*Research article*

# Hydro- and Osmo-priming Effects on Upland Rice Exposed to Drought Conditions at Vegetative and Reproductive Stages

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**Abstract** Drought stress during vegetative and/or generative stages could cause massive reduction in rice yield. This study evaluated effectiveness of hydro- and osmo-priming on improving seed germination, growth and development of upland rice under drought stress during late vegetative, booting, or heading stage. Treatments consisted of hydro-priming and osmo-priming consisted of three polyethylene glycol (PEG) concentrations i.e. 10%, 15%, and 20%. Results showed that application of osmo-priming at 10% PEG required longer time (21.93 hours) to reach 50% germination, lower germination and lower vigor index. However, after germination, seeds primed with 10% PEG exhibited better seedling growth than other seed priming treatments. Effects of seed priming on yield components were overshadowed by drought exposures. Drought imposed during vegetative stage did not significantly affect yield; however, regardless of seed priming treatments, yield reduction was inevitable in rice plants exposed to drought during booting or heading stages. Rice plant shortened time to reached physiological maturity as an adaptive mechanism if drought treatment was applied during heading stage.

**Keywords:** Germination, Polyethylene glycol, Seedling growth, Seed priming, Yield component

# INTRODUCTION

Rice cultivation at irrigated paddy fields has been well established with high productivity. Meanwhile, average productivity of upland rice is only about half of irrigated rice. Expanding paddy fields for rice production is costly, required major infrastructure development, and not always be ecologically and/or economically feasible. However, massive cultivation of upland rice by large number of smallholder farmers could collectively contribute to efforts in increasing food availability for continuously rising population in developing countries.

Natural constraint in upland rice cultivation is reliable sources of water. Water is essential for seed germination, vegetative growth stage, and especially during booting stage to few days before harvest. Massive yield loss is mostly attributed to drought occurrence during seed germination and reproductive organ development.

Drought inhibited some plant physiological and biochemical processes which, in turn, restraining plants growth and development (Larkunthod et al., 2018; Kartika et al., 2020; Lakitan et al., 2020). Drought stress caused disruption of plant metabolic processes, including nutrient absorption (da Silva et al., 2011), cell division and enlargement (Avramova et al., 2015), enzyme activity (Abedi and Pakniyat, 2010), and stomatal conductance (Kartika et al., 2020). Stomatal closure is the first reaction of plants in water deficient condition which inhibited gas exchange, including photosynthesis, transpiration, and related carbohydrates metabolism (Pirasteh-Anosheh et al., 2016). Alteration in plant physiological and biochemical under drought stress condition resulting in yield reduction. Besides affecting quantitative yield, drought also lowered grain quality in rice, including grain shape, percentage of brown rice, chalkiness, and nutrient content (Yang et al., 2019).

Seed priming has been used for enhancing germination and improving seedling quality (Ria et al., 2020). Seed priming stimulated metabolic process at pre-germinative stage but disrupted actual radicle and plumule emergence (Raj and Raj, 2019). Recently, there are many priming techniques, i.e. hydro-, osmo-, halo-, matrix-, hormone-, and nutria-priming. However, hydro- and osmo-priming are affordable, simple, and more likely to be adopted by smallholder farmers. Hydro-priming utilizes pure water, while osmo-priming uses chemical solution. Seed priming process was managed for specific time under controlled condition and was dehydrated and stored until it was ready to be planted (Pawar and Laware, 2018). Both hydro- and osmo-priming had been reported to have positive impact for improving germination (Ghassemi-Golezani et al., 2008; Lemmens et al., 2019) and seedling growth (Oliveira et al., 2019).

Although the main effect is an acceleration of germination, priming treatment has also been reported to improve vegetative growth and grain yield in rice (Mahajan et al., 2011) and ability of rice crop to tolerate drought condition (Hussain et al., 2017). However, response among rice varieties might be different to hydro- and osmo-priming treatments. Therefore, this research was conducted to evaluate effectiveness of hydro- and osmo-priming on seed germination, vegetative growth and yield of Situ Patenggang, an upland rice variety, exposed to drought stress during late vegetative, booting, and heading stages.

# MATERIALS AND METHODS

## Seed material and priming procedure

Upland indica rice studied was Situ Patenggang variety. Seeds with pretested germination rate higher than 90% and initial moisture contents of 10.03% were used in this study. Seeds were treated with hydro-priming or osmo-priming with different concentrations of polyethylene glycol (PEG 6000), i.e. 10%, 15%, and 20%. Seeds treated with hydro-priming were soaked in aerated distilled water. Seeds of hydro- and osmo-priming were soaked for 12 hours in complete darkness at $28 \pm 2^{\circ}$C. The primed seeds were rinsed with distilled water for 3-4 times; then, air dried at $28 \pm 2^{\circ}$C for restoring seed moisture to the pre-treatment level.

## Germination and seedling growth

Primed seeds were arranged in petri dish and soaked with distilled water for 32 hours.  Germination was defined by the emergence of the radicle from the seed coat. Seed emergence were counted every 4-hour intervals from 12 until 32 hours, after no further additional seed germination was detected. Time taken to 50% germination ($T_{50}$) was calculated based on the formula of Coolbear et al. (1984). Mean germination time (MGT) was calculated according to Ellis and Roberts (1981). Germination index (GI) was calculated using the formula proposed by Khan and Ungar (1998). Vigor index (VI) was calculated based on percentage of germination and seedling length at 7 days after sowing (DAS) as described by Abdul-Baki and Anderson (1973).

Formula for calculating $T_{50}$ was:

$$T_{50} = t_i + \frac{\left(\frac{N}{2} - n_i\right)(t_j - t_i)}{(n_j - n_i)}$$

where $N =$ the final number of germinations; $n_i$ and $n_j$ = cumulative numbers of seeds germinated by adjacent counts at times $t_i$ and $t_j$ when $n_i < {N}/{2} < n_j$

MGT was calculated as follow:

$$MGT = \frac{\sum(n\,T)}{N}$$

where $n =$ number of seeds newly germinated at time $T$; and $T$ = hours from the beginning of the germination test.

Furthermore, GI was calculated based on the following formula:

$$GI = \sum \frac{\%\ germinated\ seed}{germination\ periode}$$

and VI was calculated using this formula:

$$VI = \%\ germination \times seedling\ length\ at\ 7\ DAS$$

Germinated seeds were sown in seedling tray filled with soil as growing substrate. Seedling shoot length was measured at 7 DAS and 14 DAS by selecting 3 seedlings randomly for each treatment.  Seedling data were destructively collected at 14 DAS for measuring length as well as fresh and dry weight of shoot and roots.

## Pot experiment and drought stress treatment

One seedling of two weeks old were transplanted into pot filled with soil-manure mixed growing substrate (2:1 v/v). Pots were 26 cm in height, 22 cm at base diameter and 30 cm at upper diameter. Plants were supplied with enough water at early vegetative stage. Drought stresses initiated at three different stages, i.e. late vegetative stage, started at 49 days after sowing (DAS); booting stage, started at 63 DAS; or heading stage started at 91 DAS.

Plants were placed in a plastic house to avoid unexpected rainfall during drought treatment period. Drought treatment was conducted by halting water supply and prevent the plants from rain until soil moisture was dropped to less than 10%; required 14 consecutive days during late vegetative stage, and 12 consecutive days during booting and heading stages. Soil moisture was monitored daily using soil moisture meter (Lutron PMS-714) at 10 cm depth during treatments.

Measured parameters during vegetative growth and reproductive stage included leaf area (LA), shoot length (SL), root length (RL), shoot fresh weight (SFW), root fresh weight (RFW), shoot dry weight (SDW), root dry weight (RDW), root length (RL), leaf rolling score (LRS), number of tillers (NoT), time of physiological maturity (TPM), panicle length (PL), weight of 100 grains (W100), number of spikelet (NoS), and yield. LRS was

observed during last two days of drought stress according to the Standard Evaluation System for Rice (IRRI, 2002). At termination of drought treatment, all plants were re-arranged to their original positions at the open field area and generously re-watered until harvest.

## Experimental design and statistical analysis

This study was arranged in a randomized block design for germination and seedling growth; but, factorial randomized complete block design for pot experiment. Observed variables were measured with three replications. Each hill is assigned as replication. Collected data were subjected to ANOVA using SAS 9.0 statistical analysis software (SAS Institute Inc., Cary, North Carolina, US). Significant differences between treatments were tested using the least significant difference (LSD) test at $P \le 0.05$.

## RESULTS

### Effect of seed priming on germination and early vegetative growth

The application of hydro- and osmo-priming with application rate up to 20% did not indicate significant effect on percentage of seed emergence (Table 1). However, mean comparison of priming treatments for $T_{50}$ revealed that seeds treated with 10% PEG was significantly slower to germinate than those treated with hydropriming or osmo-priming at higher PEG treatments. The application of osmo-priming decreases osmotic potential and inhibits imbibition process and metabolic activities. The application of PEG 10% took 21.93 hours to reach $T_{50}$ with MGT at 26.80 hours. Furthermore, GI and VI of seeds treated with PEG 10% were 10.37 and 100.90, respectively. These values were also lower than those treated with higher PEG concentrations (Table 2). This results indicated that hydropriming was better for stimulating seed germination of Situ Patenggang variety. Otherwise, if osmo-priming used, higher concentrations (15% or 20%) of PEG was required.

**Table 1.** Emergence percentage of hydro and PEG-primed seeds counted every 4 hours after germination initiation.

| Priming | Time after initiation (hour) | | | | | |
|---|---|---|---|---|---|---|
| | 12 | 16 | 20 | 24 | 28 | 32 |
| Hydro-priming | 10.00 [a] | 40.00 [a] | 86.67 [a] | 90.00 [a] | 90.00 [a] | 93.33 [a] |
| PEG 10% | 3.33 [a] | 10.00 [b] | 30.00 [b] | 63.33 [a] | 73.33 [a] | 86.67 [a] |
| PEG 15% | 10.00 [a] | 50.00 [a] | 76.67 [a] | 83.33 [a] | 90.00 [a] | 100.00 [a] |
| PEG 20% | 16.66 [a] | 40.00 [a] | 66.67 [a] | 83.33 [a] | 83.33 [a] | 93.33 [a] |
| LSD (0.05) | 14.89 | 23.78 | 27.66 | 38.68 | 33.39 | 31.93 |

Note: Means followed by different letters within each column indicated statistically significant differences at LSD05. There was no seed germinated at 4 and 8 hours after initiation.

**Table 2.** Time taken to 50% germination ($T_{50}$), means germination time (MGT), germination index (GI), and vigor index (VI) of hydro and PEG-primed seeds.

| Priming | $T_{50}$ | MGT | GI | VI |
|---|---|---|---|---|
| Hydro-priming | 16.40 [b] | 24.89 [b] | 17.54 [a] | 193.26 [a] |
| PEG 10% | 21.93 [a] | 26.80 [a] | 10.37 [b] | 100.90 [b] |
| PEG 15% | 17.04 [b] | 24.87 [b] | 17.60 [a] | 142.53 [ab] |
| PEG 20% | 18.22 [b] | 24.78 [b] | 16.58 [ab] | 188.89 [a] |
| LSD (0.05) | 2.49 | 1.01 | 6.25 | 55.55 |

Note: Means followed by different letters within each column indicated statistically significant differences at LSD05.

Both shoot and root growth in all priming treatments were not significantly different as indicated by their length and weight, except in leaf area and root fresh weight. Interestingly, leaf area and root fresh weight of seedlings treated with PEG 10% was the highest amongst hydro or osmo-primed seedlings at higher PEG concentrations (Table 3).

**Table 3.** Effects of seed priming on rice seedlings growth at 14 days after sowing.

| Priming | LA (cm$^2$) | SL (cm) | RL (cm) | SFW (g) | RFW (g) | SDW (g) | RDW (g) |
|---|---|---|---|---|---|---|---|
| Hydro-priming | 15.85 [b] | 21.60 [a] | 16.50 [a] | 0.126 [a] | 0.14 [ab] | 0.023 [a] | 0.020 [a] |
| PEG10% | 23.98 [a] | 21.10 [a] | 13.53 [a] | 0.126 [a] | 0.20 [a] | 0.024 [a] | 0.031 [a] |
| PEG15% | 11.97 [b] | 20.96 [a] | 18.00 [a] | 0.116 [a] | 0.09 [b] | 0.017 [a] | 0.022 [a] |
| PEG20% | 16.48 [b] | 20.33 [a] | 13.30 [a] | 0.120 [a] | 0.14 [ab] | 0.017 [a] | 0.022 [a] |
| LSD(0.05) | 6.64 | 4.21 | 9.40 | 0.040 | 0.09 | 0.007 | 0.013 |

Note: Means followed by different letters within each column indicated statistically significant differences at LSD[05].

During vegetative stage at 5 WAT and 6 WAT, PEG-priming at concentration of 15% produced significantly taller plants than hydro and other osmo-priming treatments. However, the differences in plant height were diminished at 7 WAT. This phenomenon suggested that at 7 WAT height of Situ Patenggang variety had reached its near maximum height and vegetative growth progressively shifted from increasing height to developing tillers. Rice plant has been well known for having a maximum height. However, number of tillers per hill may variably depending on genetics, environmental conditions, and cultivation practices. In this study, highest number of tillers produced by rice plants treated with hydropriming (Figure 1). However, it should be kept in mind that not all tillers were productive.
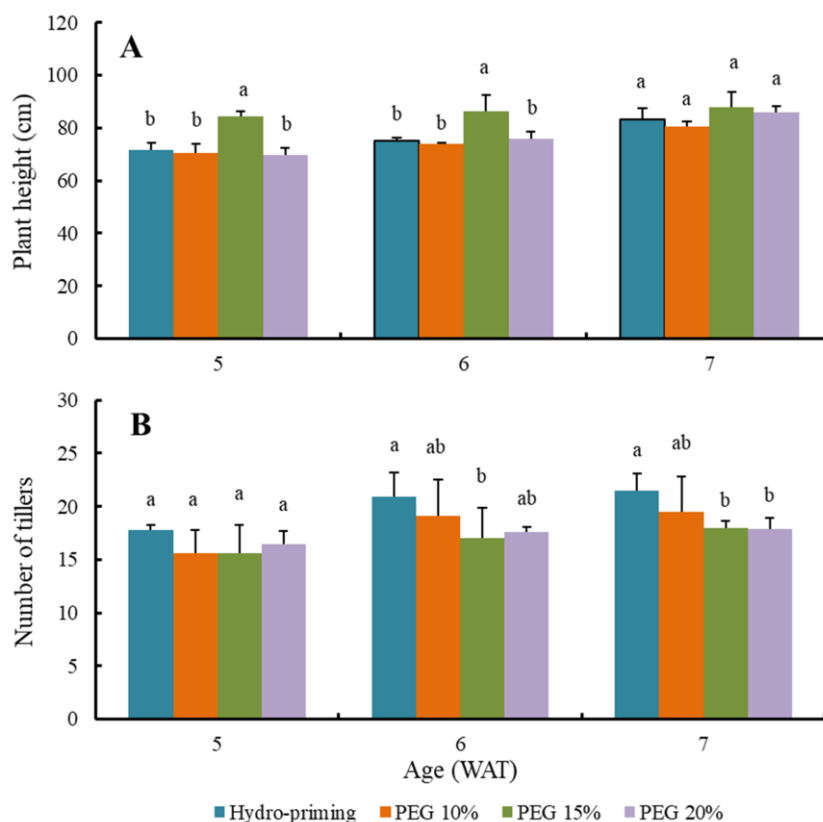


**Figure 1.** **Effect of seed priming on plant height (A) and number of tillers (B) at 5, 6, and 7 week after transplanting.**

## Effect of seed priming and drought stress on growth and yield components

Drought stress was initiated at 3 different growth stages i.e. late vegetative, booting, and heading stage. Drought stress indicated by decline of water availability in the soil. The amount of water absorbed by plants was higher during generative phase including booting and heading stages. Thus, soil moisture decreased faster at generative phase. The application of different seed priming methods did not affect soil moisture at booting and heading stages. Furthermore, leaf rolling score did not affected by seed priming, but the score was higher if plant imposed to drought stress during generative phase (Figure 2).
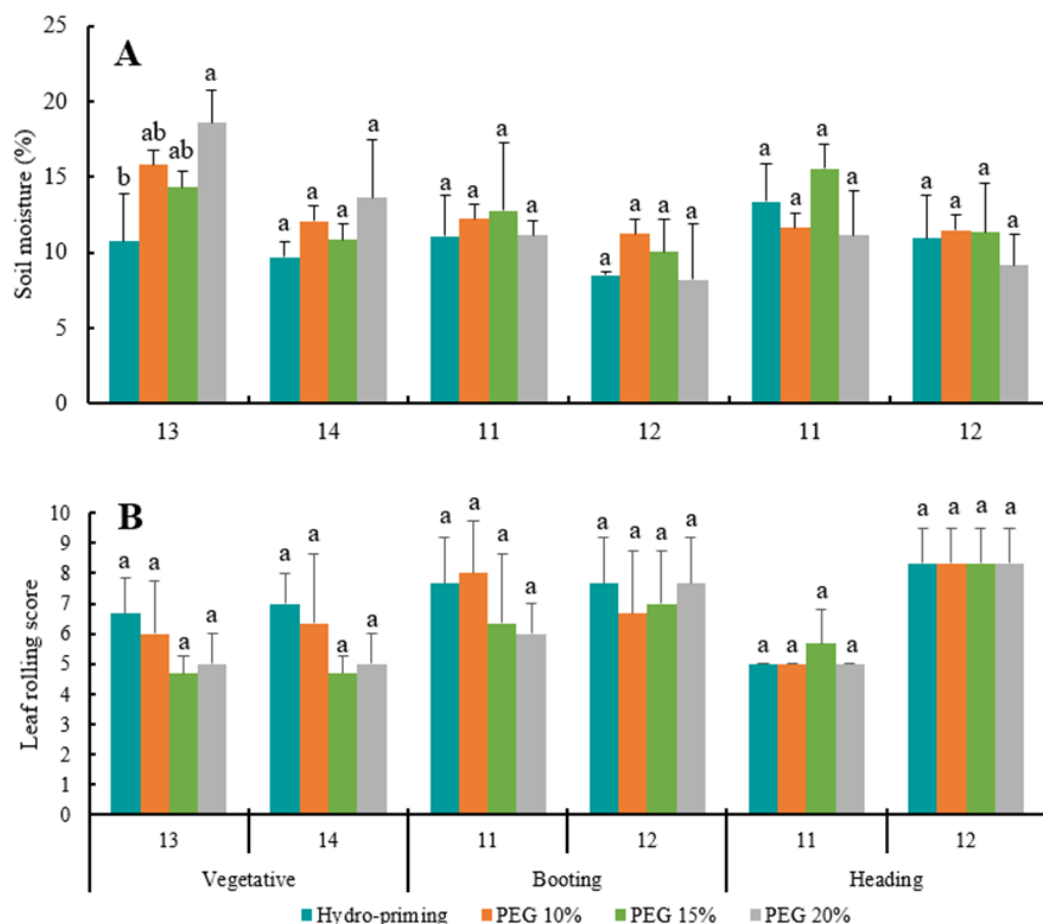


**Figure 2. Moisture of growing substrate (A) and leaf rolling score (B) at the last two days of drought stress during vegetative, booting, and heading stage.**

Application of seed priming did not significantly affect plant biomass except for stem fresh weight (Table 4). Plant treated with PEG 10% and PEG 15% exhibited heavier stem fresh weight of 119.22 g and 117.56 g, respectively. Plants imposed to drought stress during booting stage showed the highest weight of leaf, root, and stem. However, plant biomass significantly decreased when drought stress occurred during heading stage. Overall, the highest plant biomass obtained in plants treated with hydro-priming and experienced less stress during heading stage. Longest root was obtained by plants primed with 10% PEG priming. Plants treated with PEG10% were more able to endure drought stress during heading stage, as indicated by significant root length compared to plants treated with other priming procedures.

**Table 4.** Effects of seed priming, drought stress, and their interactions on the growth of vegetative organs.

| Treatment | LFW (g) | RFW (g) | SFW (g) | LDW (g) | RDW (g) | SDW (g) | RL (cm) |
|---|---|---|---|---|---|---|---|
| *Priming* | | | | | | | |
| Hydro-priming($P_0$) | 15.72 [a] | 117.11 [a] | 107.78 [ab] | 4.89 [a] | 23.93 [a] | 23.64 [a] | 47.23 [a] |
| PEG 10% ($P_{10}$) | 17.17 [a] | 115.11 [a] | 119.22 [a] | 5.25 [a] | 23.64 [a] | 24.76 [a] | 49.68 [a] |
| PEG 15% ($P_{15}$) | 15.72 [a] | 101.78 [a] | 117.56 [a] | 5.15 [a] | 20.06 [a] | 28.20 [a] | 45.63 [a] |
| PEG 20% ($P_{20}$) | 13.87 [a] | 102.78 [a] | 86.22 [b] | 6.36 [a] | 22.86 [a] | 25.18 [a] | 44.43 [a] |
| LSD (0.05) | 4.78 | 22.07 | 31.17 | 2.88 | 6.30 | 8.77 | 5.37 |
| *Drought stress* | | | | | | | |
| Vegetative (V) | 15.96 [ab] | 98.92 [b] | 104.67 [a] | 4.85 [ab] | 18.84 [b] | 22.38 [b] | 45.05 [a] |
| Booting (B) | 18.32 [a] | 124.17 [a] | 121.83 [a] | 7.14 [a] | 26.95 [a] | 31.84 [a] | 46.80 [a] |
| Heading (H) | 12.59 [b] | 104.50 [b] | 96.58 [a] | 4.25 [b] | 22.08 [ab] | 22.12 [b] | 48.38 [a] |
| LSD (0.05) | 4.14 | 19.11 | 26.99 | 2.49 | 5.46 | 7.59 | 4.65 |
| *Interaction of priming and drought stress* | | | | | | | |
| $P_0V$ | 14.49 [abc] | 100.33 [bc] | 89.67 [bcd] | 4.11 [b] | 17.85 [b] | 18.20 [bc] | 44.63 [ab] |
| $P_0B$ | 22.61 [a] | 145.33 [a] | 153.33 [a] | 5.92 [b] | 31.46 [a] | 36.97 [a] | 47.73 [ab] |
| $P_0H$ | 10.05 [c] | 105.67 [bc] | 80.33 [cd] | 4.64 [b] | 22.49 [ab] | 15.75 [c] | 49.33 [ab] |
| $P_{10}V$ | 19.33 [ab] | 92.00 [c] | 126.67 [abcd] | 5.88 [b] | 18.33 [b] | 26.17 [abc] | 45.73 [ab] |
| $P_{10}B$ | 17.46 [abc] | 130.67 [ab] | 136.00 [ab] | 5.29 [b] | 26.12 [ab] | 27.15 [abc] | 52.93 [a] |
| $P_{10}H$ | 14.73 [abc] | 122.67 [abc] | 95.00 [bcd] | 4.58 [b] | 26.48 [ab] | 20.96 [bc] | 50.37 [ab] |
| $P_{15}V$ | 15.71 [abc] | 96.33 [bc] | 119.67 [abcd] | 4.61 [b] | 17.53 [b] | 21.40 [bc] | 44.17 [ab] |
| $P_{15}B$ | 17.63 [abc] | 114.33 [abc] | 100.00 [abcd] | 5.59 [b] | 24.81 [ab] | 32.74 [ab] | 43.67 [ab] |
| $P_{15}H$ | 13.83 [bc] | 94.67 [bc] | 133.00 [abc] | 5.26 [b] | 17.84 [b] | 30.47 [abc] | 49.07 [ab] |
| $P_{20}V$ | 14.30 [bc] | 107.00 [bc] | 82.67 [bcd] | 4.79 [b] | 21.64 [ab] | 23.75 [abc] | 45.67 [ab] |
| $P_{20}B$ | 15.57 [abc] | 106.33 [bc] | 98.00 [bcd] | 11.77 [a] | 25.40 [ab] | 30.49 [abc] | 42.87 [b] |
| $P_{20}H$ | 11.75 [bc] | 95.00 [bc] | 78.00 [d] | 2.51 [b] | 21.53 [ab] | 21.29 [bc] | 44.77 [ab] |
| LSD (0.05) | 8.28 | 38.23 | 53.99 | 4.99 | 10.92 | 15.19 | 9.03 |

Note: Means followed by different letters within each column for each treatment and their interaction indicated significant differences at LSD.05. Abbreviations have been mentioned in Materials and Methods section.

The effects of seed priming were visible in rice generative traits (Table 5). Hydro- and osmo-priming with PEG application rate up to 15% resulted heavier grains. Meanwhile, high concentration of PEG application at 20% decreased grain size as indicated by lowest weight of 100 grains. Grain weight was reduced by drought exposure. The reduction was higher if drought was imposed at generative stage. Drought stress at vegetative stage did not affect grain weight and number of filled spikelet regardless of variable priming treatments. Drought stress during late vegetative stage and booting stage decelerated maturity process and grain filling as indicated by smaller grains. Moreover, plants treated with drought stress at heading stage shortened life cycle of rice plants as indicated by earlier time of physiological maturity yet produced lower yield (Table 5).

**Table 5.** Effects of seed priming, different time of drought exposure, and interactions between priming and drought on yield components.

| Treatment | Time of physiological maturity (DAT) | Panicle length (cm) | Weight of 100 grains (g) | Yield (Ton ha$^{-1}$) |
|---|---|---|---|---|
| *Priming* | | | | |
| Hydro-priming (P0) | 147.00 [a] | 22.87 [a] | 2.21 [ab] | 2.72 [a] |
| PEG 10% (P10) | 143.22 [a] | 23.47 [a] | 2.30 [ab] | 3.06 [a] |
| PEG 15% (P15) | 133.66 [a] | 24.39 [a] | 2.47 [a] | 2.80 [a] |
| PEG 20% (P20) | 139.66 [a] | 23.81 [a] | 2.16 [b] | 3.37 [a] |
| LSD (0.05) | 15.15 | 1.77 | 0.28 | 0.86 |
| *Drought stress* | | | | |
| Vegetative (V) | 142.66 [ab] | 24.06 [a] | 2.42 [a] | 3.67 [a] |
| Booting (B) | 149.25 [a] | 22.89 [a] | 2.32 [ab] | 3.14 [a] |
| Heading (H) | 130.75 [b] | 23.95 [a] | 2.12 [b] | 2.15 [b] |
| LSD (0.05) | 13.12 | 1.53 | 0.24 | 0.74 |
| *Interaction of priming and drought stress* | | | | |
| $P_0V$ | 152.00 [a] | 22.64 [ab] | 2.27 [ab] | 3.09 [abc] |
| $P_0B$ | 152.00 [a] | 22.80 [ab] | 2.26 [ab] | 2.76 [abcd] |
| $P_0H$ | 137.00 [ab] | 23.18 [ab] | 2.10 [bc] | 2.31 [bcd] |
| $P_{10}V$ | 151.67 [a] | 24.96 [ab] | 2.60 [a] | 4.14 [a] |
| $P_{10}B$ | 152.00 [a] | 21.99 [b] | 2.52 [ab] | 3.47 [ab] |
| $P_{10}H$ | 126.00 [ab] | 23.47 [ab] | 1.77 [c] | 1.58 [d] |
| $P_{15}V$ | 141.00 [ab] | 25.28 [a] | 2.50 [ab] | 3.84 [a] |
| $P_{15}B$ | 141.00 [ab] | 22.91 [ab] | 2.45 [ab] | 2.90 [abcd] |
| $P_{15}H$ | 119.00 [b] | 24.98 [ab] | 2.46 [ab] | 1.65 [cd] |
| $P_{20}V$ | 126.00 [ab] | 23.38 [ab] | 2.30 [ab] | 3.60 [ab] |
| $P_{20}B$ | 152.00 [a] | 23.88 [ab] | 2.04 [bc] | 3.43 [ab] |
| $P_{20}H$ | 141.00 [ab] | 24.19 [ab] | 2.14 [abc] | 3.07 [abc] |
| LSD (0.05) | 26.24 | 3.07 | 0.49 | 1.48 |

Note: Means followed by different letters within each column for each treatment and their interaction indicated significant differences at LSD.05.

The highest number of panicles collected from three consecutive harvests (Harvest 1, 2, and 3) was found in rice plant pre-treated with hydropriming and exposed to drought at booting stage. Meanwhile, the lowest was found in plant pre-treated with PEG 15% and exposed to drought at heading stage. Conversely, percentage of filled spikelet was the lowest in the plant exhibited the highest number of panicles (Figure 3).
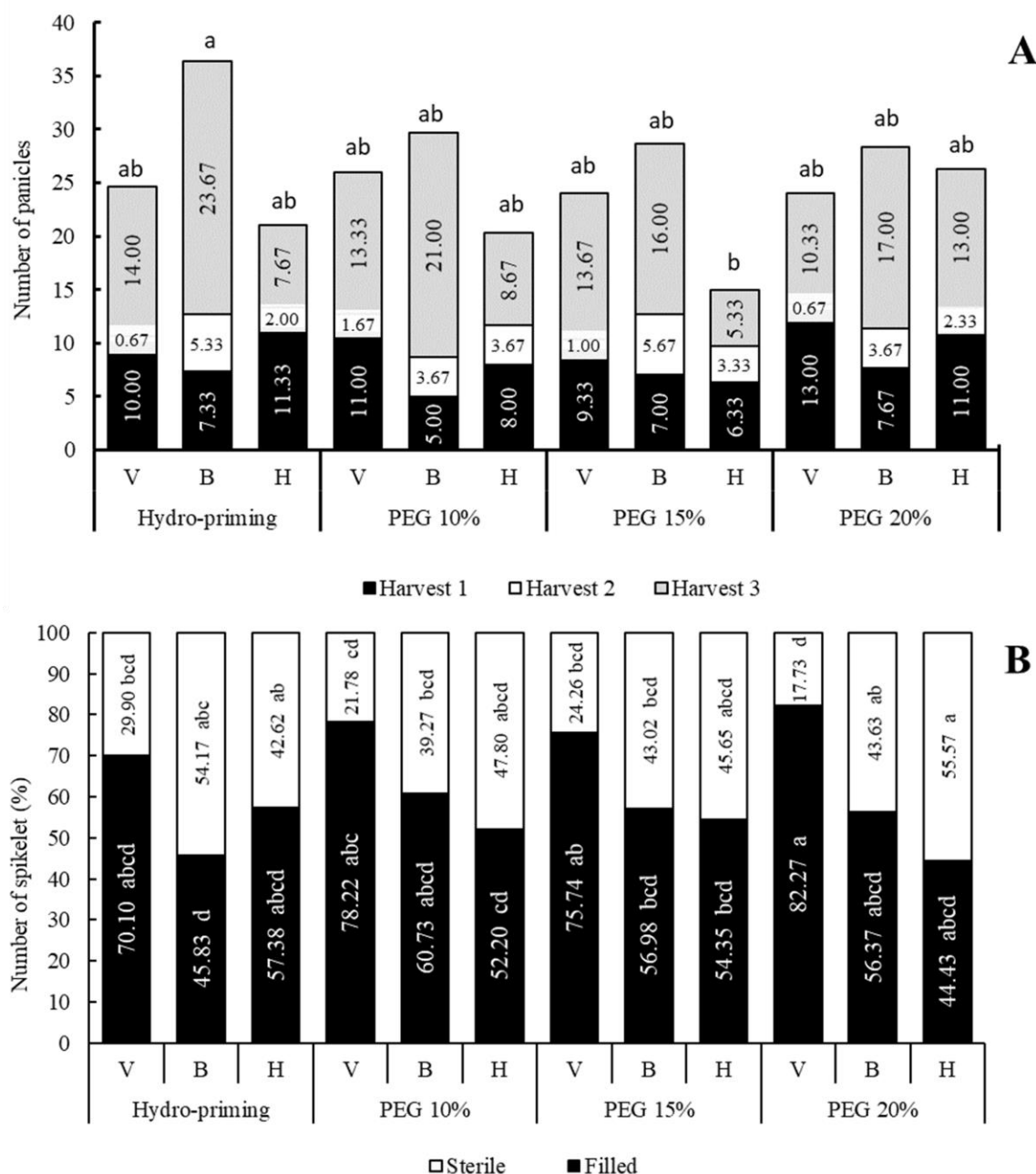
**Figure 3.** Interaction between seed priming and drought stress at different growth stages for number of panicles (A) and percentage of filled spikelet (B).

## DISCUSSION

### Seed priming on germination and seedling growth

Seed priming was implemented to improve germination rate, seedling quality which determines growth and yield of plant. The present study compared hydro-priming with 3 different concentrations of PEG used in osmo-priming treatments. Results of the study revealed that seed priming treatments caused different effects on germination, including time to 50% germination, mean germination time, germination index, and vigor index. Germination of Situ Patenggang rice variety treated with hydro-priming was comparable with osmo-priming with the application rate at 15% and 20% PEG; meanwhile, germination and vigor indexes of seeds treated with 10% PEG were significantly lower (Table 1 and 2). Similar result was previously reported in Batu Tegi,

another Indonesian upland rice variety (Abdallah et al., 2016). Singh et al. (2015) recognized that effects of seed priming might be varied as affected by some factors, including specific germplasm of plant species, growing media, kind and concentration of chemical used, duration of priming treatment, and seed storage conditions after priming treatment.

Prior to germination, imbibition and activation phases occurred. Water quickly uptake during imbibition phase and causing activation of metabolic and physiological activities (Pawar and Laware, 2018). Hydro- and osmo-priming stimulates those phases but prevent radicle protrusion. Immediate germination occurs once seeds are sown. Enhanced germination is associated with several process during priming treatment, including enzyme activation, synthesis of primary metabolites i.e. proteins, nucleic acids, and ATP (Oliveira et al., 2019). The physiological changes increased starch hydrolysis and sugars availability for enhancing germination and seedling growth (Hussain et al., 2017).

Growth of seedlings treated with hydro-priming and osmo-priming at 15% and 20% PEG was not significantly different. Interestingly, seeds primed with 10% PEG exhibited better seedling growth than other priming treatments, as indicated by higher leaf area and root fresh weight (Table 3). This result was in line with report of Faijunnahar et al. (2017) that the best seedling growth was obtained when wheat seed was treated with 10% PEG compared to non-priming, hydropriming, and seedling growth gradually decreased in higher PEG concentrations. The suppression of seedling growth at high PEG concentrations has been reported as due to nutrient imbalance, mobility of toxic ions, and reduction in solubility of applied solutes (Abiri et al., 2016).

Rice seedling predominantly uses available nutritions in cotyledon during their early growth stage. At later stage, seedling growth depends on ability of roots in absorbing water and soil nutrients and harvesting light energy by newly developed leaves via photosynthesis (Artadana et al., 2019). As measured at 14 DAS, higher leaf area and longer roots of seedlings primed with 10% PEG are beneficial for supporting growth and development. Seedling growth was completely dependent on its roots for absorbing water and nutrients and leaves for capturing $CO_2$ and light energy, since cotyledon had been fallen off the mesocotyl.

## Carry-over effects of seed priming on vegetative growth

The effect of priming treatment on rice vegetative growth was mainly exhibited on development of tillers. Number of tillers is important since it determines number of panicles produced and potential yield in rice crop (Kartika et al., 2018a). Higher number of tillers was produced in hydro-priming treatment compared to osmo-priming with 10% to 20% PEG concentrations (Figure 1). Tillers initiation seemed to be triggered by drier growing substrate, since moisture dropped to a much lower level (around 10%) in the substrate grown with hydro-primed rice plant; meanwhile, all recorded moisture levels were consistently higher in substrate grown with osmo-primed plants, varied from 10% to 20% PEG concentrations (Figure 2). Alam et al. (2015) mentioned some genes that could contribute to increase of photosynthesis and tiller numbers in rice plants exposed to salinity and drought conditions. Further research is needed to prove whether Situ Patenggang variety also contains the drought tolerant genes.

All rice plants exposed to 14 days drought treatments reached maximum leaf rolling index regardless of seed priming treatment selected if drought was exposed at heading stage (Figure 2). Leaf rolling index has been used for visual indicator for drought stress in rice plant (Kartika et al., 2018b). As for plant biomass, higher stem fresh weight identified in 10% and 15% of PEG priming application and less reduction in biomass of vegetative organs was observed if drought was exposed during booting stage (Table 4). Drought during booting stage severely suppressed newly developed water-sensitive inflorescence. This might cause relocation of assimilates produced by photosynthesis to further support growth of vegetative organs. Kobata et al. (1994) reported that drought during booting stage did not reduce assimilate production but did increase sterility in spikelet. These findings support the possibility of assimilate relocation to vegetative organs since the damaged reproductive organ lose its role as a dominant sink.

### Drought effects on yield

Larger grain size was obtained in rice plant grown using seeds pre-treated with 15% PEG; however, the plant did not produce higher yield (Table 5). This indicated that the plant produced lower total grains per hill than plants treated with other priming treatments. In this case, low total grains were consistently associated with lower number of tillers developed during vegetative stage, as shown at Figure 1. Despite showing positive effects on germination and early stage of seedling growth, the effects of osmo-priming did not go beyond four weeks after seedlings were transplanted. Instead, drought exposure during late vegetative stage restrained number of tillers produced per hill (Ria et al., 2020). Kang et al. (2017) had identified specific gene related to tiller development in drought tolerant rice. During recovery after drought treatment was terminated, assimilates were mainly allocated for supporting development of reproductive organs within the limited number of tillers, resulting larger grain size.

Among seed-primed plants exposed to drought, the highest yield was attained in plants pre-treated with 10% PEG and exposed to drought only during vegetative stage. In this study, panicle length, grain size, and yield in rice plants exposed to drought during vegetative stage were less severely affected compared to those exposed to drought during booting or heading stage. Similar results were reported by Zhang et al. (2018) who found that drought had larger detrimental influences during the reproductive phase, e.g., blooming stage, filling stage, and maturity.

Drought exposure during heading stage enhanced maturity but produced less yield (Figure 5). Drought stress reduced endogenous auxins production but increased abscisic acid and ethylene, which in turn, fastened flowering and grain maturity (Abobatta, 2019). Shortened time to physiological maturity potentially reduced grain yield due to short available time for accumulation of photosynthates (Shavrukov et al., 2017).

Plant biomass and yield components were more significantly affected by drought stress condition. As rice plants shifted from vegetative to generative stage, the plants had reached maximum vegetative organ growth. Situ Patenggang rice variety was able to maintain their yield if drought stress occurred during vegetative stage. Similar results reported in another upland rice varieties i.e Inpago 4, Inpago 10, and Rindang 1 (Ria et al., 2020). Reduction of grain weight and number of filled spikelet occurred when plant imposed to drought stress during generative stage. Farooq et al. (2009) reported that yield reduction under drought stress was due to reduction of photosynthetic rate. Akram et al. (2013) estimated that drought stress reduced photosynthetic rate by 30.69% at panicle initiation, 28.00% at anthesis, and 12.26% at grain filling. In addition, reproductive abortion also happened as the capacity of sinks to utilize assimilates declined.

## CONCLUSION

Seed priming exhibited positive effects on germination and early growth of rice seedlings. The effects of seed priming gradually diminished soon after the plants were exposed to drought at late vegetative stage. Development of reproductive organs were predominantly regulated by drought during booting and heading stages. Drought during late vegetative stage suppressed tillers development but the plants were able to recover and produced comparable yield with their significantly higher percentage of filled spikelet and larger grain. Rice plants exposed to drought at booting stage were inherited with higher number of tillers per hill since the plants had produced optimum number of tillers before drought exposure; however, they produced higher percentage of sterile spikelet. Physiological maturity in rice plants was enhanced by drought exposure during heading stage but this survival mechanism was at cost of yield reduction.

## ACKNOWLEDGEMENTS

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