

Interactive effect of duration of complete submergence in two -leaves stage on seedling growth of five rice (*Oryza sativa* L.) varieties in greenhouse

Behnaz Shabani Moziraji¹, Allahyar Fallah²

1. Former M. Sc. Student of Agronomy of Azad University of Damghan Branch.
2. Professor assistant of rice research institute of Iran Amol.

Corresponding Author email: a.fallah@areo.ir

ABSTRACT: In order to investigating the effect of complete submergence on seedling growth of five rice cultivars in transplanting boxes, a greenhouse test was carried out in the Deputy of Rice Research Institute of Iran (Amol). Experiment was done as a split plot, based on a randomized complete plot design with two factors of cultivar and duration of complete submergence. Flooding periods with zero, 12, 24 and 48 hours were considered as the main plot and cultivars of Shiroodi, Tarom Mahali, Fajr, Kouhsar and Keshvari as the sub plots. Complete submergence were performed for 12, 24 and 48 hours in two leaves stage of seedling. The results showed that interactive effect was significant on seedling height, root length, leaf area, dry weight of shoot, roots, seeds, total dry weight and entire volume of roots and shoots, SPAD value, chlorophyll and nitrogen content at 1%. Seedling of Kouhsar had higher seedling height, roots volume, leaf area and total dry weight, SPAD value, chlorophyll and nitrogen content. With the increase of submergence duration, seedling height, roots volume, SPAD value, chlorophyll and nitrogen content were increased, but seeds and total dry weights decreased. Complete submergence was caused increasing of 10% of seedling height and decreasing of 50% seed and total dry weight.

Key words: Chlorophyll, Nitrogen, Rice seedling, Submergence, SPAD value

INTRODUCTION

Rice plant is a semi-aquatic plant in which it can grow in irrigation and upland condition (Ismail et al., 2009). Chen and Qualls (2003) were mentioned that the plants are growing in flooded soil, they have aerenchyma tissue in roots and it continues to shoot. Thomson et al., (1992) were carried out a same condition of flooded stress for wheat, triticale, and rice plants. They are cited that shoot growth of rice plants was better than wheat and triticale. The ratio of root to shoot was lower in wheat and triticale, as a result, decreasing of root growth caused to limitation of stem elongation. Pezeshki (2001) concluded that flooding stress in aerobic plants decreased stem growth, leaf development, and leaves abscission. Horiguchi (1995) conceded that rice seedling grown in a nursery under upland conditions, the shoot part lower in height than seedling grown in a nursery under flooded conditions.

Plant roots are required oxygen for growth and the availability of oxygen is depended to soil condition and climate factors (Pezeshki, 2001). The researches show that diffusion of oxygen is 10000 times lower in water than air. Under soil condition with ample oxygen, roots grow favorably and under condition of oxygen deficiency, the roots growth is just reverse. Rice roots have well-developed aerenchyma within the cortex, and it supplies oxygen to the root from the aerial part (Horiguchi, 1995). Colmer (2003) cited that activity of microorganism became limited due to water deficiency in soil.

Among of cereal crops, only rice crop is germinated and growth of coleoptiles will be well in flooded soil compared to other cereal crops (Stetter and Waters, 2003). Brisson et al., (2002) were concluded that respiration rate was decreased by low temperature condition due to oxygen deficiency. Therefore, seed germination and growth of seedling in flooded soil related to normal soil, will has more damages. Abdelbagi et al., (2009) have shown that effect of flooding was more in directed culture for seed germination and seedling growth. Ismail et al.,

(2009) was obtained flooding decreased rate of germination of rice. Fukao et al., (2003) have reported that the plant under flooding alcohol dehydrogenase enzyme activity was increased. Formation of aerenchyma tissue for plants under flooding is a way to water deficiency, especially in flooding soils (Musgrave and Ding, 1998). Ashraf (2003) concluded that the rate of photosynthesis decreasing in sensitive crops, such as, wheat and tomato were higher than resistance crop like rice. There is no information about of submergence effects on seedling of different rice varieties of Iran. Sometimes, there are flooding in rice nursery north of Iran and it caused to damage of rice seedling. Therefore, the target of this research was evaluation of five rice seedlings under complete submergence in two- leaves stage.

MATERIALS AND METHODS

In order to study of interactive effect of complete submergence in two- leaves stage, a greenhouse experiment was carried out in deputy of rice research of Iran (Amol) in 2012. A split plot, based on completely designs with two factors were done which complete submergence was main plot in four levels (0, 12, 24 and 48 h), and Shiroodi, Tarom Mahali, Fajer, Keshvari, Kohsar were sub-plot. Seed boxes were 30 x 60 cm wide and length with 3 cm depth. Soil was put in seed boxes with silty loam tissue. One gram nitrogen was used per kilo gram soil and half gram of P_2O_5 and K_2O were used as well. For each variety was used 30g germinated seed. The five varieties were randomly seeded in each seed box. There were 12 seed boxes in experiment. The seed boxes were control in greenhouse up to the two- leaves stages. This stage 3 seed boxes put down in box with 2x2 m wide and length with 30 cm depth for 12h and 3 seed boxes complete submergence for 24h. Finally, 3 seed boxes put down in box for 48h. There were 3 seed boxes in greenhouse. As soon as possible, seedling characteristics, such as, seedling height, root length, leaf area, dry weight of roots, shoot, seeds, total dry weight and entire volume of roots and shoots, root-shoot ratio were measured. We measured SPAD value, chl_a, chl_b, chl (a+b) and N concentration of leaves as well. For each variety and trait, 10 seedlings were selected and measured. Seedling and root length were measured by ruler. The leaf area was obtained by measure of wide and length of all leaves and calculated by times in 0.67 (Yoshida et al., 1976). The root and shoot volume was measured in inserted tube. The dry weight was obtained by put of roots, shoots and seeds in oven with 70^oc for 48h. SPAD value was measured by SPAD 502 tool. Chlorophyll a, b and (a+b) were measured by method of Arnon. The percent of nitrogen was measured by Kjeldahl Method. The average of each characteristic was used to analysis by SAS program. The mean comparisons were done by MSTATC program at 5% probability level in DMRT method.

RESULTS

Seedling height

Table (1) showed that expect of the Shiroodi seedling, other rice seedling was increased by increasing of duration of complete submergence. Seedling height was increased about 10- 15 percent compared to check (without submergence). The highest height was related to seedling of Kouhsar variety in which it was equal 28.13cm.

Root length

The reactions of varieties were different to duration of complete submergence. The highest of root height was belonged to seedling of Kouhsar variety in which it was equal 6.93cm (Table 1). 12h complete submergence was increased root height of rice seedling about 8 percent compared to check.

Leaf area

The leaf area for each variety was different and seedling of shiroodi, Fajer and Kouhsar were increased by increasing of duration of complete submergence (Table 1). The Kouhsar variety had more leaf area than the Fajer and Tarom Mahali. The leaf area of Kouhsar was about 42.4 cm² under 48h submergence but, Fajer variety had 46 cm² leaf area per seedling in 24h complete submergence (Table 1).

Root volume

The highest root volume related to shiroodi variety in 48h complete submergence in which it was equal 0.43 cm³. Overall, except Kouhsar variety, with increasing of complete submergence was increased root volume. The root volume of Kouhsar variety was decreased about 63 percent in 48h complete submergence compared to check treatment (without submergence (Table 1).

Shoot volume

The highest shoot volume related to Kouhsar and Keshvari varieties in 24h complete submergence in which it was equal 0.36 cm³. In all varieties, except Tarom Mahali, the highest shoot volume was produced in 24h complete submergence. However, with increasing of 48h complete submergence, shoot volume was increased (Table 1).

Root and shoot dry weight

There are not different between treatments about root dry weigh but the trend to be increasing (Table 1). The highest of root dry weight related to shiroodi variety in 12h complete submergence. The shoot dry weight of the Fajer variety was highest in 24h and 48h complete submergence (Table 1). The lowest of shoot dry weight was belonged to Tarom Mahali variety in 12h complete submergence, that is, equal 0.01 g. The shoot dry weight of all of these varieties, except Keshvari variety, were increased by increasing of duration of complete submergence (Table 1).

Seed dry weight

The highest of seed dry weight was in check treatment (Table 1). The complete submergence compared to without submergence caused 50 percent of decreasing. The highest and lowest of seed dry weights were related to Shiroodi and Tarom Mahali varieties, respectively (Table 1).

Total dry weight of seedling

The seedling of Kouhsar variety had highest total dry weight. The more decreasing of total dry weight was observed in 12h complete submergence and with increasing of duration of complete submergence; there was different reaction between five rice seedlings (Table 1).

Table 1. Mean comparison of interactive effect of rice (*Oryza sativa* L.) variety and duration of complete submergence on seedling height, root length, leaf area, dry weight of shoot, roots, seeds, total dry weight and entire volume of roots and shoots of one rice seedling

	seedling height (cm)	root length (cm)	leaf area (cm ²)	Root Volume (cm ³)	Shoot Volume (cm ³)	Root Dry Weight (g)	shoot Dry Weight (g)	Seed Dry Weight (g)	Total Dry Weight (g)
V ₁ h ₀	16.51jk	5.62bcde	21.53k	0.07fg	0.23bcde	0.01cd	0.04bc	0.05a	0.10bc
V ₁ h ₁	18.91gh	5.70bcd	18.67l	0.10fg	0.23bcde	0.06a	0.05ab	0.02bc	0.13ab
V ₁ h ₂	18.87hi	5.75bcd	24.25ij	0.10fg	0.26bcde	0.01cd	0.05ab	0.03b	0.09bc
V ₁ h ₃	18.36ghi	6.19bc	23.46jk	0.43a	0.16de	0.04b	0.05ab	0.03b	0.10bc
V ₂ h ₀	20.25ef	5.11de	35.29e	0.10fg	0.20cde	0.02cd	0.05ab	0.05a	0.12a
V ₂ h ₁	24.07b	3.33i	41.62cd	0.13ef	0.30bcd	0.015d	0.015d	0.03b	0.06d
V ₂ h ₂	21.56cd	4.32fgh	33.31ef	0.10fg	0.53abcd	0.015d	0.055ab	0.03b	0.10bc
V ₂ h ₃	22.40c	3.89hi	31.78f	0.36b	0.26bcde	0.015d	0.045bc	0.03b	0.10bc
V ₃ h ₀	17.39ij	4.16gh	29.07g	0.10fg	0.26bcde	0.025cd	0.04bc	0.045a	0.11ab
V ₃ h ₁	17.90hi	6.24b	27.10gh	0.13ef	0.20cde	0.015c	0.045bc	0.01c	0.07d
V ₃ h ₂	20.93de	4.20gh	46.37a	0.10fg	0.30bcd	0.025d	0.075a	0.01c	0.11ab
V ₃ h ₃	20.79de	4.06h	44.06b	0.36b	0.1e	0.02d	0.07a	0.01c	0.10bc
V ₄ h ₀	21.62cd	5.06def	32.18f	0.36b	0.33bcd	0.015d	0.055ab	0.05a	0.12ab
V ₄ h ₁	24.90b	4.86efg	41.17cd	0.16de	0.26bcd	0.02cd	0.05ab	0.03b	0.10bc
V ₄ h ₂	28.13a	6.05bc	40.34cd	0.23c	0.60a	0.025cd	0.065ab	0.03b	0.11ab
V ₄ h ₃	27.46a	5.94bc	42.42bc	0.13ef	0.30bcd	0.02d	0.05ab	0.02bc	0.10bc
V ₅ h ₀	16.02k	4.90efg	26.96gh	0.13ef	0.26bcde	0.01d	0.04bc	0.05a	0.10a
V ₅ h ₁	18.10ghi	6.93a	39.54d	0.16de	0.16de	0.01d	0.04bc	0.01c	0.06d
V ₅ h ₂	18.84gh	5.43cde	26.26hi	0.06gh	0.36bc	0.03bc	0.02cd	0.02bc	0.07d
V ₅ h ₃	19.26fg	5.04def	25.67hig	0.20cd	0.13de	0.025cd	0.025cd	0.01c	0.06d

For each parameter, means followed by different letter (with a column) are significantly different at the 0.05 probability level according to DRMT. V₁= Shiroodi, V₂= Tarom Mahali, V₃= Fajer, V₄= Kohsar, V₅= Keshvari h₀=Cheek, (without submergence), h₁= 12h, h₂=24h and h₃=48 complete submergence

SPAD value

Table (2) showed that the highest SPAD value was related to Kouhsar variety. The SPAD value was decreased with increasing of duration of complete submergence. The value of decreasing was between 1-7 units. The highest of decreasing was obtained in Keshvari variety in 48h complete submergence compared to normal situation (Table 2).

Table 2. Mean comparison of interactive effect of rice (*Oryza sativa* L.) variety and duration of complete submergence on seedling SPAD value, chl_a, chl_b, chl(a+b) and percent of leaves nitrogen

	SPAD Value	Chl _a (mg.g ⁻¹) Fresh weight	Chl _b (mg.g ⁻¹) Fresh weight	Chl(a+b) (mg.g ⁻¹) Fresh weight	Leaves N (%)
V ₁ h ₀	16.4cd	0.558d	0.589b	1.147bc	1.64c
V ₁ h ₁	15.2d	0.561d	0.558b	1.119cd	1.73c
V ₁ h ₂	18.4c	0.665c	0.426c	1.090c	1.30d
V ₁ h ₃	16.6c	0.910a	0.712a	1.620a	1.71c
V ₂ h ₀	19.2bc	0.527d	0.512c	1.039c	2.23b
V ₂ h ₁	19.4bc	0.527d	0.511c	1.039c	1.37de
V ₂ h ₂	20.3ab	0.445de	0.429c	0.878d	1.06e
V ₂ h ₃	13.8de	0.550d	0.537bc	1.080c	1.31e
V ₃ h ₀	18.7c	0.822b	0.753a	1.575a	2.41a
V ₃ h ₁	17.2cd	0.821b	0.758a	1.579a	2.45a
V ₃ h ₂	13.8de	0.813b	0.588b	1.401b	2.51a
V ₃ h ₃	13.9de	0.557d	0.571b	1.128cd	2.41a
V ₄ h ₀	21.3a	0.558d	0.589b	1.147bc	1.91bc
V ₄ h ₁	19.9ab	0.231f	0.140f	0.373f	0.98e
V ₄ h ₂	20.8ab	0.302ef	0.249ef	0.551e	1.24d
V ₄ h ₃	16.5cd	0.557d	0.543bc	1.100cd	2.13b
V ₅ h ₀	20.2ab	0.493de	0.355d	0.824d	1.23c
V ₅ h ₁	20.1ab	0.493de	0.365d	0.824d	1.014e
V ₅ h ₂	15.7de	0.363e	0.317de	0.671de	1.21c
V ₅ h ₃	12.7e	0.633cd	0.557bc	1.19c	2.49a

For each parameter, means followed by different letter (with a column) are significantly different at the 0.05 probability level according to DRMT.

V₁= Shiroodi, V₂= Tarom Mahali, V₃= Fajer, V₄= Kouhsar, V₅= Keshvari
 h₀=Cheek, (without submergence), h₁= 12h, h₂=24h and h₃=48 complete submergence

Chlorophyll content

The highest of chlorophyll a, b and (a+b) was belonging to Kouhsar variety and decreasing of chlorophyll related to other rice seedling was lower if duration of complete submergence became higher (Table 2). The amount of chlorophyll a, b and (a+b) were decreased in 24h complete submergence (Table 2). Except Shiroodi variety, other rice seedling, amount of chlorophyll a, b and (a+b) were decreased in 48h complete submergence (Table 2).

Nitrogen content

The highest of nitrogen content belong to Kouhsar variety in which it was not affected by complete submergence, however, four others rice seedling, the nitrogen content decreased by increasing of duration of complete submergence (Table 2). As results, there is positive effect between SPAD values, chlorophyll content with nitrogen content of rice seedling.

DISCUSSION

The results of this experiment showed that complete submergence in two- leaves stage of rice seedling was caused to increasing of seedling height, root height, leaf area but decreased seed dry weight and total dry weight of seedling. The SPAD value, chlorophyll and nitrogen content were decreased by complete submergence as well. Water itself is chemically harmless to plants. All rice cultivar are damaged under submergence for several days. Setter and Laureles (1996) cited that after phenotypic screening of 18115 lines of rice seedling to possess about a few days to submergence, they concluded that only 2 percent was resistance to complete submergence. The effects of water in this situation are drive from physical properties. The seedling of rice in waterlogged, to face with slow gas diffusion of O₂ and CO₂. Visible symptoms are faster elongation and one or two leaves more with yellowing of older leaves (Jackson and Ram, 2002). The deep water rice plants to escape aerobically from slowly rising water by means of accelerated stem extension that maintains apical parts above the water (Catling et al., 1988). The rice plant was not well adapted to sudden and total in undulation when this is sustained for several days and the effect can be fatal, especially when plants are small (Jackson and Ram, 2002). Yamanda (1959) cited that rice plant under waterlogged for 12 days, showed increasing of plant height, however, this variety of rice was not resistance to submergence. Singh et al., (2001) were obtained same results. They above discussion show that submergence increased plant height.

Yamanda (1959) provided that extensive data showing the rapid loss of starch and total carbohydrate in leaves and roots (Jackson and Ram, 2002), as a result, dry weight of shoot and root were decreased. Insufficiency of light in complete submergence, and low diffusion of CO₂, was caused decreasing of photosynthesis and, decreasing of dry matter production. Soltani et al., (2006) concluded that decreasing of seedling dry weight under

submergence was caused by decreasing efficiency of transport changes of photosynthesis production to shoot and root.

Chaturvedi et al (1995) observed the higher total nitrogen content in leaves of submergence – tolerance than susceptible rice genotypes. Yamanda (1959) cited that submergence rapidly depletes protein reserves through hydrolysis to amino acids and other soluble nitrogen compounds. Yordanova and Popova (2007) concluded that decreasing of photosynthesis process under submergence was due to by decreasing of some enzyme activity such as RUBP and PEP carboxylase enzymes.

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