

Regrowth of Lowland Ecotype *Cyperus rotundus* L. in Response to Soil Depth, Shoot Clipping and Flooding Depth Interventions

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Abstract

Lowland ecotype Cyperus rotundus L. (LE-CYPRO) has tubers that can grow and survive under flooded conditions. However, information on the responses of its tubers to the combination of soil depth or shoot clipping with flooding is unavailable. Two experiments were conducted to determine the germination and regrowth of LE-CYPRO tubers in response to soil depth, shoot clipping and flooding depth interventions. Experiment 1 involved pre-sprouted (PS) and non-sprouted (NS) tubers subjected to three soil depths (0, 5 and 10 cm) and three flooding depths (0, 3 and 5 cm); Experiment 2 used PS tubers subjected to shoot clipping at periods of 10, 20 and 30 days after planting (DAP) under three water conditions (saturated, early flooding and late flooding). Irrespective of soil depths, PS and NS tubers had 100% germination and regrowth without flooding (0-cm flooding depth). When planted at the soil surface (0-cm soil depth) and flooded by 3- to 5-cm, only 50 and 40% of PS while 20% of NS tubers germinated. No germination was seen on tubers buried under 5- to 10-cm soil depths and flooded by 3- to 5-cm depths. Revived PS tubers buried and flooded for 100 days at 0-, 5- and 10-cm depths had 20 to 60% germinations at 3-cm flooding depth and 20 to 70% at 5-cm flooding depth; NS tubers had 40 to 50% germinations at 3-cm flooding depth and 40% at 5-cm flooding depth. Growth variables of the weed at shoot clipping periods of 10, 20 and 30 DAP were comparable to the control under saturated conditions. Growth variables were reduced by 68 to 97.3% and 21.7 to 100% when aided by early and late flooding, respectively. This information could be used in the formulation of effective and comprehensive weed management for LE-CYPRO.

Keywords: Cyperaceae, purple nutsedge, weed control, weed ecology

1. Introduction

Cyperus rotundus L. (CYPRO), commonly known as purple nutsedge, is a plant belonging to the family of Sedge. It is an upright perennial with triangular, smooth stems; linear and dark green leaves; reddish-brown spikelets when mature; achene, ovate to oblong seeds; white, fleshy and thin rhizomes. When young, its rhizomes are covered with scale leaves and when old, it is brown and fibrous. Its tubers are white and succulent when young; brown, fibrous and in-chained as they mature (Moody *et al.*, 2014).

CYPRO has been considered the world's worst weed because of its reproductive behavior in the soil, resistance to harsh environments and infestations on many primary crops around the globe (Holm *et al.*, 1977). When allowed to compete at 100 to 1,000 density m^{-2} under the upland conditions, CYPRO decreases the grain yield of rice by up to 53, 44 and 47% at nitrogen levels of 0, 600 and 120 $kg\ ha^{-1}$, respectively (Okafor and De Datta, 1976). On irrigated rice, the weed reduced the grain yield of transplanted and wet direct-seeded rice by 14 to 38% and 11 to 28%, respectively, when allowed to compete at initial tuber densities of 22 to 88 m^{-2} (Donayre *et al.*, 2022).

The tuber is the main organ of CYPRO for proliferation, spread and survival. This organ can germinate even in total darkness within four to six weeks (Betria and Montaldi, 1975). Although it can regrow at temperatures from 4 to 45 °C, it has better germinations within 20 to 40 °C by way of daily alternating and high-temperature pulses (Dor and Hershenhorn, 2013; Wallace *et al.*, 2013). From a single fragment, the tuber could multiply into more than a thousand pieces per square meter in just three months (Islam *et al.*, 2009).

Prior studies showed that CYPRO has numerous variants worldwide (Wills 1998; Tachie-Menson *et al.*, 2021). In the Philippines, it was reported to have a variant that can grow both in irrigated and rainfed-lowland ricefields (Baltazar *et al.*, 1999). The variant was identified as a lowland ecotype CYPRO (LE-CYPRO) that can grow and develop under flooded conditions (Casimero *et al.*, 1999). Morphological features in the greenhouse showed that LE-CYPRO was taller and had more off-shoots, heavier biomass, and larger and heavier tubers than its upland ecotype. Under field conditions, Donayre *et al.* (2015) also reported that LE-CYPRO was taller (81.4 cm) than the cultivated rice plants infesting both the transplanted and direct-seeded rice areas. Baltazar *et al.* (1997) theorized that the survival of lowland ecotype in

flooded conditions was a result of selection pressure in response to alternate wet-dry moisture regimes that were continuously repeated each year and over the years. Peña-Fronteras *et al.* (2009) and Fuentes *et al.* (2010), on the other hand, stated that the survival of the lowland ecotype under such conditions was due to morphological and physiological adaptive mechanisms.

Growth of CYPRO in response to soil burying, shoot clipping and flooding had been already reported in previous studies. For example, Baltazar *et al.* (1997) revealed that the lowland and upland types of CYPRO, buried at 1-cm depth, sprouted and emerged at all water levels (1, 3 and 5 cm). Meanwhile, Bangarwa *et al.* (2012) showed that shoot clipping-soil disturbance intervention at biweekly or weekly intervals effectively reduced the proliferation of CYPRO regardless of its initial tuber size. Despite the findings, the regrowth of LE-CYPRO tubers in response to soil burying or shoot clipping in combination with flooding was not considered. There is also a need to update information on the ecology of LE-CYPRO under flooded conditions. Thus, this study was conducted to determine the germination and regrowth of LE-CYPRO tubers in response to soil burying, shoot clipping and flooding interventions.

2. Methodology

2.1 Collection of Tubers

Tubers of LE-CYPRO were collected in January 2018 in paddy fields of Aliaga, Nueva Ecija, Philippines (15° 30' 59" N, 12° 49' 44" E). This area was chosen based on a study by Donayre *et al.* (2015). Purple nutsedge plants, at the maturity stage, were carefully drawn out from the soil using a trowel. Using a pair of scissors, tubers attached to their mother plants were separated and placed inside a plastic container. Mature tubers were taken into the Weed Science Laboratory of the Philippine Rice Research Institute (PhilRice), Science City of Muñoz, Nueva Ecija for washing and planting.

2.2 Growing of Tubers

In the laboratory, cleaned tubers were placed and sealed inside moistened and autoclavable plastics (30.48 x 20.32 cm LW). Next, the autoclavable plastics were then placed inside a temperature-controlled oven (DN83, Yamato

Scientific Co., Ltd., Japan) for subjecting all the tubers to 40 °C for 4 h. The tubers were then transferred into a 50-L capacity plastic box with a lid for a five-day incubation at 25 °C. Tubers that produced new shoots were selected and planted in plastic containers (30.48-cm diameter and 25.4-cm deep) filled with 5 kg of sterilized moist soil. All planted tubers were allowed to grow and multiply into new plants in the greenhouse. After 60 days, all matured mother plants were drawn out. All aged tubers were harvested, washed and placed in plastic containers.

2.3 Experimental Treatments, Design and Analysis

2.3.1 Experiment 1: Response to Soil and Flooding Depths

This experiment was conducted in two trials in PhilRice Central Experiment Station (CES) greenhouse from February to May and July to October 2018. A round, black plastic container (25.4-cm diameter and 22.86-cm deep), filled with 5 kg of sterilized soil (pH = 5.8, 1.9% organic matter, 113-ppm phosphorus and 200-ppm nitrogen) was used as the experimental unit to determine the regrowth of two types of LE-CYPRO tubers (pre-sprouted [PS] and non-sprouted [NS]) subjected to three soil depths (0, 5 and 10 cm) and three flooding depths (0, 3 and 5 cm). PS and NS tubers were used to simulate the effects of tillage before flooding under field conditions. PS tubers were prepared following the procedure of growing tubers described above. Tubers that produced new shoots (1-cm length) were selected for planting. NS tubers were prepared by keeping the newly harvested ones inside a dry plastic container for five days at 25 °C room temperature and 56% relative humidity. A recent study has shown that this procedure is the most effective and efficient method for preparing NS tubers in the laboratory (Donayre *et al.*, 2022). In each experimental unit, both tubers were simultaneously planted and allowed to grow at different soil and water depths for 100 days under greenhouse conditions. To maintain the desired flooding depths, the water level in each experimental unit was measured daily using a ruler. Water was added immediately as needed.

The experiment was arranged in randomized complete block design with five replications. After 100 days, the height of the weed in each container was recorded by measuring the distance between the basal stem and the tallest leaf of the plant. The number of off-shoots and tubers was manually counted in each container. Shoot and root biomasses were determined by drying the fresh biomasses in an oven for 48 h at 70 °C and weighing the dried biomasses using a digital weighing balance (FX-3000, A&D Co., Ltd., Japan). The data

collected were subjected to analysis of variance (ANOVA) using the Statistical Tool for Agricultural Research (STAR) (version 2.0.1.) (International Rice Research Institute, 2014). The square-root transformation method [$\sqrt{(X + 0.5)}$] was utilized whenever there were heterogeneous data detected (Gomez and Gomez, 1984). Since prior analysis showed no significant interactions between trial and treatments, all data from the two trials were pooled into one and re-subjected to ANOVA. All the means were compared using the LSD at a 5% level of significance. In addition, *t*-Test was also utilized to compare the growth variables between PS and NS tubers of LE-CYPRO.

2.3.2 Experiment 2: Response to Shoot Clipping and Flooding

This experiment was also conducted in two trials at PhilRice CES from July 2018 to February 2019. A cylindrical clay pot (15.24-cm wide by 5-cm deep) filled with 2 kg of sterilized moist soil, and planted with one PS tuber was the experimental unit used. Each tuber was subjected to shoot clipping done at three growing periods (10, 20 and 30 days after planting [DAP]) under three moisture conditions (saturated, early and late flooding). Early flooding (5-cm depth) was initiated at seven DAP and maintained until 60 DAP. Late flooding (5 cm) was initiated simultaneously with shoot clipping. The times of flooding were simulated to farmers' practices under field conditions. Meanwhile, other growing tubers were subjected to no-clipping treatment and served as the control for the entire experiment.

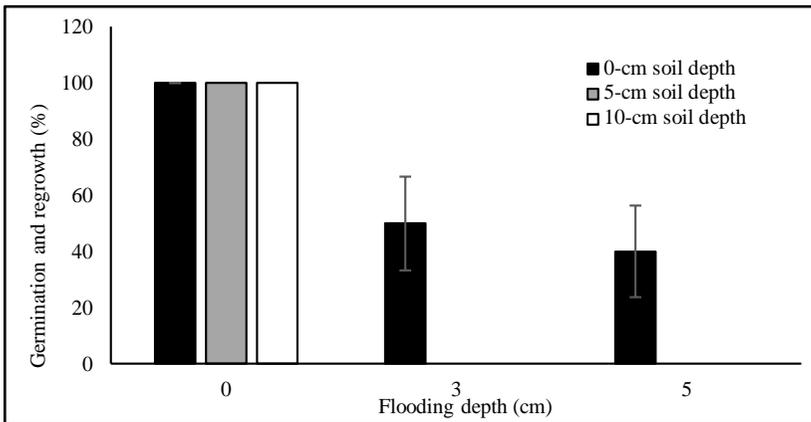
The experiment was arranged in completely randomized design with three replications. Data on height, numbers of off-shoots and tubers and shoot and root biomasses were recorded at 60 DAP. Prior analysis showed no significant interaction between trial and treatments. Thus, all data from the two trials were pooled together and subjected to one-way ANOVA. The square-root transformation method was again utilized whenever there were heterogeneous data detected. All treatment means were compared through LSD at a 5% level of significance.

3. Results and Discussion

3.1 Response to Soil Burying and Flooding

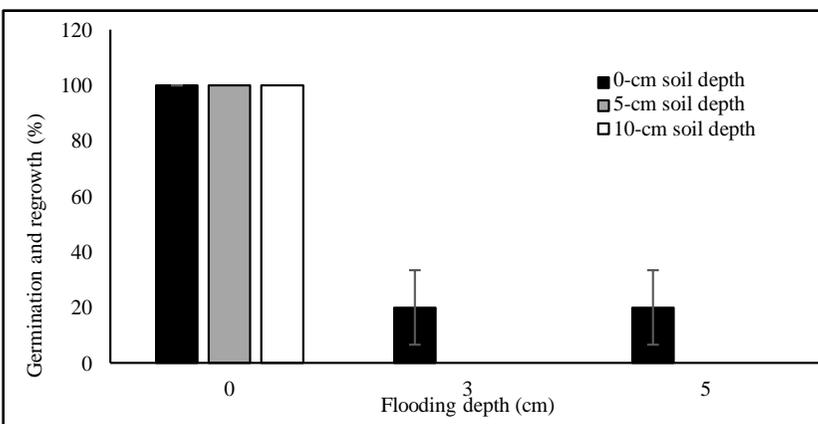
PS tubers, at different soil depths, completely germinated and regrew (100%) into new plants without flooding (0-cm flooding depth, saturated conditions)

(Figure 1). The tubers, however, had 50 and 40% germination and regrowth when planted at the soil surface (0-cm soil depth) and flooded by 3- and 5-cm depths. No regrowth was observed on PS tubers buried at 5 to 10 cm and flooded by 3- to 5-cm depths. NS tubers also had complete germination and regrowth (100%) into new plants without flooding regardless of soil depths (Figure 2). However, only 20% of the tubers germinated and regrew when planted in the soil surface at 5 to 10 cm and flooded by 3- to 5-cm depths. No regrowth occurred when buried at 5 to 10 cm and flooded by 3- and 5-cm depths.



Vertical lines are +SE of the means.

Figure 1. Germination and regrowth of pre-sprouted tubers of LE-CYPRO into new plants



Vertical lines are +SE of the means.

Figure 2. Germination and regrowth of non-sprouted tubers of LE-CYPRO into new plants

PS tubers planted at the soil surface without flooding produced higher means of height, numbers of off-shoots and new tubers, and heavier shoot and root biomasses (Table 1). The values, however, were not significantly different from the growth of PS tubers buried at 5 and 10 cm. Similar results were also observed in NS tubers although the means were higher at 5- and 10-cm soil depths. Growths of LE-CYPRO that came from PS tubers had higher means of height, number of off-shoots, dry weight of shoots and roots, and a new number of tubers than that from NS tubers. Statistical analysis, however, showed that growth and development of the weed from PS tubers was not significantly different from NS tubers in terms of height at 0, 5 and 10 cm; shoot biomass at 5 and 10 cm; and root biomass and the new number of tubers at 10-cm soil depths (Table 2). Growths of LE-CYPRO from PS tubers were significantly different from NS tubers in terms of the number of off-shoots at 0, 5, and 10 cm; shoot biomass at 0 cm; and root biomass and the new number of tubers at 0- and 5-cm soil depths.

Table 1. Growth and development of LE-CYPRO at 0-cm flooding depth (saturated conditions)

Soil depth (cm)	Height plant ⁻¹ (cm)	No. of off-shoots plant ⁻¹	Shoot biomass (g plant ⁻¹)	Root biomass (g plant ⁻¹)	No. of tubers plant ⁻¹
Pre-sprouted tubers					
0	115.3	29.7	40.7	16.7	38.1
5	126.7	25.5	38.2	12.4	32.1
10	138.6	25.0	31.9	12.3	27.6
LSD	ns	ns	ns	ns	ns
Non-sprouted tubers					
0	92.8	10.4	11.5	5.0	11.6
5	110.4	16.8	26.3	5.9	18.9
10	112.9	15.4	20.8	6.0	17.9
LSD	ns	ns	ns	ns	ns

Values in columns with ns at the bottom are not significantly different at 5% level of significance using LSD.

PS tubers that germinated and regrew at the soil surface and flooded by 3- and 5-cm water depths had higher means of height, number of off-shoots, number of new tubers, shoot biomass and root biomass than NS tubers (Table 3). Meanwhile, PS tubers that germinated and regrew at the soil surface and flooded by 3-cm water depth had higher means of height, number of off-shoots and new tubers, as well as shoot, and root biomass than the PS tubers at the soil surface but flooded by 5-cm water depth. Similar trend was also observed

in NS tubers that germinated and regrew at the soil surface and flooded by 3-cm depth.

Table 2. Summary of P-values for the comparison of pre-sprouted and non-sprouted tubers at 0-cm flooding depth (saturated conditions)

Soil depth (cm)	Height plant ⁻¹ (cm)	Pre-sprouted vs. non-sprouted tubers			
		No. of off-shoots plant ⁻¹	Shoot biomass (g plant ⁻¹)	Root biomass (g plant ⁻¹)	No. of tubers plant ⁻¹
0	.227 ^{ns}	.003 ^{**}	.008 [*]	.007 [*]	.004 ^{**}
5	.368 ^{ns}	.015 [*]	.094 ^{ns}	.014 [*]	.031 [*]
10	.189 ^{ns}	.044 [*]	.066 ^{ns}	.055 ^{ns}	.065 ^{ns}

P-values were calculated from transformed data using TTEST; * - $p < 0.5$, ** - $P < 0.005$, ns - not significant at a 5% level of significance.

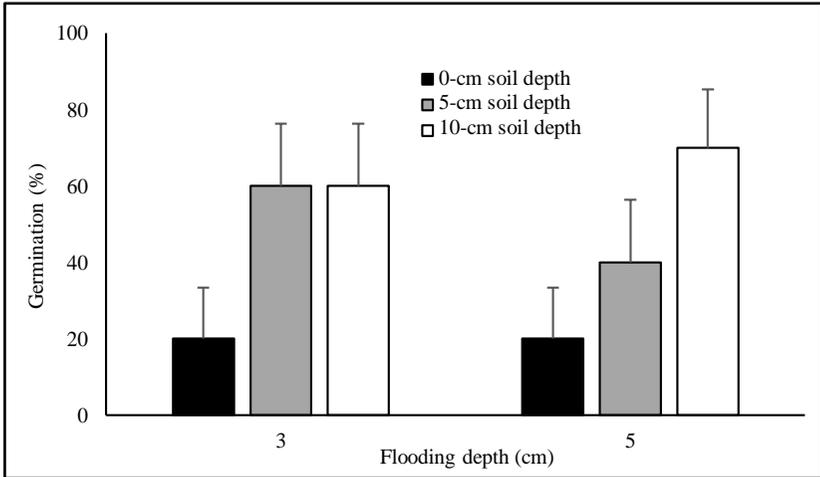
Table 3. Growth and development of LE-CYPRO planted at soil surface (0 cm)

Flooding depth (cm)	Height plant ⁻¹ (cm)	No. of off-shoots plant ⁻¹	Shoot biomass (g plant ⁻¹)	Root biomass (g plant ⁻¹)	No. of tubers plant ⁻¹
Pre-sprouted tubers					
3	132.9	54.3	51.7	85.9	40.8
5	117.2	47.8	28.2	28.2	53.3
Non-sprouted tubers					
3	30.3	3.7	4.1	0.9	3.2
5	25.3	3.3	2.3	3.8	1

PS tubers buried for 100 days at 0-, 5- and 10-cm soil depths had 20 to 60% germinations when flooded by 3-cm depth; 20 to 70% when flooded by 5-cm depth (Figure 3). NS tubers buried at the same span of days and depths had 40 to 50% germinations at 3-cm flooding depth; and 40% at 5-cm flooding depth (Figure 4).

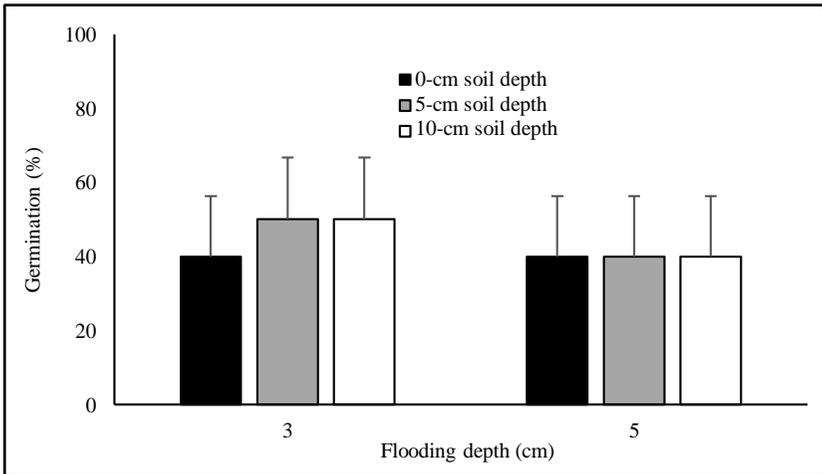
The discovery that LE-CYPRO regrows and propagates to new plants in the absence of flooding and regardless of soil depths presents two implications. First, starting tillage operations in moist or saturated conditions without considering it as a way of control will only intensify the infestation of the weed either before or after rice establishment. In this scenario, effective and economical weed control and better yields are difficult to achieve later. The second implication is an advantage when tillage is considered as a control measure against the weed, particularly with the use of the stale seedbed technique. To illustrate, weed control happens by allowing the tubers of LE-CYPRO to sprout under moist or saturated conditions after first harrowing

then later eradicated by repeated harrowings, or by applying non-selective herbicides. This type of control has been shown to be effective against CYPRO infesting rice-based cropping systems in the Philippines (Baltazar *et al.*, 2003; Islam *et al.*, 2009).



Vertical lines are +SE of the means.

Figure 3. Germination of pre-sprouted tubers after 100 days of exposure to flooding and soil depth



Vertical lines are +SE of the means.

Figure 4. Germination of non-sprouted tubers after 100 days of exposure to flooding and soil depth

Water management by flooding is one of the most recommended approaches for controlling many rice weeds under lowland conditions. Flooding the paddy field at the right time, duration and depth puts many weeds such as *Borreria ocymoides* (Burm.f.) DC., *Echinochloa colona* (L.) Link., *E. crus-galli* (L.) P. Beauv., *E. glabrescens* Munro ex Hook. f., *Heliotropium indicum* L., *Ischaemum rugosum* Salisb., *Leptochloa chinensis* L., weedy rice (*Oryza sativa* L.), *Cyperus difformis* L., *C. iria* L., *Fimbristylis miliacea* (L.) Vahl., *Ludwigia hyssopifolia* (G. Don) Exell, *L. octovalvis* (Jacq.) Raven and *Murdannia nodiflora* (L.) Brenan under stressful conditions due to oxygen deficiency and rapid biomass loss. Accordingly, these weeds had none to less seed and seedling germinations from 0.5- to 5-cm flooding depths (Rao and Moody, 1995; Moon *et al.*, 1999; Begum *et al.*, 2006; Chauhan and Jonhson, 2010; Estioko *et al.*, 2014; Ahmed *et al.*, 2015). In this experiment, however, it was found that flooding had less effect on the growth of LE-CYPRO particularly its tubers that were on the soil surface. The weed surpassed the highest flooding depth (5 cm) used despite the reduction of its germination by 20 to 50%.

Baltazar *et al.* (1997) also had similar conclusions when they examined the characteristics of CYPRO from the rice-onion cropping system. They found that the lowland and upland types of CYPRO, buried at 1-cm depth, sprouted and emerged in any water levels (1, 3 and 5 cm). The height and fresh weight of the lowland type did not decrease much with 5-cm flooding. The adaptive mechanism of LE-CYPRO under flooded conditions was studied by Peña-Fronteras *et al.* (2009). According to them, the resistance of the weed to flooding was due to its high carbohydrate content, amylase activity and ability to maintain high levels of soluble sugar in the tubers during germination and early growth. These activities were improved with the modulation of alcohol dehydrogenase and pyruvate decarboxylase to control the utilization of carbohydrate reserves and sustain substrate supply to avoid starvation and death of seedlings during long-term flooding. In a separate study, Fuentes *et al.* (2010) also expounded that the adaptation of the weed in flooded conditions was due to the following: a) bigger tubers that have high levels of carbohydrate and soluble sugar contents, larger stems and air spaces (aerenchyma) that allow oxygen diffusion to its submerged parts; b) ability to mobilize and utilize carbohydrate reserves for energy production through anaerobic respiration and to optimize the use of carbohydrate reserves through regulation of key enzymes like the alcohol dehydrogenase; and c) down-regulation of lactate dehydrogenase, probably to prevent lactate accumulation and cellular acidosis.

Plants use the escape and quiescence strategies to cope with flood stress (Mommer and Visser, 2005; Bailey-Serres and Voesenek, 2008; Striker, 2012; Van Veen *et al.*, 2014). In the escape strategy, plants do rapid elongation of shoots toward the water surface to restore contact of leaves for photosynthesis and re-aeration of underwater organs via aerenchyma tissues. In the quiescence strategy, plants reduce their energy by stopping some processes (e.g., shoot elongation) to conserve carbohydrates that can be used for regrowth later once the flooding conditions recede. The results of this experiment suggested that LE-CYPRO used both strategies. The weed, at the soil surface, elongated its shoots and surpassed the 5-cm flooding depth (escape). It also produced numerous off-shoots which could mean more leaves to capture sunlight for photosynthesis, as well as more stems, that contain air spaces for oxygen diffusion. Meanwhile, its tubers that did not germinate on the first day of planting but survived and re-sprouted after 100 days of burying and flooding suggested the quiescence strategy.

3.2 Response to Shoot Clipping and Flooding

Height and numbers of off-shoots and tubers of LE-CYPRO were not significantly affected by shoot clipping under saturated conditions (Table 4). Its growth at different clipping periods was comparable to control (unclipped treatment). Yet, its height was reduced by 19.2 to 36.5%, the number of off-shoots by 5 to 50% and the number of tubers by 40.9%. Its shoot and root biomasses were significantly reduced by 28.7 to 71% and 22 to 76%, respectively, when shoot clipping was done at 10, 20 and 30 DAP under saturated conditions. On the other hand, the height of LE-CYPRO was significantly reduced by 68 to 96.6%, the number of off-shoots by 89.1 to 95.7%, shoot biomass by 84.9 to 99.3%, root biomass by 96.2 to 96.7% and the number of tubers by 79.7 to 87.5% when shoot-clipping was aided by early flooding. The growth of the weed did not significantly differ in all clipping periods. Meanwhile, its growth response to shoot clipping aided by late flooding varied at different clipping periods. For example, the height, the number of off-shoots and shoot biomass were highly reduced (99, 96.2 and 100%) when clipping was done at 30 DAP; slightly reduced at 10 DAP (21.7, 50.0 and 61.4%) and 20 DAP (56.5, 73.1 and 81.4%). The root biomass that was reduced by 68 to 79.3% and the number of tubers by 62.1 to 79.3% were not significantly different among the three clipping periods.

Table 4. Growth of LE-CYPRO as influenced by shoot clipping at different periods

Clipping period (DAP)	Height plant ⁻¹ (cm)	No. of off-shoots plant ⁻¹	Shoot biomass (g plant ⁻¹)	Root biomass (g plant ⁻¹)	No. of tubers plant ⁻¹
Saturated conditions					
No clipping	86.1	6.7	3.4 (2.0) ^a	4.7 (2.2) ^a	7.3
10	69.6	7.2	2.4 (1.7) ^b	3.6 (1.9) ^{ab}	7.5
20	49.5	6.3	1.0 (1.2) ^c	1.1 (1.2) ^b	4.3
30	54.7	3.3	1.0 (1.2) ^c	1.6 (1.4) ^b	4.3
LSD	(ns)	(ns)	(0.22)	(0.71)	(ns)
Early flooding					
No clipping	102.5 (10.2) ^a	7.7 (2.7) ^a	6.4 (2.6) ^a	6.5 (2.4) ^a	10.7 (3.2) ^a
10	32.8 (4.1) ^b	0.8 (1.7) ^b	1.0 (1.2) ^b	0.2 (0.8) ^b	2.2 (1.5) ^b
20	0.8 (1.1) ^c	0.5 (0.9) ^b	0.2 (0.8) ^b	0.2 (0.8) ^b	1.5 (1.4) ^b
30	3.5 (1.4) ^{bc}	0.3 (0.9) ^b	0.0 (0.7) ^b	0.2 (0.8) ^b	1.3 (1.3) ^b
LSD	(2.9)	(0.73)	(0.49)	(0.73)	(0.72)
Late flooding					
No clipping	104.5 (10.2) ^a	4.3 (2.2) ^a	3.6 (2.0) ^a	1.4 (1.3) ^a	4.8 (2.3) ^a
10	81.8 (9.0) ^a	2.2 (1.6) ^b	1.4 (1.4) ^b	0.5 (1.0) ^b	1.8 (1.5) ^b
20	45.5 (6.0) ^b	1.2 (1.2) ^b	0.7 (1.1) ^c	0.3 (0.9) ^b	1.8 (1.5) ^b
30	1.0 (1.0) ^c	0.2 (0.8) ^c	0.0 (0.7) ^d	0.3 (0.9) ^b	1.0 (1.2) ^b
LSD	(2.3)	(0.42)	(0.27)	(0.28)	(0.35)

DAP – days after planting; values outside parenthesis original data while inside are transformed data; values in a column with the same letter are not significantly different at a 5% level of significance using LSD.

Previous studies have also reported the response of other variants of CYPRO to shoot clipping interventions. For example, Sierra (1974) revealed that regular shoot clipping of upland type CYPRO at five and 10 DAP for a month under saturated conditions, respectively, reduced the number of off-shoots by 93.2 and 40.9%, number of tubers by 98.1 and 64.5%, shoot biomass by 89.6 and 78% and tuber biomass by 96.4 and 88.5%. Marambe (1996) also found out that repeated shoot clippings at two and four weeks after emergence (WAE) reduced the regrowth ability of CYPRO under field capacity compared with shoot clipping done once either at two or four WAE. In addition, repeated shoot clipping reduced the leaf area, leaf biomass, mother tuber biomass, total number of tubers, endoamylase activity and starch content by 56, 69, 41, 63, 72 and 56%, respectively, than the control (no shoot clipping). In another study, Santos *et al.* (1997) also pointed out that tuber weight, shoot number and dry biomass are affected if the first shoot clipping is done six or 12 days after transplanting (DAT). In their study, they found that initial shoot clipping at six DAT resulted in total tuber depletion of 0.25 g after the fifth shoot clipping and 0.50 g after the seventh shoot clipping. Delaying the initial clipping until 12 DAT resulted only in depletion of 0.25 g after the sixth shoot

clipping. No tuber depletions (> 0.75 g tubers) were observed at any initial shoot clipping times. Bangarwa *et al.* (2012), on the other hand, had other findings stating that shoot clipping-soil disturbance intervention at biweekly or weekly intervals effectively reduced the proliferation of CYPRO regardless of initial tuber size.

Manual and mechanical weeding are two of the recommended control options against many weeds. Many farmers in Asian countries use these weed control methods because of their availability and suitability in the field (Heong and Escalada, 1997; Donayre *et al.*, 2014). Manual weeding is best done by cutting the shoots and stems, or by pulling the entire plant with the hands or sharpened tools. Mechanical weeding is best utilized by cutting, trampling and burying shoots and stems into the soil. The results of this experiment suggested that the growth of LE-CYPRO may not be effectively controlled by either manual or mechanical weeding if the only aim is to destroy the shoots without the aid of flooding. Although reduced growth of the weed can be expected, the consequences of implementing these techniques are not significant without flooding. Weed growth is suppressed by manual and mechanical weeding if supported later by flooding at the right time, depth and duration. Repeated initiation of these techniques with flooding could also help further reduce the growth of the weed as suggested by Marambe (1996) and Bangarwa *et al.* (2012).

4. Conclusion and Recommendation

This study concluded that LE-CYPRO, irrespective of soil depths, can germinate, regrow and multiply better without flooding, but it can also germinate and regrow even with the presence of flooding provided that its tubers are on soil surfaces. However, it cannot germinate when buried and flooded to at least 3-cm depth; but can germinate and regrow after prolonged exposure to deeper soil and flooding depths. Its growth was better reduced when shoot clipping was done under flooded conditions. The combination of soil burying and flooding, as well as shoot clipping and flooding controlled the growth of LE-CYPRO in this study. Despite the significant findings, however, it is recommended that further research must be conducted on the effects of the three interventions. For example, there is a need to look at the effects of different flooding durations on the growth of the weed and yield of rice; investigate the effects of shoot clipping or manual weeding on its growth

and yield of rice; and determine the effects of different tillage methods on regrowth and development of the weed under field conditions. The information from the proposed research could further improve the development of holistic weed management for LE-CYPRO.

5. Acknowledgement

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6. References

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