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ORIGINAL ARTICLE

Critical Growth Stage of Weed Removal in Cowpea under Water Stress

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This study aimed at investigating the critical growth stage of weed removal in cowpea in order to prevent unacceptable yield loss during water deficit. The study was carried out under a screenhouse to minimize extraneous factors such as pests and rodents using a complete randomized design (CRD) with five replicates. Seeds of cowpea and early germinating seedlings of *Tridax procumbens* and *Chromolaena odorata* were used for this study and were stressed for five days. Samplings were carried out at vegetative, flowering and fruiting stage. Growth indices such as relative growth rate, net assimilation rate, leaf area ratio and tissue water of contents of cowpea were determined from plant biomass. The photosynthetic pigments were determined spectrophotometrically at different stages of growth. Weed interference index such as relative crowding coefficient and land equivalents ratio were also determined from the plant biomass. The results indicated that the growth indices of cowpea interfered by *Chromolaena odorata* and *Tridax procumbens* were greatly reduced at flowering stage compared with the growth indices at vegetative and fruiting stage. Photosynthetic pigments such as chlororphyll a, chlorophyll b, carotenoids and total chlorophyll of cowpea as interfered by *Chromolaena odorata* and *Tridax procumbens* were also lowest at flowering stage. Interference index of cowpea were lowest at flowering stage as to the vegetative and fruiting stage of cowpea. There was significant difference in the growth indices, photosynthetic pigments accumulation and interference index of at the different growth stages of cowpea at p≤0.05. From the results obtained, flowering stage is the most critical period of growth stage to remove weed in cowpea under water stress in order to prevent an unacceptable yield loss.

Key words: Growth indices, Growth stages, Pigments, Water Stress, Weed, Yield

Globally, weeds constitute a major constraint to crop production. Problems caused by weed in cowpea production include reduction in crop yield, less efficient land use, higher cost of production due to insects and plant disease control, reduction in crop quality, water management problems, and less efficient utilization of labor (Singh *et al.,* 2017). Primarily, weed reduced crop yield through competition for water, soil nutrient, light, and space (Akobodun, 1987; Osipitan, 2017). Weeds may also reduce crop yield by releasing allelopathic compounds such as phenol, flavonoid, steroid, etc into the environment (Marinov-Serafimov, 2007) and by providing a conducive environment for pest infestation and virus (Fisichelli *et al*., 2014).

Critical stage of Weed Control has been developed to determine the most efficient period of growth stage to remove weed in order to prevent an unacceptable yield loss in the period of water deficit. Critical stage of weed control is recently being developed to manage herbicide resistant weeds in crops (Osipitan *et al*., 2016). However, the critical stage of weed control might vary with environmental conditions, level of weed infestations, composition of weed population, pre-crop emergence management practices, soil moisture and fertility level, cowpea cultivar and plant density (Adigun *et al*., 2014; Osipitan *et al.,* 2016).

Cowpea (*Vigna unguiculata* L. Walp) is a warm-season, annual, herbaceous legume belonging to Fabaceae family. Cowpea is one of the most drought-resistant food legumes (Dadson *et al*., 2005), sensitive and well adapted to drought, high temperatures and other biotic stresses compared with other crop species. Because of that it is primarily grown in drier regions of the world where is one of the most drought-resistant food legumes (Dadson *et al*., 2005). In cowpea cultivation, yield losses cause by weeds alone can be as high as 76% depending on the cowpea cultivar, environment and weed management practices. A timely weed removal at the critical stage under water deficit would help to prevent unacceptable yield loss. There are limited numbers of selective herbicides with wide spectrum for weed control in cowpea. Therefore, there is need to investigate the critical growth stage of weed infestation in cowpea for optimum yield under water deficit.

MATERIALS AND METHODS

Plants Materials and Collection: Seeds of cowpea (*Vigna unguiculata* L. (Walp)) IT99K-573-3-1 variety) and early germinating seedlings of *Tridax procumbens* and *Chromolaena odorata* were used in this study. Seeds of cowpea were collected from the Department of Crop Production and Protection, Obafemi Awolowo University, Ile-Ife. Earling germinating seedlings of *Tridax procumbens* and *Chromolaena odorata* were collected behind Department of Botany Building, Obafemi Awolowo University, Ile-Ife.

Raising of Seedlings: The seedlings were raised in the screenhouse of Department of Botany, Obafemi Awolowo University, Ile-Ife. Mean daily temperature of the screenhouse was taken with the aid of thermometer and the mean temperature was 29ºC. The intensity of light was also determined using a digital luxmeter LX 1000 and the light intensity was 5360 lux. Forty five planting bags (of about 20 cm in diameter and 10 cm in heights) were obtained having several perforations to allow proper drainage and prevent logging during the course of the experiment. The bags were filled with 10 kg collected soil. Seeds of cowpea were planted at a depth of about 3 cm below the soil. The seeds were sown at the rate of two seeds per with *Tridax procumbens* and *Chromolaena odorata*. The bags were then supplied with 200 mL of tap water in the morning and 200 mL of tap water in the evening until they were fully established, thereafter the seedlings were made to received 200 mL of tap water every five days.

Experimental Layout: The experiment was laid out in a completely randomized design (CRD) with five replicates.

Growth Indices: Samplings were carried out at vegetative, flowering and fruiting stage. Growth indices such as relative growth rate (RGR); net assimilation rate (NAR), leaf area ratio (LAR) and tissue water contents (TWC) were determined according to Hunts (1978) using leaf area and dry matter data collected at 4(t1) and 6 (t2) WAP as follows:

RGR = NAR X LAR;

NAR = (W2-W1) (lnA2- lnA1)/ (A2-A1) (t2-t1);

LAR = (A2-A1) (lnW2-lnW1)/ (W2-W1) (lnA2-lnA1);

TWC = Fresh weight-dry weight/fresh weight ×100 (Black and Pritchard, 2002)

W1and W2 is weight at t1and t2, while A1 and A2 are the respective leaf area. Leaf area was calculated from leaf length and width as follows:

LA= L ×W × CF. Where CF is the correction factor

CF=0.64 for cowpea (Awal *et al.,* 2004)

Weed Interference Indices: The interference of *Chromolaena odorata* and *Tridax procumbens* on cowpea were calculated using Land Equivalent Ratio (LER) and Relative Crowding Coefficients (RCC).

$$LER = \frac{Ya\ mix}{Ymono} + \frac{Yb\ mix}{Yb\ mon}$$

$$RCC = \frac{Pcontrol – Pmix}{x}$$

Where Y is the performance per unit area, Y = density x biomass, P = Planting density, x = number of species used in the experiment.

RESULTS

Critical stage of weed removal on the Growth Indices of cowpea interfered by *Chromolaena odorata* under Water Stress

The critical stage of weed removal of cowpea interfered by *Chrormolaena odorata* on the growth indices under water stress is shown on Table 1. Net Assimilation Rate (NAR) of cowpea interfered by *Chromolaena odorata* was highest at the fruiting stage, followed by at vegetative stage and lowest at flowering stage. There was significant difference in the Net Assimilation Rate (NAR) of cowpea interfered by *Chromolaena odorata* at different stages of growth at $p \leq 0.05$.

Tissue Water Contents (TWC) of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage and fruiting stage with the same value, and lowest at flowering stage. The Tissue Water Contents (TWC) of cowpea interfered by *Chromolaena odorata* at flowering stage was significantly different from the Water Contents (TWC) at vegetative stage and fruiting stage of growth at $p \leq 0.05$.

Leaf Area Ratio (LAR) of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the Tissue Water Contents (TWC) of cowpea interfered by *Chromolaena odorata* at different stages of growth at $p \leq 0.05$.

Relative Growth Rate (RGR) of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the Relative Growth Rate (RGR) of cowpea interfered by *Chromolaena odorata* at different stages of growth at $p \leq 0.05$.

Critical stage of weed removal on the Growth Indices of cowpea interfered by *Tridax procumbens* under Water Stress

The critical stage of weed removal of cowpea interfered by *Tridax procumbens* on the growth indices under water stress is shown on Table 1. Net Assimilation Rate (NAR) of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage and fruiting stage with the same value of NAR and lowest at flowering stage. The Net Assimilation Rate (NAR) of cowpea interfered by *Tridax procumbens* at flowering stage was significantly different from vegetative stage and and at fruiting stages at $p \leq 0.05$. There was no significant difference in the Net Assimilation Rate (NAR) of cowpea interfered by *Tridax procumbens* at the vegetative stage and fruiting stage at $p \leq 0.05$.

Tissue Water Contents (TWC) of cowpea interfered by *Tridax procumbens* at vegetative stage and fruiting stage with the same value, and lowest at flowering stage. The Tissue Water Contents (TWC) of cowpea interfered by *Tridax procumbense* at flowering stage was significantly different from the Water Contents (TWC) at vegetative stage and fruiting stage of growth at $p \leq 0.05$. There was no significant difference in the Net Tissue Water Contents (TWC) of cowpea interfered by *Tridax procumbens* at the vegetative stage and fruiting stage at $p \leq 0.05$.

Leaf Area Ratio (LAR) of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the Tissue Water Contents (TWC) of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Relative Growth Rate (RGR) of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the Relative Growth Rate (RGR) of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Critical stage of weed removal on the Photosyntheic Pigments Accumulation of cowpea interfered by *Chromolaena odorata* under Water Stress

The critical stage of weed removal of cowpea interfered by *Chromolaena odorata* on the photosynthetic pigments accumulation under water stress is shown on Table 1. Chlorophyll *a* accumulation of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the chlorophyll *a* accumulation of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Chorophyll b accumulation of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the chlorophyll b accumulation of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Carotenoids content of cowpea interfered by *Chromolaena odorata* was highest at the fruiting stage stage, followed by at flowering stage and lowest at vegtative stage. There was significant difference in the carotenoids content of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Total chorophyll content of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the chlorophyll b accumulation of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Critical stage of weed removal on the Photosynthetic Pigments Accumulation of cowpea interfered by *Tridax procumbens*

The critical stage of weed removal of cowpea interfered by *Tridax procumbens* on the photosynthetic pigments accumulation under water stress is shown on Table 1. Chlorophyll a accumulation of cowpea interfered by *Tridax procumbens* was highest at the fruiting stage, followed by at vegetative stage and lowest at flowering stage. There was significant difference in the chlorophyll a accumulation of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Chorophyll b accumulation of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the chlorophyll b accumulation of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Carotenoids content of cowpea interfered by *Tridax procumbens* was highest at the fruiting stage stage, followed by at vegetative stage and lowest at flowering stage. There was significant difference in the carotenoids content of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Total chorophyll content of cowpea interfered by Tridax procumbens was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. There was significant difference in the chlorophyll b accumulation of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Weed Interference Index of Cowpea under Water Stress

The interference index of cowpea interfered by *Chrormolaena odorata* and *Tridax procumbens* is shown on Table 1. Relative Crowding Coefficients (RCC) of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. The Relative Crowding Coefficients of cowpea at the different growth stages was greater than one. There was significant difference in the Relative Crowding Coefficients of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Land Equivalents Ratio (LER) of cowpea interfered by *Chromolaena odorata* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering tage. The Land Equivalents Ratio (LER) of cowpea at the different growth stages was greater than one, except at flowering stage which equals one. There was significant difference in the Land Equivalents Ratio (LER) of cowpea interfered by *Chromolaena odorata* at different stages of growth at p≤0.05.

Relative Crowding Coefficients (RCC) of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowering stage. The Relative Crowding Coefficients of cowpea at the different growth stages was greater than one. There was significant difference in the Relative Crowding Coefficients of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Land Equivalents Ratio (LER) of cowpea interfered by *Tridax procumbens* was highest at the vegetative stage, followed by at fruiting stage and lowest at flowerig stage. The Land Equivalents Ratio (LER) of cowpea at the different growth stages was greater than one, except at flowering stage which is less than one. There was significant difference in the Land Equivalents Ratio (LER) of cowpea interfered by *Tridax procumbens* at different stages of growth at p≤0.05.

Table 1. Critical stage of weed control on the Growth Indices of cowpea interfered by *Chromolaena odorata* under Water Stress

	Growth Stages			
Growth Indices	Vegetative stage	Flowering Stage	Fruiting stage	P-Value$_{0.05}$
NAR	0.07±0.01b	0.03±0.00c	0.15±0.00a	<.0001
TWC	0.93±0.49a	0.89±0.43b	0.93±0.49a	<.0001
LAR	2.80±0.004a	1.70±0.00c	1.80±0.003b	<.0001
RGR	0.26±0.006a	0.054±0.006c	0.065±0.33b	<.0001

Means with the same letter along the same rows are not significantly differet at p≤0.05. NAR=Net assimilation rate, TWC=Tissue water content, LAR=Leaf area ratio, RGR=Relative growth rate.

Table 2. Critical stage of weed control on the Growth Indices of cowpea interfered by *Tridax procumbens* under Water Stress

	Growth Stages			
Growth Indices	Vegetative stage	Flowering Stage	Fruiting stage	P-Value$_{0.05}$
NAR	0.10±0.002a	0.06±0.02b	0.10±0.01a	<.0001
TWC	0.93±0.04a	0.92±0.06b	0.93±0.02a	<.0001
LAR	4.96±0.06a	1.24±0.05c	3.10±0.11b	<.0001
RGR	0.49±0.12a	0.13±0.00c	0.18±0.00b	<.0001

Means with the same letter along the same rows are not significantly differet at p≤0.05 NAR=Net assimilation rate, TWC=Tissue water content, LAR=Leaf area ratio, RGR=Relative growth rate.

Table 3. Critical stage of weed control on the Photosyntheic Pigments Accumulation of cowpea interfered by *Chromolaena odorata* under Water Stress

	Growth Stages			
Photosynthetic Pigments	Vegetative stage	Flowering stage	Fruiting Stage	P-Value$_{0.05}$
Chlorophyll a	8.92±0.012a	8.87±0.06b	6.79±0.05c	<.0001
Chlorophyll b	12.95±0.03a	11.44±0.01b	821±0.04c	<.0001
Carotenoids	2.05±0.03c	2.95±0.04a	2.59±0.00b	<.0001

Total Chlorophyll	24.50±0.31a	22.76±0.04b	16.79±0.06c	<.0001

Means with the same letter along the same rows are not significantly different at p≤0.05

Table 4. Critical stage of weed control on the Photosyntheic Pigments Accumulation of cowpea interfered by *Tridax procumbens* under Water Stress

	Growth Stages			
Photosynthetic Pigments	Vegetative stage	Flowering Stage	Fruiting stage	P-Value$_{0.05}$
Chlorophyll a	7.91±0.04b	6.74±0.01c	8.28±0.08a	<.0001
Chlorophyll b	13.67±0.01a	6.89±0.03c	11.57±0.05b	<.0001
Carotenoids	2.91±0.05b	2.86±0.01c	3.88±0.31a	<.0001
Total Chlorophyll	24.18±0.39a	15.21±0.00c	22.50±0.00b	<.0001

Means with the same letter along the same rows are not significantly differet at p≤0.05

Table 5. Weed Interference Index of Cowpea under Water Stress

		Growth Stages			
Weed Species	Interference Index	Vegetative stage	Flowering Stage	Fruiting Stage	P-Value$_{0.05}$
Chromo	RCC	11.20±12a	3.30±0.19c	10.20±0.00b	<.0001
	LER	1.43±0.00a	1.00±0.00c	1.33±0.02b	<.0001
Tridax	RCC	3.87±0.11a	2.45±0.03c	3.45±0.03b	<.0001
	LER	1.44±0.02a	0.71±0.006c	1.03±0.02b	<.0001

Means with the same letter along the same rows are not significantly different at p≤0.05, Chromo- *Chromolaena odorata,* Tridax- *Tridax procumbens*, RCC-Relative crowding coefficients, LER- Land equivalents ratio

DISCUSSION

The lowest plant growth indices observed in cowpea interfered by *Chromolaena odorata* and *Tridax procumbens* at flowering stage under water stress may be attributed to intense competition for growth resources such as solar energy, soil nutrients and water condition among the components plants. This showed that weeds (*Chromolaena odorata* and *Tridax procumbens*) influences plant allocation patterns and architecture mostly at flowering stage (Andrade *et al.,* 2005).

The lowest value of relative crowding coefficients (RCC) and land equivalents ratio (LER) of cowpea *Chromolaena odorata* and *Tridax procumbens* observed at flowering stage of growth under water deficit indicated that cowpea exhibits weak interspecific competitiveness in the presence of its competitor at this stage of growth.

The decreased contents of chlorophyll a, chlorophyll b, total chlorophyll and carotenoids at flowering stage may be probably due to the result of mineral deficiencies. Certain minerals such as magnesium, nitrogen and iron are important in photosynthetic pigments formation (Olowolaju and Adelusi, 2017). When these minerals becomes limiting, pigments formation becomes inhibited. This may be attributed to the fact that other factors at flowering stage become limiting at the expense of competition of shared use of the available resources.

CONCLUSION

From the results obtained, all the parameters measured were greatly reduced at flowering stage of weed interference under water deficit. Therefore, in order to realized acceptable yield in cowpea, flowering stage is the most efficient period of growth stage to remove weed in cowpea under water deficit in order to prevent an unacceptable yield loss

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