Maize recovery to waterlogging as influenced by nitrogen fertilization regimes

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Abstract The result showed that maize lines can survive up to eight consecutive days of waterlogging when imposed at the V2 and V7 leaf stages. Exposure to waterlogging twice during the life cycle (at the V2 and V7 leaf stages) had the most significantly adverse affected on the growth and yield, followed by exposure at the V2 and V7 leaf stages individually. Waterlogging is triggered the early development of nodal roots and the formation of surface rooting as an adaptive response to waterlogging. Applying 50% of the RR of nitrogen at sowing and 50% at the ten-leaf stage improved the shoot and root growth, and shortened the days to anthesis and days to silking, even maize experienced waterlogging stress. Splitting the RR of nitrogen improved the growth and yield of maize when experiencing waterlogging stress. The screening identified the BRK and Sige-sige (Milako) maize lines were tolerant, while the Tiniguib (Monkayo) line was the most susceptible to waterlogging. These lines showed waterlogging tolerance which may serve as the parent materials for developing more resilient varieties for excessive moisture conditions.

Keywords: Corn, Excess soil moisture stress, Nitrogen fertilizer, Recovery

Introduction

Maize (*Zea mays* Linn.) is an essential component of agricultural food, livestock feed, and industrial products (Gazal *et al.*, 2017). This crop is a major source of food and feed in Asia, providing a significant source of income and energy for millions of farmers (Shiferaw *et al.*, 2011). The crop is the second most productive and important crop in the Philippines, after rice (Alcantara *et al.*, 2021). Maize production in the Philippines reached 759,578 million metric tonnes in November 2020 on 245,271 million hectares of harvest area, a continuous increase in productivity since 2003 (Philippines Statistics Authority, 2020). With the continuous increase of maize production in the Philippines, it become among Asia's seven major maize producers (Gerpacio and Pingali, 2007;

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Wada *et al.*, 2008). As a result, maize demand remains extremely high and has been continuously increasing over the years, both for human consumption and livestock.

However, sustaining productivity remains a major challenge. According to Rosegrant *et al.* (2009), the world's population will reach 9.3 billion by 2050, and becomes a major challenge faced by the agriculture industry. Aside from that, the crop is adaptable to a wide range of agro-climatic zones, ranging from subtropical to colder temperate regions (Lone *et al.*, 2018). As a result, the plant is inevitably exposed to various biotic and abiotic stresses. Excess soil moisture (ESM) is caused by temporary waterlogging caused by heavy rains, high groundwater tables, or dense soil texture, among other abiotic stresses (Lone *et al.*, 2018).

Waterlogging affects approximately 12% of agricultural land on a global scale (Li *et al.*, 2011). In South and Southeast Asia alone, floods and waterlogging affect 15% to over 18% of the total maize-growing area (Lone and Warsi, 2009; Zaidi *et al.*, 2009), resulting in annual losses of 25-30% of maize production (Rathore *et al.*, 1998). Moreover, maize yields would be reduced by more than 40% if waterlogging occurred for more than three days (Li *et al.*, 2011. Furthermore, Li *et al.* (2011) noted that yield reduction increased with the increasing duration of waterlogging.

Until now, the majority of research has focused on the effects of waterlogging on maize growth responses (Lone and Warsi, 2009; Liu *et al.*, 2010; Zaid *et al.*, 2003, 2004, 2007, 2010, 2012, Esteban and Solilap, 2016; Bin *et al.*, 2010. The recovery of plants following waterlogging has been overlooked (Malik *et al.*, 2002, Striker, *et al.*, 2011; 2012). Striker (2012) emphasized the importance of accurately estimating tolerance to waterlogging stress and considering plant performance during flooding and recovery periods. Additionally, there is a dearth of literature on the crop's ability to recover from waterlogging. However, few data were available on nitrogen uptake recovery (Kaur *et al.*, 2018; Zhou *et al.*, 2000).

The majority of previous maize waterlogging research has focused on hybrid selection (Lone *et al.*, 2018; Zaidi *et al.*, 2003; Thirunavukkarasu *et al.*, 2013; Souza *et al.*, 2012), compensation effects of biochemical preparation on growth and production(Ren *et al.*, 2016; 2017), and new planting management techniques such as ridge tillage or different drainage measurement to overcome the waterlogging. These studies were significant, and the majority of them focused on maize's response to waterlogging. Also, drainage management is an excellent method for draining water from cornfields. These strategies, however, are insufficient because no significant improvement in maize growth and yield performance was observed after waterlogging. Another feasible method of increasing maize productivity after waterlogging is to provide the maize plant with optimal nitrogen nutrients. This could be accomplished by fertilizing with sufficient nitrogen (Ren *et al.*, 2017; Wu *et al.*, 2018).

The available nitrogen in nitrate (NH3) is significantly reduced in waterlogged soil due to anaerobic denitrification. This loss of available nitrogen in the soil may result in soil and plant nitrogen deficiency. In addition, nitrogen deficiency in the soil will affect the uptake of other plant nutrients (Smethurst *et al.*, 2005), most notably P, K, S, Ca, and Mg (Wilkinson *et al.*, 2000). Optimum nitrogen fertilization must be equally important. As a result, physiological mechanisms for waterlogging tolerance must investigate the effects of waterlogging on maize growth and yield and the plant's ability to recover, and nitrogen efficiency. It is essential to include nitrogen because it has contributed to cellular acclimation to low oxygen stress in plants (Bailey-Serres *et al.*, 2012). Thus, the correct amount and timing of nitrogen applications are critical for alleviating the adverse effects of waterlogging and enhancing maize growth and yield.

To have a consistent maize, it is critical to screen various potential lines and evaluate various optimal nitrogen regimes to alleviate the adverse effects of waterlogging on maize productivity. Hence, the objectives of the study were to determine the effects of temporary waterlogging on growth and yield performance of traditional maize lines;to determine the effects of nitrogen fertilization regimes on the recovery period of maize after exposure to waterlogging; and to determine the maize lines that are tolerant/sensitive to temporary waterlogging.

Materials and methods

The study was determined the effect of different waterlogging conditions, amount, and period of nitrogen application on the growth and yield of maize under rainout shelter conditions at the CMU experimental station from October 2021 to June 2022. Four maize lines were used, including three traditional maize genotypes and one open-pollinated variety. The experiment followed a split-split plot design in a randomized complete block design (RCBD) with three replications. The main plot treatments were waterlogging conditions at the second (V2) and seventh (V7) leaf stages of maize, and the subplots were the amount and period of N application based on the recommended rate (RR) of nitrogen from soil analysis. The sub-sub plot treatments were the maize lines. The study used the screening technique described by Zaidi *et al* (2003) to identify waterlogging tolerant lines and traits, which were modified to meet the experimental requirements.

The growing medium used was regular garden soil, and the RR of NPK based on the result of soil analysis was used for fertilization. The seeds were sown and thinned seven days after emergence, and waterlogging treatments were established by filling the plastic containers with approximately 3cm of water above the soil surface with water in all maize lines during the V2 leaf stage, V7 leaf stage, and in double waterlogging treatment. The nitrogen application period was done at sowing and at ten leaf stage of maize following the treatment rates. The study was conducted at the Agricultural Experiment Center (AEC) in Musuan, Maramag, Bukidnon, Philippines, using rainout shelter conditions constructed of wood and bamboo, with thick transparent plastic cellophane roofing to protect plants from rain. The net was draped on each side of the shelter to ensure proper air circulation. The data collected from the study were analyzed using analysis of variance (ANOVA), and the means were compared using Tukey's Honest Significance Difference (Tukey's HSD) test at a 5% probability level.

Results

The study found that the maize V2 and V7 leaf stages can withstand up to eight days of waterlogging without causing mortality. However, waterlogging significantly affected growth parameters compared to no waterlogging stress. Multiple waterlogging events during the life cycle (at V2 and V7 stages) resulted in shortened plant height, reduced root volume, root and shoot dry weight, and total dry matter. Moreover, the plant height and root length of V2 leaf stage exposed to waterlogging were comparable to waterlogging imposed at V2 and V7 leaf stages (Table 1).

On the other hand, The results showed that waterlogging during the older vegetative stage did not significantly affect the reproductive phases and harvest maturity of maize (Table 2). It indicated that older growth stages may be more tolerant to the adverse effects of stress on reproductive phases and harvest maturity compared to earlier growth stages.

The different maize lines exhibited varietal differences in root length, nodal root count, and root volume when exposed to waterlogging stress The variety Tiniguib (Monkayo) had the longest root, while USM Var 10 had the shortest root expansion. Meanwhile, the BRK lines had the highest nodal root number compared to other maize lines, and developed a prolific number of nodal roots throughout their life cycle when exposed to waterlogging stress. Tiniguib (Monkayo) had the highest root volume, but was comparable to BRK. These findings suggest that maize lines exhibited varying responses to waterlogging stress in terms of root length, nodal root count, and root volume.

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					Root	Shoot	Total
		Plant	Root	Root	Dry	Dry	Dry
Waterlogging	%	Height	Length	Volume	Weight	Weight	Matter
Conditions	Survival	(Cm)	(Cm)	(cm^3)	(g)	(g)	(g)
Normal condition	100.00	142.74 ^a	45.67 ^b	11.15 ^a	3.88 ^a	34.40^{a}	38.28ª
Waterlogged at	100.00	98.27°	51.41ª	5.29°	1.98°	13.02°	14.99°
2 nd leaf stage							
Waterlogged at 7th	100.00	122.85 ^b	42.67 ^b	8.64 ^b	2.63 ^b	20.52 ^b	23.15 ^b
leaf stage							
Waterlogged at	100.00	99.66°	50.80ª	3.95 ^d	1.32 ^d	10.58 ^d	11.90 ^d
2^{nd} leaf and on 7^{th}							
leaf stage							
F-Test	ns	**	**	**	**	**	**
CV (%)	0.00	10.02	11.22	6.83	12.08	11.13	10.73

Table 1. Effects of different waterlogging conditions on maize growth

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

Waterlogging Conditions	Days to Anthesis (Day)	Days to Silking (Day)	Anthesis Silking Interval	Harvest Maturity (Day)	Grain Yield (g/plant)	Yield Reduction (%)
Conditions	(Day)	(Day)	Interval	(Day)	(g/plain)	(70)
Normal condition	59.97 ^b	67.22 ^b	7.25 ^b	102.53 ^b	9.24 ^a	0.00°
Waterlogged at 2 nd leaf stage	65.44 ^a	79.86ª	14.18 ^a	113.36 ^a	1.31 [°]	84.68 ^a
Waterlogged at 7 th leaf stage	61.56 ^b	70.69 ^b	9.14 ^b	105.97 ^b	4.22 ^b	54.01 ^b
Waterlogged at 2 nd leaf and on 7 th leaf stage	66.28 ^a	81.56ª	15.28ª	115.67ª	1.17 [°]	87.31 ^ª
F-test	**	**	**	**	**	**
CV (%)	4.09	5.83	9.52	5.76	30.72	25.22

Table 2. Effects of different waterlogging conditions on maize yield and yield components

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

In addition, the root dry weight, shoot dry weight, root shoot ratio, and total dry matter of different maize lines at harvest were significantly differed from each other. BRK had the heaviest root dry weight, while Sige-sige (Milako) had the lowest root dry weight at harvest when exposed to waterlogging stress.

BRK and Tiniguib (Monkayo) had the heaviest shoot dry weight among lines, while USM Var 10 had the lowest shoot dry matter at harvest. Moreover, the root shoot ratio of Tiniguib (Monkayo) significantly varied from Sige-sige (Milako) at harvest. BRK had the heaviest total dry matter, indicating efficient photosynthesis even under waterlogging stress.

The days to anthesis, days to silking, anthesis silking interval, and harvest maturity of maize lines were significantly affected by waterlogging (Table 4). The results show that Sige-sige (Milako) obtained the lowest days to silk and shortest days to harvest, while Sige-sige (Milako) and USM Var 10 obtained the earliest days to silk. In contrast, BRK and Tiniguib (Monkayo) had longer days to anthesis, and Tiniguib (Monkayo) obtained the most delayed appearance of silk among the maize lines. Additionally, USM Var 10 and Tiniguib (Monkayo) had the most prolonged period of anthesis silking interval, and Tiniguib (Monkayo) had the highest days of harvest maturity (Table 4).

	% Survi	Root Length	Nodal Root Numb	Surface Rooting	Root Volum	Root Dry Weigh	Shoot Dry Weigh	Total Dry
Maize Lines	val	(cm)	er	(%)	$e(cm^3)$	t (g)	t (g)	Matter
USM Var. 10	100.0 0	45.50 ^b	30.50 ^b	31.48°	7.08°	2.24 ^{bc}	16.41°	18.66°
BRK	100.0	48.20 ^{ab}	33.24 ^a	52.68ª	7.60 ^{ab}	2.84 ^a	23.28ª	26.13ª
Sige-sige (Milako)	0 100.0 0	48.10 ^{ab}	29.22 ^b	40. ^{bc}	6.31°	2.09°	19.03 ^b	21.11 ^b
Tiniguib (Monkayo)	$\begin{array}{c} 100.0\\ 0\end{array}$	49.00 ^a	28.27 ^b	35.00 ^{bc}	8.10 ^a	2.63 ^{ab}	19.80 ^a	22.43 ^b
F-test	ns	*	**	**	**	**	**	**
CV (%)	0.00	12.10	11.93	10.34	11.18	27.84	20.81	20.30

Table 3. Effects of waterlogging on maize lines growth

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

The shorter days to silking of Sige-sige (Milako) could be attributed to a shorter vegetative phase, resulting in an earlier reproductive stage than other maize lines. The results showed that Sige-sige (Milako) obtained the shorter period to anthesis and shorter period of anthesis silking interval. This earlier period to anthesis indicated a shorter period for the vegetative growth stage and earlier reproductive phase. Furthermore, this early reproductive stage means faster growth of the male and female reproductive organs. It is shown that the anthesis silking interval of Sige-sige (Milako) appeared the earliest among the maize lines, which could contribute to its higher grain yield per plant. On the other hand, Tiniguib (Monkayo) obtained the most delayed appearance of silk among the maize lines (Table 4).

Differences in root dry weight, shoot dry weight, and total dry matter were observed at harvest due to various amounts and periods of nitrogen application. The findings indicate that the combination of 50% RR nitrogen at sowing and 50% at the ten-leaf stage resulted in the highest root dry weight, shoot dry weight, and total dry matter among the different nitrogen application rates and periods (Table 5). Also, splitting the RR of nitrogen into different growth stages (75% at sowing and 25% at V10 leaf stage; and 50% at sowing and 50% at tenth leaf stage) reduced the number of days to anthesis (Table 6).

The crop susceptibility index (CSI) and waterlogging tolerance coefficient (WTC) were utilized to evaluate the susceptibility and tolerance of different maize growth stages and lines to waterlogging stress. The CSI and WTC values were based on the dry matter and grain yield data. A higher CSI value indicates greater susceptibility to waterlogging stress, while a lower value implies greater tolerance to the stress. Conversely, a lower WTC value indicates greater susceptibility, while a higher value indicates greater tolerance to waterlogging.

			Anthesis		Grain	
	Days to	Days to	Silking	Harvest	Yield	Yield
Maize Lines	Anthesis	Silking	Interval	Maturity	(g/plant)	Reduction (%)
	(1.1.4h		10.000	100 0140	4.0 5 .0h	
USM Var. 10	61.14 ^b	74.17 ^{bc}	13.03 ^a	108.31 ^{bc}	4.07^{ab}	56.18 ^{ab}
BRK	65.05ª	75.67 ^{ab}	10.61 ^{ab}	110.50 ^{ab}	3.83 ^{ab}	56.41 ^{ab}
Sige-sige	62.22 ^b	71.69°	9.47 ^b	107.67°	4.77 ^a	49.02 ^b
(Milako)						
Tiniguib	64.83 ^a	77.81ª	12.47 ^a	111.06 ^a	3.26 ^b	64.40^{a}
(Monkayo)						
F-test	**	**	**	**	*	*
CV (%)	5.66	3.48	14.09	3.48	44.59	31.99

Table 4. Effects of waterlogging on maize line's yield and yield components

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

Amount and Period of N Application	Shoot Dry Weight (g/plant)	Root Volume (g/plant)	Total Dry Matter (g/plant)
1000/ CDD CN	10.0 0 h	(Joh	
100% of RR of N at sowing period	18.02 ^b	6.73 ^b	20.28 ^b
50% of RR of N at sowing; 50% at ten leaf stage	21.63 ^a	8.03ª	24.30 ^a
75% of RR of N at sowing; 25% at ten leaf stage	19.24 ^b	7.00 ^{ab}	21.67 ^b
F-Test	*	*	**
CV (%)	20.05	14.72	18.94

Table 5. Maize growth response on different amount and period of nitrogen as influenced by waterlogging imposed at V2 and V7 leaf stage

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

		2	88 8			8
Amount and Period of	Dava to	Dava to		Harvest	Grain	Percentage Yield Reduction
	Days to	Days to	ACT			
N Application	Anthesis	Silking	ASI	Maturity	Yield	(%)
100% of RR of N at sowing period	64 ^a	75.88ª	11.86ª	109.77 ^a	3.33°	60.22 ^c
50% of RR of N at sowing; 50% at ten leaf stage	62.88 ^b	74.31 ^b	11.44 ^a	109.27ª	6.53ª	50.24ª
75% of RR of N at sowing; 25% at ten leaf stage	63.06 ^{ab}	74.31 ^b	11.25ª	109.10ª	4.08 ^b	55.05 ^b
F-Test	*	*	*	*	*	*
CV (b)%	3.07	3.90	9.22	3.77	18.18	16.80

Table 6. Maize yield and yield components response on different amount and period of nitrogen as influenced by waterlogging imposed at V2 and V7 leaf stage

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

The results showed that waterlogging during the V2 leaf stage and waterlogging during both the V2 and V7 leaf stages had the highest CSI values and the lowest WTC values for grain yield (Table 7). This finding suggested that these growth stages are the most susceptible to waterlogging stress in terms of grain yield. Moreover, the CSI grain yield exhibited varietal differences, where Sige-sige was found to be more tolerant than the other maize lines tested, while

Tiniguib (Monkayo) was found to be the most susceptible in terms of grain yield (Table 8).

Waterlogging Conditions	CSI Grain Yield	WTC Grain Yield
Normal condition	0.00°	1.00^{a}
Waterlogged at 2 nd leaf stage	0.85^{a}	0.15°
Waterlogged at 7 th leaf stage	0.54 ^b	0.46^{b}
Waterlogged at 2 nd leaf and on 7 th leaf	0.87^{a}	0.13°
stage		
F-test	**	**
CV (%)	25.34	32.88

Table 7. Tolerance rating of different growth stages experienced waterlogging

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

Table 8. Tolerance rating of different maize lines experienced waterlogging

Maize Lines	CSI Grain Yield		
USM Var. 10	0.56^{ab}		
BRK	0.56^{ab}		
Sige-sige (Milako)	0.49^{b}		
Tiniguib (Monkayo)	0.64ª		
F-test	*		
CV (%)	38.05		

*, ** indicates statistical significance at $P \le 0.05$ and $P \le 0.01$, respectively, Means with the same letter in the column are not significantly different

Discussion

Effects of waterlogging condition on maize growth

Waterlogging at early stages and multiple times during the life cycle led to the development of longer roots than in normal conditions. This finding confirms the previous studies by Dill *et al.* (2020) that flooding leaded to increase total root area, suggesting greater root exploration to escape flood conditions or increase nutrient capture. The study also found that waterlogging reduced root volume, indicating that growth stages experiencing waterlogging stress cannot recover from the adverse effects of waterlogging.

In addition, growth stages exposed to waterlogging exhibited a significant reduction in root dry weight, shoot dry weight, and total dry matter. Waterlogging

at the V2 and V7 leaf stages resulted in the lowest root dry weight, shoot dry weight, and total dry matter compared to other growth stages exposed to waterlogging. Moreover, the adverse effects of waterlogging persisted even after the waterlogging stress was removed, indicating that growth stages experienced waterlogging could not recover from it.

Furthermore, the study revealed that multiple waterlogging events during the life cycle caused the most significant reduction in root dry weight, shoot dry weight, and total dry matter, followed by the V2 and V7 leaf stages. The study also found that the early seedling stage was much more sensitive than the older growth stage when exposed to waterlogging, which is consistent with previous studies by Ren *et al* (2016) and Liu *et al.* (2010) that the V2 leaf stage is much more sensitive than other growth stages exposed to waterlogging.

The results suggested that waterlogging significantly affects maize plant growth parameters, with multiple waterlogging events during the life cycle causing the most severe adverse effects. Furthermore, the study findings indicated that the early seedling stage and the V2 and V7 leaf stages are more sensitive to waterlogging stress, and such growth stages cannot recover from the adverse effects of waterlogging. These findings were significant implications for maize growers and plant breeders in developing waterlogging-tolerant maize cultivars.

Effects of waterlogging condition on maize yield and yield components

These longer days to harvest maturity are attributed to delay in the reproductive phase. This findings are consistent with those of Ren *et al* (2014), who reported that waterlogging can lead to a longer period of harvest. Also, the results indicate that waterlogging during the early growth stage and multiple times in the life cycle of maize obtained a longer period to reach the reproductive phase. This suggests that a longer period to reach the productive stage means a longer vegetative growth stage, resulting in late harvest maturity. Ren *et al* (2014) also found that waterlogging can delay maize growth and development, which may result in delayed pollen shedding and lower yields. Similarly, Ren *et al* (2016) reported that waterlogging at vegetative growth stages (V3 and V7) significantly delayed growth processes, resulting in delayed days of silking and lower yields.

On the other hand, the results showed that waterlogging during the V2 leaf stage and waterlogging during the V2 and V7 stages obtained the highest reduction of percentage yield loss. It indicated that waterlogging during early growth stages and multiple times in the life cycle can significantly increase percentage yield reduction. Shin *et al.* (2016) also found that the percentage yield

loss at the V2 growth stage is about 80.00%. Furthermore, our study shows that older growth stages, such as the V7 leaf stage, have a lower percentage yield reduction than the early growth stage that experienced waterlogging stress. This is consistent with the findings of Shin *et al.* (2016) that older growth stages exposed to waterlogging have a lower percentage of yield reduction.

It is worth noting that a reduction in yield on different growth stages that experienced waterlogging stress may be attributed to multiple factors, such as the widening of the anthesis-silking interval, which results in a lower chance of successful pollination of maize (Paril *et al.*, 2014). This, in turn, can lead to the barrenness of summer maize due to waterlogging stress, as noted by Ren *et al* (2014) resulting in increased yield loss. Ren *et al* (2014) also reported that lower maize yield is contributed by a reduction in plant height, ear height, leaf area index, ear characteristics (grains per ear and 1000-grain weight), grain filling period, and dry matter accumulation and distribution due to waterlogging stress.

The findings suggested that waterlogging stress can have significant adverse effects on maize growth and yield, particularly when it occurs during early growth stages and multiple times in the life cycle. Therefore, it is important to implement appropriate management practices to minimize the negative impacts of waterlogging stress on maize production.

Effects of waterlogging on maize lines growth

Interestingly, the evaluated maize lines exhibited surface rooting when exposed to waterlogging stress, with BRK having the highest surface rooting and moderate white root tips that emerged due to waterlogging. On the fourth day after waterlogging, BRK had the highest surface rooting compared to other lines. This development of white root tips is triggered by waterlogging, which serves as an extension of maize lines during anaerobic conditions. Surface rooting has been identified as an adaptive strategy for coping with flooding in many wetland species, as reported by Armstrong and Drew (2002) and cited by Zaidi *et al.* (2007). Kaur *et al.* (2020) also observed a proliferation of surface roots in some corn hybrids as a response and possible tolerance mechanism to extended soil saturation.

The survival of maize lines during waterlogging stress at the V2 and V7 leaf stages could be related to the development of nodal roots and surface rooting. It was noted that maize lines subjected to waterlogging stress developed nodal roots and surface roots. The formation of adventitious roots or nodal root development is associated with waterlogging tolerance in plants. Furthermore, adventitious root and surface root formation during waterlogging enable maize to breathe, avoiding anaerobic respiration and allowing metabolic processes to

continue (Cañete and Baldo, 2019). Therefore, maize lines can withstand up to eight days of waterlogging on the V2 and V7 leaf stages, and no mortality occurred after waterlogging was removed with the help of nodal roots and surface roots. These traits are considered to be tolerant traits of maize to adapt to waterlogging conditions, allowing maize to survive.

Effects of waterlogging on maize line's yield and yield components

This delayed silking of Tiniguib (Monkayo) is attributed to a more extended period of anthesis and anthesis silking interval. Delayed days to anthesis mean a prolonged vegetative growth stage, which results in a delayed reproductive stage of Tiniguib (Monkayo). This delayed reproductive stage of Tiniguib (Monkayo) also affects the days of anthesis and silking emergence. Furthermore, Tiniguib (Monkayo) also obtained the most delayed anthesis silking interval, resulting in late silking emergence. The prolonged days to harvest of Tiniguib (Monkayo) are due to the adverse effects of waterlogging, as reported in previous studies (Ren *et al.*, 2014).

The results showed that Sige-sige (Milako) obtained the heaviest grain yield per plant and significantly differed from Tiniguib (Monkayo). Moreover, Sige-sige (Milako) had a shorter period of anthesis silking interval, contributing to high grain yield. A shorter anthesis silking period had a high chance of successful pollination, contributing to a high number of kernels per row and many kernel rows per ear, eventually resulting in a high grain yield per plant. In contrast, maize lines with low grain yield per plant had a long period of anthesis silking interval, which reduces the grain yield per plant. Moreover, waterlogging stress can cause barren maize, contributing to lower yield (Ren *et al.*, 2014).

Effects of different amount and period of nitrogen application on maize growth and =yield as influenced by waterlogging stress

The result suggested that supplementing 50% of nitrogen at the ten-leaf stage significantly increases root dry weight, shoot dry weight, and total dry matter compared to other nitrogen application rates and periods. Moreover, higher nitrogen application at the ten-leaf stage enhances root dry weight, shoot dry weight, and total dry matter revealed to be more than lower nitrogen application rates. Additionally, splitting nitrogen application into various growth stages enhanced maize's dry shoot weight.

Moreover, applying higher amounts of nitrogen at the ten-leaf stage was found to increase dry matter accumulation. This finding is consistent with Nelson *et al.* (2011) report, which stated that nitrogen application after flooding can help

to overcome nitrogen deficiencies induced by floods, improved grain yields, and increased dry matter. This implies that the addition of nitrogen after waterlogging can enhance photosynthetic activity, leading to higher dry matter accumulation.

This finding is consistent with Nelson *et al.* (2011) study, which reported that in-season rescue nitrogen applications could improve grain yields by mitigating flood-induced nitrogen deficiencies. In contrast, applying the entire amount of RR of nitrogen at sowing did not reduce the days to anthesis and silking, as the nitrogen may have been lost due to denitrification and leaching in flooded or saturated soil. Additionally, applying a larger amount of nitrogen during sowing can still result in nitrogen deficiency during waterlogging, as reported by Kaur *et al.* (2020).

Furthermore, the study evaluated different maize lines and their response to waterlogging stress. The results revealed that Sige-sige (Milako) had the heaviest grain yield per plant and the shortest anthesis silking interval, contributing to successful pollination and eventually a high grain yield. In contrast, Tiniguib (Monkayo) had the smallest ear length among other lines. The findings suggested that waterlogging stress can lead to a number of barren maize plants, which reduced the grain yield per plant (Ren *et al.*, 2014). Previous studies have also reported that waterlogging or flooding can lead to reduced maize grain yields (Howell and Hiler, 1974).

Thus, the present study is provided an evidence that splitting the RR of nitrogen into different growth stages can mitigate the negative impact of waterlogging stress on maize development, leading to a reduced number of days to anthesis. Also, the study highlights the importance of choosing the appropriated maize lines that are resulted to be more resilient to waterlogging stress to increase grain yield per plant. These findings can be useful for maize growers and researchers interested in developing strategies to reduce the impact of waterlogging stress on crop yield.

Tolerance rating of different maize growth stages and lines that experienced waterlogging stress

These results showed that certain maize growth stages and lines are found to be more vulnerable to waterlogging stress than others, which is significant impact on grain yield. Therefore, it is important for farmers to carefully consider the timing of planting and other management practices to minimize the risk of waterlogging stress on maize crops. Additionally, plant breeders can use the information from this study to develop maize varieties with improved tolerance to waterlogging stress, which could help to increase yield and mitigate the negative effects of waterlogging stress on maize production.

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