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Ameliorating Morphological, Physiological and Biochemical Attributes of *Vigna mungo* by Using Polyacrylamide under Water Stress

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ABSTRACT: Legumes belong to second most popular family after poaceae. Black gram is a summer crop grown for its nutritive protein rich seed. Drought is a major abiotic stress on black gram. A pot experiment was carried out in the natural conditions of net house to resolve whether the basal application of polyaccryamide might enhance the growth, physiology of black gram.Pots were irrigated with ground waterregularly as a control and in other pots water was given as 15 and 30 DID at 20 DAS. A uniform basal dose of Pam_{1792} was applied at the vegetative stage on each and every pot excluding control plants. Water stress significantly reduces the growth attributes, biochemical attributes.Basal dose application of Pam_{1792} reduced the damaging effect of stress on plantand accelerated the restoration of growth processes. The findings prove that the application of the Pam_{1792} overcame the adverse effect of stress in *V. mungo*.

KEYWORDS: Drought stress, Black gram, Polyacrylamide, Growth, Protein content

I. INTRODUCTION

Now a day's water stress is the major abiotic stress, crores of pulse cropsreduces yield per year [1].Black gram (*Vigna mungo*) is a leguminous crop and occupied fourth position in the entire pulse crop both in acreage and production [2]. Water stress is a major abiotic stress on black gram. It is cultivated in rabi season and a cheap source of protein approximately 25-28%, ash 5-5.5%, oil 0.5-1.5%, fibre 3.5-4.5% and carbohydrate 6.2 -6.5% on dry weight basis [3]. Soil water is another important factor. Its adequate supply helps in obtaining the optimum growth and development of plants by maintaining proper water potential of cells. Due to the deficiency and access of water it may cause water stress but the majority is due to deficiency of water termed as water stress or drought stress [4, 5]. Water stress is the form of loss of moisture content in cellular termedwhich causes two types of damages *viz.* uncontrolled causes ultimate death of plant and other controlled causes physiological and morphological changes *i.e*large capacity of storing water, deep tap root, wide spreading roots near the soil surface and few stomata to reduce transpiration [6,4].Due to this the rate of transpiration exceedsthan the moisture absorption causes diminished of relative water content, water potential, turgor pressure and osmotic potential of cell increased of ions and other solutes conclude the changes in morphological and physiology of plants [7,8, 9,10].The moisture index in plants in this range becoming too low that natural precipitation is highly inadequate to support plant life.

The soil moisture may be lost due to several factors including run off, erosion and infilteration. To check the moisture loss to a great extent, application of polyacrylamide, i.e. Pam to the soil may be an effective soil moisture conservation practice [11]. It absorbs the water by osmosis forms a soft gel which preserves 70% of water and maintain moisture in soil which laterally reduces water stress [12, 13, 14]. It develops soil aggregation, reduce crust hardness, increase infilteration rate and prevent erosion [15, 16]. It is a long chain, synthetic, high molecular weight polymer made by the polymerization of acrylamide monomers with each other or with other vinylmonomers to yield products with extremely high molecular weight in the properconditions [17]. It may be of three types due to charge characteristics. These include anionic, cationic and non-ionic. The degree of positive and negative charge is called charge density. These polymer macromolecules reactive the linear particles of the soil, aerate the soil and provide water to the plants[18, 19].



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Keeping in mind the meticulous importance of Pam and black gram a potent pulse crop an experiment was conducted to work out the effects of Pam on crop productivity, protein content, and enzyme activities of black gram under water stress condition.

II. MATERIALS AND METHODS

Plant materials and growth condition

Healthyseeds of black gram were obtained from IARI, NewDelhi, and surface sterilized with 0.2% HgCl₂ solutionfor 5 min with frequent shaking and then a thoroughwashing with deionised water was done. Prior to seedsowing, 5kg homogenous mixture of soil was filled in the earthen pots (25 cm diameter). Six seeds weresown in each pot. All the pots were watered with ground water up to tri-foliate leaf formation at 20 days aftersowing (DAS). Control pots were irrigated with ground water regularly. Waterwas given *viz.* 15 and30 days interval drought (DID) after 20 DAS. The potswere covered with a rain out shelter, made up of plasticsheets, whenever rainfall was anticipated. Immediately after rain, rain out shelter was pulled back so thatpots may receive maximum sunlight. Further, the potswere regularly covered with rain out shelter duringnight time. Using this system the pots were protected from rainfall and from any external moisture entry. Plants were uprooted randomly at 45 and 60 DAS, washed carefully and separated into root, stem and leavesfor analyses. A uniform concentration Pam 0.4 g i.e. 1792 kg Pam/hawas applied as basal dosefrom the sowing of seeds. Total four treatments of Pamwere doneas a basal dose after every 10 day of interval.

Experimental design and growth and biochemical analysis

The experiment was conducted in pots in the nethouse at the Botany Department, A.M.U., Aligarh. Growth and biochemical attributes of black gramwere determined at 45 and 60 days after sowing(DAS). The experiment was conducted using a simplerandomized block design. Each treatment was replicated three times. At each stage, plants from each treatment were harvested and washed carefully with tap water to remove all adhering foreign particles, surface drying the plants thereafter using blotting paper. Growth attributes, *viz.* shoot length, root length, number of nodules, shoot fresh weight and root fresh weight were recorded. Plant samples were dried at 80°C for 24 h using a hotair oven to obtain shoot and root dry weights.

Total chlorophyll and carotenoid content

Totalchlorophyll and carotenoid content in fresh leaveswere estimated using the method of [20].

Protein content

The seed and leaf protein content was estimated by adopting the methodology of [21]. *Carbonic anhydrase (CA) activity* The activity of the enzyme will be determined by the method of [22]. *Estimation of proline (PRO) content* The PROcontent was estimated by the method of [23]. *Leaf Nitrogen (N)content* N content will be estimated according to the method of [24].

III. RESULTS

Growth Parameters

All the parameters of growth showed the adverse in both stages (45 and 60 DAS) in drought stressed black gram when compared with well-watered control plants. But, there was significant improvement by the application of Pam in stressed plants when compared to without Pam application (Table 1). By the application of Pamshoot length, root length, number of nodules is increased by 30.86%, 89.01%, 73.33% at 45 DAS and 22.94%, 30.60%, 34.78% at 60 DAS as compared to drought stressed plants. Pam also proved best in raising the shoot fresh weight, shoot dry weight, root fresh weight and root dry weight by 31.86%, 89.01%, 73.33% at 45 DAS and 22.94%, 30.60%. 34.78% at 60 DAS respectively as compared to drought stressed plants (Table 2).



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Parameter	Control	Drought	Drought + Pam		
45 DAS					
Shoot length per plant(cm)	23.78±0.08	15.78±0.12	20.65±0.32		
Root length per plant (cm)	10.70±0.04	4.55±0.28	8.60±0.14		
Number of nodules per plant	29.00±0577	15.00±.408	26.00±0.408		
60 DAS					
Shoot length per plant (cm)	35.52±0.25	26.37±.205	32.42±.188		
Root length per plant (cm)	19.05±.170	13.17±.154	17.20±.158		
Number of nodules per plant	35.75±.250	23.00±.577	31.00±.577		

Table 1: Effect of basal application of Pam₁₇₉₂ on shoot length per plant, root length per plant and number of nodules per plant of *V. mungo* grown under drought stress. The data mean of three replicates \pm SE.

Parameter	Control	Drought	Drought+ Pam	
45 DAS				
Shoot fresh weight per plant (g)	11.55±0.028	5.42±0.062	9.32±0.028	
Root fresh weight per plant (g)	5.42±0.408	2.72±0.629	4.27±0.750	
Shoot dry weight per plant (g)	4.80±0.070	2.15±0.028	3.85±0.028	
Root dry weight per plant (g)	4.22±0.047	1.00±0.032	2.82±0.25	
60 DAS				
Shoot fresh weight per plant (g)	19.27±0.039	11.56±0.151	17.1±0.029	
Root fresh weight per plant (g)	12.63±0.127	7.63±0.039	10.70±0.029	
Shoot dry weight per plant (g)	11.72±0.150	7.77±7.53	9.85±9.74	
Root dry weight per plant (g)	10.75±0.066	6.75±0.033	8.76±0.035	

Table 1: Effect of basal application of Pam_{1792} on shoot fresh weight per plant, root fresh weight per plant, shoot dry weight per plant and root dry weight per plant of *V. mungo* grown under drought stress. The data mean of three replicates \pm SE.

Biochemical Parameters

Drought stress caused a decrease in photosynthetic pigment *viz*. chlorophyll and carotenoid content as compared to the watered control plants. Total chlorophyll content and the carotenoid content decrease by 14% and 19.10% at 45 DAS and 24.49 and 22.38% at 60 DAS severally due to drought but the effect of the Pam it significantly increases 9.75% and 13.01% at 45 and 16.73% and 16.67% at 60 DAS respectively as compared to water stressed plant.

Water stress also reduces the protein content in seeds and protein content in leaves at both the stages as compared to the control plants. There is an increase of protein content in leaves and seedsby the application of Pam₁₇₉₂over the stressed black gram plant by 9.81% and 6.10% at 45 DAS while 11.24% and 6.94% by 60 DAS severally.

In comparison to control, there is reduction of the CA activity in both stages 45 DAS and 60 DAS observed anincrease after the application of the Pam. The effect of the basal application of Pam₁₇₉₂ significantly increases by 8.27% at 45 DAS and 9.49% at 60 DAS respectively on water stressed plants.



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Fig 1: Effect of basal application of Pam₁₇₉₂ on chlorophyll content, carotenoid content, carbonic anhydrase activity, protein content in leaves and protein content in seeds of *V. mungo* grown under drought stress. Each value represents the mean of three replicates with SE (T) determined.

The proline content increased in both stages under drought condition. The higher value for proline content was noted in 60 DAS by as compared to 45 DAS.Proline content increased by 20.73% at 60 DAS and 20.32% at 45 DAS respectively. Pam application under drought stress also alters at both the stages.

Leaf N content is also decreased by the drought stress at both the stages but by the application of Pam_{1792} it increased upto 16.15% and 17.03% as compared to drought stressed plant at 45 and 60 DAS respectively.



Fig 2: Effect of basal application of Pam_{1792} on proline content and leaf nitrogen content of *V. mungo* grown under drought stress. Each value represents the mean of three replicates with SE (τ) determined.

IV. DISCUSSION

There is no scientific literature available regarding the effect of Pam on enhancement of growth and biochemical parameters under drought stressed condition. However, the reduction caused by drought stress on shoot length, root length and number of nodules can be ascribed as the loss of cellular water mainly from vacuoles, closing of stomata pore, decrease in cell enlargement, wilting, reduction of turgor pressure, reduction of water potential, plasmolysis, reduced cyclin-dependent kinase activity ultimately causes inhibition of cell division and shrinkage of whole plant



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growth[25, 26]. These results coincide withthose of, [27, 28, 10] on Vigna sinensisCymbopogon flexuosus and Catharanthus roseus respectively. The shoot fresh and dry weight, root fresh and dry weights are also reduces in drought stress condition. The reason of this the reduction of shoot length, root length and number of number of nodules directly effect on fresh and dry masses of shoot and root and due to the deficiency of water in the plant causes the senescence. The following results are collaborated by the effect of drought on various crops [29, 30, 31]. However, by the application of Pam increases the shoot length, root length, number of nodules, shoot fresh weight, root fresh weight, shoot dry weight and root dry weight under drought stress conditions. When Pam is applied to the soil, it gets adsorbed water 500 times more than its own weight [32, 33, 34] thereby improving the porosity, aeration, thermo and water regime, water holding capacity, fertility and stability of soil, which in turn delays senescence, improve the growth and development of plants [18,19]. The other reason for the improvement in these parameters may be due to the nutrients made readily available by this polymer, which have been reported to increase the activity of cell division, cell expansion and cell elongation [35]. These results are collaborated on soyabeen [36].

The photosynthetic pigments are decelerate in water stress conditions, the basic reason for this during water deficiency activity of chlorophyllase enzyme accelerate which causes degradation of chlorophyll content and protein degradation. The present study showed that enzyme activity is reduced at both the stages due to water stress. Water stress supress the activity of key enzymes of photosynthesis namely Rubisco and PEP carboxylase [37, 28] consequently it results in decrease in CA activity. However, the protein content (leaves and seeds) and N content (leaves) are reduced due to water stressed conditions as compared to well watered plants. The decrease in protein content observed due to paralysation of protein biosynthesis. Water deficiency results in retardation of the membrane permeability which causes limits in nutrient uptake. The results are also confirmed by [38]. The increase in proline on leaves is a good indicator of the exposure of plant to water stress [39]. Water deficit condition generated a physiological drought that led to activation of the enzymes of proline biosynthesis and suppression of those of its degradation and consequently the accumulation of higher level of proline. The results coincided with earlier reports of the research under water stress in tomato [40, 10]. However, there is no scientific literature available regarding the effect of Pam on these biochemical parameters under water stress condition. But the author say that Pam may have favorable effect on soil conditions which may be helpful for optimum metabolism of plants leading to higher values for these parameters in water stress condition.

V. CONCLUSION

In the end, the present author wishes to claim, with all the modesty at her command, that she has been able to enrich the scientific literature on Pam by contributing the following findings:

The effectiveness of basal application of Pam was studied separately on the growth, physiological and biochemical parameters of black gram under water stress and found Pam₁₇₉₂was established to overcome the effect of drought stress on black gram.

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