa 4 (2.235 ppm g^{-1}). While Venus 1 had lowest glucose content (1.521 ppm g^{-1}) that significant different to others. It suggested a correlation between lower glucose levels and reduced seed viability and vigor. Result showed a positive correlation between soluble glucose content and vigor index (0.76), speed of germination (0.83), and germination percentage (0.82) (Table 2), but the correlation among characters was not significant.

Discussion

Result showed the highlight key metrics of vigor index, speed of germination, germination percentage, and glucose content—across four cucumber seed lots. The findings indicated that lots Vanesa 2, Vanesa 3, and Vanesa 4 exhibit similar levels of vigor, with no significant differences among them. Their vigor index values ranged from 63.3% to 76.0%, demonstrating adequate potential for seed performance. Additionally, the speed of germination for these lots was relatively high, varying from 24.22 to 28.42% etmal^{-1}, and their germination percentages were impressive, ranging from 79.3% to 90.7%.

Seedling establishment in the field was highly competitive, influenced by various factors, including the seed vigor (Filgueiras, 1981). These parameters play a critical role in determining which seedlings successfully survive to maturity (Basu and Groot, 2023).

In contrast, lot Venus 1 displayed significantly lower performance metrics, with a vigor index of just 18.7%, a speed of germination of 11.35% etmal^{-1}, and a germination percentage of 58.7%. These results suggested that lot Venus 1 may possed inferior seed viability and vigor as compared to the other lots, which could hinder successful cultivation. The marked differences between Venus 1 and the other lots emphasized the importance of selecting high-quality seeds to ensure optimal germination establishment and overall crop performance. A reduction in seed vigor is demonstrated to adversely affect the uniformity, growth, development, and yield of crops (Reed *et al.,* 2022).

Correlation describes how well the relationships between variables can be defined using linear functions (Rebekić *et al.,* 2015). The correlation between soluble glucose content and vigor index, speed of germination, and germination percentage ranged from 0.76 – 0.82. This correlation ranged was categorized a strong correlation (Akoglu, 2018). While these correlations were strong but the correlation was not significant. It indicates that although higher soluble glucose content tends to be associated with increased seed vigor, faster germination, and higher germination percentage, the relationship is not definitive and could be influenced by other factors.

This study indicated that seed lots with higher viability and vigor also had greater soluble glucose content. It suggested that glucose as a potential indicator of seed quality. Although the correlation was strong, it was not statistically significant, implying other factors may influence seed quality. These findings offer a new perspective on using biochemical markers like glucose in seed testing, highlighting the need for further research to develop rapid and reliable tools for assessing seed vigor and viability.

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Conflicts of interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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