

Morpho-agronomic trait responses of maize growing in various substrates to waterlogging at the flowering stage

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Abstract: Waterlogging and types of plant growth substrates are common abiotic stresses that decrease maize (*Zea mays* L) yield and biomass production. This study aims to identify the growth and development of maize crops in two different growing mediums, assess the impact of waterlogging during the flowering stage on yield, and analyse the interactions between morpho-agronomic traits at harvest. The experiment design of this study was factorial with a randomised complete block design (RCBD) with two factors. The first factor was various plant growth substrates, and the second was the duration of waterlogging (waterlogging for seven hours during the flowering time and control). Seeds were sown in polybags containing potting mixes or a mixture of potting mix: latosol soil (1:1). Plant height was measured during the vegetative stage, and morpho-agronomic traits of stem diameter, ear diameter, fresh root weight, ear length, and yield were recorded at harvest. The potting substrate did not affect plant height. On the other hand, waterlogging for seven hours at the flowering stage significantly reduced yield by 25% compared to control. The correlation between morpho-agronomic traits during harvest showed that yield positively and significantly correlates with ear length. The positive correlation between ear length and yield was useful to indicate the maize yield. Waterlogging for seven hours during the flowering stage significantly reduced maize yield by 25%, while the potting substrate did not affect plant height.

Keywords: *Zea mays*, growing substrate, abiotic stress, maize yield.

Introduction

Agriculture faces uncertainty due to extreme weather events that can result in yield loss (Liu et al., 2023). Waterlogging is a common environmental challenge that decreases maize (*Zea mays* L) yield and biomass production (Mustroph, 2018). The frequency and duration of waterlogging are expected to increase due to global warming (Mustroph, 2018). Waterlogging reduces maize yield by 18% (Kaur et al., 2021), accounting for 25-30% of grain yield losses in South and Southeast Asia (Kaur et al., 2021). Maize is a sensitive crop to waterlogging because when the water capacity in the soil is more than 80%, plant growth and development are reduced (Tian et al., 2020).

The maize crop requires a high amount of water during sowing, emerging, flowering, silking, tasselling, jointing, maturity, and harvesting (Wang et al., 2023). However, maize growth and yield can be significantly reduced when the crop is exposed to excess water (i.e., waterlogging) during the emerging, jointing, and flowering stages. The longer the waterlogging period, the more yield reduction in maize (Huang et al., 2022; Hu et al., 2023; Rean et al., 2023). Such adverse effects of waterlogging were also observed in legumes (Wiraguna et al., 2017; 2020; 2021). Waterlogging results in the reduction of chlorophylls, carbohydrates (Dash et al., 2022), and nitrogen content in several organs throughout the plant development (Otie et al., 2019) and the waterlogging periods (Tian et al., 2020). Moreover, waterlogging can disrupt the balance of carbon-nitrogen metabolism of plant hormones, hasten leaf senescence, and

considerably decrease photosynthetic capability and maize grain yield (Ren et al., 2018; Tian et al., 2019).

The length of waterlogging during the flowering stage is crucial in determining crop yield (Tian et al., 2021; Meta et al., 2022). Studies have demonstrated that extended periods of waterlogging can cause significant yield reductions across various crops (Anuradha et al., 2013; Tian et al., 2021; Meta et al., 2022). Ma et al. (2022) observed increased grain yield loss in wheat from 3.7% to 28.8% as the waterlogging period extended from 3 to 9 days. Similarly, Tian et al. (2021) found consistently declined grain yields in maize, rice, and wheat with extended waterlogging periods, regardless of the cultivation method. In tobacco, 24 hours of waterlogging negatively affected physiological characteristics and reduced yield and quality (Anuradha et al., 2013). However, the effects of brief waterlogging during the flowering period on the maize yield have not been studied.

Waterlogging in the potting mix (growth substrate) can significantly impact plant growth and development (Khabaz-Saberi et al., 2006; Alluri, 2024). Despite efforts to regulate irrigation, the potting mix in sub-irrigated containers often becomes waterlogged (Alluri, 2024). This waterlogged condition can change redox potentials, increase nutrient toxicities, and reduce plant yield grown in the potting mix (Setter et al., 2009). Thus, understanding the effects of waterlogging in different potting mixes is crucial for ensuring optimal plant growth and development.

Table 1. Multiple comparisons (Tukey HSD) of morpho-agronomic traits (maize diameter, stem diameter, maize length, root weight and yield) at harvest. The morpho-agronomic traits of stem diameter (mm), maize diameter (mm), root weight (g/plant), maize length (mm), and yield (g/plant) were tested using one-way ANOVA based on waterlogging treatment.

Treatment	Stem diameter		Maize diameter		Root weight		Maize length		Yield	
Waterlogging	21.5	± 1.0	27.0	± 2.9	1.2	± 0.1	107.1	± 18.1	388.5	± 27.8 ^b
Control	23.1	± 0.7	24.6	± 2.3	1.0	± 0.1	133.2	± 15.3	521.3	± 50.7 ^a
P value	Ns		Ns		Ns		Ns		<0.05	

Note: Values within a column followed by different letters indicate a significant difference with the Tukey HSD at $P < 0.05$. Data are means followed by the error of three replicates. The yield was identified as a total above-ground fresh biomass, and the root weight was identified as a root fresh biomass. The maize diameter and maize length were measured with the husk still attached. Differences between waterlogged treatments were significant for only yield. The maize is grown on a potting mix or a mixture of soil and potting mix, and is either exposed to waterlogging during flowering or not.

Table 2. Pearson Correlation (r) between morpho-agronomic traits from maize at harvest– stem diameter (mm), maize length (mm), maize diameter (mm), root weight (g/plant) and yield (g/plant).

Morpho-agronomic traits	Stem Diameter (mm)	Maize Length (mm)	Maize Diameter (mm)	Root Weight (g/plant)	Yield (g/plant)
Stem Diameter (mm)	1				
Maize Length (mm)	0.55	1			
Maize Diameter (mm)	0.09	0.45	1		
Root Weight (g/plant)	-0.55	0.08	-0.25	1	
Yield (g/plant)	0.45	0.68*	0.02	-0.09	1

Note: The morpho-agronomic traits were collected at harvest from the total sample ($n=3$). The yield was identified as a total above-ground fresh biomass, and root weight as a fresh root weight. The yield was identified as a total above-ground fresh biomass, and the root weight was identified as a root fresh biomass. The maize diameter and maize length were measured with the husk still attached.

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

This study aims to determine the effects of (1) growing substrates on plant growth at the vegetative stage, (2) waterlogging during the flowering stage on maize yield, and (3) the correlation between maize yield and the morpho-agronomic traits.

Results

Vegetative responses on two different maize-growing substrates

The maize growing in two growing substrates did not differ in their vegetative growth. The evaluated plant height of maize growing in the potting mix medium and in the mixture of potting mix and latosol soil (1:1) showed sigmoid growth patterns (Figure 1). The height of maize growing in the potting mix medium at the R2 growth stage (12 weeks after sowing) reached 179 mm and in the mixture of potting mix and latosol soil (1:1) medium reached 187 mm (Figure 1).

Impact of waterlogging during flowering on various growing substrates

Yield (Above-ground fresh biomass)

The results of two-way ANOVA indicated there are no significant interaction effects between growing substrates and waterlogging treatments on yield (above-ground fresh biomass). Moreover, the yield did not differ between the plants growing in a potting mix and in a mixture of potting mix and latosol soil (1:1). On the other hand, there was significant yield reduction associated with seven hours of waterlogging treatment during the flowering stage (R5 stage) than the control. The yield of maize plants exposed to waterlogging for seven hours during the flowering stage (R5) was 25% lower than that of the control one (Table 1).

Multiple comparisons of morphological traits at harvest

Analysis of two-way ANOVA in all morpho-agronomic traits demonstrated that the interaction between maize growth substrate and waterlogging treatment; and the main effect of soil substrates were not significant. However, there were significant differences in waterlogging treatments for yield ($P < 0.05$) (Table 1). Waterlogging did not have a significant effect on stem diameter, maize diameter, root weight (fresh root biomass), and maize length (Table 1).

Responses among morpho-agronomic traits at harvest

The PCA (Principal Component Analysis) explained 69.6% of the variation of maize morpho-agronomic traits at harvest (Figure 2). The first principal component (PC1), which explained 42.3% of the variance, showed positive loadings for stem diameter, maize length, and yield, with root weight displaying a negative loading (Figure 2). The second principal component (PC2), accounting for 27.3% of the variance, is primarily associated with positive loadings for maize diameter and root weight and negative loadings for yield and stem diameter (Figure 2; Table 3). The Pearson Correlation (r) identified that the yield of maize was significantly positively correlated with maize length, while yield was not significantly correlated with other morpho-agronomic traits (Table 2). The significant positive correlation between yield and maize length was 0.68 (Table 2). The person correlation did not identify a significant correlation for other traits (Table 2; Fig 3).

Discussion

The plant growth substrate does not influence maize growth and development, but waterlogging at flowering time significantly reduces yield (Figures 1 and Table 1). The finding is similar to a previous experiment in cotton (*Gossypium herbaceum* L.), where cotton yield dropped by 26% relative to control after waterlogging for ten days during flowering time (Zhang et al., 2015). The correlation between morpho-agronomic traits during harvest showed that yield positively and significantly correlates with maize length (Table 2). Moreover, the result of PCA demonstrated that yield is positively influenced by maize diameter, maize length, and stem diameter at harvest but negatively influenced by root weight (Figure 2; Table 3). The findings of Pearson correlation and PCA are similar to previous studies by Tian et al. (2020), where waterlogging reduces yield. The growth of maize grown under 100% potting mix did not differ significantly from maize grown in a mixture of a potting mix and soil (1:1) (Figure 1). This growth similarity is probably caused by comparable nutrient content in the soil and potting mix, similar to that shown in spinach and tomatoes (Adediran et al., 2003). This finding suggests that compost in the potting mix can substitute plant growth medium of maize crops from soil to support gardening for families with small yards in big cities (Schröder et al., 2021).

Table 3. Loading contributions in Principal Component Analysis (PCA) for five morphological traits in maize.

Morphological traits	PC1	PC2	PC3	PC4	PC5
Stem Diameter	0.816	-0.326	-0.233	0.411	0.023
Maize Length	0.793	0.558	0.121	-0.0730	-0.199
Yield	0.776	-0.532	0.580	-0.175	0.167
Maize Diameter	0.200	0.711	-0.658	-0.058	0.138
Root Weight	-0.417	0.662	0.536	0.313	0.043

Stress caused by waterlogging is recorded to have a significantly negative impact on maize yield (Huang et al., 2022; Wang et al., 2023). The findings in this experiment, where waterlogging reduced yield by 25% compared to the control (Table 1), align with previous studies by Huang et al. (2022) and Wang et al. (2023). The reduction in yield due to waterlogging can be caused by low oxygen concentrations in the roots, a reduction in nutritional intake, and an increase in free radical concentration in plants (Yang et al., 2024). Moreover, low nutritional intake can lead to pigment degradation and reduced photosynthetic activity (Tian et al., 2019). In this study, the main impact of waterlogging was observed as a reduction in yield caused by a decrease in maize length (Table 2).

Interestingly, despite the yield decline due to waterlogging during flowering, root growth can increase during recovery following waterlogging stress, as observed in previous experiments by Lee et al. (2010) and Ploschuk et al. (2017). In this experiment, the root biomass of maize at harvest increased by 17% for roots exposed to waterlogging during flowering compared to the control (Table 1). The increased root biomass in the waterlogged treatment may be caused by a high growth rate of secondary roots during recovery, as shown in *Phalaris aquatica* (Ploschuk et al., 2017). Thus, maize yield under waterlogging during flowering is reduced probably to compensate for an increased root growth during recovery.

The previous report indicated that morpho-agronomic traits correlate with maize yield (Wiraguna et al., 2023). In this experiment, the ear length positively correlates with maize yield at 0.68 (Table 2). The correlation between ear length and yield can help to indicate the maize yield reduction before harvest. Therefore, the anticipation of minimizing yield reduction due to waterlogging can be applied.

Material and Methods

The experiment was conducted at Gunung Gede, School of Vocational Studies, IPB University, located at Babakan, Bogor, from 2 September 2023 to 23 December 2023. The experiment was conducted in an open area, where the sides of the open area were covered with a screen net to minimize pest infestation. Throughout the experiment, the average daily temperature fluctuated between 21°C to 28°C, with daily rainfall varying from 5 to 20 mm.

Randomised complete block design (RCBD) was used during the experiment. The experiment consisted of 2 treatments (waterlogging for 7 hours during flowering time and various plant growth substrates) in three replicates (3 polybags for each replicate). The waterlogging was applied at the flowering time of R2 (Raun et al., 2005). The various substrates were applied from sowing to harvesting.

Plant material and growing conditions

The commercial seeds of sweet maize cv. Pertiwi was used in this experiment. Five seeds were sown in each polybag (350 x 350 mm). After two weeks, all seedlings were thinned, leaving one seedling per polybag (Wiraguna et al., 2017; 2020). Each polybag contained approximately 3 kg of either a potting mix comprised of organic matter, including leaves, straw, grasses, rice bran, maize stoves, vines, tendrils, and animal manure (1:1:1:1:1:1:1), or mixtures of potting mix and latosol soil (1:1). NPK 15–15–15 at 5 g per polybag was applied at sowing, and Urea (45% nitrogen) at 5 g per polybag was applied at four weeks after sowing. The appropriate pesticide was applied when required to control

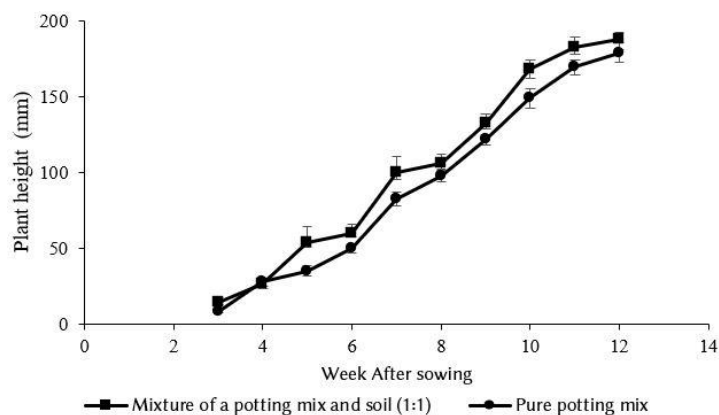


Fig 1. The growth of maize when it is cultivated in pure potting mix and a mixture of a potting mix and soil (1:1).

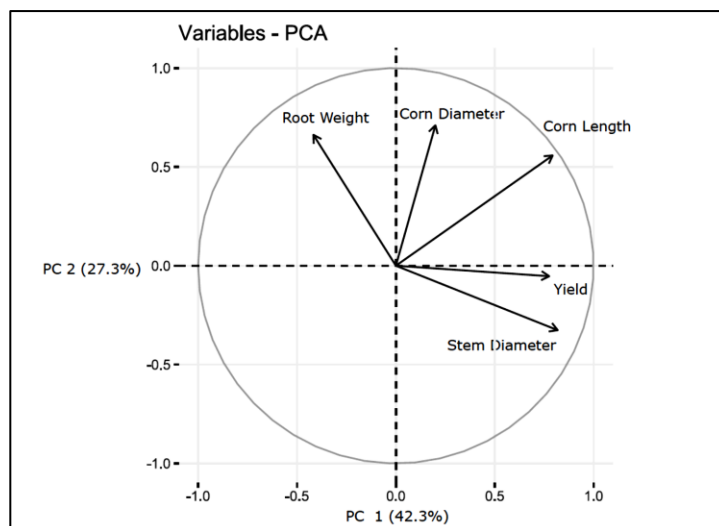


Fig 2. Principle component analysis (PCA) of morpho-agronomic traits (maize diameter, stem diameter, maize length, root diameter, and yield) at harvest. The yield was defined as a fresh weight of above-ground biomass. The root weight was identified as a root fresh biomass. The maize diameter and maize length were measured with the husk still attached. The two principal components (PC1 and PC2) explain a significant portion of the total variance, with PC1 accounting for 42.3% and PC2 accounting for 27.3%. The yield, maize length, and stem diameter are the major contributions in PC1, and root weight (fresh root biomass) and maize diameter are the major contributions in PC2.

insect pests. The growing plants were watered once a day during the first four weeks of development (up to the V6 stage) and twice a day during the later growth stage (V7 to R4 stages) (Raun et al., 2005). Watering was not applied when there was more than 10 mm of rainfall or the medium in the polybag was 80% of water capacity (Wiraguna et al., 2021). The watering routine was stopped when the plants reached the R5 growth stage (Raun et al., 2005).

Growing media and waterlogging treatments

The evaluated maize plants were grown in two growing media and exposed to waterlogging or non-waterlogging (control)

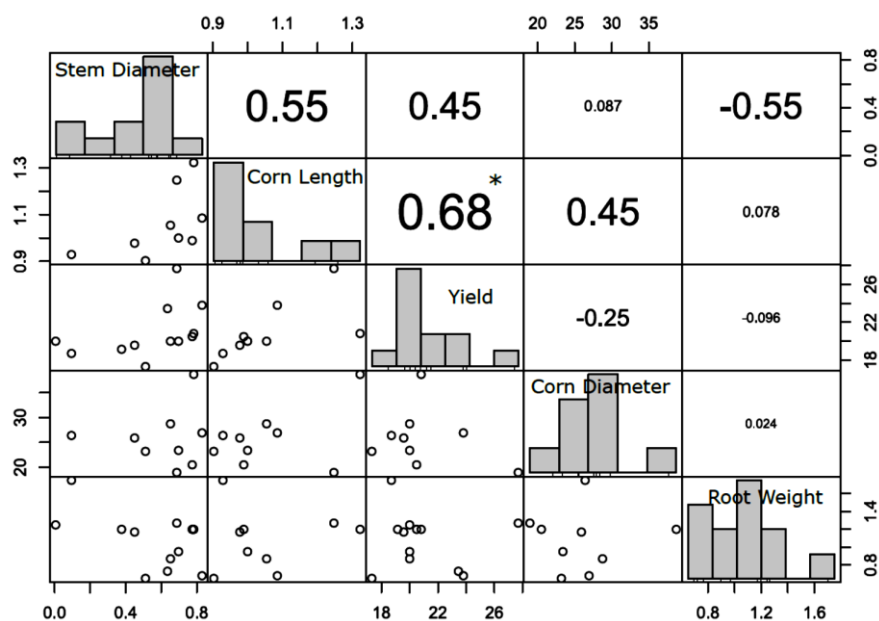


Fig 3. Correlation graph of morpho-agronomic traits at harvest.

treatments. The growing medium compositions comprised (1) a potting mix made from mixtures of the same amount of leaves, straw, grasses, rice bran, maize stalks, vines, tendrils, and animal manure (Wiraguna et al., 2017) and (2) a mixture of potting mixes and latosol soil (1:1). The waterlogging treatment included exposing the maize plant at flowering stage to seven hours of waterlogging, while the control was not. Polybags were submerged in a large container (82 liters, measuring 687 mm x 478 mm x 390 mm) filled with tap water for 7 hours to apply the waterlogging treatment. The water level was maintained 2 cm above the polybags (Ploschuk et al., 2020).

Agro-morphological observations and statistical analyses

Plant height was measured during vegetative stages (V3 to V11) (Raun et al., 2005). Stem diameter, yield (above-ground fresh biomass), root weight (fresh root biomass), ear length, and maize diameter were recorded at harvest (Wiraguna et al., 2023).

The recorded data from each treatment were the mean of three samples and were analysed using analyses of variance (ANOVA) with a randomized complete block design (RCBD) and three replications per treatment. Multiple comparison (Tukey HSD) was used to test for significant differences between treatments. Data normalisation ($W \geq 0.05$) was carried out before testing the Principal Component Analysis (PCA) and Pearson's correlation between agro-morphological traits and yield. The R-studio version 2023.03.1 was used to calculate the significant effect, and significant differences were tested at $P = 0.05$.

Conclusion

The growth and development of maize crops are not affected by plant growth substrates. Waterlogging treatment at flowering for 7 hours does not significantly affect morpho-agronomic traits at harvesting; however, the waterlogging treatment reduced yield significantly by 25% relative to the control. The yield had a significant positive correlation to maize length. Therefore, the morpho-agronomic traits of maize length can be used to indicate yield reduction affected by waterlogging at flowering.

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