EFFECTS OF SUBMERGENCE DEPTH ON THE GROWTH AND TUBERIZATION OF *Eleocharis ochrostachys* Steud.

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Abstract

Eleocharis ochrostachys is considered to be a native plant in Tram Chim National Park. This study determined the effects of soil moisture and submergence conditions on the growth and tuberization of the plant. The results showed that the development of the plant was the most suitable at soil moisture of 80 -100% and submergence of 15 cm, while it poorly grew and did not produce any tuber at soil moisture of 45 - 50%. Eleocharis ochrostachys still produced tubers at continuously submergence depth of 5 cm, 10 cm and 15 cm, but its tubers were waterlogged. These results show that a suitable water level should be controlled so that the plant can grow well and produce vital tubers.

Keywords: *Eleocharis ochrostachys, soil moisture, submergence depth, growth, tuber.*

ẢNH HƯỞNG CỦA ĐIỀU KIỆN NGẬP NƯỚC LÊN SỰ TĂNG TRƯỞNG VÀ TẠO CỦ CỦA NĂNG KIM (*Eleocharis ochrostachys* Steud.)

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Tóm tắt

Năng kim (Eleocharis ochrostachys) mọc tự nhiên và được coi là loài thực vật bản địa ở Vườn Quốc gia Tràm Chim. Nghiên cứu này nhằm đánh giá sự ảnh hưởng của độ ẩm đất và điều kiện ngập nước đến sự sinh trưởng và tạo củ của chúng. Kết quả cho thấy, Năng kim tăng trưởng và tạo củ thích hợp ở độ ẩm đất 80 - 100% và mức ngập sâu của nước là 15 cm. Ở mức ẩm độ đất 45 - 50% Năng kim tăng trưởng kém và không tạo củ. Việc duy trì ngập nước liên tục ở mức 5 cm, 10 cm và 15 cm thì Năng kim vẫn tạo củ, nhưng củ nhanh chóng bị úng. Từ kết quả này có thể đề xuất biện pháp quản lý nước phù hợp để Năng kim có thể sinh trưởng và tạo củ tốt.

Từ khóa: *Năng kim, sinh trưởng, độ ẩm của đất, ngập nước, củ.*

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1. Introduction

Tram Chim National Park is a wetland ecosystem in Dong Thap province, and it is also known to be the habitat of many species of plants and animals. In Tram Chim, *Eleocharis ochrostachys* grassland is one of the main ecosystems and its tubers are considered to be a main food source for the crane named *Grus Antigone sharpie* (Huynh et al., 2016). The crane usually migrates there in the dry season. In the flood season, most areas of *Eleocharis ochrostachys* ecosystem were submerged, so the plant did not produce any tuber (Nguyen, 2012). During the dry season, the water level around canals was dried up, and the plant produced tubers (Nguyen, 2010).

In recent years, the natural and ecological conditions have been changed by human activities such as maintaining water in the park to control fire, which may be somewhat unsuitable for the ecology. The water control may affect the growth and tuberization of *Eleocharis ochrostachys*, such as poorly producing tubers. The area of *Eleocharis ochrostachys* grassland has been narrowed, affecting the habitat and feeding of birds and some other animals, especially *Grus antigone sharpie*. However, the effects of submergence depth on the growth and tuberization of the plant have not been investigated in detail yet. Therefore, this study aims to determine the effects of soil moisture and submergence depth on the growth and tuberization of *Eleocharis ochrostachys*, from which an appropriate water level is proposed to reduce the negative effects and improve the growth and ability to produce tubes of the plant.

2. Materials and methods

Tuber and soil collection: *Eleocharis ochrostachys* tubers were collected from area A5 in Tram Chim Nation Park - Tam Nong District, Dong Thap Province. The tuber dimension was about 7.0 - 9.0 mm × 4.0 - 6.0 mm.

Topsoil samples were taken from the surface layer (0 - 15 cm deep) in *Eleocharis ochrostachys* field in Tram Chim National Park. Significant debris was removed from the soil, and the soil was homogenized, dry naturally in the air for 710 days (Figure 1A). Soil was transferred into pots mulched with a plastic sheet (Figure 1B). Each pot was added with 3.5 kg soil and placed

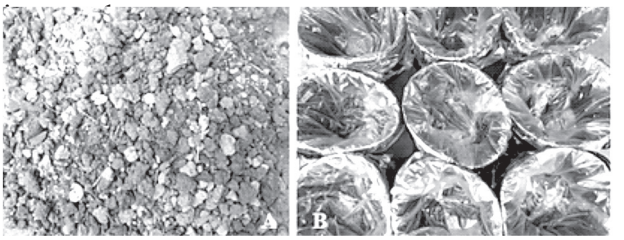

Figure 1. Soil (A) and pots used to plant tubers (B)

for cultivating

Soil moisture and water conditions: after planting, soil in all pots was controlled at the moisture of 80-100% to ensure the survival of *Eleocharis ochrostachys* because high soil moisture could stimulate root development. After 15 days, soil moisture and water levels were set up at different treatments. In the control, water was added to a submergence depth of 20 cm at the beginning, then reduced the level by 5 cm every 15-day period until none-flooding condition, and subsequently controlled at soil moisture of 80-100%. The control depicted the water variation in Tram Chim National Park after flooding season. The treatment was comprised of soil moisture of 45-50% (treatment 1), soil moisture of 80-100% (treatment 2), submergence depth of 5 cm (treatment 3), submergence depth of 10 cm (treatment 4), and submergence depth of 15 cm (treatment

5). Soil moisture was measured using Moisture Tester DM15 (Takemura, Japan). The plant was cultured for 5 months.

Size and weight measurement of the plant: Plant height (above soil surface) was measured after 45, 75, 105 and 150 days of cultivation, while root length was measured after harvesting (150 days). The fresh and dried leaves, roots and tubers were also weighed. The fresh weight was measured after harvesting several hours, while dried weight was determined after being dried using Memmert UN110 (Germany). The drying process was controlled at 50^{o}C for 30 minutes, increased to 105^{o}C for two hours and then controlled at 75^{o}C until constant weight.

Figure 3. ***Eleocharis ochrostachys*** **Steud.**

Statistical analysis

All obtained data from all experiment replicates are shown as the mean ± standard deviation. Significant differences among means were statistically analyzed using one-way Duncan's test ($p < 0.05$) in SPSS program version 22.0.

3. Results and discussion

3.1. Effects of submergence depth on the growth of *Eleocharis ochrostachys*

3.1.1. Effects of submergence depth on the plants height and roots length of Eleocharis ochrostachys

After 45 days, the height of the plant growing at the depth of 10 and 15 cm was 10.20 and 11.20 cm on average, respectively, and not significantly different compared to the control ($p < 0.05$). The height values of plant in treatments 2 and 3 were 6.40 and 8.82 cm, respectively. The height of the plant in the treatment 3 was statistically higher than that of treatment 2.

After 75 days old, the height values of the plant in treatment 0 and 5 reached the highest with 17.80 and 17.52 cm, respectively. The average plant height in treatments 3 and 4 were statistically higher compared to those of experiments 1 and 2. Data in treatment 1 showed the lowest value.

After 105 and 150 days old, the average plant height of experiments 5 was the highest with 26.08 cm, and was significantly higher compared to those of other treatments as well as the control.

The plant root in treatment 5 was the longest with 16.90 cm, which was significantly higher than those compared with the other treatments and the control. Root length values in experiments 3 and 4 were 13.32 and 14.44 cm respectively, significantly higher than those of other experiments. The root length in treatment 2 was significantly higher compared with treatment 1, showing the lowest value. All results were presented in Table 1.

The study results showed that soil moisture and submergence depth affected the plants height and root length of *Eleocharis ochrostachys*, which is completely consistent with the judgment of Le and Nguyen (2005) who showed that lacking of water might cause the plant to stop growing completely. Through field surveys, when the water level rose, the plant growth was stimulated. In submergence conditions, the plants elongated and rose above the water level. The flood water was a factor that stimulated the growth of *Eleocharis ochrostachys*. When soil moisture was controlled at 45-50%, the plant height and root length poorly grew.

Table 1. Plants height and roots length of *Eleocharis ochrostachys*

Treatments	Plants height (cm)				Roots length (cm)
	45 days	75 days	105 days	150 days	150 days
0	10.86±0.32^{a}	17.80±0.44^{a}	21.56±0.56^{c}	21.90±0.48^{d}	9.20±0.51^{cd}
1	4.80±0.25^{d}	6.30±0.25^{e}	8.42±0.52^{d}	8.70±0.46^{e}	7.30±0.25^{d}
2	6.40±0.50^{c}	10.86±0.54^{d}	20.80±0.54^{c}	21.16±0.47^{d}	10.84±1.07^{c}
3	8.82±0.14^{b}	15.84±0.39^{c}	23.26±0.53^{b}	25.16±1.22^{c}	13.32±1.14^{b}
4	10.20±0.26^{a}	16.40±0.42^{bc}	24.58±0.78^{ab}	28.80±0.76^{b}	14.44±0.61^{b}
5	11.20±0.40^{a}	17.52±0.32^{ab}	26.08±0.33^{a}	31.86±0.38^{a}	16.90±0.67^{a}

Different small superscript letters indicate statistically significant differences ($p < 0.05$) in the same treatment groups within a column.

3.1.2. Effects of submergence depth on stem number of Eleocharis ochrostachys

After harvesting, the numbers of plants per pot in each treatment were counted. Treatment 5 showed the highest density with 225.20 plants/pot on average, and significantly higher than those of other treatments. The numbers of plants per pot cultured at the depth of 15 cm and 10 cm were higher than plants growing in soil without submergence. The plant cultured in soil moisture of 45-50% resulted in the lowest plant numbers in a pot. These results provide valuable information which can be applied to control water level to stimulate the growth of *Eleocharis ochrostachys*. All results of plant numbers are shown in table 2.

3.1.3. Effects of submergence depth on the plant weight

The fresh and dry weight values of *Eleocharis ochrostachys*'s roots in treatment 5 were the highest value with 7.84 g and 1.90 g, respectively, which were significantly higher than those of other treatments (Table 3). Root weight of the plant growing at the depth of 10 cm and 15 cm was not statistically different. The fresh and dry weight of roots gradually decreased in the treatments with lower moisture contents and the lowest at treatment 1. The result showed that higher soil moisture and submergence water affected the growth.

Table 2. The number of plants/pots of *Eleocharis ochrostachys*

Treatments	Growth conditions	Number plants/pots
0	Control	173.80±4.22^{c}
1	45-50% soil moisture	80.40±4.79^{d}
2	80-100% soil moisture	168.80±4.64^{c}
3	5 cm submergence depth	204.60±1.60^{b}
4	10 cm submergence depth	208.00±2.07^{b}
5	15 cm submergence depth	225.20±4.06^{a}

Different small superscript letters indicate statistically significant differences ($p < 0.05$) in the same treatment groups within a column.

The fresh and dry weight of leaves in submergence depth of 10 cm and 15 cm were the highest values, while data obtained at treatment of 45-50% showed the lowest value. The fresh and dry weight values of *Eleocharis ochrostachys* in the treatment with a submergence depth of 15 cm were the highest values with 77.78 g and 12.09 g on average, respectively. The plant

growing in soil moisture had the lowest fresh and dry weight.

The Table 3 showed that the plant growing in soil moisture at a low level (45 – 50%) poorly accumulated biomass. The plants grew well at 80% - 100% soil moisture and submergence conditions. *Eleocharis ochrostachys* was cultured at high soil moisture and suitable submergence depth increased the plant height, root length and plant biomass. Moreover, the ability of water storage in the plant when it grew in the flood season helped the plant to survive in the dry season. At low soil moisture contents during the dry season, water storage plays an important role for the plant to survive. When it rains, *Eleocharis ochrostachys* will recover and grow.

Table 3. The fresh and dry weight of roots, leaves of *Eleocharis ochrostachys*

Treatments	Roots (g/plant)		Leaves (g/plant)		All of plant (g/plant)	
	Fresh weight	Dry weight	Fresh weight	Dry weight	Fresh weight	Dry weight
0	5.33±0.47^{bc}	1.19±0.11^{b}	11.49±1.19^{b}	1.84±0.22^{c}	60.27±3.33^{b}	10.39±0.23^{a}
1	2.97±0.15^{d}	0.32±0.03^{c}	4.21±0.55^{c}	0.62±0.05^{d}	14.98±1.82^{c}	2.73±0.29^{b}
2	4.70±0.21^{c}	1.01±0.13^{b}	8.57±0.46^{b}	1.46±0.12^{cd}	64.46±4.86^{b}	10.96±1.14^{a}
3	5.57±0.58b^{c}	1.17±0.27^{b}	11.74±1.69^{b}	2.01±0.27^{bc}	56.46±2.88^{b}	9.43±1.17^{a}
4	6.50±0.63^{ab}	1.07±0.13^{b}	17.58±0.79^{a}	3.46±0.61^{a}	62.17±2.01^{b}	12.16±1.46^{a}
5	7.48±0.56^{a}	1.90±0.41^{a}	16.88±0.86^{a}	2.90±0.24^{ab}	77.78±4.05^{a}	12.09±0.76^{a}

Different small superscript letters indicate statistically significant differences ($p < 0.05$) in the same treatment groups within a column.

3.2. Effects of submergence depth on the tuberization of *Eleocharis ochrostachys* and tuber anatomy

3.2.1. Effects of submergence depth on the formation of Eleocharis ochrostachys tuber

Table 4 shows that the plant growing in soil with moisture of 45%-50% did not produce any tubers. Lower biomass accumulated in plant tissue might be accounted for non-tuberization. In other treatments, the plant produced tubers and the tuber numbers were not statistically different among treatments. The tuber formation of the plant growing at the depth of 10 cm and 15 cm was 142 days after culturing, which was longer than that of other treatments. However, all tubers in treatments 3, 4, 5 were waterlogged (Figure 4D, E, F), which affected next generation because the tubers might not produce offsets. These results suggested that soil moisture of 80%-100% was the best condition for *Eleocharis ochrostachys.*

Table 4. Effects of submergence depth on the formation of *Eleocharis ochrostachys* tuber

Treatments	Soil moisture and submergence depth	Number of tubers/plant	Time to start produce tubers	Condition tubers
0	Control	13.40±1.08^{a}	135 days old	Normal
1	45-50% soil moisture	0.00±0.00^{b}	-	-
2	80-100% soil moisture	14.20±1.02^{a}	136 days old	Normal
3	5 cm submergence depth	14.00±0.71^{a}	139 days old	Waterlogged.
4	10 cm submergence depth	14.80±0.86^{a}	142 days old	Waterlogged
5	15 cm submergence depth	16.00±0.71^{a}	142 days old	Waterlogged

Different small superscript letters indicate statistically significant differences ($p < 0.05$) in the same treatment groups within a column.

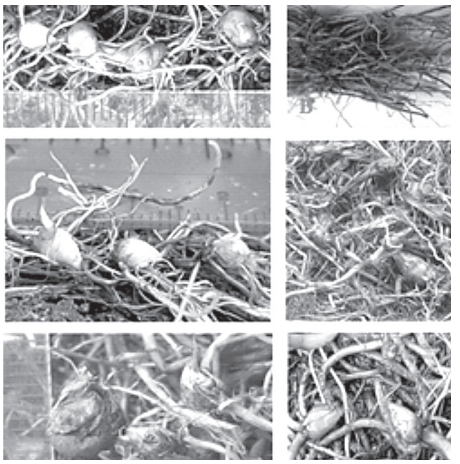

Figure 4. *Eleocharis ochrostachys* produced tubers in soil after 150 days at the treatments: (A) control, (B) 45%-50% soil moisture, (C) 80% - 100% soil moisture, (D) 5 cm submergence depth, (E) 10 cm submergence depth and (F) 20 cm submergence depth

3.2.2. Effects of submergence depth on the fresh and dry weight of Eleocharis ochrostachys tuber

The results showed that, the fresh and dry weight of *Eleocharis ochrostachys*'s tubers at treatment of 80-100% soil moisture was the highest with 1.19 g/tuber and 0.34 g/tuber, respectively, which were higher than that of the control.

3.2.3. Cross section of tubes

The tubers of plant cultured in soil with 80%-100% humidity used to investigate inside structure. The cross sectional anatomy of tubers clearly showed vascular bundles, axial parenchyma and ray parenchyma. Vascular bundle distributed dispersedly in the tubes similar to stems of monocotyledon plants (Figure 5).

Table 5. Effects of submergence depth on the fresh and dry weight of *Eleocharis ochrostachys* tubers

Treatments	Soil moisture and submergence depth	Fresh weight (g/tuber)	Dry weight (g/tuber)
0	Control	0.70±0.04	0.18±0.02
1	80-100% soil moisture	1.19±0.15	0.34±0.03

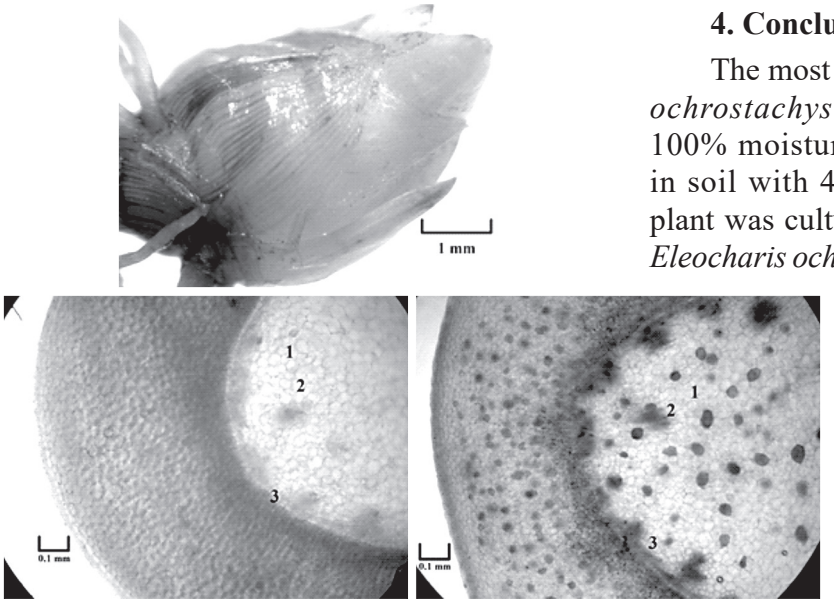

Figure 5. Tuber morphology of *Eleocharis ochrostachys* (A), cross sections of *Eleocharis ochrostachys*'s tuber without staining with methylene blue (B) and staining with methylene blue (C). Axial parenchyma (1), vascular bundles (2), and ray parenchyma (3) is shown in the picture

4. Conclusion

The most suitable condition for *Eleocharis ochrostachys* to grow is in soil with 80% - 100% moisture, while the plant poorly grew in soil with 45% - 50% moisture. When the plant was cultured in submergence conditions, *Eleocharis ochrostachys* also grew and produced tubers well; however, all tubers were waterlogged. This study provides valuable information which can be applied to control water level to stimulate the growth and production of tubers.

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