PHYSIOLOGICAL RESPONSE OF COWPEA IN A RAINFED ALFISOL ECOSYSTEM TO THE IMPULSE OF SOIL MOIS-TURE CONSERVATION PRACTICES

T.Ramesh^{*}, P.Devasenapathy^{**}

Department of Agronomy, Tamil Nadu Agricultural University, Coimbatore 641 003, India.

Received 5 April 2006

Summary. Proven techniques of soil moisture conservation were imposed in two consecutive seasons (2002 and 2003) under a rainfed alfisol ecosystem in order to investigate the physiological response of cowpea as the test crop. The impulsive variations in soil moisture status with reference to these techniques were found to influence the physiological and biochemical parameters of cowpea significantly. Mulching was applied alongside the moisture conservation techniques for a complement analysis. Ridges and furrows with mulching (2002) and tied ridges with mulching (2003) led to higher chlorophyll content (1.88 and 1.54 mg g⁻¹ FW) and chlorophyll stability index (87.28 and 83.97%) when compared to the traditional farmers' practice. Leaf relative water content was higher (by 6.24%) in tied ridges with mulching. Accumulation of proline was observed during the second year mainly due to soil moisture stress induced by poor rainfall distribution. Mulch treatments resulted in a lower proline content (132.53 and 130.44 μ g g⁻¹ DW) while the farmers' practice showed higher proline accumulation (214.33 µg g⁻¹ DW). Ridges and furrows resulted in a higher grain yield (715.9 kg ha⁻¹) when coupled with mulching (2002). Tied ridges with mulching (2003) also produced nearly the same effect (297.4 kg ha⁻¹) over farmers' practice.

Key words: chlorophyll, cowpea, mulching, proline, tied ridges.

Abbreviations: DAS - days after sowing, DW – dry weight, CSI - chlorophyll stability index, FW- fresh weight, RWC- relative water content.

^{**} Corresponding author, e-mail: devasenapathy@tnau.ac.in

INTRODUCTION

Soil moisture is the foremost factor that alters the physiological processes of rainfed pulses thereby influencing the productivity. From the soil perspective, alfisols possess inherently low water retention capacity on account of their particle size distribution and mineralogical composition, which in turn causes a serious threat of soil moisture deficit even after relatively short spells of rain. There are research evidences that deficit soil moisture conditions and poor soil fertility directly influence the physiological parameters such as chlorophyll, chlorophyll stability index, proline and relative water content and eventually the yield of rainfed crops. Continuous moisture stress leads to a decline in leaf chlorophyll and relatively wild stress would inhibit chlorophyll synthesis in wheat (Singh et al., 1985). Relative water content (RWC) is a physiological parameter to assess the drought tolerant capacity and photosynthetic efficiency of plants. Jiang and Huang (2002) reported that RWC decreased under drought stress. Proline accumulation occurred in leaves of plants under moderate to severe water stress, salinity, high temperature, nutrient deficiency and due to heavy metals and high acidity (Oncel et al., 2000). An increase in proline content in soil moisture stress conditions was reported by Unyayar et al. (2004) in sunflower. For rainfed crops soil moisture conservation is of paramount importance for maintaining an optimal stress environment. Any soil moisture conservation measure, mechanical or biological, or both coupled together will have an impact on the soil moisture stress thereby influencing upon the crop physiological behaviour. The present study was envisaged on the premise that soil moisture conservation techniques can help maintaining favourable physiological status of crops, so that rainfed farming can be sustained. Cowpea was selected as a test crop due to its compatibility to rainfed farming.

MATERIALS AND METHODS

The experimental platform was shallow to moderately deep, medium textured, acidic in reaction and non-calcareous soil, which was derived from a lateritic parent material. Physical properties of the soil such as bulk density, field capacity and permanent wilting point were 1.42 g cc⁻¹, 21.4% and 10.3 %, respectively. The soil was acidic (pH 5.32) with 111 mg kg⁻¹ available nitrogen (alkaline permanganate method as described by Subbiah and Asija, 1956), 7 mg kg⁻¹ available phosphorus (calorimetric method as suggested by Bray and Kurtz, 1945) and 45 mg kg⁻¹ available potassium (flame photometer method as suggested by Stanford and English, 1949), respectively. The experiment was carried out during 2002 incorporating four different *in situ* soil moisture conservation practices: M₂ - ridges and furrows (R and F), M₃ - compartmental bunding (CB), M₄ - ridges and furrows + mulching, M₅ - compartmental bunding + mulching and keeping M_1 farmers' practice of moisture conservation (disc ploughed once during pre-monsoon showers and subsequently country plough tillage was given once during sowing) as control. Formation of ridges and furrows were done with a spacing of 60 cm in between two ridges. Based on the results obtained during the first year, compartmental bunding, which produced a comparatively low yield, was replaced by tied ridges as suggested by the project committee during 2003. Tied ridges were formed by blocking the furrows manually with earthen bunds randomly at 1.5 m intervals. Mulching was done with locally available crop residues of groundnut, horse gram and green gram and sugarcane trash at a rate of 2.5 t ha⁻¹ on a dry weight basis 15 days after sowing. The total rainfall received during the cropping period in cowpea was 162 and 132.4 mm with distribution of 7 and 7 rainy days during 2002 and 2003, respectively. Physiological parameters were estimated at days 25 and 50 after sowing and grain yield was recorded at the time of harvest.

Estimation of relative water content in leaves

Relative water content was measured in the fully expanded leaves using the method suggested by Weatherly (1950). To determine plant RWC, nine leaves were weighed (fresh weight) immediately after harvest and placed in distilled water for 2 h at 25° C and then their turgid weight (TW) was recorded. The samples were then dried in an oven at 110 °C for 24 h to obtain their dry weight (DW). The RWC was calculated by the following formula:

$$RWC (\%) = \frac{Fresh weight (g) - Dry weight (g)}{Turgid weight (g) - Dry weight (g)} x 100 (1)$$

Estimation of proline content

Proline content in the leaves was estimated by the method of Bates et al. (1973). Leaf samples (250 mg) were extracted with 3 % sulphosalicylic acid. Extracts (2 ml) were held for one hour in boiling water by adding 2 ml of acid ninhydrin, 2 ml of glacial acetic acid and finally, cold toluene (4 ml). Proline content was measured using a spectrophotometer at 520 nm and calculated as $\mu g g^{-1}$ DW against standard proline.

Estimation of chlorophyll content

Chlorophyll content was estimated in the fully expanded trifoliate leaf in the main branch of crop growth (Yoshida et al., 1971). A leaf sample of 250 mg was macerated with 10 ml of 80% acetone. The supernatant was transferred to a 25ml volumetric flask and the volume was made up to 25 ml using 80 % acetone. Then the colour intensity was read at 652 nm using a spectrophotometer. Total chlorophyll content was expressed as mg g⁻¹ FW.

Total chlorophyll = $\frac{\text{OD at 652 nm}}{34.5 \text{ x FW}}$ x V (2) Where, FW- Fresh weight of the leaf sample V- Volume of the supernatant

OD – Optical Density

Estimation of chlorophyll stability index

Two leaf samples of 250 mg each were put in two test tubes containing 10 ml of distilled water. One of the test tubes was placed in a water bath and heated at 65°C for 30 minutes while the other was kept as a control. Then total chlorophyll content was estimated using a spectrophotometer at 652 nm (Koleyoreas, 1958). CSI was calculated using the following formula:

$$CSI (\%) = \frac{\text{Total chlorophyll content (heated)}}{\text{Total chlorophyll content (control)}} \times 100 \quad (3)$$

For soil moisture estimation, soil samples from each plot were drawn at a weekly interval from sowing to harvest at a 0-30 cm depth using a screw auger. The gravimetric method was used for estimating soil moisture. Soil moisture measurements were based on the presumption that the effective root zone depth of cowpea is normally confined to a depth of not more than 30 cm below ground level. Besides, the effective plough sole depth for the experimental study could be extended up to 30 cm depth only, below which a relatively impervious stratum was encountered inhibiting root development and penetration.

Statistical analysis

Data obtained were subjected to ANOVA and Student's t-test.

RESULTS AND DISCUSSION

Available soil moisture under moisture conservation practices

Mulching treatments along with ridges and furrows or tied ridges were found to be superior in soil moisture conservation than the treatments without mulching and farmers' practice of moisture conservation during both years (Fig 1). At the stage of flow-



Fig 1. The effect of *in situ* soil moisture conservation practices on available soil moisture at different developmental stages of cowpea: A (2002); B (2003).

ering, ridges and furrows with mulching resulted in 4.4 % and 3.3 % higher soil moisture over farmers' practice of moisture conservation during both years, respectively. Mulching produced greater infiltration by runoff reduction and subsequent evaporation suppression of the infiltrated water that apparently contributed to soil moisture gains (Unger et al., 1997) and reflected on favourable plant physiological parameters in cowpea. The increased soil water accumulation under mulching in comparison with no mulching treatment has been reported by Sharma and Parmar (1998).

Chlorophyll content under moisture conservation practices

In situ soil moisture conservation practices have shown significant variations in total chlorophyll content of cowpea during both years. Total chlorophyll content was significantly higher under ridges and furrows with mulching (1.88 mg g⁻¹ FW) in 2002 (Table 1) and tied ridges with mulching (1.54 mg g⁻¹ FW) in 2003 over farmers' practice of soil moisture conservation. Availability of soil moisture and nutrients for a longer period in moisture conserved plots, which resulted in increased chlorophyll synthesis. A reduction in chlorophyll content (1.33 and 1.08 mg g⁻¹ FW) was noticed under farmers' practice during both years. In water stressed plants, loss in chlorophyll is associated with a reduction in the flux of nitrogen into the tissue as well as alterations in the activity of enzyme systems such as nitrate reductase (Begaum and Paul (1993).

Treatments	Total chlorophyll		CSI		RWC		Grain yield
	(mg g ⁻¹ FW)		(%)		(%)		(kg ha ⁻¹)
	25	50	25	50	25	50	
	DAS	DAS	DAS	DAS	DAS	DAS	
2002							
Farmers' practice	1.04	1.33	67.45	59.87	85.67	80.53	572.2
Ridges and furrows	1.29	1.60	78.15	73.42	86.61	81.41	662.8
Compart. bunding	1.15	1.61	81.01	74.36	88.34	83.48	627.8
R and F + Mulching	1.46	1.88	87.28	83.80	90.34	85.65	715.9
CB + Mulching	1.36	1.83	86.97	85.33	89.88	86.02	688.9
C.D. at 5% P	0.18	0.23	9.07	6.40	4.26	3.86	49.5
2003							
Farmers' practice	0.72	1.08	65.49	58.13	84.83	74.22	169.7
Ridges and furrows	0.78	1.31	75.87	71.31	88.38	77.33	208.5
Tied Ridge	0.81	1.32	78.58	72.17	89.80	78.58	223.2
R and F + Mulching	0.89	1.19	83.62	80.55	91.25	79.84	279.0
TR + Mulching	0.94	1.54	83.97	82.06	91.96	80.46	297.4
C.D. at 5% P	NS	0.18	8.80	8.59	2.25	2.51	29.1

Table 1. The effect of *in situ* soil moisture conservation practices on some physiological parameters and grain yield of cowpea. NS – non significant at 5% level.

Relative water content under moisture conservation practices

Mulching treatments led to higher relative water content of 85-86% during 2002 and 80% during 2003 whereas farmers' practice of moisture conservation resulted in only 80.53% and 74.22% during both years at day 50 after sowing (Table 1). This could be mainly due to the presence of higher soil moisture combined with low evaporation loss under these treatments. Similarly, higher plant water status under mulching has been reported by Gupta and Gupta (1986) in cowpea. Lower relative water content noticed in farmers' practice was possibly due to lower soil moisture, the leaves experiencing the soil moisture stress earlier than roots, with lower RWC. This result is in accordance with the findings of Unyayar et al (2004) who reported a sharp decline in relative water content in sunflower under water deficit conditions.

Proline content under moisture conservation practices

The effect of *in situ* soil moisture conservation practices on proline content of cowpea showed that ridges and furrows with mulching during 2002 and tied ridges with mulching during 2003 resulted in lower proline content (64 and 130.44 μ g g⁻¹, respectively) at day 50 after sowing (Fig 2), whereas farmers' practice of no soil moisture conservation showed increased proline content at all stages of observation in both years. Under stress conditions, the amino acid metabolism is largely altered, protein synthesis impaired and proteolysis increased. As a consequence proline synthesis might be promoted by increasing the concentrations of related metabolites such as polyamines, ammonia, arginine, ornithine, glutamine and glutamate. The increase in the concentrations of metabolites involved in the production of proline precursors might be the main cause for proline accumulation in plant tissues exposed to environmental stress (Silveira et al., 2001). Similarly, accumulation of free proline in water stressed leaves of many crops has been reported by Schurr et al. (2000).

Chlorophyll stability index under moisture conservation practices

Higher values for CSI were noted under mulching along with ridges and furrows (87.28%) or tied ridges (83.7%) mainly due to adequate availability of soil moisture. The higher CSI indicates the tolerance of plants under water stress condition. Significantly lower CSI was observed at farmers' practice moisture conservation during both years.

Grain yield under moisture conservation practices

Our results revealed a significantly higher grain yield of cowpea due to the applied land configuration techniques like ridges and furrows, compartmental bunding and tied ridges as compared with farmers' practice of moisture conservation (Table 1). A



Fig 2. The effect of *in situ* soil moisture conservation practices on proline content in cowpea leaves at 25 and 50 DAS: A (2002); B (2003).

higher grain yield was obtained under ridges and furrows with mulching (by 25.1 %) during 2002 and tied ridges with mulching (by 75.3 %) during 2003, respectively. Availability of higher soil moisture, enhanced infiltration of rain water by restricting the surface flow, reduced evaporation and suppression of weeds under ridges and furrows or tied ridges with mulching contributed collectively to better plant physiology and grain yield. Xiao-Yan Li (2000) reported that the ridges and furrows technique combined with mulching maximized the utilization of rainwater in arid and semi arid areas. These findings are in accordance with the results of Gargi Das and Gautam (2003) who reported that the favourable effect of straw mulch on yield of pearl millet was mainly due to conservation of rainwater in the soil profile.

To sum up, sowing of cowpea on either ridges and furrows or tied ridges along with mulching by locally available crop residues at the rate of 2.5 t ha⁻¹ could be a viable moisture conservation technology for better crop physiology and grain yield of cowpea in the rainfed *alfisol* ecosystem.

Acknowledgements: The authors are grateful to M. S. Swaminathan Research Foundation, Chennai, India for providing financial assistance for this project.

References

- Bates L.S., Waldren R.P., Teak T.D., 1973. Rapid determination of free proline for water stress. Plant and Soil, 39, 205-207.
- Begum F.A., Paul N.K., 1993. Influence of soil moisture on growth, water use and yield of mustard. J. Agron. Crop Sci., 170, 136-141.
- Bray R.H., Kurtz L.T., 1945. Determination of total, organic and available forms of phosphorus in soils. Soil Sci., 59, 39-45.
- Gargi Das, Gautam R.C., 2003. Yield and water use efficiency of pearlmillet as influenced by moisture conservation methods under rainfed conditions. Annals of Arid Zone, 24(1), 78-81.
- Gupta J.P., Gupta G.K., 1986. Effect of tillage and mulching on soil environment and cowpea seedling growth under arid conditions. Soil Tillage Res., 7, 233-240.
- Jiang Y., Huang B., 2002. Protein alterations in tall fescue in response to drought stress and abscisic acid, Crop Science, 42, 202-207.
- Koleyoreas S.A., 1958. A new method for determining drought resistance. Plant Physiol., 33, 232-233.
- Oncel I., Keles, Y., Ustun, A.S., 2000. Interactive effects of temperature and heavy metal stress on the growth and some biochemical compounds in wheat seedling. Environ Pollut, 107, 315-320.
- Schurr U., Heckenberger U., Herdel K., Walter A., Feil R., 2000. Leaf development in *Rici-nus communis* during drought stress: dynamics of growth processes, of cellular structure and of sink-source transition, Journal of experimental Botany, 51, 1515-1529.
- Sharma P.K., Parmar D.K., 1998. Effect of phosphorus and mulching on the uptake of sec-

ondary nutrients and productivity of wheat at different growth stages on an *alfisols* from western Himalayas. J. Indian Soc. Soil Sci, 46, 67-70.

- Silveira J.A.G., Melo A.B., Viégas R.A., Oliveira J.T.A., 2001 Salt- induced effects on the nitrogen assimilation related to growth in cowpea plants. Environ. Exp. Bot., 46, 171-179.
- Singh K., Verma H.D., Singh P.P., Jasmini D.K., 1985. Effect of sowing dates and nitrogen on growth and yield of some new wheat varieties. Indian J. Agron., 30, 72-74.
- Stanford S., English L., 1949. Use of flame photometer in rapid soil tests for potassium and calcium. Agron. J., 41, 446-447.
- Subbiah B.V., Asija C.L., 1956. A rapid procedure for estimation of available nitrogen in soils. Current Sci., 25(8), 259-260.
- Unger P.W., Schmberg H.H., Dao T.H., Jones O.R., 1997. Tillage and crop residue management practices for sustainable dryland farming system. Annals of Arid Zone, 36, 209-232.
- Unyayar, S., Keles Y., Unal E., 2004. Proline and ABA levels in two sunflower genotypes subjected to water stress. Bulg. J. Plant Physiol, 30 (3-4), 34-47.
- Weatherly P.E., 1950. Studies on water relation of the cotton plant. I. The field measurement of water deficit in leaves. New Physiol., 49, 91-97.
- Xiao-Yan Li., 2000. Soil and water conservation in arid and semi arid areas. The Chinese experience. Annals of Arid Zone, 39(4), 377-393.
- Yoshida S., Forno D.A., Cook H.H., Gomez K.A., 1971. Laboratory manual for physiological studies of rice. IRRI. Los Banos. Philippines.