Effect of foliar spray of humic acid on root growth, yield components and quality in Redgram *(Cajanus cajan)*

Abstract

The present study examined the effect of foliar spray of humic acid on root Growth, yield components and quality in Redgram (*Cajanus cajan*).The study was conducted during Kharif 2015-16 at Main Agricultural Research Station, UAS, Raichur by using randomized block design. The effectiveness of humic acid was studied with different levels as T_1 - Humic acid liquid 15% @ 1.0 ml/l of water, T_2 - Humic acid liquid 15% @ 1.5 ml/l of water, T_3 - Humic acid liquid 15% @ 2.5 ml/l of water, T_4 - Humic acid liquid 15% @ 4.0 ml/l of water, T_5 - Planofix 4.5 % @ 20ppm and T_6 - as a control. Redgram root growth including root length, shoot length, root dry weight , shoot dry weight , leaf area, dry matter production, flower drops, minerals content (quality), and yield components were measured at 60,90 DAS and at harvest, respectively. Significant differences ($p<0.05$) were observed for all the above mentioned parameters across the humic acid levels. Based on this study, the foliar application T_4-Humic acid liquid 15% @ 4.0 ml/l of water might be recommended to improve growth physiology, quality and yield components of Redgram in similar environmental conditions. Further, research is required in diverse plant environments to determine economically feasible application levels of Humic acid while comparing it with other plant growth regulators sources.

Keywords: Humic acid, Growth Physiology, flower drop, mineral content and yield

Introduction

Pulses as one of the most important plant resources are full of protein and after grains are considered as the second most important source of food for human being. The rate of protein in legumes grains is twice or three times more than that of grain cereals and 10 to 20 times more than that of tuberous crops like potatoes (Kouchaki and Banayan, 2007). Pigeonpea (*Cajanus cajan*) is cultivated on an about 4.83 million hectares in the world with annual production of 2.98 million tonnes and a productivity of 700 kg ha^{-1}. It is an important pulse crop in India, which accounts for an about 90 per cent (3.88 m ha) of the total world area and production (2.92 m tons) with a productivity of 860 kg ha^{-1}. In Karnataka, pigeonpea occupies second place in area (0.78 m ha) and production (0.38 m tons) with a productivity of 760 kg per ha (Anon., 2015). Gulbarga called as dalbowl, is a very potential district in the Northern Karnataka state for extensive cultivation of pigeonpea. Pigeonpea is intrinsically perennial, but it is generally grown as an annual crop. The initial vegetative growth take place during the monsoon and floral initiation to end of grain filling phase occurs in winter season; which is generally dry and the pigeonpea crop depend for their continued development on stored moisture. As a result, redgram consumption in most of the low income countries like India has increased from 22% - 66%. Despite all these achievements, yields for the rainfed area are generally low and variable due to sparse, erratic rainfall and marginal soils.

Humic acids (HAs) are the main fractions of humic substances (HS) and the most active components of soil and compost organic matter. They exert indirect and direct effects on plants (Chen *et al.* 2004), and this action of HS is dose dependent and high concentrations of HS are inhibitory for nutrient accumulation (Chen and Aviad 1990). Some plant hormone-like substances seem also to be present in the HS, thus exerting a possible stimulating effect on growth (Pizzeghello *et al.* 2002). Humic acid is a commercial product contains many elements which improve the soil fertility and increasing the availability of nutrient elements and consequently affected plant growth and yield. Humic acid particularly is used to remove or decrease the negative effects of chemical fertilizers and some chemicals from the soil. The major effect of humic acid on plant growth has long been reported. There is basic agreement on the benefits of humus, but there is quite a controversy on the benefit of application of applied humate (the deposits containing the humic acids). Humic acid is extracted from different sources such as soil, humus, peat, oxidized lignite, and coal. Humic acid can directly have positive effects on plant growth and increases the growth of shoots and roots, absorption of nitrogen, potassium, calcium, magnesium, and phosphorus by plant. Humic acid is consistent with nature and is not dangerous for the plant and environment (Haghighi *et al.*, 2011 in horse been). Abdel Mawgoud *et al.,* (2007) states that humic acid increases plant growth through chelating different nutrients to overcome the lack of nutrients, and has useful effects on growth increase, production, and quality improvement of agricultural products due to having hormonal compounds. Among legume family plants, humic acid foliar spray has remarkable effects on vegetative growth of plant and increases photosynthetic activity and leaf area index. Ghorbani *et al.*(2010) in corn. The results of the research on wheat showed that the interactive effect of different concentrations of humic acid at three foliar spraying times on leaf area was significant

(Masciandaro *et al*., (2002)). Sharif *et al.* (2002) stated that humic acid could sustain photosynthetic tissues and thus total dry weight would increase. To manage agriculture production in unfavorable soil conditions by enriching their organic matter, various options are found in literature for example, crop rotation, green manures, residue and humic acid application (Delfine *et al*., 2005 in wheat ; Selim *et al*., 2009 in potato ;Johnson *et al*., 2012 in sweet corn; Ludibeth *et al*., 2012). Keeping in consideration the magnitude for shipment and universal availability humic acid seems a choice amongst the various options to improve the yield and quality of crop plant with foliar application. Many studies have demonstrated the foliar application importance of humic acid in agriculture for example Nardi *et al*.,(2002) in higher plant, Buyukkeskin & Akinci (2011) in broadbean , Çelik *et al*.,(2011) in maize, Tahir *et al*., (2011)in wheat and Humintech (2012) have reported beneficial effects of foliar application of humic acid substances on plant growth physiology, mineral nutrition, seed germination, seedling growth, root initiation, root growth, shoot development, yield and the uptake of macro-and microelements. Masciandaro *et al*., (2002) have indicated that humic substances might counteract abiotic stress conditions e.g., un-favorable temperature, pH, and salinity enhancing the uptake of nutrients and reducing the uptake of some toxic elements. However, Hartz &Bottoms (2010) have reported that humic acid neither improves crop nutrient uptake nor productivity in vegetable crops. Ayuso *et al.* (1996) in barely investigated the effect of humic substances originating from various organic materials on the growth and nutrient absorption of barley during hydroponic cultivation. They found that doses representing less than 10 mg L^{-1} carbon favored plant growth, while higher doses sometimes inhibited it. The absorption of macronutrients was significantly affected by the addition of humic substances but differed for each nutrient. Tufencki *et al.(*2006) applied increasing doses of humic acids, varying from 500 to 2000 mg per kg, at different times before lettuce seedling transplantation, to experimental soil placed in pots. Especially early application of humic acids had positive impacts on the plant growth and nutrient contents of lettuce plants with a short growing period. Also, no comprehensive study is available on optimization of humic acid for any crop especially for redgram flower drop management and enhancing the productivity and production. The present study for that reason explore full potential of foliar application of humic acid on growth physiology, mineral content and yield component redgram seed production-with optimization of foliar application levels of Humic acid .The research findings of this study are based on the key parameters necessary for evaluation of redgram growth physiology, quality and yield, and hoped to be valuable information for farmers and researchers.

Materials and methods

A field experiment was conducted to find out effect of foliar spray of humic acid on root Growth physiology, yield components and quality in Redgram during Kharif 2015-16 at Main Agricultural Research Station, UAS, Raichur. The data of prevailing climatic parameters were collected from research centre meteorological station which is located within one kilometer from experimental area. The crop was sown on 17th July, 2015 by manual line sowing 90 cm row spacing and 30 cm between plants. The redgram variety selected for the study was TS-3R released by University of Agricultural Sciences, Raichur. It is a high yielding variety. The humic acid was sprayed three times at 60-90 days after sowing, at the time of flowering and pod development stage. The concentration of Humic acid for each treatment was sprayed at different levels as T_1 - Humic acid liquid 15% @ 1.0 ml/l of water, T_2 - Humic acid liquid 15% @ 1.5 ml/l of water, T_3 -Humic acid liquid 15% @ 2.5 ml/l of water, T_4 - Humic acid liquid 15% @ 4.0 ml/l of water, T_5 - Planofix 4.5 % @ 20ppm and T_6 - control. The observations on various root growth physiological parameters viz., root length was measured by meter scale . While the fresh weight and dry weight of roots, the root sample was placed in the oven for 48 hours at 75°C and then it was weight by a digital scale with accuracy of 0.01 g balance. Root volume was measured by the water displacement method. The dried plant seeds material was ground and digested with a diacid 2:1mixture of nitric acid (HNO_3) and perchloric acid ($HClO_4$) to determine the various macro and micro nutrient content from redgram seeds with Atomic absorption spectroscopy for Fe, Cu, Zn and flame photometry for K.

Result and Discussions

Leaf area (cm^2 per plant) and total dry matter production (g/plant)

The data pertains to leaf area total dry weight trend (Fig. 1 & 2) shows that at different growth stages, total dry weight of plant has increased gradually and all the treatments differ significantly to each other. As it is observed, total dry weight of redgram plant in treatment with 4.0 ml /l of water humic acid is more than that of other treatments. This shows that as humic acid concentration increases, total dry weight also increased. The results of this study are conformity with the findings of [Haghighi, 2013] in Horsegram , [Turkmen,2004] in tomato stated that humic acid could improved the activity of photosynthetic tissues in crop plant and thus leaf area & total dry weight would increases at all the stages . All levels of humic acid 98 days after sowing maximized leaf area & dry matter accumulation and then they showed a descending trend. The plantsown its accumulated dry matter into reproductive organs, and the loss of leaves led to decrease of dry matter accumulation. The highest descending trend was observed in control treatments due to lack of absorption of humic acid by the leaves . [Majedi,2006] showed that application of humic acid foliar sprays had a key role in increasing the yield. The results were consistent with the findings of [Xiumei and Yaping,2003] in potato and [Ziyaeyan,2004] in maize and [Azizi M,1998] in soybean

Root growth parameters

The data on root development at harvest presented in Table 1 indicated significant differences between the treatments. The all the treatments differed significantly in root length(cm) , root fresh weight(mg) , root dry weight (mg) and T_4 recorded significantly higher root length(cm) , root fresh weight(mg) , root dry weight (mg) **(**24.25, 27.12, 9.93, respectively) as compared to all other treatments. While significantly lower root length (cm), root fresh weight (mg), root dry weight (mg) was recorded in control (10.92, 19.7, 4.60, respectively), but it was onpar with T_1. These findings are good agreement with the growth promoting results of humic substances those reported for a wide number of plant species (Visser, 1986; Chen and Aviad, 1990). The good results of the potato field trial correspond with the conclusions of a study in 2005 from the Potato Research Institute in Finland (Kuisma, 2005). In this study Humifirst also had a positive effect on total tuber yield (+ 17% compared to control) and marketable yield (+ 24% compared to control). The best response was obtained when Humifirst was applied to the soil just before seed bed tillage, which is similar to our experiment, compared to later application on planting and hilling. Other positive effects of Humifirst on potato yield were found at Gembloux (+ 25%) and Geer (+ 11%) both located in the southern part of Belgium (Anonymous, 2002). Eyheraguibel (2004) detected that humic substances accelerated both vegetative and reproductive growth of maize plants and thus stimulated optimal production of plant biomass (shoot and cobs). Root growth was stimulated as well with more fine lateral and secondary roots in the humic substances treatments. In line with these results While , Sharif *et al.* (2002) also reported that sprayed 50 to 300 mg per kg humic acids on the soil in a pot experiment with maize and found that the addition of 50 and 100 $mgkg^{-1}$ caused a significant increase of 20 and 23% in shoot and 39 and 32% in root dry weight. The incorporation of humic substances in the soil stimulated root mass of creeping bent grass with 45% in the 0 to 10 cm depth and with 38% in the 10 to 20 cm depth (Chen *et al*., 2004). Above ground biomass was only slightly promoted and was attributed by the authors to a sufficient nutrient supply.

The data on flower drops (%) at flowering development presented in Table 3 indicated significant differences between the treatments. The all the treatments differed significantly in flower drops and T_4 recorded significantly lower flower drops **(**45.8 %) as compared to all other treatments. While significantly higher flower drops was recorded in control (65.4%). These results are good agreement with the findings of Haghighi *et al.*, 2011 in horse been; Hossain *et al*., 2007 in groundnut and Turkmen *et al.,* (2005) in tomato. Similalarly, Albairak and Camas (2005) stated that that humic acid increases plant growth through chelating different nutrients to overcome the lack of nutrients, and has useful effects on growth increase, production, and quality improvement of agricultural products due to having hormonal compounds. Ghorbani *et al.,* (2010) stated that in legume family plants, humic acid foliar spray has remarkable effects on vegetative growth of plant (plant height, number of branches) and increases photosynthetic activity and leaf area index. Haghigh *et al.*, (2011) in horse been investigated the effect of humic acid on growth parameters of cowpea and found that humic acid would increase leaf area , total dry matter and leaf area index. Abdel- Al., (2005) and Erik *et al*., (2000), on onion plant and Hafez, (2003), on squash reported that humic acid applications led to a significant increase in soil organic matter which is improves plant growth and crop production. Tahir *et al*., (2011) with study effects of mineral fertilizers and humic substances on growth and yield of cowpea were reported that, combination of chemical fertilizer with application of humic substances improve growth and yield of cowpea.

Seed quality parameters

The data on seed quality i.e. macro-nutrient (%) and micro-nutrients content (ppm) of redgram seeds at harvest presented in Table 2 indicated significant differences between the treatments. The all the treatments differed significantly in Macro-nutrient (%) and micro-nutrients content (ppm) and T_4 recorded significantly higher Macro-nutrient (4.92 ,0.952, 3.80 %, N P K, respectively) and micro-nutrients content (2.98, 10.60, 5.59 ppm Cu, Zn, Fe, respectively) as compared to all other treatments. While significantly lower macro-nutrient (%) and micro-nutrients content (ppm) was recorded in control (1.87, 0.259, 1.13 %, NPK, respectively) and micro-nutrients content (1.50, 6.65, 3.50 ppm Cu, Zn, Fe, respectively) , but it was onpar with T_1. Our results are supported by Delfine *et al*. (2005), Morard *et al*. (2011) who have reported that humic substances provoked a better efficiency of plant water uptake and improved the mineral nutrition and grain protein content. Similalarly, our results are further supported by Turan *et al*. (2011) that salinity had negative impacts on the dry weight and the N, P, K,Ca, Mg, Fe, Cu, Zn and Mn uptake of maize plants, the humic acid mitigate salinity and increase dry weight and nutrients composition of plants. Similar to this the foliar application of humic acid affected the uptake of P which was statistically significant in the uptake of Na, K, Cu, and Zn. However, its amounts were not found statistically significant with other nutrients. The highest dry weight and nutrients uptake were obtained with 0.1% dose of humic acid. Nevertheless, the dry weight and nutrients uptake were decreased at 0.2% dose of humic acid,but the amounts except for Fe, Cu, and Mn were found higher than in the control (Hussein and Hassan, 2011). Similarly, Fernandez-Escobar et al. (1996) studied the effect of foliar application of humic acid extracts to young olive plants in greenhouse and infield experiments. Under field conditions, shoot growth and accumulation of potassium (K), boron (B), magnesium (Mg), calcium (Ca), and iron (Fe) in leaves was promoted. The effects of humic substances on plant production and nutrient absorbance generally depends on their origin, type and concentration and on the species and variety of the plant treated (Visser, 1986; Chen and Aviad, 1990).

Yield and yield components

The data on pods and yield of redgram was significantly influenced by Humic acid Liquid 15% application (Table 3). Significantly higher redgram pod yield was recorded with application of 4.0ml/L of 15% of Humic (2,154 kg/ha) followed by application Humic acid liquid 15% @ 2.5 ml/l of water (1323.02 kg/ha). However, lower pod yield was recorded in untreated control (1073.88 kg/ha) extent of reduction in pod yield was 32 % University check Planofix 4.5 % @ 20ppm (1288.66 kg/ha). Similarly higher number of pods and pods weight were also recorded in T_4 (126.88 and 75.45, respectively) while lower number of pods and pods weight were recorded in control (89.38 and 44.88, respectively). Similar results were obtained by Raj and Rao, 1996; Hafez and. Magda, 2003; Hossain *et al*., 2007, Amiri and Gohari, 2010 and Bozorgi *et al*., 2011 in peanut. Khan *et al.*, 2009; Hartz and Bottoms, 2010; Abdzad Gohari and Noorhosseini Niyaki,2010 in peanut.

Conclusions

Application of humic acid substances at the start of the growing season induced an overall positive effect on growth, root development, seed quality and yield of redgram in the field. It was also observed that the foliar application of all the doses of Humic acid Liquid 15% on redgram significantly increased the root length per plant , root dry weight per plant root volume, number of pods/plant and yield/ha. Further, there was a significant reduction in the flower and pod drop compared to control. The seeds/plant analysis for quality aspects indicated significantly higher content of macro (N, P.K) and micronutrients (Cu, Mn, Zn, and Fe) with the foliar application of Humic acid Liquid 15% @ 4.0ml/L over the control. The application of Humic acid Liquid 15% @ 4.0ml/L at flower bud formation stage may reduce flower drops in redgram compared to control. Increment in Humic acid concentration increased root growth and quality of redgram in the present study. Based on the present study findings Humic acid Liquid 15% @ 4.0ml/l foliar application to may be recommended. Further research is required in diverse plant environments to determine economically feasible application level of Humic acid while comparing it with other manures and organic fertilizer sources.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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Table 1. Effect of foliar application of Humic acid on root characteristics and Flower drops (%) at flowering development in Redgram

Treatments	Root length (cm)	Root fresh weight(g)	Root dry weight (mg)	Root volume (cc)	Flower drops (%)
T_1 = Humic acid liquid 15% @ 1.0 ml/l of water	15.24	21.67	5.83	45.82	58.2
T_2 = Humic acid liquid 15% @ 1.5 ml/l of water	18.81	23.58	7.48	48.55	57.4
T_3 = Humic acid liquid 15% @ 2.5 ml/l of water	21.98	25.64	8.63	49.50	53.2
T_4 = Humic acid liquid 15% @ 4.0 ml/l of water	24.25	27.12	9.93	52.08	45.8
T_5 = Planofix 4.5 % @ 20ppm	18.52	24.87	8.78	45.39	55.3

T_6 = Control	10.92	19.07	4.60	43.80	65.4
S.Em (±)	**1.86**	**1.60**	**1.59**	**1.78**	**2.57**
C. D. (5%)	**5.60**	**4.84**	**4.78**	**5.38**	**7.74**

DAS = Days after sowing

Table 2. Effect of foliar application of Humic acid on macro and micronutrient at harvest in Redgram

Treatments	Macronutrient			Micronutrient		
	N (%)	P (%)	K (%)	Cu (ppm)	Zn (ppm)	Fe (ppm)
T_1 = Humic acid liquid 15% @ 1.0 ml/l of water	2.69	0.437	1.28	1.93	7.48	3.88
T_2 = Humic acid liquid 15% @ 1.5 ml/l of water	2.92	0.575	1.37	2.31	7.93	4.55
T_3 = Humic acid liquid 15% @ 2.5 ml/l of water	3.62	0.765	2.73	2.42	8.78	5.03
T_4 = Humic acid liquid 15% @ 4.0 ml/l of water	4.92	0.952	3.80	2.98	10.60	5.59
T_5 = Planofix 4.5 % @ 20ppm	3.75	0.521	1.58	2.35	8.40	4.73
T_6 = Control	1.87	0.259	1.13	1.50	6.65	3.50
S.Em (±)	**0.454**	**0.147**	**0.34**	**0.19**	**0.67**	**0.39**
C. D. (5%)	**1.382**	**0.444**	**1.04**	**0.57**	**2.02**	**1.18**

DAS= days after sowing

Table 3. Effect of foliar application of Humic acid on yield and yield components at harvest in Redgram

Treatments	Yield components			
	Number of pods per plant	Pod weight (g)	Test weight (g)	Yield (kg/ha)
T_1 = Humic acid liquid 15% @ 1.0 ml/l of water	95.35	46.65	7.85	1176.98
T_2 = Humic acid liquid 15% @ 1.5 ml/l of water	102.18	51.60	8.58	1254.30
T_3 = Humic acid liquid 15% @ 2.5 ml/l of water	116.28	62.38	9.40	1323.02
T_4 = Humic acid liquid 15% @ 4.0 ml/l of water	126.88	75.45	9.78	1426.12
T_5 = Planofix 4.5 % @ 20ppm	105.10	61.85	8.40	1288.66

T_6 = Control	89.38	44.88	7.43	1073.88
S.Em (±)	**6.15**	**6.14**	**0.52**	**39.91**
C. D. (5%)	**18.53**	**18.49**	**1.56**	**120.21**

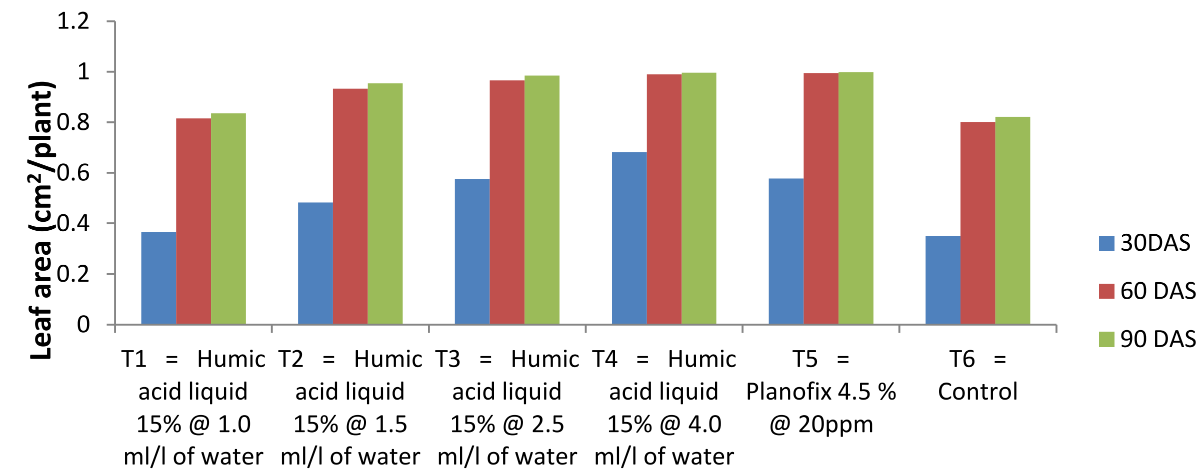

Fig. 1 Influence of foliar application of Humic acid on leaf area (dm^2/plant) at different stages of crop growth in Redgram

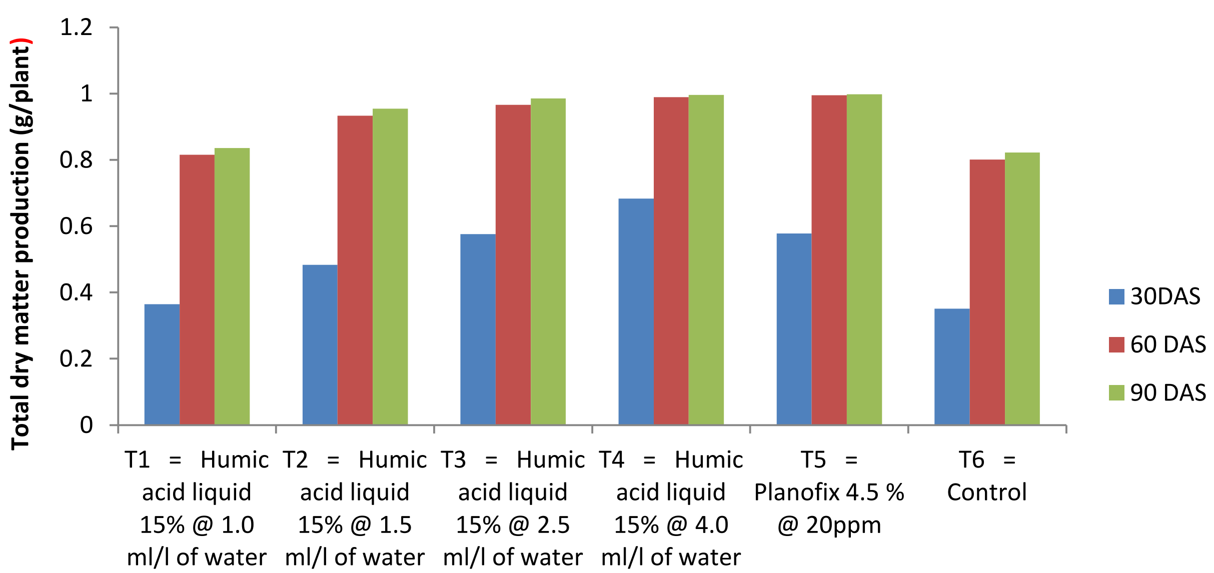

Fig. 2 Influence of foliar application of Humic acid on Total dry matter production (g/plant) at different stages of crop growth in Redgram