STUDIES ON WATER STRESS RECOVERY AND REGROWTH IN GENOTYPES OF CASTOR (*Ricinus communis* L.)

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CERTIFICATE

Ms. M. JYOTHI has satisfactorily prosecuted the course of research and that the thesis entitled "**STUDIES ON WATER STRESS RECOVERY AND REGROWTH IN GENOTYPES OF CASTOR (*Ricinus communis* L.)**" submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination. I also certify that the thesis or part thereof has not been previously submitted by him for a degree of any university.

Date : **(Dr. T. RAMESH)**
Place : Hyderabad Major Advisor

CERTIFICATE

This is to certify that the thesis entitled "**STUDIES ON WATER STRESS RECOVERY AND REGROWTH IN GENOTYPES OF CASTOR (*Ricinus communis* L.)**" submitted in partial fulfilment of the requirements for the degree of **MASTER OF SCIENCE IN AGRICULTURE** of the **Acharya N.G. Ranga Agricultural University, Hyderabad**, is a record of the bonafide research work carried out by **Ms. M. JYOTHI** under our guidance and supervision. The subject of the thesis has been approved by the Student's Advisory Committee.

No part of the thesis has been submitted for any other degree or diploma. The published part has been fully acknowledged. All assistance and help received during the course of the investigation have been duly acknowledged by the author of the thesis.

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I, **M. JYOTHI** hereby declare that the thesis entitled **"STUDIES ON WATER STRESS RECOVERY AND REGROWTH IN GENOTYPES OF CASTOR (*Ricinus communis* L.)"** submitted to the Acharya N.G. Ranga Agricultural University for the degree of **MASTER OF SCIENCE IN AGRICULTURE** is the result of original research work done by me. I also declare that any material contained in the thesis has not been published earlier in any manner.

Date : **(M. JYOTHI)**

Place: Hyderabad

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LIST OF ABBREVIATIONS

μ E m^{-2} sec^{-1}	:	Micro Einstein per meter square per second
μ g m^{-2} s^{-1}	:	Micro gram per meter square per second
Ψ_l	:	Leaf water potential
BA	:	Benzyl adenine
CGR	:	Crop growth rate
DAI	:	Days after inoculation
DAS	:	Days after sowing
g m^{-2} d^{-1}	:	Gram per meter square per day
$HgCl_2$	:	Mercuric chloride
HI	:	Harvest index
LAI	:	Leaf area index
MPa	:	Mega pascals
NAA	:	α - Napthalene acetic acid
OP, Ψs	:	Osmotic potential
PAR	:	Photosynthetically Active Radiation
PEG	:	Polyethylene glycol
PGR's	:	Plant growth regulators
RWC	:	Relative water content
S.Em$\pm$	:	Standard error mean
SCMR	:	SPAD chlorophyll meter readings
SLA	:	Specific leaf area
T	:	Transpiration
TE	:	Transpiration efficiency

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CHAPTER I

INTRODUCTION

Castor (*Ricinus communis* L.) an important oilseed crop of the family Euphorbeaceae is traditionally a long duration crop, flowering late and maturing under residual soil moisture conditions. Castor crop plays an important role in the agricultural economy of our country by earning valuable foreign exchange through the export of castor oil (Damodaram and Hegde, 2002). In India, it is cultivated under rainfed conditions in the states of Andhra Pradesh, Karnataka, Tamil Nadu and Orissa. The occurrence of rainless periods during monsoon (June to September) is a common feature, and the number of stress periods and their intensity are highly unpredictable causing significant moisture stress at different critical stages of crop at 31-60 and 61-90 days after sowing (DAS) which coincides with flowering and maturity of primaries and secondaries (Hanumantha Rao *et al*., 2003).

India ranks first in the world castor production with 8.67 lakh t from 10.7 lakh ha area. Andhra Pradesh is a major castor growing state and occupies an area of 0.4 lakh ha producing 1.3 lakh t. The crop is chiefly grown in the districts of Nalgonda, Mahaboobnagar, Warangal, Rangareddy, Medak, parts of Prakasam and Guntur. The crop is mainly grown under rainfed conditions where it suffers from intermittent drought that results in low yields. The productivity of Andhra Pradesh is only 333 kg/ha as compared to the national average of 805 kg ha^{-1} (Damodaram and Hegde, 2002). For bringing

about yield improvement, a number of castor varieties (Kranthi, Jyothi, Haritha, Kiran) and hybrids (GCH-1, 4 and DCH-177) have been released by conventional breeding approaches and these have been recommended for drylands (DOR, 2002). Despite their release the possibility of improving drought tolerance by genetic manipulation remains a challenge to modern genetics and plant breeding. So far, the response of the genotypes to water stress, their recovery and regrowth has not been characterized in castor. Hence, the traits conferring tolerance to water stress need to be studied in detail, so that the identified traits can be incorporated into future breeding programmes (Kramer, 1983).

Plants adopt different strategies to cope with drought. The strategies have been grouped into the main classes namely drought escape, drought avoidance and drought tolerance (Levitt, 1980). Under drought escape, the crop varieties with short duration show promise. However, for determinate crops that show regrowth and productivity upon recovery, unpredictability of environment poses potential danger (Villalobes – Rodriguez and Shibles, 1982). Under conditions of water stress, plants show ability to retain a high level of dehydration. The desiccation tolerant moss *Tortula ruralis* regains physiological activity (i.e. normal respiration and photosynthesis in about 30 min and photosynthesis in 2 hours) after free hydration. However, adequate explanation is not available to why protoplasm of some kinds of plants can tolerate dehydration while most others do not (Paleg and Aspinall, 1981).

The capacity of plants to withstand drought can be measured by its osmoregulation (i.e.) maintenance of turgor, by lowering of osmotic potential (O.P) by accumulating solutes in response to water deficits (Turmer and Jones, 1980). Osmotic adjustment (OA) occurs in response to lowering of soil water potential (Morgan, 1984). The turgor maintenance through OA is positively associated with yield, reduced photosynthesis upon stress recovery irrespective of stomatal activity (Lee and Asahira, 1983).

For many crop plants, the leaf water potential at which the stomata close is -0.8 to -2.8 MPa. Water loss by transpiration is the primary cause of plant water deficits. A six-fold reduction in daily mean transpiration was observed due to drought stress (Sivakumar and Sarma, 1985). Reduced transpiration due to drought stress leads to low leaf water potentials (Ramesh and Durga Prasad, 2003).

Transpiration efficiency (TE), defined as gm of dry matter produced per kg of water transpired is one such trait, which influences the performance of crops under water limited conditions. Variation in TE, has been demonstrated among genotypes within peanut species (Wright *et al*., 1994).

SPAD chlorophyll meter units are used as selection criteria for correlations with specific leaf area (SLA), TE and specific leaf nitrogen (SLN). SPAD units could provide a rapid low cost non-destructive technique to screen large breeding populations for TE (Rao *et al*., 2001). Such correlations have not yet been explored in castor.

Plants regenerated *in vitro* could be screened for drought stress by growing them in nutrient solution that decreases the OP (Bhojwani and Rajdan, 1983). Reports on multiple shoots regeneration using polyethelene glycol / mannitol that creates drought stress *in vitro* have been few. Screening procedure was previously developed to obtain stress tolerant plants in groundnut (Venkateswarlu *et al*., 1998) and callus as a system in castor (Manjula *et al*., 1999). Such techniques can be used to screen genotypes for stress and study recovery ability *in vitro*.

Therefore, this study was conducted with the following three objectives.

1. To identify castor genotypes that show quick recovery after the relief of water stress.
2. To study the physiological attributes conferring recovery and regrowth.
3. To study the recovery ability of castor genotypes *in vitro*.

CHAPTER II

REVIEW OF LITERATURE

Castor (*Ricinus communis* L.), a non edible oilseed crop is chiefly grown in low rainfall regions of semi-arid tropics and sub tropics. In Andhra Pradesh, the crop is grown under rainfed conditions in Telangana region on light textured red soil which are mainly characterised by low moisture retention capacity and poor nutritional status (Hanumantha Rao *et al*., 2003). Being indeterminate in growth habit, castor experiences moisture stress at flowering or at any stages of the spike orders leading to the significant loss of seed yield. Hence, understanding the nature and occurrence of dry spells is of paramount importance to realize higher yields. Knowledge on the physiological responses of this hardy crop is also limited. Hence, understanding of plant responses to water stress is essential to bring about crop improvement. Thus, literature is being reviewed on the influence of water stress on different parameters like crop growth, transpiration efficiency, water potential, osmotic potential, soil moisture and SPAD.

2.1 PERIODICITY OF DROUGHT

Instability in yields due to abiotic stresses are common under tropical and subtropical areas (Talwar *et al*., 2002). For improvement of yield in any region study of plant characters that confer tolerance to moisture stress and

identification of variability in physiological traits is critical (Reddy *et al*., 2000).

The periodicity of drought i.e. occurrence of dry spells, rainfall and rainy days over ten years period in Hyderabad is given in Table 1. In castor, drought due to moisture stress may occur in the beginning of season either due to late on set or early termination of monsoon. In addition, occurrence of rainless periods during monsoon period (June to September) is a common feature causing significant moisture stress at different critical crop growth stages especially from 31-60 and 61-90 days after sowing which coincides with the flowering and maturity of primaries and secondaries (Hanumantha Rao *et al*., 2003).

Table 1 : Periodicity of drought occurrence

Year	Rainfall (mm)	Rainy days	No. of dry spells	Standard weeks of occurrence of dry spells
1993-94	524	40	4	26-29, 31-35, 36-38, 41-52
1994-95	712	47	5	27-29, 31-33, 34-37, 38-39, 41-43, 40-52
1995-96	785	46	5	28-31, 32-34, 36-37, 39-41, 44-52
1997-98	384.9	28	6	28-30, 33-34, 38-40, 41-43, 45-47, 49-52
1998-99	1221.2	62	6	27-30, 32-33, 35-36, 37-38, 43-45, 46-52
1999-2000	362.0	-	7	27-28, 29-30, 31-32, 33-34, 37-38, 40-43, 44-52
2001-02	741.4	39	4	27-30, 33-35, 41-42, 43-52
2002-03	532.7	33	6	27-28, 28-30, 31-34, 36-37, 38-40, 42-44
2003-04	728.7	38	6	28-30 (4*), 32-33 (6*), 34-35 (8*), 36-37(13*). 41-42(10*), 44-52 (48*)

* Actual dry spell in days

In the present study, the castor crop experienced six dry spells during the crop growth. The dry spells matches the crop growth stage at 31-60, 75-105, 120-135 DAS. At 105 DAS, the crop recovered from drought.

2.2 SOIL MOISTURE AND CROP RESPONSE

Veihmeyer and Hendrickson (1927) claimed that soil water is uniformly available throughout a definable range of soil wetness, from an upper limit (FC) to a lower limit (PWP), both of which are characteristic and constant for a given soil. It is postulated that plant functions remains unaffected by an decrease in soil wetness until PWP is reached, at which plant activity is curtailed.

Richards and Wadleigh produced evidence indicating that soil water availability to plants actually decreases with decreasing soil wetness and that a plant may suffer water stress before wilting point is reached. The soil water potential at field capacity (FC) ranges from -0.1 to -0.3 bars or -0.01 to -0.03 MPa and -15 bars or -1.5 MPa at PWP (Reddy, 1999).

Crops with extensive and dense roots can utilize soil moisture more effectively than crops with sparse and shallow roots (Reddy, 1999). Optimal soil moisture for plant growth varies with the stage of the crop growth. Inadequate water supply during moisture sensitive periods will irrevocably reduce the yield. Provision of adequate water and other management practices at other growth stages will not help in recovering the yield lost.

Table 2 : Moisture sensitive periods of different crops

Crop	Moisture sensitive periods
Groundnut	Rapid flowering, peg penetration and early pod development
Sesame	Flowering to maturity
Sunflower	Flowerbud initiation, head initiation flowering and milky stages
Soybean	Flowering and seed formation
Cotton	Flowering and boll development

The soil moisture content values during dry spell ranged from 6.5 to 14.7 per cent in groundnut. At the beginning of dry spell (70 DAS), RWC ranged from 56 to 84 per cent and during dry spell RWC was 43 per cent (77 DAS) and decreased to 38 per cent (84 DAS) indicating that crop sensitiveness to drought. On the relief of water stress, groundnut cultivars recovered and recorded increased RWC values 60-70 per cent at 91 DAS and 70 to 85 per cent at 98 DAS. During dry spell the leaf water potential ranged from -8 to 19 bars. The high water status of the cultivars during the dry spell indicated their ability to tolerate drought (Ramesh *et al*., 2003).

Table 3 : Available soil moisture in various soil types

Soil texture	FC (%)	PWP (%)	Available moisture
Sandy	9 (6-12)	4 (2-6)	5 (4-6)
Sandy loam	14 (10-18)	6 (4-8)	8 (6-10)
Loam	22 (18-26)	10 (8-12)	12 (10-14)
Clay loam	27 (23-31)	13 (11-15)	14 (12-16)
Silty clay	31 (27-35)	15 (13-17)	16 (14-18)
Clay	35 (31-39)	17 (15-19)	18 (16-20)

Moisture stress reduces net photosynthesis (PN). This decrease in PN was in the order maize followed by pearl millet and sorghum. Recovery of PN was lowest in maize and highest in sorghum (Singh and Singh, 1993). The moisture sensitive stages of crops are given below in Table 2.

During the period of sprouting, castor seed requires a comparatively small amount of water which is due to small quantity of gel (non-oil part) in the seeds. The water content in leaves during the period from sprouting to formation of the central spike does not exceed 76-77 per cent as against 83-84 per cent in sunflower. Before flowering per cent moisture decrease to 70-73 per cent and during flowering and fruiting to 67-70 per cent.

Under irrigated conditions, castor seeds germinate at a moisture level of 40 per cent of FC or less, and quick swelling and germination occurs at 60-80 per cent. For castor, moisture is most important in the phase of flowering when the dry mass increases rapidly. If under conditions of insufficient water supply pollen movement decreases, female flowers abortion increases and seeds become undersized with low oil content. The optimum soil moisture for castor 70 per cent FC or more (Moskhin, 1986). The available moisture in various types of soils is given in Table 3 (Reddy *et al*., 1999).

The excised leaf water retention capacity of castor genotypes showed that VP-1, female parent of GCH-4 lost less moisture compared to other upto 45 min. after excision (Gangadhar Rao *et al*., 1998).

In castor, growing in dry areas, occurrence of non rainy periods and their intensity are highly unpredictable causing significant moisture stress at different critical stages of the crop especially between 31-60 and 61-90 DAS coinciding with flowering, maturity of main raceme (primaries) and second order racemes (secondaries) causing significant yield losses (Singh, Harvir and Hegde, 2003).

2.3 WATER DEFICIT AND PLANT RESPONSES

Water deficit effects of the growth. Developing cells are the most affected in two ways one size and two expansion. The effect on plant depends on the phenology and crop growth stage. If crop is affected at the young stage or at the beginning of the growth cycle, the effects are as follows. Plants developed smaller leaves with reduced leaf area, reduced `C' gain and show a variation in the nutrient and water absorption pattern. If water stress occurs at inflorescence development, continuation, plant biomass is unaffected. Flowering number is reduced and reproductive structures are aborted. If stress occurs at fruit maturation, plant biomass is unaffected. Inflorescence is unaffected. Seed filling is inhibited while fruit abscission is enhanced. (Nilsen and Orcutt, 1996).

Drought limits the agricultural production by preventing the crop plants from expressing their full genetic potential.

Drought resistance can be grouped into three categories viz., drought escape, drought avoidance and drought tolerance (Jiban Mirtra, 2001).

Drought avoidance is performed by maintenance of turgor through increased rooting depth, increased hydraulic conductance and by reduction of water loss through reduced epidermal stomatal conductance, reduced absorption of radiation by leaf rolling or folding and reduced evaporation surface (leaf area).

The mechanisms of drought tolerance are maintenance of turgor through osmotic adjustment, increase in plasticity in cell and decrease in cell size and desiccation tolerance by protoplasmic resistance.

Drought resistance being a complex trait, its expression depends on action and interaction of different morphological (earliness, reduced leaf area, leaf rolling, wax content, efficient rooting system, awn, stability in yield and reduced tillering), physiological (reduced transpiration, high WUE, stomatal closure and OA) and biochemical traits (accumulation of proline, polyamine, trehalose etc., increased nitrate reductase activity and increased storage of carbohydrate (Jiban Mitra, 2001).

Stress triggers a wide range of plant responses from altered gene expression and cellular metabolism to changes in growth rates and crop yields. The duration, severity and rate at which a stress occurs influences the plant response (Elizabeth A Bray *et al.*, 2000). Physiological change can result in decreased growth, yield or acclimation or adaptation, such as wilting under stress. As a short term measure, plants under stress can loose water resulting in decreased carbon leading to death of plants. As a long term measure wilting

brings down the leaf temperature by way of less absorption of light and protects the plants from permanent damage (Nilsen and Orcutt, 1996).

Plants loose lot of water by transpiration in the morning. The lose of water is compensated by the water present in the parenchyma cells of the tissues. By noon time, the stomata close, thereby it is reduced. The water in parenchyma cells are refilled with water absorbed from the roots. As a result Ψ increases and provides a driving force for water movement (Nilsen and Orcutt, 1996).

During stress, the plant process actually suffers from lack of water rather than limitation in the CHO supply. Starch in the chloroplast gets metabolized into sugars. If stress occurs at vegetative stage, more CHO are partitioned into roots, thereby altering shoot : root ratio. If stress occurrs at later stages, it affects reproductive parts or seed filling. Under water stress situations the N in nitrate and ammonical form is accumulated in the roots itself and their flow or translocation is reduced.

Under stress situations, protein hydrolysis coupled with increase in polyribosome and ribonucleases results in proline degradation. Amino acids like glutamine or arginine and non aminoacids like proline and glycine betaine get accumulated. Utilizing the new molecular biology techniques, transgenics are produced which show drought and salt tolerance.

Transgenic over expressing	Plant	Claim	Reference
Glycine - betaine	Tobacco chloroplast	Marker for osmoprotectant	Rathinasabapathi *et al*.,1994
Fructan	Tobacco	Resistance to drought	Pilon – Smits *et al*.,1995
Proline	Tobacco	Tolerance to osmotic stress	Kavikishore *et al*.,1995
LEA	Rice	Tolerance to water deficit and salinity	Xu *et al*., 1996

2.4 WATER STRESS AND CROP GROWTH

Water stress is an important environmental variable influencing plant growth and yield (Hsiao *et al*., 1976). It affects many aspects of plant growth by modifying their anatomy, morphology, physiology and biochemistry. The prominent effects on crop plants are reduction in plant growth coupled with changes in various physiological processes (Nandwal *et al*., 1991). Stem length and dry weight of groundnut plants decreased markedly due to drought and the decrease was entirely dependent upon the availability of soil water to the plants (Orcutt and Hopkins, 1988). Similar decreases were reported in cotton (Taylor and Klepper, 1974). Cornor and Jones (1985) reported that sunflower plants watered for the first time at late budding (50 DAS) or at anthesis (70 DAS) maintained more number of leaves than control during seed filling period. Moisture stress reduced dry matter accumulation in groundnut (Ramesh *et al*.,2003) and in sunflower (Prabhudeva *et al*., 1998).

Gangadhar Rao and Sinha (1988) showed that leaf area produced has a direct bearing on the Ψ under limited soil moisture status. Sarkar (1994) showed a correlation between leaf area and drought tolerance and further stated that this character could be used to select genotypes for drought tolerance. Under stress situation, smaller thick leaves having well developed palisade with smaller cell is considered desirable (Bhardwaj *et al*., 1988).

In groundnut, during the post stress period both LAI and biomass production rates lagged behind the control plants for about a fortnight. After alleviation of stress, LAI and biomass production exceeded that of control plants. This recovery of the plant processes for production of fast vegetative cover condition the crop for subsequent survival, growth and development (Ramesh Babu *et al*., 1984).

A strong negative relationship existed between WUE and SLA (cm^2g^{-1}) and between Δ and SLA, indicating that genotypes with thicker leaves had greater WUE (Wright *et al*., 1994). In pearl millet and groundnut SLW increased significantly under drought, but there was a significant reduction in SLA (Suvarna *et al*., 2001). Asalatha *et al*., 1999 reported that SLA is positively correlated with partitioning, suggesting that selection for low SLA might result in production of more total dry matter.

Fischer and Hagon (1965) revealed that CGR is sensitive to water stress mainly because leaf area was sensitive to water stress. The reduction in cell expansion due to drought may reduce the size of photosynthesizing surface

and thereby CGR unless leaf area was not limiting NAR (Slatyer, 1970). LAI was the characteristic which reduced CGR in water deficit and dry land treatments when compared to irrigated conditions (Cox and Jolliff, 1986). Water stress decreased RGR and NAR which ultimately resulted in poor dry matter accumulation (Bhan, 1980) and seed yields (Sharma and Arvind Kumar, 1989).

Reddy *et al*. (1997) reported that maximum dry matter production in castor cultivar Aurna is achieved at optimum LAI of 0.85 and 0.62 at 90 DAS and at maturity respectively indicating that the dry matter produced on the plant is in excess. Sung (1982) reported that the translocation rate was slowed down under stress conditions in cotton. Prabowo *et al*. (1990) reported that the DMP and pod yield were extremely low under stress conditions in groundnut.

Water stress of -14 and -16 bars was induced during vegetative and peg formation in groundnut, reduced leaf number by 70.9 per cent, total leaf area by 59.9 per cent, DMP by 61.8 per cent, LAI by 62.9 per cent and HI by 6.9 per cent over normal soil moisture levels (Dhopte *et al*., 1991).

2.5 RELATIVE WATER CONTENT (RWC)

Relative water content is the ratio of actual water content to water content at saturation (fully turgid) and expressed in percentage. RWC is often considered as an appropriate measure of plant water status in terms of the physiological consequence of cellular water deficit. Decline in RWC as a result of water stress brings about changes in several plant processes

particularly nitrogen assimilation in groundnut (Umar *et al*., 1991) and sunflower (Plesnicar *et al*., 1995). The cultivar which maintained high RWC and shows positive turgor inspite of reduced Ψ during stress had optimum photosynthesis and solute accumulation as evident from osmotic potential (Rama Krishnayya and Murthy, 1991). RWC is thus reported to be a sensitive index of plant water content and any fluctuations in the plant water status influence the leaf temperature, leaf diffusive resistance and net photosynthetic rate primarily through their direct effects on stomatal regulation and mesophyll resistance (Sandhu and Horton, 1978). The manner in which water deficits effect the functioning of various biophysical processes and their relationship with yield is not well established.

The water stress of -16 bars reduced leaf relative water content (LRWC) by 6.47 per cent over control at 60 DAS in groundnut (Dhopte *et al*., 1991). Shallow soil induced water stress reduced RWC of leaf in cotton (Ingole, 1989).

2.6 TRANSPIRATION EFFICIENCY

Transpiration efficiency (TE) is an important physiological trait that influences adaptation of crop plants TE/WUE is one character which can improve productivity when available moisture levels are low (Wright *et al*., 1994). Under water limited conditions, the crop dry matter (DM) produced is a product of the amount of water transpired (T) and the ratio of DM produced per unit T. Transpiration ratio is given as water used in T (ml) / Dm

production in g. At leaf level, it is defined as the ratio of CO_2 assimilated to water lost in transpiration.

Transpiration ratio is the reverse of WUE.

$$TE = \frac{\text{Millimole of carbon assimilated}}{\text{Mole of water transpired}}$$

Water loss by transpiration is the primary cause of plant water deficits. The increased ratio of roots to shoots, smaller leaves, thicker cutin and denser venation found in plants subjected to water stress are desirable. Such characteristics probably provide a better water supply to the leaf tissue and lower transpiration per unit of leaf surface, when the stomata are closed by water stress. Thus, plants previously subjected to water stress have better control over water loss than those not previously stressed. Soybean, transpires more slowly after being subjected to water stress because stressed plants have more lipids on the leaf surface (Clark and Levitt, 1956).

TE or WUE could not be exploited as it is often associated with reduced dry matter accumulation and yield. Passioura (1986) proposed a yield model wherein seed yield = WUE x T x HI. Selection for high WUE often resulted in decrease in CGR. Variations in WUE are brought about by stomatal diffusive characteristics (gs) and / or intrinsic photosynthetic capacity (gm). Most often plants have evolved to maximize WUE through a reduction in T that is linked with the g_s. Since DMP is strongly associated with total T, any reduction in T results in reduced CGR. Since of g_s is associated both with

T and internal CO_2 partial pressure (Pi), WUE and T become strongly interdependent. Moreover, the variation in Pi and hence WUE are brought about by gm, the inter dependency between T and WUE will result in higher CGR. The two traits WUE and T are strongly interrelated which is not desirable. Therefore identification of types with lower interdependency are sought (Udaykumar *et al*., 1998).

WUE and carbon isotope discrimination ratio (Δ) are related. WUE, which ranged from 1.81 to 3.15 g/kg was negatively correlated with Δ, which ranged from 19.1 to 21.8 per cent. Δ is a useful trait for selecting genotypes of peanut with improved WUE (Table 4). A strong negative relationship existed between WUE and SLA and between Δ and SLA, indicating that genotypes with thicker leaves (lower SLA) had greater WUE. SLA used as a rapid inexpensive selection index for high WUE in peanut (Wright and Nageswara Rao, 1994).

Transportation rate was lower and TE was higher during early than during late reproductive phase when PAR was above 1500 μ mol $m^{-2}s^{-1}$ (Table 4) due to any developmental effect in castor (Balasubramanian and Venkateswarlu, 1995).

The transpiration rate in castor increased even when LWP decreased indicating environmental control of water loss. LWP was found almost constant over a wide range of T (Jones, 1983). Under low soil moisture

situation also, T was enhanced to meet the atmospheric demand (Carlson and Lynn, 1991).

Water stress reduced WUE in groundnut (Dhopte *et al*., 1991). Rasario and Fujardo (1988), observed significant differences in root weight under stress with decrease in WUE.

The chlorophyll content exhibited a strong positive relationship with TE (8.71 mg/g) in groundnut (Reddy *et al*., 2000). Increase in total chlorophyll content under moisture stress in groundnut has been reported earlier (Reddy *et al*., 1980), but this trait was not studied in relation to the TE of a genotype.

The hypothesis postulated by Nageswara and Wright (1994) reported that genotypes with lower SLA had more photosynthetic machinery, supporting strong positive relationship between the leaf chlorophyll content and the TE.

Table 4 : Influence of morphophysiological parameters on total water transpiration

S.No.	Plant characters influenced by T/TE	Values (Range)	Crop	Reference
1.	When TE is 3.2 g kg^{-1} SLA is	117.3 cm^2 g^{-1}	Groundnut	Reddy *et al*., 2000
2.	When WUE is 1.81 to 31.5 g/kg, then carbon isotope discrimination	1.91 to 21.8 % (per mil)	Groundnut	Nageswara Rao and Wright, 1994
3.	TE at PAR > 1500 μ mol m^{-2} s^{-1} in early development stage	0.72 ± 0.044 (mg CO_2 gH_2O^{-1})	Castor	Balasubramanian *et al*., 1995
4.	TE at PAR > 1500 μ mol m^{-2} s^{-1} in late development stage	0.63 ± 0.037 (mg CO_2 gH_2O^{-1})	Castor	Balasubramanian *et al*., 1995
5.	Stomatal conductance	664 ± 58.1 (m mol m^{-2} S^{-1})	Castor	Balasubramanian *et al*., 1995
6.	Total chlorophyll content	8.71 mg g^{-1}	Groundnut	Reddy *et al*., 2000

S.No.	Plant characters influenced by T/TE	Values (Range)	Crop	Reference

2.7 LEAF WATER POTENTIAL (Ψ_1)

The water potential of plant tissue is a standard means of expression of plant water status. Ψ describes the relative movement of water and it is defined as the amount of work that would be required to move the water from solution state to pure water at a potential of zero (Nilsen and Orcutt, 1996).

When a plant is subjected to water stress, stomatal closure accounts for most of the decline in the photosynthetic rate in short term water stress experiments, while non stomatal inhibition is a major factor for longer periods of stress (Ackerson *et al*., 1977).

Water potential is useful in dealing with water transport in the soil plant atmosphere continuum (SPAC). The stomata during the nights close. Plants will try to recover the lost water from the soil. Upon recovery, the water potential of the plant is equal to the soil WP. When soil supply is not adequate plant shows wilting symptoms but when plant WP is equal to soil WP and plants do not recover during the night it cause permanent wilting (PWP) in plants which occurs at 1.5 MPa. At dawn, plant WP are not affected by the low atmospheric vapour pressure and by soil hydraulic water conductance (Nilsen and Orcutt, 1996).

Water moves in a plant in a continuous column from root surface to mesophyll cells. The driving force for the water is the difference between Ψ at atmosphere and soil.

Various plant spp. show difference in their tolerance to stress, dehydration and their recovering ability. Tolerant plant (viz., *Tortula*) should regain Ψ within 30 minutes of dehydration and this happens by converting its cytoplasm into a glass state by accumulation of a low molecular weight carbohydrate, raphanose (Nilsen and Orcutt, 1996).

Maize seedlings upon exposure to drought for 0, 48, 72 h. RWC and Ψ deteriorated. At 48 h, after withholding irrigation, RWC decreased about 12.9 per cent and Ψ became -0.24 MPa. At 72 h, RWC and Ψ declined to 33.3 per cent and -0.76 MPa respectively, leaf water status recovered to the control level water stressed seedlings were rewatered for 24 h (Hsiao *et al*., 1973).

Studies in groundnut indicate reduced transpiration due to drought stress which could lead to Ψ of -3.0 to -4.5 MPa (Bhagsari *et al*., 1976) and in irrigated plants at around -1.2 to -1.3 MPa (Pallas *et al*., 1979). Patel *et al*., showed that Ψ decreased from -1.0 to -1.8 MPa with a decrease in soil water potential from -0.05 to -2.0 MPa. When seasonal evapotranspiration of 47 mm, the Ψ reached to -6.3 MPa in groundnut (Sarma, 1984).

Stomatal conductance and photosynthetic responses decreased the Ψw during the water stress in genotypes of sunflower. Acclimation to drought resulted in the maintenance of the same levels (50 %) of photosynthesis at Ψ

that were 0.43 and 0.45 MPa lower than in hybrids. WUE was highly improved at Ψw values between -1.2 and -1.8 MPa (Fahima Mojayad *et al*., 1994).

Table 5 : Effects of LWP on different parameters

S.No.	Plant characters influenced by LWP	Values (Range) MPa	Crop	Reference
1.	LWP in stress conditions	-1.0 to -1.5 MPa	Castor	Babitha, 2003
2a.	When T was reduced, Ψ are	-3.0 to -4.5 MPa	Groundnut	Bhagsari *et al*., 1970
b.	In frequently irrigated plants Ψ	-1.3 MPa	Groundnut	Allen *et al*., 1976
c.	when ET was 47 mm, Ψ was	-6.3 MPa	Groundnut	Sarma, 1984
3.	WUE increased at Ψ_w	-1.2 and -1.8 MPa	Sunflower	Fahima Mojayed *et al*., 1994
4a	At 1800 μ mol quantum and low VPD	-13.3 bars	Castor	Ziyu Dai *et al*., 1994
b	When VPD was increased	-13.6 bars	Castor	Ziyu Dai *et al*., 1994
5.	During dry spell, LWP are	-8 to -19 bars	Groundnut	Ramesh *et al*., 2003

In castor, at 1800 μ mol m^{-2} s^{-1} quantum, 30^{o}C and low VPD, the Ψw was -13.3 bars. Gradually increasing the VPD over about 3 to 4 hours to 36 m bar, the Ψ was -13.6 bars. Thus, the Ψ was only slightly more negative after exposure of the leaf to a high VPD (Ziyu Dai *et al*., 1994).

During the stress period moisture stress significantly decreased Ψ_1 of all genotypes of castor compared to irrigated conditions. Genotypes showed a

slightly declining trend in Ψ_1 with the age of the plant and as the stress progressed. The values were -0.5 to 1.0 MPa in irrigated and -1.0 to 1.5 MPa in stress conditions (Babitha, 2003).

During dry spell, leaf water potential in groundnut cultivars varied between -8 and -19 bars. The high water status of the cultivars during the dry spell indicated their ability to tolerate drought (Ramesh *et al*., 2003).

2.8 OSMOTIC ADJUSTMENT

Osmotic adjustment or accumulation of solute by cells is defined as a process by which water potential (Ψ) decreased without an accompanying decrease in turgor (Levitt, 1980).

The term OA is widely used to describe ormoregulation in response to water stress in higher plants wherein it is used to describe changes in solute content after plants recovered from water stress (Turner and Jones, 1980).

In addition, OA increases HI by maintaining assimilate transfer to the grain and delaying senescence (Turner, 1997). OA benefits grain yield by reducing the likelihood of ABA induced pollen sterility (Morgan, 1984).

OA arises from the decreased leaf growth at higher leaf water potential, thereby leading to a passive accumulation of solutes (Turner and Jones, 1980). Cowpea in which photosynthesis is as sensitive to water deficits, leaf expansion is unlikely to osmotically adjust unless hydrolysis of reserves occurs (Nagarajah and Schulze, 1983).

Considerable evidence suggests that OA allows photosynthesis to occur at lower leaf water potentials (Gunasekara *et al*., 1994). Munns (1988) point out that OA has no direct control upon growth and stomatal conductance but appears to be important in drought resistance mechanism for maintaining productive process survival of the developing apex and leaves.

Babita (2003) observed that in castor OA increased from 11 to 33 days after imposing moisture stress. Maximum OA has recorded at 33 days after imposing stress and declined there after even after imposing stress. OA of castor genotypes was found to have a significant positive correlation with total dry matter (r = 0.87) and CGR (r = 0.92) under moisture stress treatment at 33 days and thereafter (r = 0.076). OA has negative correlation with SLA and significant positive correlation with total seed yield (Table 6) (Babitha, 2003).

Solute accumulation enables cell enlargement and growth to proceed at lower water potentials. Major osmotic constituents responsible for decrease in Ψs are increased levels of soluble sugars, amino acids and organic acids accompanied with change in potassium (Ford and Wilson, 1981).

Osmtic potential, presence of solutes reduce free energy of water, primarily because the solute ions or molecules attract the water molecules. OP has little effect on mass movement of water in soils. In soil high in soluble salts, OP may be greater in the soil solution than in the plant root cells, thus leading to constraints in water uptake by plants. As the plant take up water due

to OP differences, its movement is restricted by the elasticity of cell walls (Reddy, 1999).

The active reduction of Ψs in response to water deficits facilitate maintenance of turgor potential (Hsiao *et al*., 1976). Decrease in OP maintained positive turgor in sorghum and cotton (Ackerson *et al*., 1977). Resistant cultivars of oilseed *Brassica juncea* developed a significantly lower Ψs after 8 days of water stress at the seed initiation stage. This contributed towards better OA, less inhibition of photosynthesis and higher WUE of the resistant cultivars. Susceptible cultivar showed a greater decline in rate of photosynthesis than the rate of transpiration. Thus, the poor performance of susceptible cultivar under moisture deficiency could be a more extensive stress induced damage to the photosynthetic apparatus (Veena Sawhney *et al*., 1996).

No significant differences were observed in the OP (Ψs) of castor genotypes under irrigated conditions. Ψs gradually decreased with age and under stress and reached to minimum by the end of stress period (-1 to 1.5 in irrigated and -1.0 to -2 MPa in stress) (Babitha, 2003).

The magnitude of OA depends on the degree of water stress. Plant leaves that are capable of OA can maintain turgor at low water potentials than non adjusted leaves (Levitt, 1980).

To maintain water potential equilibrium of cytoplasm outside the vacuole during stress some other organic solutes called as compatible solutes or osmolytes like prolime accumulate (Levitt, 1980).

The degree of osmoregulation is affected by both the rate of stress and stress preconditioning. Solutes which accumulate as the result of pre-stressing are eventually dissipated if no further water stress occurs. The rate of dissipation depends on the drought hardening conditions (Wilson *et al*., 1983).

Plants with high osmoregulation showed higher dry weight per unit ground area of grain, stem and root. The increased values were due to the maintenance of higher leaf area index and net photosynthesis per unit leaf area during water stress (Wright *et al*., 1983).

The additional dry matter produced with higher osmoregulation will depended upon the additional water extracted from the soil, because of higher root length density (Passioura, 1982).

Table 6 : Influence of morphophysiological parameters on OA/osmoregulation

S. No.	Plant characters influenced by OA	Values (range)	Crop	Reference
1.	Solute accumulation diurmal values	0.4 MPa	Soyabean	Wenkert *et al*., 1978
		0.6 MPa	Sunflower	Taxami *et al*., 1982
2.	Change in osmotic potential	0.1 to 0.4	Sunflower	Turner *et al*., 1978
3.	Reduction in PAR	1500 $\mu E\ m^{-2}\ S^{-1}$ to 30-50 $\mu E\ m^{-2}\ S^{-1}$	Sorghum	Acevedo *et al*., 1979
4.	At full turgor water potential	0 to -1.5 MPa	Wheat	Morgan J M., 1980
	At zero turgor Ψ	-2.0 MPa		

S. No.	Plant characters influenced by OA	Values (range)	Crop	Reference
	Ψ at stomatal closure	0.6 MPa	Sorghum	Wright *et al*., 1983
5.	At high osmoregulation yield	1.5 t/ha	Wheat	Morgan J M., 1980
	At low osmoregulation yield	1.0 t/ha	Wheat	Morgan J M., 1980
	Total dry matter	r = 0.87 (70-200 gm^{-2})	Castor	Babitha, 2003
	SLA	r = 0.71 (200-160)	Castor	Babitha, 2003
	CGR	r = 0.92 (0 to 3 g m^{-2} d^{-1})	Castor	Babitha, 2003
	LAI	r = 0.76 (0 to 1.5)	Castor	Babitha, 2003
	Total seed yield	r = 0.908 (100-200 g m^{-2})	Castor	Babitha, 2003
	Primary spike yield	r = 0.821 (10-25 g m^{-2})	Castor	Babitha, 2003
	Secondary spike yield	r = 512 (25-50)	Castor	Babitha, 2003
	Tertiary spike yield	r = 0.497 (50-100)	Castor	Babitha, 2003

2.9 SPAD CHLOROPHYLL CONTENT

SPAD-Chlorophyll meter has proven effective in determining the N status of rice (Turner and Jund, 1991). A close correlation was shown between maximum single leaf net photosynthetic rate under saturating light and leaf N content per unit leaf area under field conditions in tropics (Peng *et al*.,1995). Photosynthetic rates and radiation use efficiency increase with higher concentration of leaf nitrogen but this relationship changes with crop ontogeny (Hasegawa and Horie, 1996).

Photosynthetic activities, specific leaf weight (SLW), chlorophyll contents in terms of SPAD value and nitrogen concentrations were greater in the uppermost most compared to lower leaves in rice (Sato and Kim, 1980).

Saitoh *et al*. (1991) reported significant correlation between Pn with nitrogen concentration and chlorophyll content in the leaves of various positions on the stem and of various ages. SLW and N per cent were non significant and relation of chlorophyll in terms of SPAD value to leaf N contents (% DW) however was highly significant (Turner and Jund, 1991).

Chlorophyll content (SPAD value) measured by the chlorophyll meter gave best indication of Pn as compared to other parameters. It is therefore, possible that a constant SPAD value could be used directly as a threshold for timing of N top dressing.

The effects of sampling (leaf position, time of sampling and leaf water status) and climatic factors (solar radiation and vapour pressure deficit) on SLA and SPAD chlorophyll meter reading (SCMR) were studied in a range of peanut genotypes. Solar radiation and VPD during the sampling period had a significant influence on the relationship between SLA and SCMR, largely through their effects on SLA. However, standardization of SLA for these environmental factors significantly improved the relationship between SLA and SCMR from -0.50 to -0.80 suggesting that SCMR can be a surrogate measure of SLA. The overall relation between SLA and SCMR was negative but not highly significant, in groundnut (Rao *et al*., 2001).

2.10 WATER STRESS AND YIELD

2.10.1 Yield components

Yield is a complex character, highly influenced by the environment. In castor, length of main spike increases at higher levels of irrigation which ultimately resulted in higher yields (Rao, 1979; Sailasree, 2001). Increased yield was due to increase in number of spikes per plant at higher levels of irrigation in castor (Sailasree, 2001). In sunflower, there was a reduction in the number of seeds per plant under stress (Cox and Jolliff, 1986; Balasubramian and Maheswari, 1991). In other water stress treatments, number of seeds per inflorescence were not attested though it resulted in significantly smaller heads and seeds in sunflower (Human *et al*., 1990) and castor (DOR, 1994). The reduction in seed number was apparently compensated for by the increase in seed weight in sunflower (Eck *et al*., 1987).

Kernal weight was reduced under stress in groundnut (Ramana Rao, 1994). In some cases of castor, seed weight increased but this could not fully compensate for the decrease in the number of seeds and thus total yield reduced (Weiss, 1971). However, irrigation treatments did not significantly influence 100 seed weight in sunflower (DOR, 1987), groundnut (Gowda and Megde, 1986) and castor (DOR, 1994).

2.10.2 Seed yield

Prolonged water shortages affect virtually all metabolic processes and often result in severe reductions in plant productivity (Bohnert *et al*., 1995). The capacity for DMP and its subsequent allocation to economically important plant parts are two major determinants of yield in a crop (Watson, 1952). The effects of water stress on growth and yield depend both on stress and on the stage of growth at which stress occurs (Garrity and Oille, 1994). The final yield in crop plants is the result of complementary functioning and relation of source and sink components (Sinha and Khanna, 1975). under stress significant reduction in seed yield has been reported in sunflower (Teama and Mahmoud, 1994), groundnut (Senthong 1989) and in castor (Wali *et al*, 1991).

2.10.3 Harvest index

Moisture stress resulted in a decrease in HI of groundnut (Samsukumar, 1991). On the other hand water stress did not affect HI of sunflower (Cox and Jolliff, 1986) and sometimes even resulted in higher HI in sunflower (Baldini *et al*., 1999) and mungbean genotypes (Rao *et al*., 2001). Such reduction in dry matter under stress if coupled with increased seed weight could increase HI.

Growth stages of castor

In castor, there are distinct developmental stages like formation of the central spike, flowering and ripening of seeds of the central and lateral spikes of the first, second and subsequent order (Moshkin, 1986). Castor, crop

showed 12 different stages which could be subjected to stress (Rzhanova *et al*., 1962) (Table 7).

Table 7 : Different growth stages of castor

Stage No.	Name of the stage
1	Germination (9-18 days)
2	Formation of two opposite true leaves (7-17 days)
3	Segmentation of axis of raceme, with the initiation of lateral axillary buds
4	Differentiation of primary meristem and formation of rudimentary raceme lasts for 7-18 days
5	Differentiation of floral parts (10-17 days)
6	Formation of pollen and embryonic Sac
7	Growth of the raceme
8	Budding (Spike emerging out of the covering leaves)
9	Flowering and pollination (10-14 days)
10	Formation of capsules and seeds (8-10 days)
11	Beginning of waxy maturity (12-16 days)
12	Onset of complete maturity (14-18 days)

Solar radiation and air humidity influence growth and water relations of crops. Fluctuation in solar radiation integrity is large during rainy season. The reproductive phases, particularly the development of secondary and tertiary racemes, depends on residual soil moisture and unseasonal rainfall, if any when PAR was above 1500 μ mol m^{-2} s^{-1}. Transpiration rate was lower and TE was higher during early than during late reproductive phase. This was due to larger VPD during post rainy period, rather than due to any development

effect. LWP was higher during early than during late reproductive phase (Balasubramanian *et al*., 1995).

Varietal Development (AP and India level)

The local varieties of castor HC-1 to HC-8 traditionally grown prior to 1960's in Telangana (Andhra Pradesh) region were flowering in the rainy season and maturing with residual moisture. In 70's, development of Aruna variety through irradiation of local variety HC-6 enabled to identify a mutant and thereby reducing days to floral initiation of primaries from 120 to 45 days and maturity from 200 to 90 days. Aruna is the first variety that tolerates to drought and thus replaced local varieties completely in the entire rainfed areas of India. A short duration variety, SA_2 flowering and maturing in 95 and 100 days, respectively was developed in Tamil Nadu. Other short duration varieties like Bhagya (R-63) and V1-9 developed at Hyderabad and SK Nagar showed highest moisture use efficiency in Parbhani of Western Maharastra due to their efficient utilisation of soil moisture during capsule development. Entire crop improvement was based on early maturity leading to the release of varieties like Jyothi and Kranthi in A.P, TMV-4 and TMV-5 in Tamil Nadu and Vijapur inbred – 9 (VI-9) and GC-2 in Gujarat.

Release of the hybrid GAUCH-1 in the rainfed areas further improved and stabilized yields. In loam soils of Saurashtra region of Gujarat, GAUCH-1 and GAUC the male parent of the hybrid gave highest yield, utilizing rainfall

and conserved moisture during the post rainy season. Hybrid GCH-4 was more responsive to flavourable environment and highly predictable across environments while GAUCH-1 was found suitable for all environments, while varieties SA 2, Aruna, 48-1, Kranthi, DCS-33, GAUCH-1, DCH-158 and DCH-141 were found relatively tolerant to salinity (Hanumantha Rao and Lavanya, DOR, 2003).

IN VITRO STUDIES

The development of first completely defined medium provided the basis for the rapid development of tissue culture studies (Murashige and Skoog, 1962).

2.11 CULTURING OF SHOOT TIPS

Shoot tip shows potential for unlimited growth because of the presence of apical meristems constantly undergoing cell division and cell differentiation. Commonly a single shoot arises from culture of one shoot tip, but can be made to produce multiple shoots in special instances on excision and removal of these shoots from the proliferating area and upon transfer to a microenvironment. The isolated new shoots can be rooted and complete plants recovered in large numbers and in quicker time (Narayanaswamy, 1994).

Various factors of an explant tissue. Those are physiological age of tissue, size of the explant and quality of source plant material influence the growth *in vitro*.

Explants from embryo axes and shoot tips were cultured on MS medium supplemented with 0.5-10 mg/l adenine, BA, Kinetin, thiadiazuron (TDZ). TDZ @1.0-10.0 mg/l gave the maximum number of shoots (37.8 to 40.0) from embryo axis, while BA (2.0 mg / lit) was found superior to other cytokinins for obtaining higher number of shoots (47.7) from the shoot apex (Sujatha and Reddy, 1998).

2.12 DIRECT SHOOT REGENERATION

Multiple shoots could be proliferated directly using shoot tip as explants in linseed (Lane, 1979), castor (Prasanna Athma and Reddy, 1983, Sujatha and Reddy, 1998, Biju *et al*., 2002 and Rampal *et al*., 2003).

In sunflower, Lupi *et al*. (1987) reported that shoot apices cultured on MS + 2 mg / lit kin + 5 mg / lit GA_3 resulted in direct regeneration of 9.8 shoots / apex. Weber *et al*. (2000) reported that shoot apices cultured on MS + 0.1 mg /l. BAP resulted in direct regeneration of 3.5 shoots / apex. Multiple shoots could be proliferated in castor when cytokinin was supplied singly as BA 0.5 to 2 mg/l (Prasanna Athma and Reddy, 1987), 5.0 mg/l (Biju *et al*., 2002), 12 mg/l (Sujatha and Reddy, 1998). GA_3 reduces the total number of shoots per explant (Sujatha and Reddy, 1998).

2.13 CULTURING OF APICAL MERISTEMS

Apical meristem is the tissue located within shoot tip and appears as a dome like structure distal to youngest primordium. Meristems measure less than 0.1 mm in length and is the site of apical divisions (Phillips and

Gamborg, 1995). The culture of excised apical dome with one or two leaf primordial and isolated from the terminal shoots has a better chance of survival and in development of plants. Meristem offer scope for experimental interaction in the production of more shoot meristems either by the development of multiple shoot apices, lateral buds or by somatic embryogenesis (Narayanaswamy, 1994).

2.14 DIRECT ORGANOGENESIS

MS medium was found more suitable for direct regeneration of shootlets and a maximum of 173 shoots were produced in groundnut (Radha Krishnan *et al*., 2000).

Phytoharmones like BA was more effective than any other cytokinin in direct organogenesis in most oilseed crops like castor (Sarvesh and Ram Rao, 1992), niger (Sarvesh *et al*.,1993), groundnut (Radhakrishna *et al*., 2000), sunflower (George *et al*., 1989, Shiv *et al*., 2000).

Apart from plant growth regulators, addition of some inorganic compound like KNO_3 in sunflower (Berrios *et al*., 1999) and cassien hydrolysate in groundnut (Nikam and Shitole, 1999) showed synergistic effect on multiple shoot induction.

2.15 STUDIES ON DROUGHT STRESS IN CULTURES

2.15.1 Development of stress tolerant cell lines

A potential application of tissue and cell culture is for the production of novel genotypes with valuable attributes for agriculture particularly in water and salt stress tolerance (Srivastava *et al*., 1995).

A gradual exposure to higher levels of stress is more promising for raising stress tolerant plants rather than through shock treatment (Srivastava, 1998). Development of stress tolerate cell lines by somaclonal selection *in vitro* is a promising technique as these lines may adjust by their cellular mechanisms to those hostile environments (Purushotham *et al*., 1998). However, water stress tolerant cell lines also represent ideal system for assessing the physiological effects of water stress at cellular levels. Further such cell lines when compared with the normal sensitive callus can provide a useful means of measuring the capacity and range of stress tolerance and may be used for elucidating tolerance mechanism (Srivastava *et al*., 1995).

There are several organic and inorganic solutes, which are used to induce drought stress in media viz., polyethelen glyclol, mannitol and sodium chloride. Among these PEG is the mostly used stress inducing agent.

2.15.2 Polyethylene glycol (PEG)

PEG, a drought stress inducing agent is of several types based on its molecular weight. The mode of action is, it induces water stress in the medium by acting as a non-penetrating osmotic agent that lowers the water potential of the medium thus creating water stress in explants or callus. Explants exposed

to increasing concentration of PEG can tolerate increased drought stress levels (Srivastava *et al*., 1995).

2.15.2.1 Studies on regeneration under induced drought stress

Hypocotyl explants of castor genotypes produced callus in large amounts in control and also in PEG induced stress treatments of -3 bars. At increased stress of -6 and -9 bars explant growth was effected resulting in small sized callus. The protein content and total sugar content increased with increase in PEG concentration from -3 to -6 bars and declined at -9 bars as compared to control (Manjula *et al*., 2001). Enhanced levels of praline in the callus under PEG induced stress was also reported in groundnut (Purushotham *et al*., 1998). The increased praline content helps in tolerating water stress either by rehydration of the protoplasm or by providing energy for recovery from stress (Khidse *et al*., 1982).

Osmotic stress of -3, -6, -9 bars induced by incorporating polyethylene glycol into Murashige and Shoog (MS) medium containing various cytokinins revealed that increased stress levels of -6, -9 bars decreased the number of castor shoots produced *in vitro* (Rampal *et al*., 2003).

Number of shoots initiated varied with respect to genotype and PGR's used. The shoot tip explants of kranthi produced a mean number of 4.4 axillary shoots as compared to 3.7 shoots in GCH-4. Such differential response between genotypes was also observed in sunflower crop, where in C-108 and C-96 produced 27 and 2 shoots on MS + BA 2.0 mg/lit respective

(Berrios *et al*., 1999). The PGRs incorporated into the medium in term influenced the genotypic response. Mollina and Schobert (1995) reported that, combination of NAA + BA was more effective in inducing multiple shoot buds in castor. Sujatha and Reddy (1998) however, suggested that exogenous supply of auxin was not required as the shoot tip explants themselves are the sites of auxin synthesis. These findings in castor indicate that the effect of both PGR and genotype are important in determining the shoot bud induction.

Table 8 : Inorganic and organic constituents of MS medium

Stock solution	Constituents	Mg/lit and magnification	Stock solution prepared (ml)	Volume of stock solution used (ml)
A	$NH_4 NO_3$	1650 x 10	200	20
1	KNO_3	1900 x 10	200	20
	$MgSO_4 7H_2O$	370 x 10	200	20
	KH_2PO_4	170 x 10	200	20
B	$CaCl_2 2H_2O$	440 x 10	200	20
C	KI	0.83 x 100	200	20
D	H_3BO_3	6.2 x 10	200	20
	$MnSO_4 4H_2O$	22.3 x 10	200	20
	$ZnSO_4 7H_2O$	8.6 x 10	200	20
E	$Na_2MoO_4 7H_2O$	0.025 x 100	200	2
	$CuSO_4 5H_2O$	0.025 x 100	200	2
	$CaCl_2 6H_2O$	0.025 x 100	200	2
F	Na_2 EDTA	37.3 x 10	200	20
	$FeSO_4 7H_2O$	27.8 x 11	200	20
G	Glycine	2 x 25	250	10
	Thiamine Hcl	0.1 x 25	250	10
	Nicotinic acid	0.5 x 25	250	10
	Pyridioxime Hcl	0.5 x 25	250	10
	Myo-inositol	100	Added directly	
	Sucrose	30 g/lit	Added directly	
	Agar Agar	8 g/lit	Added directly	
	pH	5.8		

CHAPTER III

MATERIALS AND METHODS

This chapter elicits the information about materials used and methods adopted to achieve the set forth objective of the study titled as "Studies on water stress recovery and regrowth in genotypes of castor (*Ricinus communis* L.)".

3.1 LOCATION

The present research work was conducted at Agricultural College Farm, Rajendranagar, Hyderabad during the kharif season of 2003-04. *In vitro* studies were carried out in the Department of Plant Physiology, Tissue Culture Lab.

3.2 EXPERIMENTAL DETAILS

The experiment was laid out in a Randomised Block Design with four replications and six genotypes viz., PCS-124, VP-1, Kranthi, PCS-136, PCS-137, PCS-138. The seed material was obtained from Regional Agricultural Research Station, Palem.

The seed was treated with Bavistin @ 3 g/kg to protect them from seed borne diseases. Seed was sown at a depth of 5 cm by adopting recommended spacing of 90 x 30 cm in plot area of 4.5 x 4.5 m.

3.2.1 Fertilizer application

Fertilizer was applied to the crop as per the recommended dose @ 40 : 40 : 30 kg N, P_2O_5 and K_2O ha^{-1}. Half of the recommended dose of N along with the entire dose of P and K was applied as basal dose. Remaining N was applied at 30 DAS.

3.2.2 Plant protection

The crop was protected from most common pests like Spodoptera, Helicoverpa and castor semilooper by spraying monocrotophos @ 0.1 per cent as and when pest attacked.

3.2.3 Harvesting

Harvesting was done three times at maturity of primary spike and subsequently at secondary and tertiary spike maturity. The harvested spikes were heaped, sundried and threshed manually by beating with sticks. The threshed produce was winnowed and seed was separated from stalk and the seed yield and stalk yield per each plot were recorded separately after drying.

3.3 PHYSIOLOGICAL AND BIOCHEMICAL OBSERVATIONS

3.3.1 Soil moisture content

Soil moisture content was measured gravimetrically following the procedure of Dastane (1972). Soil samples were taken from profile depth of 15-30 cm from each plot with the help of soil augar. The collected soil was placed in alluminium tins and weighed to get the moist weight of the soil. Later, the samples were dried in an oven at 105^{o}C for 24 hours or more till

constant weight was obtained. The moisture content in the soil was calculated by the formula.

$$Pw = W_w - W_d / W_d \times 100$$

Where Pw is the moisture percentage on dry weight basis.

W_w is the weight of wet soil.

W_d is the weight of dry soil

3.3.2 Leaf water potential (Ψ_l)

Leaf water potential expressed in Mega Pascals was measured using the Pressure bomb apparatus following the procedure of Scholandar *et al*. (1965), For measurement, the fully opened 2nd and 3rd leaf was selected and inserted into the Chamber top with the help of rubber stop, pressure released and the xylem tap exudation was observed. The pressure recorded gave water potential of leaf.

3.3.3 Osmotic potential (OP)

Osmotic potential of cell sap extracted from index leaf (2nd or 3rd) from top and estimated using Wescor vapour pressure osmometer. The values were expressed in Mega Pascals. The 2nd or 3rd leaf from the top used in measuring water potential was utilized for recording OP. The sample was initially blotted on a filter paper to wipe off the moisture if any. Later wrapped in an aluminium foil and finally kept in the plastic cover and labeled separately. The samples were placed in the ice box and preserved in dry freeze having -80^{o}C temperature. The leaf samples were crushed in between the fingers. For cell

sap extraction, the exuded extract was blotted on filter discs and osmotic potential estimated by osmometer.

3.3.4 Chlorophyll content (SPAD)

The chlorophyll content of the leaf was estimated using the SPAD meter (SPAD-502, Minolta Camera Corp, Ramsey, NJ). SPAD meter readings are based on the difference between light attenuation at 430 nm (the peak waved length for chlorophyll a and b) and that 750 nm (near infrared) with no transmittance. Data collection involved placing of second leaf from top below the apex on the main axis of the plant intact in the SPAD meter and recording the reading following the procedure of Nageswara Rao and Wright (2001).

3.3.5 Transpiration

Transpiration was measured using steady State Porometer (LI 1600). The readings were taken on the day time when there was no shade. The sensor head was connected to instrument and null adjustment was made. The selected leaf was placed on sensor head to record the readings of transpiration.

3.4 GROWTH PARAMETERS

3.4.1 Total dry matter

Five plants from each plot were uprooted at 15 day interval for estimation of dry matter per plant. The roots of the selected plants were separated and the remaining haulms were cleaned and separated into different plant parts viz., stem, leaf and spike. The separated plant parts were dried in a hot air oven at 70^{o}C for 48 hours and the weights recorded.

3.4.2 Leaf area (cm^2)

Leaf area was measured at 15 days interval from five randomly selected plants using LI-3100 leaf area meter (LICOR, Lincoln, Nebraska, USA). The average leaf area was reported and expressed as leaf area per plant. LAI was computed and expressed as LA/m^2.

3.4.3 Growth analysis

From the measured total dry matter and leaf area, the following growth parameters were calculated.

Crop growth rate (CGR) g m^{-2} d^{-1} = $(W_2 - W_1) / (T_2 - T_1)$ x $\frac{1}{P}$ (Watson, 1952). Where W_1 and W_2 are plant dry weights at times T_1 and T_2 respectively and P is Land area.

Leaf Area Index (LAI) = Leaf area (m^2) / ground area (m^2) (Ashley., 1963)

Specific leaf area (SLA) = Leaf area / leaf dry weight (cm^2 g^{-1})

3.4.4 Yield components and yield

In each treatment, 1 m^2 area was labeled. These plants were harvested separately and combined with net plot yield to obtain data on total yield, spike wise viz., primary, secondary and tertiary orders and expressed in kg ha^{-1}.

3.4.4.1 No. of spikes per plant

Total number of spikes of the five tagged plants of each treatment was counted and the mean expressed as number of spikes per plant.

3.4.1.2 Total and effective spike length

Total spike length (cm) was measured from the peduncle node to the top of the spike on the main axis. The length of the spike from the peduncle node to the point where the capsules appeared at the top of the spike was considered as the effective spike length.

3.4.1.3 100-seed weight

Hundred seeds were collected randomly and weight was recorded.

3.4.1.4 Harvest Index (%)

Harvest index (HI) was calculated following the formula.

HI (%) = Seed yield (kg ha^{-1}) / Biological yield (seed + stalk yield in kg ha^{-1}) x 100.

3.5 STATISTICAL ANALYSIS

The results were analysed using the factorial RBD method as described in Panse and Sukhatme (1985).

IN VITRO STUDIES

Two cultivars of castor viz., Kranthi and Aruna were used in the present study to assess the stress tolerance and recovery ability in terms of regenerants produced. The seed material was obtained from Regional Agricultural Research Station, Palem and Directorate of Oilseeds Research (DOR), Hyderabad.

3.6 RAISING OF CASTOR SEEDLINGS UNDER *IN VIVO* CONDITIONS

Castor seeds of Kranthi and Aruna were washed under running tap water. The seeds were then sterilized with 0.1 % $HgCl_2$ for 10 minutes followed by rinsing with distilled water thrice and sown in plastic trays filled with acid washed sand for germination.

3.7 COLLECTION OF EXPLANTS FROM THE SEEDLINGS FOR *IN VITRO* CULTURE

Nine day old seedlings were utilized for the study. Shoot tips from seedlings consisted of shoot apical meristem and one to several primordial leaves were excised. Shoot tips of 0.1 to 1 mm length were utilized for *in vitro* culture.

3.8 PREPARATION OF NUTRIENT MEDIUM FOR REGENERATION

In the present investigation for regeneration studies Murashige and Skoog (MS) medium, 1962 was used (Table 8).

3.8.1 Preparation of stock solutions of plant growth regulators

Stock solution of plant growth regulators

BA (Benzyl Adenine) 50 mg was weighed and dissolved in a few drops of 0.1 N HCl and the final volume was made upto 50 ml with distilled water to obtain 1 mg l^{-1} concentration.

3.8.2 Steps involved in media preparation

Stock solution A, B, C, D, E, F and G as described in table 8 were mixed in sequence in distilled water to get the required volume and concentration. Myoinositol 100 mg l^{-1} and sucrose 30 g l^{-1} were added directly to the media. The final volume was made to one litre and plant growth regulators added. The pH of the solution was adjusted finally to 5.8 $\pm$ 2. Agar agar @ 8.0 g l^{-1} was added to boiling solution 20 ml of the medium was dispensed to the culture tubes and closed tightly with cotton plugs and sterilized in autoclave at 1.06 kg/cm^2 pressure at 121^{o}C for 15 min. These tubes were kept slant for gelling. The surface sterilization and inoculation of explants were carried out under the laminar air flow cabinet.

3.8.3 Induction of water stress in MS medium

To study the effect of water stress on regeneration of castor genotypes, various concentrations of polyethelene glycol (PEG) solution were prepared as given in Table 9 following Saxena (1976).

Table 9 : Induction of water stress by PEG in MS media

Water stress (bars)	PEG 6000 (g / 100 ml)
-3	7.78
-4	10.34
-5	12.29
-6	15.55
-9	23.33
-12	31.10

3.8.3.1 Preparation of polyethylene glycol (PEG) media

MS media was prepared in 500 ml of distilled water. Similarly desired amount of PEG 6000 was dissolved in 500 ml distilled water. The two solutions were autoclaved separately. The two solutions were combined aseptically with stirring under laminar airflow cabinet. Then the final medium was distributed in pre-sterilized culture tubes as described by Philips and Gamborg (1995).

3.9 *IN VITRO* CULTURE OF SHOOT TIPS

Shoot tips were excised from 9 day old seedlings. The dissected shoot tips obtained from *in vivo* grown seedlings were surface sterilized with 0.1 % $HgCl_2$ for 10 min and rinsed with sterile double distilled water thrice and then transferred to the culture tubes.

3.10 GROWTH QUANTIFICATION

The growth of shoot tips was quantified by taking observations on various parameters like colour and number of multiple shoots at 14 and 28 days after inoculation (DAI).

3.10.1 Colour of the explant

Changes in the colour of shoot tip was observed visually and grouped as light green, greenish yellow and brown.

3.10.2 Number of multiple shoots per explant

Multiple shoots were observed visually and recorded at 14 and 28 DAI and expressed as number of shoots per shoot tip.

3.11 STATISTICAL ANALYSIS OF THE RESULTS

The results were analysed using two factorial Randomized Block Design following by Panse and Sukhatme (1985).

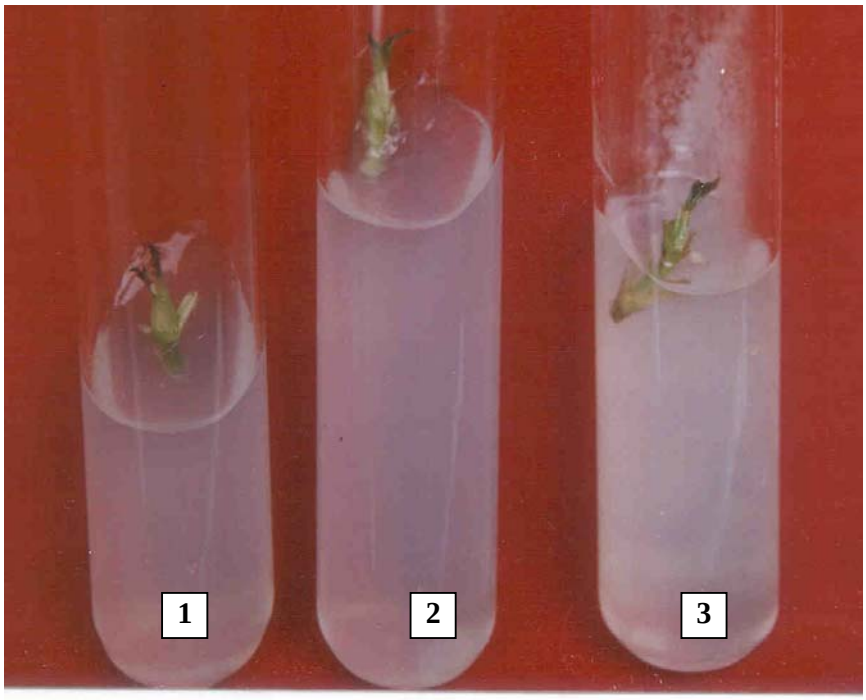

Fig. 3 : Shoot tips of Kranthi cultured on MS + BA 5 mg/l + PEG at -3bars (1), -4 bars (2), -6 bars (3) showing multiple shoots at 28 DAI

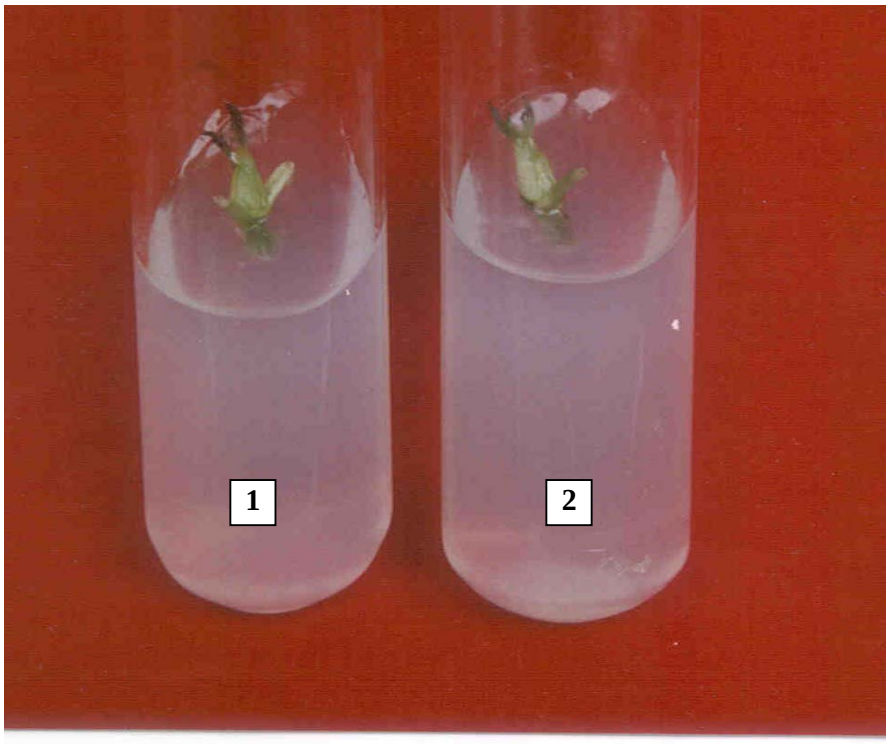

Fig. 4: Shoot tips of Aruna cultured on MS + BA 2 mg/l + PEG at -3bars (1), -4 bars (2) showing multiple shoots at 28 DAI

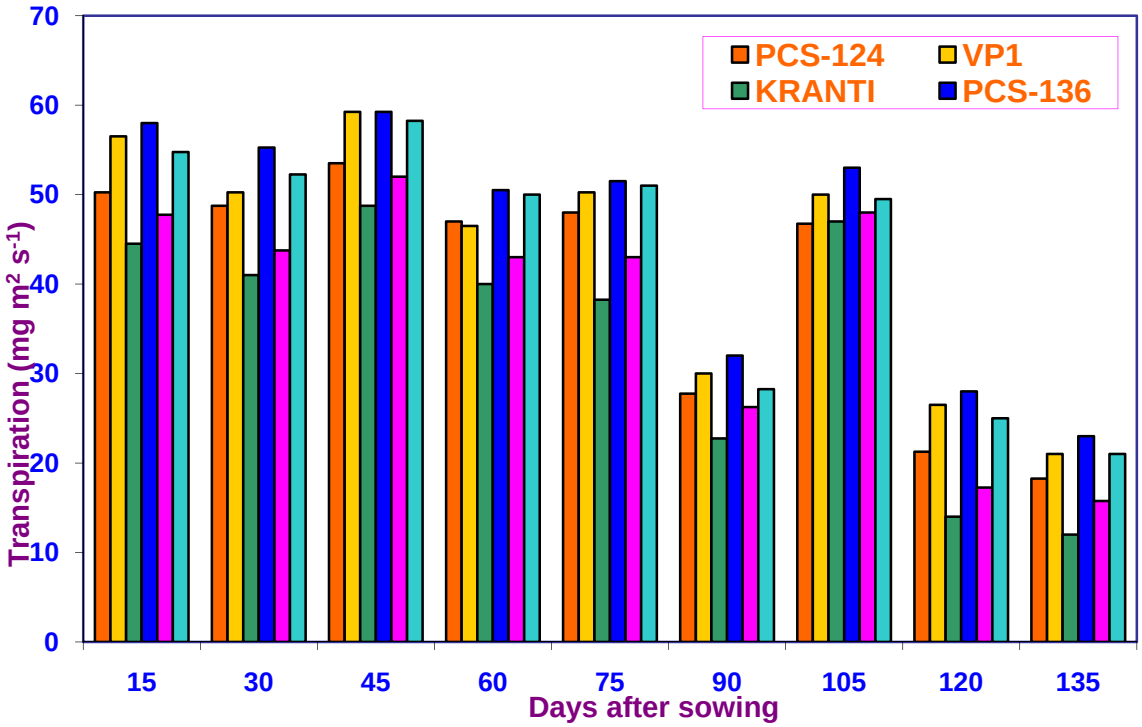

Fig. 1 : Transpiration rate at different days after sowing in six

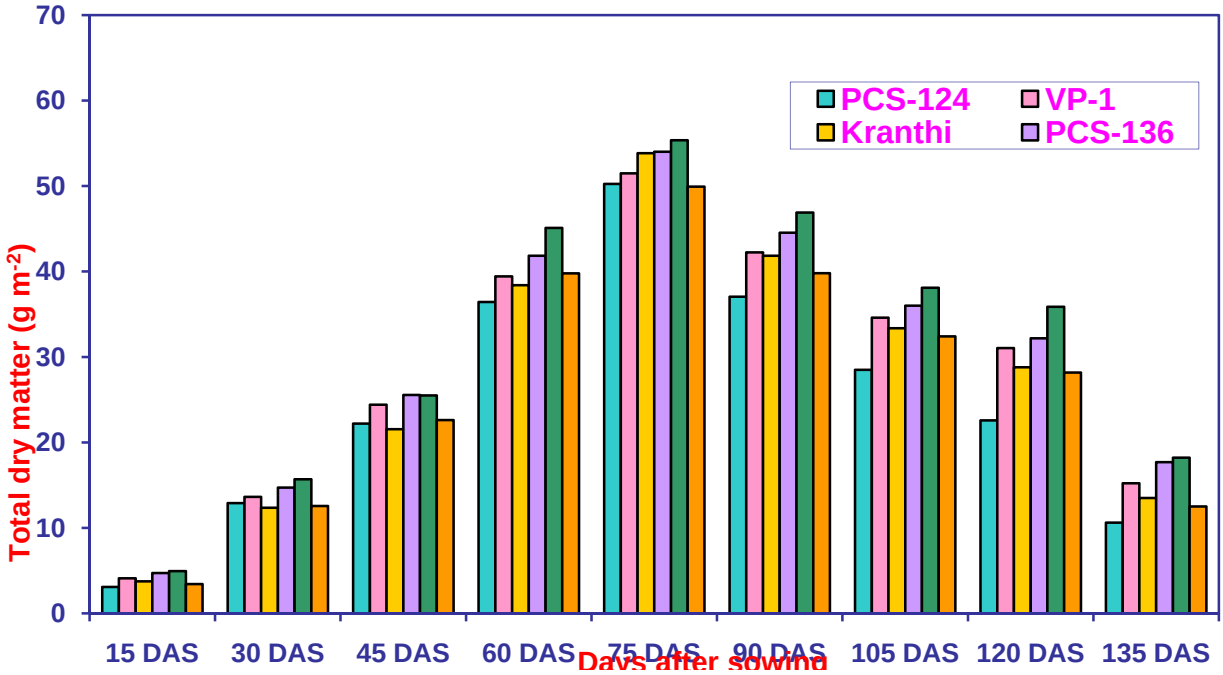

Fig. 2: Total dry matter (g m^{-2}) at different days after sowing in six castor

CHAPTER IV

RESULTS

4.1 SOIL MOISTURE

The mean soil moisture content during the crop growth ranged from 5.00 to 15.47 per cent (Table 10). Two dry spells occurred during crop growth season i.e. between 30-44 DAS and 75-104 DAS wherein the mean soil moisture content was 6.14 per cent and 6.00 per cent respectively indicating that the crop was under stress. The soil moisture content improved with the receipt of rain, consequently at 45 and 105 DAS, the soil moisture values recorded were 13.31 and 8.68 per cent, respectively. During this period recovery and regrowth of genotypes was studied. Scanty rainfall was received after 105 DAS. As a result the soil moisture values were 6.33 and 6.22 per cent, respectively at 120 and 135 DAS.

The mean soil moisture content in the treatments (genotypes) ranged between 7.38 to 7.85 per cent. Maximum soil moisture was observed in the plot with genotype VP-1 (7.85 %) which was on par with PCS-138 (7.78 %) and PCS-124 (7.71 %). The plots containing genotype PCS-137 recorded lowest soil moisture content (7.38 %) which was on par with treatments PCS-136 (7.42 %) and Kranthi (7.50 %).

Treatment x DAS interaction revealed the soil moisture content ranged between 4.75 to 16.25 per cent. During the first dry spell (30-44 DAS), the plot with genotype PCS-136 showed high soil moisture content (6.75 %),

which is on par with PCS-124. The plot having genotype Kranthi recorded lowest soil moisture content (5.75 %) which is on par with PCS-137 (5.75 %) and PCS-138 (5.75 %). During the second dry spell (75-104 DAS), the plot having the genotype PCS-136 recorded lowest soil moisture content (5.25 %) and was on par with PCS-137 (5.37 %). During the extended period of dry spells (90-104 DAS). The plot with the genotypes Kranthi, PCS-124 and PCS-136 recorded low moisture content.

Stress recovery could be quantified at two periods of growth. During the initial period at 45 DAS wherein a mean soil moisture of 13.31 per cent was recorded. Plot with PCS-124 recorded high soil moisture and was on par with plots PCS-136. Later during the second dry spell (75-104 DAS) a mean of 8.68 per cent moisture was recorded. Plots with VP-1 showed high moisture values.

Results indicate that plot containing Kranthi, PCS-137 and PCS-138 recorded lowest soil moisture content at 30 DAS. PCS-136 and 137 at 75 DAS and Kranthi, PCS-124 and 136 at 90 DAS. Soil moisture improved at 45 and 105 DAS indicating relief from stress. Plots with PCS-124 and VP-1 recorded quick recovery.

4.2 TRANSPIRATION

The effect of water stress and recovery on the transpiration in the castor genotypes was recorded using steady state porometer. During the entire stress period the transpiration values decreased significantly in all the genotypes.

The mean values of transpiration ranged from 18.5 μg m^{-2} s^{-1} to 55.16 μg m^{-2} s^{-1} across the days after sowing (Table 11). At 90, 120, 135 DAS, the transpiration values decreased indicating dry spell response. The transpiration values during the period ranged between 18.5 t 27.8 μg m^{-2} s^{-1}. At 105 DAS, the mean transpiration (49.04 μg m^{-2} s^{-1}) increased from 90 DAS (27.83 μg m^{-2} s^{-1}) showing the recovery from water stress.

The mean transpiration values among the genotypes ranged from 34.25 to 45.61 μg m^{-2} s^{-1}. The genotype PCS-136 recorded high transpiration value of 45.61 μg m^{-2} s^{-1} which was on par with VP-1 (43.36 μg m^{-2} s^{-1}) and PCS-138 (43.33 μg m^{-2} s^{-1}). Incidentally two of these genotypes PCS-136 and 138 recorded low soil moisture values during dry spell. During the dry spell all the genotypes showed low transpiration values (Table 10).

Genotypes x DAS interaction showed that the transpiration values ranged between 14.0 and 59.25 μg m^{-2} s^{-1}. During the dry spell at 90-120 DAS, the genotypes, PCS-136 and VP-1 recorded maximum transpiration rate and were on par. At 135 DAS, PCS-136 continued to record maximum transpiration (23 μg m^{-2} s^{-1}) and was on par with VP-1 (21.0 μg m^{-2} s^{-1}) and PCS-138 (21 μg m^{-2} s^{-1}).

Transpiration rate improved at 45 DAS (55.16 μg m^{-2} s^{-1}) and 105 DAS (49.04 μg m^{-2} s^{-1}). Two genotypes PCS-136 and VP-1 showed high transpiration rates at 45 DAS and at 105 DAS. The transpiration rates after

relief from dry spell are comparable to the values before the dry spell i.e. at 15 (51.95 μg m^{-2} s^{-1}) or 75 DAS (47 μg m^{-2} s^{-1}).

Results indicate that during the dry spell (70-104 and 120-135 DAS), PCS-136 followed by VP-1 and PCS-138 showed high transpiration (Table 11). Incidentally these treatments recorded low soil moisture status (Table 10). Maintenance of the maximum transpiration rate was also reflected in the means. The genotypes PCS-136 and VP-1 showed quick recovery after the dry spell ceased at 105 DAS.

4.3 LEAF WATER POTENTIAL (Ψ_l)

The effect of water stress and recovery in terms of leaf water potential during the crop growth period was recorded. The mean values of Ψ_l ranged from -0.62 to -1.33 MPa. During the dry spell at 30 DAS and 75-104 DAS, the Ψ_l values were low. At 30 DAS, the Ψ_l was -0.88 MPa. At the end of crop growth i.e, at 120 and 135 DAS due to scanty rainfall, the Ψ_l values decreased to -1.06 and -1.04 MPa respectively (Table 12). Ψ_l of leaf improved after relief of water stress, consequently high Ψ_l of -0.62 and -0.70 was recorded at 45 and 105 DAS.

The mean Ψ_l in the genotypes ranged between -0.75 to -0.99 MPa. Among the genotypes studied the maximum or higher Ψ_l was recorded in PCS-136 (-0.75 MPa) and was found to be on par with VP-1 (-0.83 MPa) indicating maintenance of high water status during the crop growth period.

The genotype x DAS interaction revealed Ψ_l values of -0.5 to -1.55 MPa. During the dry spell at 30, 75-104 DAS, the genotypes recorded low water potential. At 30 DAS, the Ψ_l values were on par in genotypes and ranged from -0.80 to -0.95 MPa. During the second dry spell at 75 DAS, the Ψ_l ranged from -0.85 to -1.15 MPa. The highest Ψ_l was observed in the genotype PCS-136 (-0.85 MPa) which is significantly different from others.

As the dry spell continued at 90 DAS, the Ψ_l further decreased and ranged between -1.05 to -1.55 MPa. High Ψ_l was recorded by two genotypes. Leaf Ψ_l of PCS-136 (-1.05 MPa) was on par with VP-1 (-1.15 MPa). By the end of crop growth period (120-135 DAS), the occurrence of dry spell was still conspicuous. At 120 DAS, Ψ_l ranged between -0.90 and -1.15 MPa. The genotype PCS-136 recorded low Ψ_l of -0.90 MPa and was on par with PCS-138 (0.90 MPa). At 135 DAS Ψ_l ranged from -0.87 to -1.17 MPa. PCS-136 recorded high Ψ_l (-0.87 MPa) which was on par with VP-1 (-0.9 MPa).

Water stress recovery was understood by high Ψ_l at 45 and 105 DAS. Ψ_l of -0.62 and -0.70 were recorded. Genotypes PCS-136, 124 and VP-1 at 45 DAS and PCS-136, 138, 124 at 105 DAS recorded low Ψ_l indicating quick recovery.

Results reveal that during the dry spells, PCS-136 followed by VP-1 recorded higher Ψ_l. These also confirm the genotype means represented in

Table 12 indicating that these genotypes maintained high water status during dry spell. In terms of recovery PCS-136 and 124 consistently recorded high Ψ_l indicating maintenance of high Ψ_l values.

4.4 OSMOTIC POTENTIAL (Ψs)

The effect of water stress and recovery in terms of osmotic potential was recorded. The mean value of Ψs across the dates after sowing ranged from -1.10 to -1.64 MPa. During the dry spell, the Ψs values decreased at 30 DAS, 75-104 and 120-135 DAS. At 30 DAS, the Ψs was -1.16 MPa and at 75-104 DAS it ranged between -1.10 and -1.64 respectively. At the end of crop growth i.e. at 120 and 135 DAS the Ψs decreased to -1.23 and -1.18 MPa respectively. During the dry spell at 30 DAS, the mean Ψs was -1.16 MPa and at 75-104 DAS ranged between -1.10 to -1.64. The Ψs decreased after the dry spells not immediately at 45 DAS (-1.16 MPa) but at 60 DAS (-1.13 MPa). Similarly the Ψs dropped by 120 DAS (-1.23) as compared to at 105 DAS (-1.64 Ψs).

The mean Ψs in the genotypes at different DAS ranged between -1.23 to -1.33 MPa. The genotypes PCS-136 recorded lowest Ψs of -1.33 MPa which is on par with VP-1 (-1.31 MPa).

The genotype x DAS interaction showed Ψs values ranged from -0.98 to -1.87 MPa. The Ψs decreased during the dry spell. At 30 DAS, the Ψs of genotypes were on par. At 45 DAS, PCS-136, Kranthi, VP-1 recorded low Ψs

values. At 90 DAS, Kranthi, PCS-124, 137 recorded high Ψs. At 105 DAS, PCS-136, 137 recorded high Ψs. In terms of recovery at 60 DAS, PCS-136, PCS-138 and at 120 DAS, PCS-136 recorded low Ψs (-1.30 MPa).

Results revealed that there exist differences in Ψs values during and after dry spells i.e. in terms of recovery from stress. PCS-136 consistently recorded lower Ψs at 30 DAS (-1.25 MPa) and 90 DAS (-1.52 MPa). Even after recovery from stress also, lower Ψs values were recorded by PCS-136 at 45 DAS (-1.30 MPa) and 120 DAS (-1.30 MPa).

4.5 SPAD CHLOROPHYLL METER READINGS (SPAD)

The SPAD values decreased with the age of the crop. The mean values of SPAD across the dates after sowing ranged between 27 to 36.40. During the dry spell between 30-45 DAS, SPAD units were 35.40 – 35.14. Between 75-104 DAS, SPAD units ranged between 27.95 to 29.97. Upon relief from dry spell, the SPAD units at 45 DAS (35.14) remained similar to those as in dry spell at 30 DAS (35.40). The SPAD units declined in the dry spell at 90 DAS (29.97) to 105 DAS (27.95). Even after receipt of rains, SPAD units at 120 DAS was low (27.8).

The genotype means ranged from 28.56 to 34.25 SPAD units. The genotypes PCS-137 recorded high SPAD value (34.25) followed by PCS-136 (32.63) and PCS-124 (32.49).

The genotype x DAS interaction showed the SPAD values of 23 to 38.02. The SPAD values during the initial dry spell at 30 DAS revealed that

PCS-138 (37.35) followed by PCS-124 (37.2) recorded high values. In terms of recovery PCS-137 (37.93) followed by PCS-124 (37.19) recorded high SPAD units. During the second dry spell PCS-137 recorded high SPAD units at 75 DAS (36.65), 90 DAS (33.5). Upon recovery from stress PCS-137 and 136 recorded high SPAD values at 105 DAS (31, 30.5), 120 DAS (30.5, 30) and 135 DAS (29.5, 29.0), respectively.

Results indicate that throughout the dry spells, the genotypes PCS-138 and 137 recorded high SPAD value which also seen in the genotype means represented in Table 14. In terms of recovery from stress, PCS-137 and 136, showed high SPAD units.

4.6. WATER STRESS AND CROP GROWTH

4.6.1 Leaf dry matter

The mean leaf dry matter during the crop growth period of six castor genotypes ranged from 2.78 to 27.96 g m^{-2}. The leaf dry matter increased upto 75 DAS and thereafter declined by 135 DAS (Table 15). The dry matter accumulation increased by 25 per cent (15-30 DAS), 87 per cent (30-45 DAS), 28 per cent (45-60 DAS) and 28 per cent (60-75 DAS). After 90 DAS the leaf dry matter accumulation decreased (Table 16).

The mean values of leaf dry matter in the genotype ranged from 14.52 to 18.30 g m^{-2}. The maximum leaf dry matter was recorded in the genotype PCS-137 (18.30 gm^{-2}) which is significantly different from the other genotypes. This is followed by PCS-136 (16.83 g m^{-2}) and VP-1 (15.74 g m^{-2}).

The genotype x DAS interaction showed values from 2.04 to 29.82 g m^{-2}. The leaf dry matter which increased upto 75 DAS showed a variation among the genotypes. During the initial dry spell at 30 DAS, three genotypes PCS 137 (10.12 g m^{-2}), PCS-136 (9.69 g m^{-2}) and VP-1 (9.21 g m^{-2}) were superior. During the second dry spell (75-104 DAS), two genotypes viz., PCS-137, 136 recorded superior values (29.82, 28.10 g m^{-2}) at 75 DAS, at 90 DAS (22.65, 21.52 g m^{-2}) and at 105 DAS (19.30, 18.6 g m^{-2}).

The genotypes which recorded high leaf dry matter were also superior in dry matter production after the dry spell. At 45 DAS, the dry matter increased by 87 per cent. Among the genotypes PCS-136, 137 and VP-1 showed 85, 87 and 91 per cent increased leaf dry matter accumulation. Two of these genotypes PCS-137 and PCS-136 recorded superior dry matter (17.38, 15.89 g m^{-2}) at 120 DAS and (15.26, 13.12 g m^{-2}) at 135 DAS.

The results on leaf dry matter accumulation revealed the dry matter increased upto 75 DAS and declined thereafter. During the dry spell and in course of their recovery and regrowth, three genotypes PCS-137, 136 and VP-1 proved superior in accumulating leaf dry matter.

4.6.2 Stem dry matter

Stem dry matter accumulation was recorded from 15 to 135 DAS during the crop growth period in different castor genotypes and presented in Table 16.

The mean values of stem dry matter ranged from 1.35 to 18.44 g m^{-2}. The stem dry matter showed continuous increase upto 75 DAS and thereafter declined. The rate of increase in stem dry matter was in the order of 39 per cent during the 1st dry spell between 30-44 DAS. Dry matter accumulation decreased by 31 per cent during the second dry spell between 75 and 90 DAS.

The genotype means for stem dry matter ranged from 6.95 to 8.83 g m^{-2}. Among genotypes VP-1 showed high stem dry matter (8.83 g m^{-2}) followed by PCS-137 (8.36 g m^{-2}) and PCS-136 (8.25 g m^{-2}) which were on par.

The genotype x DAS interaction showed varied values from 0.97 to 19.07 g m^{-2}. At first dry spell (30-44 DAS) two genotypes PCS-136 and PCS-137 recorded maximum dry matter (5.58 and 5.19 g m^{-2}). During the second dry spell (75-104 DAS) the dry matter reserves increased at 75 DAS indicating preferential partitioning, but showed a decline thereafter. At 90 DAS, PCS-137 (15.04 g m^{-2}) followed by PCS-136 (14.85 g m^{-2}) and VP-1 (14.69 g m^{-2}) were superior. These three genotypes recorded superior values even at 105 DAS.

Upon recovery from stress, the dry matter accumulation in stem showed a continuous increase by 31 per cent (30-45 DAS), 105 per cent (45-60 DAS), 34 per cent (60-75 DAS). Among the genotypes studied, PCS-136 (7.42 g m^{-2}) at 45 DAS and PCS-136 (14.05 g m^{-2}) and VP-1 (13.95 g m^{-2}) at 60 DAS and PCS-136 (19.0 g m^{-2}), PCS-137 (19.0 g m^{-2}) at 75 DAS

were superior. After second dry spell at 105 DAS, VP-1, PCS-136 and PCS-137 recorded higher stem dry matter.

Results reveal that PCS-136, 137 recorded high dry matter during dry spells. The same genotypes DCS-136, 137 along with VP-1 recorded high values during recovery periods from water stress.

4.6.3 Spike dry matter

The spike dry matter recorded from 60 to 135 DAS during the crop growth is represented in Table 17. The mean spike dry matter values ranged from 1.30 to 12.10 g m^{-2}. The dry matter in general increased from 60-120 DAS and thereafter declined. The rate of increase in dry matter was 36 per cent (60-75 DAS), 30 per cent (75-90 DAS), 31 per cent (90-105 DAS), 15 per cent (105-120 DAS). During the dry spell between 75-105 DAS, the rate of increase in spike dry matter declined.

The mean genotype values of spike dry matter ranged between 6.10 and 9.17 g m^{-2}. The genotype Kranthi recorded highest spike dry matter of 9.17 g m^{-2}, which is significantly different. This is followed by PCS-137 (7.73 g m^{-2}) and was on par with PCS-136 (7.51 g m^{-2}). The genotypes PCS-138, VP-1 and PCS-124 had shown less spike dry matter.

The genotype x DAS interaction showed values of 3.25 to 14.62 g m^{-2}. From 60 DAS, the spike dry matter was recorded. During the dry spell between 75-105 DAS, genotypic differences in spike dry matter accumulation revealed that Kranthi accumulated maximum dry matter of 8.26, 10.63 and

12.93 at 75, 90 and 105 DAS. Two other genotypes viz., PCS 136 and PCS-137 proved next best. The spike dry matter accumulation continued even after the dry spell upto 135 DAS indicating the partitioning of assimilates into spikes.

At 135 DAS, there is drastic reduction in the spike dry matter. The values ranged from 0.32 to 2.71 g m^{-2}. The genotype Kranthi recorded high spike dry matter of 2.71 g m^{-2}. This is followed by 1.98 g m^{-2} in PCS-136.

Results indicate that spike dry matter accumulation continued during the dry spell indicating preferential partitioning. The genotypes showed recovery after dry spell. Kranthi recorded high spike dry matter followed by PCS-137 and PCS-136.

4.6.4 Total dry matter

The mean total dry matter during the crop growth period of six castor genotypes ranged from 4.0 to 52.47 g m^{-2}. The total dry matter increased upto 75 DAS and declined thereafter upto 135 DAS (Table 18). The total dry matter accumulation increased by 240.75 per cent (15-30 DAS), 73 per cent (30-45 DAS), 69 per cent (45-60 DAS), 30.6 per cent (60-75 DAS). After 90 DAS, the total dry matter accumulation decreased (Fig. 2).

The mean values of total dry matter in the genotypes ranged from 24.84 to 31.73 g m^{-2}. The maximum total dry matter was recorded in the genotype PCS-137 (31.73 g m^{-2}) which is significantly different from the

other genotypes. This is followed by PCS-136 (30.13 g m^{-2}) and VP-1 (28.45 g m^{-2}).

The genotype x DAS interaction showed values from 3.09 to 55.36 g m^{-2}. The total dry matter which increased upto 75 DAS showed a variation among the genotypes. During the initial dry spell (30-45 DAS), the genotypes PCS-137 (15.68 g m^{-2}), PCS-136 (14.72 g m^{-2}) and VP-1 (13.62 g m^{-2}) were found to be superior. During the second dry spell (75-104 DAS) two genotypes viz., PCS-137, PCS-136 recorded superior values (55.36 g m^{-2} and 54.01 g m^{-2}) at 75 DAS, (46.89, 44.56 g m^{-2}) at 90 DAS and (38.10, 36.0 g m^{-2}) at 105 DAS.

The genotypes which recorded high total dry matter during dry spell also were superior after dry spell. At 45 DAS, the dry matter increased by 73 per cent. Among the genotypes PCS-137, PCS-136 and VP-1 showed increased total dry matter accumulation. The genotypes PCS-137 and PCS-136 recorded superior dry matter (35.86 and 32.17 g m^{-2}) at 120 DAS (18.21 and 17.69 g m^{-2}) at 135 DAS.

The results revealed that the total dry matter increased upto 75 DAS and declined thereafter. During the dry spell and in course of their recovery, three genotypes PCS-137, PCS-136 and VP-1 proved to be superior in accumulating total dry matter.

4.6.5 Leaf area

The mean leaf area during the crop growth period of six genotypes of castor ranged from 336.22 to 5779.5 cm^2. The leaf area increased upto 75 DAS and declined upto the end of the crop growth i.e., 135 DAS (Table 19).

The mean values of leaf area in the genotypes ranged from 1107.27 cm^2 to 5244.89 cm^2. The maximum leaf area was recorded in the genotype PCS-137, (5244.89 cm^2) which is significantly different from the other genotypes. This is followed by Kranthi (4276.4 cm^2) and PCS-138 (3875.5 cm^2). The lowest leaf area was recorded by the genotype VP-1 (1107.27) which is significantly different followed by PCS-136 (2659.5 cm^2) and PCS-124 (3840.8 cm^2).

The genotype x DAS interaction revealed values from 312.92 to 7544.02 cm^2. The leaf area which increased upto 75 DAS showed a genotypic variation. During the initial dry spell at 30 DAS, two genotypes PCS-137 (664.36 cm^2) and Kranthi (606.75 cm^2) recorded higher leaf area. During second dry spell (75-104), the same genotypes PCS-137 and Kranthi recorded high leaf area (7544.02 cm^2, 7374.47 cm^2) at 75 DAS (5490 cm^2 and 4846.4 cm^2) at 90 DAS and (4780.5 cm^2 and 4432.5 cm^2) at 105 DAS.

The genotypes which recorded high leaf area were also superior after the dry spell. At 45 DAS, PCS-137 recorded 2696 cm^2 and Kranthi 2616 cm^2 leaf area. At 120 DAS PCS-137 and Kranthi showed leaf area of 4152 and 4082.8 cm^2 respectively and 1644 and 1434 cm^2 at 135 DAS.

Results revealed that the leaf area increased upto 75 DAS and declined thereafter. During the dry spell and in course of their recovery from stress the two genotypes PCS-137 and Kranthi proved to be superior in leaf area.

4.6.6 Crop growth rate (CGR)

Data on crop growth rate was recorded at 15-30 to 120-135 DAS during the crop growth period. The mean CGR values ranged from -2.65 to 3.59 g m^2 d^{-1}. There is steady increase in CGR upto 60-75 DAS and thereafter i.e., from 75-90 to 120-135 DAS the CGR values decreased. At 75-90 DAS, the CGR was 3.27 g m^{-2} d^{-1} and at 90-105, 105-120 and 120-135 the CGR values were -2.65, -2.21 and -1.13 g m^{-2} d^{-1} respectively.

The mean genotype CGR values ranged from 0.578 to 0.925 g m^{-2} d^{-1}. The highest CGR was recorded in PCS-137 (0.925 g m^{-2} d^{-1}) which is on par with VP-1 (0.785 g m^{-2} d^{-1}). This is followed by PCS-136 (0.758 g m^{-2} d^{-1}) and PCS-138 (0.72 g m^{-2} d^{-1}) which are on par. The lowest CGR was recorded in PCS-124 (0.578 g m^{-2} d^{-1}) and Kranthi (0.640 g m^{-2} d^{-1}) which were on par.

The genotype x DAS interaction revealed values from -3.5 to 4.36 g m^{-2} d^{-1}. The CGR increased upto 75 DAS and then declined. During the initial dry spell at (30-45 DAS), the genotypes showed no significant differences and values ranged from 1.30 to 1.59 g m^{-2} d^{-1}. During second dry spell at 75-90, the genotype Kranthi recorded high CGR value of 4.09 g m^{-2} d^{-1}, followed by PCS-136 (3.6 g m^{-2} d^{-1}), Kranthi (3.51 g m^{-2} d^{-1}). At 90-105 DAS, genotypes

PCS-138, PCS-137 and VP-1 recorded high CGR of -2.10, -2.26 and -2.4 g m^{-2} d^{-1}.

At 45 days i.e. after dry spell (45-60 DAS), the genotype PCS-137 recorded high CGR of 3.86 g m^{-2} $d^{-1}$and was on par with VP-1 (3.44 g m^{-2} d^{-1}). At 120-135 DAS, the same genotype PCS-138 recorded high CGR of -0.52 g m^{-2} d^{-1} and was on par with PCS-136 (-1.02 g m^{-2} d^{-1}).

Results indicate that CGR increased upto 75 DAS and then decreased. During dry spell, the genotypes PCS-124, Kranthi and VP-1 were superior and showed high CGR even after dry spell, i.e., in course of recovery, the genotypes PCS-137 and PCS-136 showed high CGR values.

4.6.7 Leaf area index (LAI)

The mean leaf index during the crop growth period of six genotypes of castor ranged from 0.033 to 0.586. The LAI increased upto 75 DAS and then declined upto 135 DAS (Table 21). LAI followed same trend as that of leaf area (Table 20).

The mean values of LAI in the genotypes ranged from 0.12 to 0.53. The maximum LAI was recorded in the genotype PCS-137 (0.532) which is significantly different from other genotypes. This is followed by Kranthi (0.427) and PCS-138 (0.387). The lowest LAI was recorded by the genotype VP-1 (0.382) which is significantly different from PCS-124 (0.264) and PCS-136 (0.122).

The genotype x DAS interaction revealed values form 0.031 to 0.754. The LAI which increased upto 75 DAS showed a genotypic variation. During the initial dry spell at 30 DAS, two genotypes PCS-137 (0.163) and Kranthi (0.143) recorded higher LAI. During the second dry spell (75-104), the same genotypes PCS-137 and Kranthi recorded high LAI (0.754, 0.737) at 75 DAS (0.54, 0.48) at 90 DAS and (0.47, 0.44) at 105 DAS.

The genotypes which recorded high LAI were also superior after the dry spell. At 45 DAS, PCS-137 recorded 0.267 and Kranthi 0.260 LAI. At 120 DAS, PCS-137 and Kranthi recorded 0.414 and 0.40 and at 135 DAS, 0.037 and 0.035, respectively.

Results on LAI revealed that the LAI increased upto 75 DAS and declined thereafter. During the dry spell and in course of their recovery from stress, two genotypes PCS-137 and Kranthi proved to be superior in LAI.

4.6.8 Specific leaf area

Data on specific leaf area (SLA) was recorded from 15 to 135 DAS and given in Table 22. The mean values across the dates of sowing ranged between 88.44 and 259.82 cm^2 g^{-1}. There is increasing trend in SLA upto 135 DAS from 15 DAS except at 30 DAS.

Among the genotypes, the mean values ranged from 166.74 to 221.10 cm^2 g^{-1}. The genotype PCS-137 showed highest SLA value (221.10 cm^2 g^{-1}) followed by Kranthi (207.56 cm^2 g^{-1}) and lowest values of SLA were recorded by PCS-124 followed by VP-1 and PCS-136.

The genotype x DAS interaction value revealed values from 106.5 to 280.56 cm^2 g^{-1}. During the initial dry spell at 30 DAS, the genotype PCS-137 recorded high SLA of 98.06 on par with Kranthi (94.16 cm^2 g^{-1}) and PCS-136 (94.04 cm^2 g^{-1}). During second dry spell (75-104), the genotype Kranthi followed by VP-1 showed high SLA (191.14 and 183.06 cm^2 g^{-1}) at 75 DAS, (261.61 and 235.36 cm^2 g^{-1}) by Kranthi and PCS-137 at 90 DAS and (232.62 and 222.33 cm^2 g^{-1}) by PCS-137 and Kranthi at 105 DAS.

Results in SLA revealed that the values increased throughout the crop growth and during and after dry spell i.e. recovery from stress, the genotypes, PCS-137 and Kranthi recorded high SLA.

4.7 YIELD AND YIELD COMPONENTS

4.7.1 Length of spikes

The effective length of primary, secondary and tertiary order spikes varied significantly. The mean spike length decreased from primary to secondary to tertiary orders. The mean length of spikes was 9.2, 6.63 and 4.57 cm, respectively.

Genotypic variation was also recorded. The mean spike length ranged between 5.41 to 7.95 cm. Among the genotypes, Kranthi recorded maximum spike length (7.95 cm) which is significantly different and was followed by PCS-136 (7.39 cm) and PCS-137 (7.13 cm) which were on par. The lowest spike length was recorded in PCS-138 (5.41 cm).

Genotype x spike length interaction revealed that Kranthi recorded maximum length of primary spike (10.37 cm) which is significantly different. This was followed by spike length of PCS-137 (9.65 cm) and PCS-136 (9.43 cm) which were on par. The lowest primary spike length was recorded in PCS-138 (7.75 cm).

Among the secondary order spikes also, like in primary spikes, Kranthi recorded maximum length (7.75 cm) which was on par with PCS-136 (7.75 cm) followed by PCS-137 (7.25) which is significantly different. The minimum length of the secondary spike was recorded by PCS-138 (5.0 cm).

For the tertiary spikes the genotype Kranthi (5.75 cm) showed maximum value followed by PCS-136 (5.0 cm) and PCS-138 (3.5 cm).

Results indicate that the genotype Kranthi recorded maximum effective spike length for primary, secondary and tertiary orders, followed by PCS-136 and PCS-137 (Table 23). These three genotypes also produced high spike dry matter (Table 17).

4.7.2 Number of capsules

The number of capsules harvested from primary, secondary and tertiary harvest varied significantly. The mean number of capsules were 32.58, 34.56 and 38.06 respectively. The number of capsules were 32.58, 34.56 and 38.06 respectively. The number of capsules formed in primary order spikes was lower compared to second and tertiary spikes (Table 24).

Among the genotypes studied, the mean capsule, no. ranged between 27.0 and 42.43. Kranthi recorded highest number of capsules (42.43) which was on par with PCS-137 (40.39) followed by PCS-136 (36.91) which was on par with VP-1 (35.17). The lowest number of capsules were recorded in the genotype PCS-124 (27.05) which is on par with PCS-138.

Genotypes x capsule number interaction showed that at first harvest, the mean capsule number ranged from 24.3 to 39.89. The genotype Kranthi recorded highest number of capsules (39.89) which was on par with PCS-137 (37.6). This is followed by PCS-136 (35.2) and VP-1 (32.9) which were on par.

At second harvest, the number of capsules ranged between 27.3 and 41.2. The genotype Kranthi recorded high number of capsules (41.2) and was on par with PCS-137 (39.6). This was followed by PCS-136 (36.8) and VP-1 (34.5) which were on par.

The lowest number of capsules were recorded by PCS-124 (27.31) and PCS-138 (27.9) and were on par. The incidence of two dry spells is understood to decrease the number of capsules formed in primary spikes.

At third harvest, the number of capsules between the genotypes ranged from 29.5 to 46.2. The highest or maximum number of capsules was observed in Kranthi (46.2) which was found to be on par with PCS-137 (43.9) and followed by PCS-136 (38.7) which was on par with VP-1 (38.1).

Results indicate that capsule number increased little between first and second harvest due to dry spell. The genotype Kranthi followed by PCS-137 and PCS-136 recorded maximum number of capsules. These genotypes also recorded superior effective spike length (Table 23) and maximum spike dry matter (Table 17) possibly contributing to high capsule number.

4.7.3 100-seed weight

The mean values of 100-seed weight at first, second and third harvest were 28-36 g, 30-63 g and 36.77 gm. The 100 seed weight increased in all the harvests (Table 25).

Among the genotypes the mean 100 seed weight values ranged between 24.42 and 40.0 g. The genotypes Kranthi recorded highest 100 seed weight (40.0 g), which is significantly different. This is followed by PCS-137 (35.6 g) and PCS-136 (33.9 g) which were on par. The minimum 100 seed weight values were noted in PCS-124 (24.4 g) and PCS-138 (25.3 g) which were on par.

The genotype x seed weight interaction revealed 100 seed weight values to range from 20.65 to 45.65 g. At first harvest, the genotype showed values ranging from 20.65 to 36.5 g. The maximum 100 seed weight was recorded by genotype Kranthi (36.5 g) followed by PCS-136 (31.5 g) which was on par with PCS-137 (30.7 g).

During second harvest, the 100 seed weight values ranged from 22.4 g to 37.9 g. Kranthi recorded maximum 100 seed weight (37.9 g) followed by PCS-137 (35.0 g) which was on par with PCS-136 (33.17 g).

At harvest of tertiaries, 100 seed weight, values ranged from 29.9 to 45.6 g. Kranthi showed maximum seed weight (45.6 g) which is significant and followed by PCS-137 (41.1 g). PCS-136 (37 g) and VP-1 (36.72) were on par with each other. The lowest 100-seed weight was recorded by PCS-138 (29.9 g) which was on par with PCS-124 (30.0 g).

Results indicate that Kranthi followed by PCS-137 and PCS-136 recorded maximum 100-seed weight.

4.7.4 Seed yield

Seed yield comprises of yield data from first, second and third pickings. The mean seed yield increased picking wise and was also significantly different (Table 26).

At first picking, among the genotypes, Kranthi recorded highest seed yield (171.66 kg ha^{-1}) which is significantly different from PCS-137 (163.34 kg ha^{-1}) and which was on par with PCS-136 (160.29 kg ha^{-1}) followed by VP-1 (155.15 kg ha^{-1}). Lower seed yield was recorded by PCS-138 (142.01 kg ha^{-1}) on par with PCS-124 (139.73 kg ha^{-1}).

At second picking, among the genotypes Kranthi recorded highest seed yield of 175.73 kg ha^{-1} which is significantly different. This is followed by PCS-137 (167.65 kg ha^{-1}) which was on par with PCS-136 (164.57 kg ha^{-1})

and VP-1 (161.7 kg ha^{-1}). The occurrence of dry spell between the first and second picking possibly increased the seed yield marginally.

At third picking, the same genotype Kranthi recorded seed yield with 186.77 kg ha^{-1}. This is followed by PCS-137 (176.36 kg ha^{-1}) which is on par with VP-1 (171.19 kg ha^{-1}) and PCS-136 (170.74 kg ha^{-1}).

Yield x picking interaction revealed yield to range between 139.73 kg ha^{-1} and 186.77 kg ha^{-1} of total seed yield. Among genotypes, Kranthi recorded high seed yield of 534.16 kg ha^{-1} which is significantly different followed by PCS-137 (507.35 kg ha^{-1}) and PCS-136 (495.60 kg ha^{-1}). The least total seed yield was recorded by PCS-124 (442.76 kg ha^{-1}) and found to be on par with VP-1 (488.04 kg ha^{-1}).

Harvest Index

Harvest index values ranged between 28.0 per cent and 40.5 per cent. The highest HI was recorded in genotype Kranthi (43.75 %) which was on par with PCS-137 (41.25 %), PCS-136 (40.75 %) and VP-1 (39.25 %). The lowest HI was recorded by the genotype PCS-124 (33.50 %) which was on par with PCS-138 (34.60 %) (Table 26).

Correlation matrix for seven characters like CGR, LAI, SLA, spike length, number of capsules, 100 seed weight and yield have been worked out for the entire crop growth period. In the present study, yield contributing characters showed significant associations. Effective spike length showed significant association with capsule number (r = 0.9060) and 100 seed weight

(r = 0.9323). Capsule number in turn showed significant association with 100 seed weight (r = 0.9912). The character association needs to be worked out separately for stress and non stress situation to know the contributing effect of stress and non stress conditions.

IN VITRO STUDIES

Shoot tip explants obtained from *in vivo* grown aseptic seedlings of castor genotypes Kranthi and Aruna were cultured on MS medium with Benzyl adenine to induce multiple shoots. Observation were recorded on the colour of shoot tip explant and number of multiple shoots (Table 28).

Response of shoot tips explants on MS + BA

Shoot tips of two genotypes Kranthi and Aruna were cultured on MS medium fortified with BA ranging from 2 to 5 mg l^{-1} observations were recorded periodically at 14 and 28 DAI on color of shoot tip, number of multiple shoot produced per shoot tip (Table 28).

Shoot tips of Kranthi were light green coloured at inoculation. Shoot tips continued to remain light green at 14 DAI, turned green by 28 DAI in all the concentrations of BA and shoot tips of Aruna which were also light green at inoculation time turned green at 28 DAI.

Multiple shoots initiated from the explants around tenth day of inoculation. Mean number of shoots ranged from 2.12 to 4.0 per shoot tip. Mean maximum shoots of 4.0 per shoot tip were observed on MS + BA 2 mg l^{-1}.

Two genotypes showed significant difference in the number of shoots produced. Kranthi and Aruna initiated 4.41 and 1.5 shoot per shoot tip, respectively.

Treatment x genotype interaction revealed significant differences. Maximum shoots of 5.5 were recorded in Kranthi at 5 mg/l while Aruna recorded maximum shoots of 4.5 at 2 mg l^{-1} BA concentration.

The results reveal that the BA 5 and 2 mg l^{-1} initiated maximum shoots (5.5) in Kranthi and Aruna (4.5).

Response of shoot tips of castor genotypes on MS + BA supplemented with different levels of PEG

PEG was incorporated to induce drought stress into shoot induction medium wherein shoot tips of Kranthi were cultured on MS + BA 5 mg l^{-1} and Aruna on MS + BA with 2 mg l^{-1} concentration (Table 29).

The light green coloured shoot tips turned green by 28 DAI in Kranthi and Aruna genotypes at all the concentrations of PG stress.

Significant variation was observed in treatmental effects on the multiple shoots produced. A mean number of 1.25 to 5.0 shoots were produced per shoot tip. Mean maximum number of shoots (5.0) was recorded in control followed by -3 bars (3.5), -4 (2.25) and -6 bars (1.25). Among the genotypes, Kranthi recorded more shoots of 3.25 compared to Aruna (2.75).

Genotype x PEG stress interaction revealed a maximum of 3.5 shoots at -3 bars. As stress increased to -4 and -6 bars, there was a decrease in the

number of multiple shoot produced. At -3 bars Kranthi recorded 2.5 and Aruna 2.0 shoots. At -6 bars, 1.5 shoots were produced by Kranthi and 1.0 by Aruna (Table 29).

From the studies it appeared that stress levels of -3 bars persisted in the shoots and proved inhibitory to recovery in terms of regeneration of shoots *in vitro*.

Table 28 : Response of shoot tip explants of castor genotypes on MS + BA

	Colour of Shoot tip				No. of shoots / shoot tip		Mean
	Kranthi		Aruna		Kranthi	Aruna	
DAI / BA	14	28	14	28	28	28	
2	Light green	Light green	Light green	Green	3.50	4.50	4.00
4	Light green	Green	Light green	Green	4.25	0.00	2.12
5	Light green	Green	Light green	Green	5.50	0.00	2.75
Mean					4.41	1.50	

	S.Em ±	CD (p = 0.05)
Concentration	0.24	0.63
Genotype	0.20	0.51
Concentration x Genotype	0.34	0.89

Table 29 : Response of shoot tips of castor on MS + BA (-3 to -6 bars)

Genotype	Colour of Shoot tip				No. of shoots / shoot tip		Mean
	Kranthi		Aruna		Kranthi	Aruna	
DAI / PEG Stress bars	14	28	14	28	28	28	
0	Light green	Green	Light green	Green	5.5	4.5	5.0
-3	Light green	Green	Light green	Green	3.5	3.5	3.5
-4	Light green	Green	Light green	Green	2.5	2.0	2.25
-6	Light green	Green	Light green	Green	1.5	1.0	1.25
Mean					3.25	2.75	

	S.Em ±	CD (p = 0.05)
PEG stress	0.41	0.98
Genotype	0.29	0.69
PEG stress x Genotype	0.59	1.39

Table 10: Soil moisture content values in plots at different days after sowing in castor genotypes

Genotypes	Days after sowing										Mean
	0	15	30	45	60	75	90	105	120	135	
PCS-124	5.13	16.25	6.65	13.75	9.38	6.13	6.75	8.38	6.13	6.13	7.71
VP-1	5.12	15.75	6.25	13.25	9.75	6.25	5.25	9.25	6.38	6.35	7.85
Kranthi	4.75	15.12	5.75	13.25	9.38	6.25	6.75	8.25	6.43	6.13	7.50
PCS-136	5.25	15.25	6.75	13.63	8.13	5.26	6.25	8.20	6.48	6.12	7.42
PCS-137	4.75	14.75	5.75	12.75	9.25	5.36	5.13	8.82	6.43	6.13	7.38
PCS-138	5.00	15.75	5.75	13.25	10.0	6.25	5.38	8.75	6.38	6.38	7.78
Mean	5.00	15.47	6.14	13.31	9.64	5.91	6.00	8.68	6.33	6.22	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.08	0.13
Days after sowing (DAS)	0.09	0.17
Genotypes x DAS	0.22	0.42

Table 11: Transpiration rate at different days after sowing in six castor genotypes (μg m^{-2} s^{-1})

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	50.25	48.75	53.50	47.00	48.00	27.75	46.75	21.25	18.25	40.16
VP-1	56.50	50.25	59.25	46.50	50.25	30.00	50.00	26.50	21.00	43.36
Kranthi	44.50	41.00	48.75	40.00	38.25	22.75	47.00	14.00	12.00	34.25
PCS-136	58.00	55.25	59.25	50.50	51.50	32.00	53.00	28.00	23.00	45.61
PCS-137	47.75	43.75	52.00	43.00	43.00	26.25	48.00	17.25	15.75	37.41
PCS-138	54.75	52.25	58.25	50.00	51.00	28.25	49.50	25.00	21.00	43.33
Mean	51.95	48.54	55.16	46.16	47.00	27.83	49.04	22.00	18.50	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.32	0.63
Days after sowing (DAS)	0.39	0.77
Genotypes x DAS	0.97	1.90

Table 12: Leaf water potential (MPa) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	-0.68	-0.85	-0.55	-0.65	-1.07	-1.35	-0.60	-1.125	-1.07	-0.88
VP-1	-0.65	-0.80	-0.55	-0.60	-1.05	-1.15	-0.70	-1.15	-0.90	-0.83
Kranthi	-0.70	-0.95	-0.73	-0.75	-1.15	-1.55	-0.85	-1.10	-1.17	-0.99
PCS-136	-0.63	-0.85	-0.50	-0.55	-0.85	-1.05	-0.60	-0.90	-0.87	-0.75
PCS-137	-0.75	-0.95	-0.70	-0.80	-1.15	-1.47	-0.85	-1.15	-1.12	-0.99
PCS-138	-0.70	-0.90	-0.68	-0.70	-1.10	-1.42	-0.60	-0.90	-1.10	-0.91
Mean	-0.68	-0.88	-0.62	-0.67	-0.06	-0.33	-0.70	-1.06	-1.04	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.02	0.14
Days after sowing (DAS)	0.02	0.15
Genotypes x DAS	0.06	0.23

Table 13: Osmotic potential (MPa) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	-1.57	-1.17	-0.98	-1.02	-1.03	-1.20	-1.76	-1.25	-1.22	-1.24
VP-1	-1.48	-1.12	-1.17	-1.17	-1.12	-1.27	-1.87	-1.28	-1.28	-1.31
Kranthi	-1.52	-1.11	-1.17	-1.11	-1.07	-1.07	-1.77	-1.08	-1.12	-1.23
PCS-136	-1.65	-1.25	-1.17	-1.26	-1.20	-1.52	-1.35	-1.30	-1.29	-1.33
PCS-137	-1.52	-1.10	-1.30	-1.07	-1.07	-1.25	-1.46	-1.25	-1.12	-1.24
PCS-138	-1.14	-1.23	-1.13	-1.13	-1.10	-1.35	-1.66	-1.25	-1.07	-1.23
Mean	-1.48	-1.16	-1.16	-1.13	-1.10	-1.27	-1.64	-1.23	-1.18	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.02	0.02
Days after sowing (DAS)	0.03	0.07
Genotypes x DAS	0.06	2.80

Table 14: SPAD (SCMR) values at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	38.02	37.20	37.19	35.58	31.00	31.32	27.00	26.75	26.75	32.49
VP-1	36.34	35.86	35.13	33.48	31.87	27.50	27.00	27.75	27.00	31.32
Kranthi	33.50	34.76	31.09	31.90	29.62	25.00	24.50	23.75	23.00	28.56
PCS-136	33.55	35.15	34.56	34.37	35.12	31.50	30.50	30.00	29.00	32.63
PCS-137	35.28	36.46	37.93	37.47	36.65	33.50	31.00	30.50	29.50	34.25
PCS-138	33.24	37.35	34.97	34.29	32.87	31.00	27.75	28.50	26.75	31.85
Mean	34.98	36.13	35.14	34.51	32.85	29.97	27.95	27.80	27.00	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.29	0.58
Days after sowing (DAS)	0.36	0.71
Genotypes x DAS	0.89	0.75

Table 15: Leaf dry matter (g m^{-2}) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	2.04	8.46	15.79	19.80	27.09	20.78	16.28	13.18	10.12	14.83
VP-1	2.89	9.21	17.67	22.11	27.78	21.10	16.67	13.96	10.34	15.74
Kranthi	2.77	8.29	15.10	18.02	27.27	20.78	15.51	12.02	10.96	14.52
PCS-136	3.37	9.69	18.14	23.1	28.10	21.52	18.61	15.89	13.12	16.83
PCS-137	3.41	10.12	18.74	26.08	29.82	22.65	19.30	17.38	15.26	18.30
PCS-138	2.23	8.51	16.21	21.92	27.73	21.02	16.67	13.18	10.16	15.29
Mean	2.78	19.04	16.99	21.83	27.96	21.30	17.17	14.60	11.66	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.157	0.30
Days after sowing (DAS)	0.192	0.37
Genotypes x DAS	0.480	0.93

Table 16: Stem dry matter (g m^{-2}) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	1.07	4.89	6.41	13.37	18.59	10.49	5.19	3.26	1.46	7.19
VP-1	1.97	4.72	6.74	13.95	18.91	14.69	8.64	5.29	4.61	8.83
Kranthi	0.97	4.03	6.44	13.33	18.48	10.42	4.91	3.21	1.54	7.03
PCS-136	1.34	5.19	7.42	14.05	19.00	14.85	6.35	3.51	2.59	8.25
PCS-137	1.53	5.58	6.75	14.11	19.07	15.04	6.70	3.86	2.63	8.36
PCS-138	1.19	4.50	6.40	13.65	16.30	10.70	5.23	3.10	1.47	6.95
Mean	1.35	4.81	6.69	13.74	18.44	12.69	6.17	3.70	2.38	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.08	0.16
Days after sowing (DAS)	0.10	0.19
Genotypes x DAS	0.24	0.48

Table 17: Spike dry matter at different days after sowing in six castor genotypes

Genotypes	Days after sowing						Mean
	60	75	90	105	120	135	
PCS-124	3.25	4.56	5.78	7.02	8.13	0.97	4.95
VP-1	3.40	4.68	6.44	9.38	11.78	0.96	6.10
Kranthi	7.05	8.26	10.63	12.93	13.46	2.71	9.17
PCS-136	4.68	6.91	7.73	11.04	12.77	1.98	7.51
PCS-137	3.68	6.47	9.20	12.10	14.62	0.32	7.73
PCS-138	4.90	5.90	8.07	10.51	11.89	0.87	7.02
Mean	4.49	6.13	7.97	10.49	12.10	1.30	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.28	0.47
Days after sowing (DAS)	0.39	0.62
Genotypes x DAS	0.51	0.98

Table 18: Total dry matter (g m^{-2}) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	3.09	12.89	22.20	36.42	50.24	37.05	28.49	22.57	12.55	24.84
VP-1	4.09	13.62	24.41	39.42	51.47	42.23	34.59	31.03	15.21	28.45
Kranthi	3.74	12.36	21.54	38.40	53.83	41.83	33.35	28.79	13.50	27.48
PCS-136	4.72	14.72	25.56	41.83	54.01	44.53	36.00	32.17	17.69	30.13
PCS-137	4.94	15.68	25.49	45.09	55.36	46.89	38.10	35.86	18.21	31.73
PCS-138	3.42	12.56	22.61	39.77	49.93	39.79	32.41	28.17	12.50	26.79
Mean	4.00	13.63	23.63	40.15	52.47	42.05	33.82	29.76	14.62	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.22	0.43
Days after sowing (DAS)	0.27	0.53
Genotypes x DAS	0.67	1.31

Table 19: Leaf area (cm^2) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	312.92	471.79	1865.83	3057.48	4708.32	3974.20	3420.50	3141.32	1239.00	3840.08
VP-1	322.55	484.31	1961.87	4046.64	5175.30	4645.70	4371.30	3921.2	1379.00	1107.27
Kranthi	355.82	606.75	2616.80	4489.34	7374.47	4846.20	4432.50	4082.80	1434.00	4276.40
PCS-136	315.82	485.59	1992.46	3011.32	4873.20	4067.10	3433.96	3333.00	1227.00	2659.5
PCS-137	375.65	664.36	2696.00	4708.30	7544.02	5490.00	4780.50	4152.00	1644.00	5244.89
PCS-138	334.58	505.49	2139.94	3329.00	5001.87	4238.00	3757.50	3276.00	1263.00	3875.50
Mean	336.22	536.38	2212.15	3773.71	5779.50	4544.00	4032.70	3406.40	1364.30	

	S.Em ±	C.D (p = 0.05)
Genotypes	22.60	44.29
Days after sowing (DAS)	27.60	54.25
Genotypes x DAS	67.80	132.89

Table 20: Crop growth rate (CGR) g m^{-2} d^{-1} at different days after sowing in six castor genotypes

Genotypes	Days after sowing								Mean
	15-30	30-45	45-60	60-75	75-90	90-105	105-120	120-135	
PCS-124	1.102	1.59	2.83	3.35	3.69	-3.50	-2.28	-1.58	0.578
VP-1	1.108	1.41	3.44	3.45	3.21	-2.40	-2.03	-1.12	0.785
Kranthi	0.16	1.57	3.31	3.50	4.09	-3.19	-2.47	-1.22	0.640
PCS-136	1.08	1.33	3.37	3.60	3.24	-2.50	-2.27	-1.02	0.758
PCS-137	1.06	1.41	3.86	4.36	2.73	-2.26	-2.31	-0.52	0.925
PCS-138	1.24	1.30	3.31	3.32	2.71	-2.10	-1.95	-1.32	0.72
Mean	0.95	1.44	3.35	3.59	3.28	-2.65	-2.21	-1.13	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.08	0.15
Days after sowing (DAS)	0.09	0.18
Genotypes x DAS	0.22	0.44

Table 21: Leaf area index (LAI) at different days after sowing in six castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	0.048	0.126	0.213	0.332	0.516	0.423	0.375	0.332	0.032	0.264
VP-1	0.062	0.122	0.198	0.300	0.468	0.397	0.341	0.341	0.031	0.038
Kranthi	0.060	0.143	0.260	0.448	0.737	0.486	0.442	0.408	0.035	0.427
PCS-136	0.048	0.123	0.206	0.305	0.487	0.404	0.342	0.327	0.031	0.122
PCS-137	0.066	0.163	0.267	0.470	0.754	0.548	0.477	0.414	0.037	0.532
PCS-138	0.048	0.137	0.223	0.404	0.575	0.464	0.436	0.391	0.033	0.387
Mean	0.055	0.135	0.227	0.376	0.586	0.453	0.402	0.368	0.033	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.08	0.2
Days after sowing (DAS)	0.01	0.02
Genotypes x DAS	0.02	0.05

Table 22: Specific leaf area (SLA) (cm^2 g^{-1}) in different castor genotypes

Genotypes	Days after sowing									Mean
	15	30	45	60	75	90	105	120	135	
PCS-124	153.70	89.57	118.48	131.22	154.47	179.55	196.77	224.13	252.80	166.74
VP-1	111.917	74.56	130.96	126.15	183.06	187.57	213.84	246.62	272.48	171.93
Kranthi	106.50	94.16	129.94	139.76	191.14	261.61	222.33	232.10	280.56	207.56
PCS-136	139.22	94.04	121.56	131.18	167.03	193.42	209.74	238.90	248.18	173.95
PCS-137	112.21	98.06	151.79	144.05	172.21	235.36	232.62	242.13	239.45	221.10
PCS-138	152.40	80.28	123.19	132.01	157.02	172.24	199.77	227.04	268.45	168.04
Mean	129.32	88.44	129.20	134.06	170.84	204.90	212.51	235.16	259.82	

	S.Em ±	C.D (p = 0.05)
Genotypes	156.5	NS
Days after sowing (DAS)	191.7	NS
Genotypes x DAS	469.6	NS

Table 27 : Correlation Matrix of growth and yield contributing characters of six castor genotypes

Characters	CGR	LAI	SLA	Spike Length	Number of Capsules	100 seed weight	Yield
CGR	**1.0000**	0.1891	0.4985	0.1462	0.4537	0.3438	0.0463
LAI	0.1891	**1.0000**	0.7181	0.0494	0.2622	0.2083	0.4812
SLA	0.4985	0.7181	**1.0000**	0.6366	0.8184	0.7653	0.5559
Spike length	0.1462	0.0494	0.6366	**1.0000**	0.9060*	0.9323*	0.5963
Number of capsules	0.4537	0.2622	0.8184	0.9060*	**1.0000**	0.9912*	0.6711
100 seed weight	0.3438	0.2083	0.7653	0.9323*	0.9912	**1.0000**	0.6988
Yield	0.0463	0.4812	0.5559	0.5963	0.6711	0.6988	**1.0000**

* significant at 5 per cent level

Table 23 : Spike length in six castor genotypes

Genotypes	Spike length (cm)			Mean
	Primary	Secondary	Tertiary	
PCS-124	8.88	5.63	4.00	6.16
VP-1	9.13	6.40	4.68	6.73
Kranthi	10.37	7.75	5.75	7.95
PCS-136	9.43	7.75	5.00	7.39
PCS-137	9.65	7.25	4.50	7.13
PCS-138	7.75	5.00	3.50	5.41
Mean	9.20	6.63	4.57	

	S.Em ±	C.D (p = 0.05)
Genotypes	0.115	0.230
Spike length	0.080	0.160
Genotypes x Spike length	0.199	0.399

Table 24 : Number of capsules at different harvest in six castor genotypes

Genotypes	Number of capsules			Mean
	Harvest-1	Harvest-2	Harvest-3	
PCS-124	24.30	27.31	29.54	27.05
VP-1	32.90	34.50	38.12	35.17
Kranthi	39.89	41.20	46.20	42.43
PCS-136	35.20	36.80	38.75	36.91
PCS-137	37.60	39.67	43.90	40.39
PCS-138	25.60	27.90	31.85	28.45
Mean	32.58	34.56	38.06	

	S.Em ±	C.D (p = 0.05)
Genotypes	1.21	2.36
Capsule Number	0.82	1.70
Genotypes x Capsule Number	1.98	3.97

Table 25 : 100-seed weight (g) at different harvest of six castor genotypes

Genotypes	100 seed weight (g)			Mean
	Harvest-1	Harvest-2	Harvest-3	
PCS-124	20.65	22.43	30.18	24.42
VP-1	28.93	30.86	36.72	32.17
Kranthi	36.58	37.92	45.65	40.05
PCS-136	31.50	33.17	37.05	33.90
PCS-137	30.74	35.07	41.13	35.64
PCS-138	21.81	24.37	29.92	25.37
Mean	28.36	30.63	36.77	

	S.Em ±	C.D (p = 0.05)
Genotypes	1.11	2.22
Seed weight	0.78	1.57
Genotypes x Seed weight	1.92	3.85

Table 26 : Seed yield and harvest index of six castor genotypes

Genotypes	Seed yield (kg ha^{-1})			Total seed yield (kg ha^{-1})	Harvest Index (%)
	Primary	Secondary	Tertiary		
PCS-124	139.74	144.77	158.25	442.76	33.50
VP-1	155.15	161.70	171.19	488.04	39.25
Kranthi	171.66	175.73	186.77	534.16	43.75
PCS-136	160.29	164.57	170.74	495.60	40.75
PCS-137	163.34	167.65	176.36	507.35	41.25
PCS-138	142.01	146.53	156.83	445.37	34.60
Mean	155.36	160.15	170.02	485.50	38.89

	S.Em ±	C.D (p = 0.05)
Genotypes	2.73	4.42
Pickings	2.79	4.13
Genotypes x Pickings	4.82	8.69

CHAPTER V

DISCUSSION

Castor crop is mainly grown under rainfed conditions where it suffers from intermittent drought that results in low yields. For bringing about yield improvement, a number of castor varieties (Kranthi, Jyothi, Haritha, Kiran) and hybrids (GCH-1, 4 and DCH-177) have been released by conventional breeding approaches and have been recommended for dry lands based on relative yield advantage (DOR, 2002). Despite their release, the possibility of improving drought tolerance by genetic manipulation remains a challenge. Sofar, the response of the genotypes to water stress and their recovery and regrowth has not been characterized in castor. Hence, the traits conferring tolerance to water stress need to be studied in detail to incorporate then into future breeding programmes (Kramer, 1983). The present study was therefore carried out to identify the castor genotypes that show quick recovery and regrowth after the relief of water stress.

5.1 SOIL MOISTURE

Two dry spells occurred during the crop growing season between 30-44 and 75-104 DAS. Soil moisture content at 30 DAS was 6.14 per cent and at 75-104 DAS it was 5.91, 6.00 per cent. Hence, stress recovery could be quantified at two periods when crop showed recovery and regrowth after the occurrence of dry spell i.e. at 45 and 105 DAS, wherein a mean soil moisture

of 13.3 and 8.68 per cent was recorded (Table 10). Among the treatments, plots with genotypes PCS-124, 136 recorded low soil moisture. Later after the second dry spell (105 DAS), the plots of PCS-136, 138 and VP-1 recorded low soil moisture status. This difference in soil moisture content of plots indicated the plants ability to draw moisture from soil. Udaykumar *et al*. (1998) reports that plants with better root characteristics show ability to draw more moisture from soil.

5.2 TRANSPIRATION

The transpiration rate decreased significantly in all the genotypes during the entire stress period. Transpiration values at 30-44 and 75-104 DAS were from 48.54 and 27.83 as compared to after relief of stress at 45, 105 DAS which ranged from 55.16 and 49.04 $\mu g\ m^{-2}\ s^{-1}$, respectively. Among the genotypes PCS-136, VP-1, PCS-138 recorded high transpiration rate of 45.61, 43.36, 43.33 $\mu g\ m^{-2}\ s^{-1}$ during the stress period (Table 11). Incidentally these genotypes also recorded low soil moisture status (Table 10). Carlson and Lynn (1997) reported that under stress, transpiration was enhanced to meet the atmosphere demand.

5.3 LEAF WATER POTENTIAL

The leaf water potential (Ψ_l) values were low during the dry spell at 30 and 75-104 DAS. Ψ_l improved after the relief of water stress at 45 and105 DAS. During the dry spell Ψ_l values ranged from -0.88 to -0.33 MPa (Table 12). LWP in stress conditions in castor ranged between -1.0 and -1.5

MPa (Babitha, 2003). During dry spell Ψ_l in groundnut cultivars varied between -0.8 to -1.9 MPa. The high water status of the cultivars during the dry spell indicated their ability to tolerate drought (Ramesh *et al*., 2003). During the stress period, compared to irrigated conditions dry spell significantly decreased Ψ_l of castor genotypes. Genotypes showed a declining trend as stress progressed (Babitha, 2003). Genotypes PCS-136, 124 and VP-1 recorded higher Ψ_l during the dry spell indicating that these genotypes maintained high water stress. Incidentally these genotypes recorded high transpiration (Table 11). Water stress recovery was understood by high Ψ_l at 45 and 105 DAS. The Ψ_l ranged from -0.88 to -0.33 MPa. Genotypes PCS-136, VP-1 proved better up on relief.

5.4 OSMOTIC POTENTIAL

The osmotic potential values decreased during dry spell at 30 DAS (-1.16 MPa) and 75-104 DAS (-1.27 to -1.10 MPa). Among the genotypes PCS-136, Kranthi and VP-1 recorded low Ψs (Table 13). One genotype PCS-136 consistently revealed lower Ψs at 30 DAS (-1.25 MPa), at 90 DAS (-1.52 MPa) and even after recovery from stress also i.e. at 45 DAS (-1.30 MPa) and at 120 DAS (-1.30 MPa) (Table 13). Incidentally, the genotypes PCS-136 and VP-1 which showed low Ψs, recorded high leaf water potential (Table 12). Levitt (1980) reported that OA decreases leaf water potential by accumulating compatible solutes like proline, total soluble sugars, total free amino acids and

potassium and thereby maintain turgor. In terms of recovery from stress, lower OA was recorded at 45 DAS (-1.3 MPa) and at 104 DAS (-1.52 MPa) by PCS-136 (Table 13). Babitha (2003) reported that castor genotypes accumulated potassium and soluble sugars under stress.

5.5 SPAD CHLOROPHYLL METER READINGS (SCMR)

The SPAD units decreased with the age of the crop (35.40 to 27) and during the dry spell at 30 DAS (35.4 to 35.14) and at 75-104 DAS (32.85 to 29.97). At 30 DAS, high SPAD values were recorded by PCS-138 (37.35) followed by PCS-124 (37.20). In terms of recovery at 45 DAS, high SPAD units were recorded by PCS-137 (37.93) followed by PCS-124 (37.19). During second dry spell PCS-137 recorded high SPAD units at 75 DAS (36.65) and 90 DAS (33.5). Upon recovery from stress PCS-137 and 136 recorded high SPAD values at 105 DAS (30, 30.5), at 120 DAS (30.5, 30) and 135 DAS (29.5, 29.0) SPAD values did not increase after dry spell which is an indication that plants are still under stress (Table 14). Nageswara Rao *et al*. (2001) reports that there were significant relationship between specific leaf nitrogen (SLN) and specific leaf area (SLA) and SLN and SCMR. Hence, SCMR could be used as a reliable measure to identify genotypes with low SLA or high SLN and hence high TE in peanut.

5.6 WATER STRESS AND CROP GROWTH

The dry matter of castor genotypes increased upto 75 DAS and thereafter declined. The dry matter of stem (Table 15), leaf (Table 16), spike

(Table 17) and total dry matter (Table 18) showed a similar decreased. Orcutt and Hopkins (1988) reported that the stem length and dry matter in groundnut plants decreased markedly due to drought and the decrease was entirely dependent on the availability of soil water to the plants. Similar decreases in dry matter under stress was also reported in cotton (Taylor and Klepper, 1974), in sunflower (Prabhudeva *et al*., 1998) and in groundnut (Ramesh *et al*., 2003).

During the dry spell at 30 DAS, three genotypes PCS-137 (10.12 g m^{-2}), PCS-136 (9.69 g m^{-2}) and VP-1 (9.21 g m^{-2}) produced maximum leaf dry matter. During second dry spell (75-104 DAS), the leaf dry matter decreased during which period, PCS-137, PCS-136 recorded superior leaf dry matter at 75 DAS (29.82, 28.10 g m^{-2}), at 90 DAS (22.65, 21.52 g m^{-2}) and at 105 DAS (19.30, 18.6 g m^{-2}). Stem dry mater similar to leaf dry matter increased upto 75 DAS and thereafter declined. The accumulation increased by 31 per cent between 30-45 DAS, 105 per cent by 45-60 DAS, 34 per cent by 60-75 DAS. The dry matter reserves increased in leaf and stem upto 75 DAS indicate preferential partitioning for growth and development of plant. However, the reduction in stem dry matter under stress has previously been reported in castor (Hanumantha Rao *et al*., 1986; Raghuram Reddy *et al*., 1999). During the recovery and regrowth, two genotypes PCS-136, PCS-137 recorded high dry matter (Table 15 and 16). Spikes appeared visibly around 60 DAS. The rate of increase in spike dry matter was 36 per cent (60-75 DAS), 30 per cent

(75-90 DAS), 31 per cent (90-105 DAS), 15 per cent (105-120 DAS) indicating of preferential dry matter partitioning to reproductive parts at the cost of increase in leaf or stem dry matter. During the dry spell between 75-105 DAS, the rate of increase in spike dry matter however showed a decreasing trend (Table 17). Similar reduction of spike dry matter under stress was reported in castor (Lakshmamma *et al*., 2001). Throughout the study the genotype Kranthi recorded high spike dry matter (9.17 g m^{-2}) followed by PCS-137 (7.73 g m^{-2}) and PCS-136 (7.51 g m^{-2}) possibly due to increased maintenance of low Ψs (Table 13).

Total dry matter accumulation decreased after 75 DAS (Fig. 2). Total dry matter content of the plants is known to reduce under stress in castor (Raghuram Reddy *et al*., 1999). Sunflower (Chimenti *et al*., 2002) and soybean (Chandel *et al*., 1995). During the initial dry spell (30-45 DAS), PCS-137 (15.68 g m^{-2}), PCS-136 (14.72 g m^{-2}) and VP-1 (13.62 g m^{-2}) were found to be superior. During second dry spell (75-104 DAS), two genotypes PCS-137, 136 recorded superior values at 75 DAS (55.36 g m^{-2} and 54.01 g m^{-2}) at 90 DAS (46.89, 44.56 g m^{-2}) and at 105 DAS (38.10, 36.0 g m^{-2}). After relief from water stress the dry matter increased by 73 per cent (45 DAS), 69 per cent (60 DAS) and 30.6 per cent (75 DAS). Dry matter declined after second dry spell. Genotypes PCS-137 proved superior followed PCS-134, VP-1 and Kranthi (Table 18). The increased dry matter of PCS-137,

PCS-136 could be due to increased transpiration (Table 11), high water potential (Table 12) and SPAD units (Table 14).

During the initial dry spell (30 DAS) higher leaf area was recorded by PCS-137 (664.36 cm^2) and Kranthi (606.75 cm^2). Even during subsequent dry spell (75-104 DAS) genotypes PCS-137 and Kranthi recorded high leaf area (Table 19). Sarkar (1994) showed a correlation between leaf area and drought tolerance and opined that leaf area could be used to select genotypes for drought tolerance.

LAI increased upto 75 DAS and then declined. During the initial and second dry spell high LAI was recorded in the genotypes Kranthi (0.14) and PCS-137 (0.16). These genotypes PCS-137 and Kranthi which recorded high LAI were also superior after the dry spell at 45 DAS (0.26 and 0.26) at 120 DAS (0.41, 0.47) and at 135 DAS (0.037 and 0.035) (Table 21). The LAI decreased at the time of water stress / dry spell and recovered after the dry spell. Such decrease in leaf area index under stress has been reported in mustard (Singh and Singh, 1991) sesamum (Ayyaswamy and Kulandaivelu, 1992). In groundnut, during the post stress period both LAI and biomass production rates lagged behind but upon alleviation of stress, exceeded that of control. This recovery of the plant enabled production of fast vegetative cover condition and aided in survival growth and development (Ramesh Babu *et al*., 1984).

CGR was observed to be sensitive to water stress mainly because leaf area was sensitive to water stress (Fischer and Hagon, 1965). CGR reduced during dry spell in the genotypes Kranthi (4.09 g m^{-2} d^{-1}) and PCS-136 (3.6 g m^{-2} d^{-1}). PCS-137 (3.86 g m^{-2} d^{-1}) and PCS-136 (3.44 g m^{-2} d^{-1}) recorded high CGR (45-60 DAS) and PCS-138 (-0.52 g m^{-2} d^{-1}), PCS-136 (-1.02 g m^{-2} d^{-1}) at 120 to 135 DAS. In course of recovery and regrowth, these genotypes PCS-137, 136 and Kranthi owing to higher CGR also recorded high SLA (Table 22) and incidentally low transpiration rate during and after dry spell (Table 11).

Wright *et al*. (1994) reported that a strong negative relationship between WUE and SLA indicating that genotypes with thicker leaves (lower SLA) had greater WUE. In the present study during the dry spells, low SLA was observed in 2 genotypes viz., VP-1, PCS-138 (Table 22) and these genotypes comparatively showed high transpiration (Table 11). Genotypes which show low SLA, high transpiration would produce high CGR resulting in higher yields (Udaykumar *et al*., 1998).

5.7 MOISTURE STRESS AND YIELD

The mean spike length and number of capsules decreased from primary to secondary and tertiary orders. The occurrence of dry spell before the first picking possibly decreased the yield of primary spike and increased the seed yield of second picking marginally (Table 26). In general around 50 per cent of yield is realized by primaries (Kulkarni and Ramanamurthy, 1977). Under

stress yield reduction has been reported in castor (Baby Akula and Bapi Reddy 1988, Kumar *et al*., 1989 and Raghuram Reddy *et al*., 1999). Sailasree (2001) reports that in castor the length of main spike increases at higher levels of irrigation which ultimately resulted in higher yields.

The effective length of primary, secondary and tertiary order spikes varied significantly. Kranthi recorded maximum length of primary (10.37 cm) at secondary spike (7.75 cm) followed by longer primary spikes in PCS-136 and PCS-137 (9.65, 7.25 cm). For tertiary spike maximum length was noted in Kranthi (5.75 cm) followed by PCS-136 (5 cm) and 138 (3.5 cm) showed maximum. These genotypes also produced high spike dry matter (Table 17), high capsules number (Table 24).

In all the genotypes, the lateral (third) spikes contributed more to total yield compared to that of primary spike. Kranthi recorded high seed yield of 534.16 kg ha^{-1} followed by PCS-137 (507.35 kg ha^{-1}) and PCS-136 (495.6 kg ha^{-1}).

Decrease in yield components i.e. effective spike length, 100 seed weight and number of capsules, contributed to the reduction in seed yield. Decrease in seed yield under drought due to decrease in number of capsules has been observed in sesamum (Gupta and Gupta, 1977).

Kranthi recorded maximum seed yield. This superior performance could possibly due to low osmotic potential, high spike dry matter, effective spike length and 100 seed weight. Other genotypes PCS-136, 137 which

ranked second revealed high transpiration, high water potential, SPAD units, leaf, stem and total dry matter. Parameters which were commonly contributing to yield in Kranthi, PCS-136, 137 were SLA, LAI and capsule number during stress and upon relief from stress i.e. during recovery and regrowth of genotypes.

IN VITRO STUDIES

5.8 CHANGES IN COLOUR OF SHOOT TIP CULTURES

Shoot tips of size five mm were excised from 9 day old *in vivo* grown seedlings of castor genotypes Kranthi and Aruna and cultured on MS + BA 2-5 mg l^{-1} (Table 27 and 28). Shoot tips of Kranthi were light green at 14 DAI and turned green by 28 DAI. Aruna also showed similar change i.e., light green at 14 DAI and green at 28 DAI. The green colour development of shoot tips as such is due to addition of cytokinins making the explants autonomous for growth. The attainment of green colour is due to maturation of chloroplast and cell division by addition of cytokinins (Taiz and Zeiger, 1991). Cytokinins as such are known to induce one or more proteins to which chlorophylly bind and became stabilized thus promote chloroplast synthesis and development. Differential response of genotypes to change in colour may be due to the possible differences in uptake, recognition and action of cytokinin (Salisbury and Ross, 1986).

5.9 MULTIPLE SHOOT INDUCTION

Shoot tips obtained from *in vivo* grown seedlings of castor genotypes Kranthi and Aruna were used for the study. Available literature on shoot tip cultures of different oil seeds indicated that MS medium supplemented with BA was effective in many instances in inducing multiple shoots in castor (Prasanna Athma and Reddy, 1987, Sujatha and Reddy, 1998, Biju *et al*., 2002 and Rampal *et al*., 2003).

5.9.1 Effect of media and genotype on multiple shoot induction

Skoog and Miller (1957) suggested that initiation of organogenesis involves the critical balance between auxins and cytokinins, but a relatively higher level of auxin favoured the root differentiation, while higher cytokinin resulted in shoot development.

Cytokinin (BA) was manipulated in the present study to optimize multiple shoot induction. Shoot tips of castor genotypes Kranthi and Aruna were cultured on MS medium supplemented with BA. A mean maximum of 4.0 multiple shoots were produced per shoot tip in MS medium supplemented with BA 2 mg/l (Table 27). The results revealed that number of shoots produced per shoot tip was more in MS media containing BA indicating BA was more effective. Explants cultured on BA supplemented media showed differential response in other studies on castor (Sujatha and Reddy, 1998).

Genotypic differences were also observed in the multiple shoots initiated. A maximum of 5.5 and 4.5 multiple shoots were produced on MS +

BA 5 mg/l in Kranthi and MS + BA 2 mg/l in Aruna respectively (Table 28). Other studies on castor revealed striking genotypic differences. Molina and Schobert (1995) reported a maximum of 3.6 shoots MS + 1000 nM.

Weber *et al*. (2000) reported that shoot apices cultured on MS + 0.1 mg l^{-1} BAP resulted in direct regeneration of 3.5 shoots/apex. Multiple shoots could be proliferated in castor when cytokinin was supplied singly as BA 0.5 to 2 mg l^{-1} (Prasanna Athma and Reddy, 1987) or 5.0 mg/l (Biju *et al*., 2001) or 12 mg l^{-1} (Sujatha and Reddy, 1998).

5.9.2 Multiple shoot induction under PEG induced drought stress

To induce drought stress polyethylene glycol (PEG) and mannitol have been widely used (Srivastava, 1998). A drastic reduction in water potential and turgor potential in stress media was observed with increase in concentration of PEG in the medium (Purushotham *et al*., 1998). A reduction in water potential can affect the physiology of cells by changing the relationship among inter cellular membranes of cell organelles (Nilsen and Orcutt, 1996). In the present study -3 to -6 bars of stress was induced using PEG (Table 28). A maximum of 5.5 and 4.5 shoots in non stress media (MS + BA 5 and 2 mg/l) in Kranthi and Aruna respectively as compared to a maximum of 3.5 shoots. Per shoot in PEG stress medium at -3 bars were recorded (Table 29). As such -3 bars stress itself proved inhibitory to the production of multiple shoots. Similar to our results, regeneration frequency

under drought stress in groundnut reduced from 24.37 to 5.71 per cent when compared with 85.0 per cent in control medium (Venkateswarlu, 1998).

The hypocotyl segments of sunflower cultivars inoculated on MS medium with different levels of PEG showed a decrease in callus development at -8 bars of stress (Prakash *et al*., 1994). Srivastava (1998) reported that NaCl interferes with the regeneration process by disturbing the balance of the phytohormones. Thus, it was understood that PEG also interferes with multiple shoot induction process by changing the balance of phytohormones.

A protocol has thus been standardized for initiation of multiple shoots *in vitro* under normal (Biju, 2001) and PEG induced stress conditions (Rampal, 2003). Protocols has now been further standardized in genotype Aruna and Kranthi under PEG induced stress and plantlets regenerated in terms of stress recovery and their regrowth.

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ABSTRACT

A field experiment was conducted during the *kharif* 2003-04 at College Farm, College of Agriculture, Rajendranagar to identify the castor genotypes that show quick recovery and regrowth after the relief of water stress with six genotypes of castor viz., PCS-124, VP-1, Kranthi, PCS-136, 137, 138. The genotype were sown in four replications adapting a spacing of 90 x 30 cm in 4.5 x 4.5 m^2 plots.

Two dry spells occurred between 30-44 and 75-104 DAS during which period the genotypes were evaluated for stress. Soil moisture content at 30 DAS was 6.75 per cent. At 30 DAS, plots with genotypes PCS-124 and 136 recorded low soil moisture content. At 75-104 DAS, soil moisture was 8.68 per cent wherein PCS-136 and PCS-137 recorded lower soil moisture content. After dry spell at 45 and 105 DAS soil moisture content was 13.31 and 8.68 per cent, respectively, the recovery and regrowth of castor genotypes was quantified by recording data on parameters like transpiration, leaf water potential, osmotic potential, SPAD units, dry matter of leaf, stem, spike and total dry matter, CGR, LAI, SLA and yield components.

Mean transpiration of six castor genotypes during the crop growth ranged between 18.5 to 55.16 $\mu g\ m^{-2}\ s^{-1}$. At 30 DAS, transpiration rate was 48.54 $\mu g\ m^{-2}\ s^{-1}$. PCS-136 recorded high transpiration rate. At 75-104 DAS, transpiration was 27.83 $\mu g\ m^{-2}\ s^{-1}$. The genotypes PCS-136, VP-1 recorded high values. During the recovery period, PCS-136 recorded superior value of 59.25 and 53.00 $\mu g\ m^{-2}\ s^{-1}$ at 45 and 105 DAS.

Mean leaf water potential during crop growth ranged from -0.82 to 1.33 MPa. Ψ_l at 30 DAS was -0.88 MPa. Genotypes VP-1 and PCS-136 recorded high Ψ_l of

-0.8 and -0.85 MPa. At 75-104 DAS, Ψ_l values were -1.06 and -1.33 MPa. Genotypes PCS-136, PCS-138 and VP-1 recorded high Ψ_l . During recovery i.e. at 45 and 105 DAS, Ψ_l ranged from -0.62 to -0.70 MPa. Superior Ψ_l was recorded by the genotypes PCS-136 (-0.5 MPa) and VP-1 (-0.55 MPa) at 45 DAS and at 105 DAS high Ψ_l were recorded by PCS-136 (-0.6 MPa), PCS-124 (-0.6 MPa) and PCS-138 (-0.6 MPa).

Osmotic potential (Ψs) values ranged from -1.10 to -1.64 MPa. At 30 and 75-104 DAS, Ψs was -1.10 and -1.64 MPa. PCS-136 consistently recorded lower Ψs during dry spell and after relief of water stress.

SPAD units between 15-135 DAS ranged from 27 to 35.4. SPAD units at 30, 75-104 DAS were 35.40 and 27.95 to 29.97. PCS-138, 137 recorded high SPAD units during dry spell. Upon relief from dry spell, SPAD units were 35.14 at 45 DAS which were similar to those at 30 DAS (35.40). At 75-104 DAS, 27.95 to 32.85 values were recorded. Upon recovery, PCS-137 and VP-1 had high SPAD units (37.93 and 35.13) at 45 DAS and PCS-137 (31.00) and PCS-136 (30.5) at 105 DAS.

Dry matter of leaf, stem, spike and total dry matter declined after 75 DAS. Leaf dry matter at dry spell was 19.04 and 21.30 g m^{-2} and after relief it was 16.99 g m^{-2} and 17.17 g m^{-2}. Stem dry matter at dry spell were 4.81 and 12.69 g m^{-2}. After relief 6.69 and 6.17 g m^{-2} was recorded. During dry spell between 75-105 DAS, rate of increase in spike dry matter declined.

Total dry matter increased by 240.75 per cent (15-30 DAS), 73 per cent (30-45 DAS), 69 per cent (45-60 DAS), 30.6 per cent (60-75 DAS) and it decreased after 90 DAS. At 45 DAS after dry spell dry matter increased by 73 per cent. Leaf dry matter contributed maximum to total dry matter.

During dry spell CGR was 1.44 and 3.28 g m^{-2} d^{-1}. After dry spell, it was 3.35 g m^{-2} d^{-1} . LAI at dry spell was 0.13 and 0.40 and after dry spell, it was 0.22 and 0.4. SLA increase upto 135 DAS from 15 DAS except at 30 DAS. Effective spike length, 100 seed weight and number of capsules contributed to seed yield.

In conclusion, Kranthi recorded maximum seed yield. The superior performance could be possibly be due to low Ψs, high spike dry matter, effective spike length and 100 seed weight. Other genotypes, PCS-136, 137 ranked second revealed high T, SPAD, high Ψl, leaf, stem and total dry matter. Parameters commonly contributing to yields in Kranthi, PCS-136, 137 were SLA, LAI, capsule number during stress and upon relief from stress i.e. during recovery and regrowth of genotypes.

In vitro evaluation of two genotypes Aruna and Kranthi was carried out under normal and PEG stress. MS + BA was used to initiate multiple shoots. Kranthi and Aruna produced 5.5 and 4.5 multiple shoots on regeneration BA medium of 5 mg/l and 2 mg/l respectively. PEG was incorporated to induce stress. At -3 bars Kranthi recorded 2.5 and Aruna 2.0 shoots. At -6 bars, 1.5 and 1.0 shoots were recorded. Recovery and regrowth of genotypes was understood in terms of number of regenerated shoots.

CHAPTER VI

SUMMARY

The present investigation “studies on water stress recovery and regrowth in castor genotypes” was aimed at identifying the castor genotypes that show quick recovery and regrowth after the relief of water stress. The present study was carried out *in vivo* with six genotypes viz., PCS-124, VP-1, Kranthi, PCS-136, PCS-137 and PCS-138 at Agricultural College Farm, Rajendranagar, Hyderabad during *kharif* 2003. *In vitro* study was taken up with two genotypes viz., Aruna and Kranthi to understand the recovery response in terms of regenerants under PEG induced trust. The results obtained are summarized below.

During the crop growing season, the mean soil moisture ranged from 5.00 to 15.47 per cent. Two dry spells occurred i.e., between 30-44 and 75-104 DAS. Castor genotypes were evaluated during the stress period. Recovery and regrowth were quantified after the relief from stress at 45 and 105 DAS. The plots containing Kranthi, PCS-137 and PCS-138 recorded lowest soil moisture content at 30 DAS; PCS-136 and PCS-137 at 75 DAS and Kranthi, PCS-124 and PCS-136 at 90 DAS. Upon relief from stress at 45 DAS, a mean soil moisture of 13.31 per cent, was recorded. Plots with genotype PCS-136 recorded high soil moisture of 6.75 per cent which was on par with PCS-124 (6.25 %) during first dry spell. Later during the second dry spell a mean of 6.00 per cent, moisture content was recorded. Plots with VP-1 showed high

moisture values (5.25 %). Soil moisture improved at 45 and 105 DAS indicating relief from stress, plots with genotypes PCS-124 and VP-1 recorded quick recovery.

Crop response in terms of transpiration was studied during stress and after the relief of stress. The mean values of transpiration ranged from 18.5 to 55.16 $\mu g\ m^{-2}\ s^{-1}$ during the crop growth period. The transpiration values during the dry spell ranged between 48.54 $\mu g\ m^{-2}\ s^{-1}$ at 30 DAS and 18.5 to 27.8 $\mu g\ m^{-2}\ s^{-1}$ at 75-90 DAS, respectively. During the dry spell at 90, 120 DAS, PCS-136 and VP-1 recorded maximum transpiration rate (32, 30 $\mu g\ m^{-2}\ s^{-1}$) at 90 DAS (28, 26.5 $\mu g\ m^{-2}\ s^{-1}$) at 120 DAS. Transpiration rate improved at 45 DAS (55.16 $\mu g\ m^{-2}\ s^{-1}$) and 105 DAS (49.04 $\mu g\ m^{-2}\ s^{-1}$). Two genotypes PCS-136 (59.25 $\mu g\ m^{-2}\ s^{-1)}$ and VP-1 (59.25 $\mu g\ m^{-2}\ s^{-1}$) showed high transpiration at 45 DAS and at 105 DAS these genotypes recorded 53, 50 $\mu g\ m^{-2}\ s^{-1}$, respectively.

The mean values of leaf water potential ranged from -0.62 to -1.33 MPa. During the dry spell at 30 and 75-104 DAS, the genotypes recorded low Ψ_l. At 30 and 75 DAS, the Ψ_l were -0.8 to -0.95 and -0.85 to -1.15 MPa respectively. In the dry spell, the high Ψ_l was observed in genotype PCS-136 (-0.85 MPa). As the dry spell continued at 90 DAS, the Ψ_l further decreased and ranged between -1.05 to -1.55 MPa. During the dry spell, PCS-136 followed by VP-1 recorded higher Ψ_l indicating that these genotypes maintained high water status during dry spell. In terms of recovery after stress, PCS-136 and PCS-124 consistently recorded high Ψ_l.

The mean osmotic potential ranged from -1.10 to -1.64 MPa. During the dry spell the Ψs values decreased at 30 DAS, 75-104 DAS. At 30 DAS, the Ψs was -1.16 MPa and at 75-104 DAS it ranged between -1.10 and -1.64 MPa. At the end of the crop (120-135 DAS), the Ψs decreased to -1.23 and -1.18 MPa respectively. PCS-136 consistently revealed lower Ψs at 30 DAS (-1.25 MPa), 90 DAS (-1.52 MPa). Even after recovery from stress also, lower Ψs values were recorded by PCS-136 at 45 DAS (-1.30 MPa) and 120 DAS (-1.30 MPa).

The SPAD values decreased with the age of the crop. During the dry spell between 30-45 DAS, SPAD units were 35.4 to 35.14 and at 75-104 DAS, SPAD units ranged between 27.95 to 29.97. Upon relief from dry spell, the SPAD units at 45 DAS (35.14) continued to remain similar to those at 30 DAS (35.14). The SPAD units declined during dry spell at 90 DAS (29.97) to at 105 DAS (27.95). Even after receipt of rains, SPAD units at 120 DAS was low (27.8). During the initial dry spell, at 30 DAS, genotype PCS-138 (37.35) followed by PCS-124 (37.20) recorded high values. During the second dry spell, PCS-137 proved superior at 75 DAS (36.65) and 90 DAS (33.5). In terms of recovery, PCS-137 followed by PCS-124 and PCS-136 recorded high SPAD values at 105, 120 and 135 DAS.

The leaf dry matter increased upto 75 DAS and declined thereafter. Three genotypes PCS-137 (10.12 g m^{-2}), PCS-136 (9.69 g m^{-2}) and VP-1 (9.21 g m^{-2}) were superior at 30 DAS and PCS-137, PCS-136 recorded superior leaf

dry matter values at 75 DAS (29.82, 28.10 g m^{-2}), 90 DAS (22.65, 21.52 g m^{-2}) and 105 DAS (19.3, 18.6 g m^{-2}). The genotypes which recorded high leaf dry matter were also superior in dry matter production after the dry spell. At 45 DAS, the dry matter increased by 87 per cent. Among the genotypes, PCS-136, PCS-137 and VP-1 showed increased leaf dry matter accumulation. Two of these genotypes PCS-137 and PCS-136 recorded superior dry matter at 120 (17.38 and 15.89 g m^{-2}) and 135 DAS (15.26 and 13.12 g m^{-2}). During the dry spell and in course of their recovery, three genotypes PCS-137, PCS-136 and VP-1 proved superior in accumulating leaf dry matter.

The stem dry matter showed continuous increase upto 75 DAS and declined thereafter. At first dry spell (30-44 DAS), two genotypes PCS-136 and PCS-137 and during the second dry spell (75-104 DAS), PCS-137 (15.04 g m^{-2}), PCS-136 (14.85 g m^{-2}) and VP-1 (14.69 g m^{-2}), were superior. Upon recovery from stress, the stem dry matter accumulation showed a continuous increase by 31 per cent at 30-45 by 105 per cent at 45-60 and 34 per cent 60-75 DAS. Among the genotypes PCS-136, PCS-137 and VP-1 were superior at first and second dry spell and also recorded high values during recovery periods from water stress.

The spike dry matter increased from 60-120 DAS and thereafter declined. The mean spike dry matter was 1.30 to 12.10 g m^{-2}. During the dry spell between 75-105 DAS, the rate of increase in spike dry matter declined. Among the genotypes studied Kranthi recorded maximum dry matter of 8.26,

10.6 and 12.93 g m^{-2} followed by genotypes viz., PCS-136 (6.91 g m^{-2}) and PCS-137 (6.47 g m^{-2}). The spike dry matter accumulation continued even after the dry spell upto 135 DAS indicating the partitioning of assimilates into spikes.

The total dry matter (leaf + stem + spike) increased upto 75 DAS and declined thereafter. During the dry spells (30-44 DAS), the genotypes PCS-137 (15.68 g m^{-2}), PCS-136 (14.72 g m^{-2}) and VP-1 (13.62 g m^{-2}) were found to be superior. At 75-104 DAS, two genotypes viz., PCS-137 and 136 recorded superior values. The genotypes which recorded high total dry matter during dry spell recorded superior values even after dry spell. At 45 and at 120 DAS, the genotypes PCS-137, 136, VP-1 showed increased total dry matter accumulation.

The leaf area increased upto 75 DAS and declined thereafter during dry spell at 30 DAS and 75-104 DAS. Two genotypes PCS-137 and Kranthi recorded high leaf area (664.36, 606.75 cm^2). The genotypes which recorded high leaf area were also superior after the dry spell. At 45 and 120 DAS, PCS-137 and Kranthi recorded high leaf area (2696, 2616 cm^2) at 45 DAS (4152 and 4082 cm^2) at 120 and (1644, 1434 cm^2) at 135 DAS respectively.

The mean leaf area index ranged from 0.03 to 0.58. LAI increased upto 75 DAS and then declined. LAI followed same trend as that of leaf area.

The crop growth rate increased steadily upto 60-75 DAS and decreased there after. The mean CGR was -2.65 to 3.59 g m^{-2} d^{-1}. During dry spell

(30-45 DAS), genotypes showed no significant differences. During the second dry spell, at 75-90 DAS, Kranthi recorded high CGR values (4.09 g m^{-2} d^{-1}) followed by PCS-136 (3.6 g m^{-2} d^{-1}). After dry spell (45 DAS) genotype PCS-137 recorded high CGR (3.86 g m^{-2} d^{-1}) and was on par with VP-1 (3.44 g m^{-2} d^{-1}). At 120 to 135 DAS, PCS-138 recorded high CGR (-0.52 g m^{-2} d^{-1}) and was on par with PCS-136 (-1.02 g m^{-2} d^{-1}).

Specific leaf area increased from 15 DAS to 135 DAS except at 30 DAS. Mean SLA ranged from 88.44 to 259.82 cm^2 g^{-1}. During the initial dry spell at 30 DAS, the genotype PCS-137 recorded high SLA of (98.06 cm^2 g^{-1}) and was on par with Kranthi (94.06 cm^2 g^{-1}) and PCS-136 (94.04 cm^2 g^{-1}). At 75 DAS, Kranthi (191.14 cm^2 g^{-1}) followed by VP-1 (183.06 cm^2 g^{-1}) showed high SLA. At 90 and 120 DAS, PCS-137 and Kranthi were superior.

Yield and yield components were quantified in terms of effective spike length, number of capsules and 100 seed weight. The effective length of primary, secondary and tertiary order spikes varied significantly. Kranthi recorded maximum length of primary (10.37 cm) and second order spikes (7.75 cm) and was on par with PCS-136 (7.75 cm), PCS-137 (7.25 cm). For tertiary spikes, Kranthi (5.75 cm) followed by PCS-136 (5 cm) and PCS-138 (3.5 cm) recorded maximum length. These genotypes also produced high spike dry matter.

The number of capsules harvested varied significantly with different spike orders and the number of capsules formed in primary order spikes was

lower compared to second and tertiary spikes. The capsule no. increased little between first and second harvest due to dry spell. The genotype Kranthi (39.89) followed by PCS-137 (37.6) and PCS-136 (35.2) recorded maximum capsules. These genotypes also recorded maximum effective spike length and maximum spike dry matter possibly contributing to high capsule number.

The 100 seed weight increased in all the harvests. During the first and second harvest, dry spell occurred as a result there was marginal increase in 100 seed weight from 28.36 to 30.63 g. Kranthi followed by PCS-137 and 136 recorded maximum 100 seed weight values.

The seed yield comprises of yield data from first, second and third pickings. The seed yield increased from first to third pickings. In all the genotypes, the lateral (third) spikes contributed more to the total yield than that of primary spike. The total yield ranged from 442.76 to 534.16 kg ha^{-1}. Among genotypes, Kranthi (534.16 kg ha^{-1}) followed by PCS-137 (507.35 kg ha^{-1}) and PCS-136 (495.60 kg ha^{-1}) recorded high seed yield. The same trend was reported in the harvest index values. The highest HI was recorded in Kranthi (43.75 %) followed by PCS-137 (41.25 %), PCS-136 (40.75 %) and VP-1 (39.25 %).

Correlation matrix for seven characters like CGR, LAI, SLA, spike length, number of capsules, 100 seed weight and yield have been worked out for the entire crop growth period. In the present study, yield contributing characters showed significant associations. Effective spike length showed

significant association with capsule number (r = 0.9060) and 100 seed weight (r = 0.9323). Capsule number in turn showed significant association with 100 seed weight (r = 0.9912). The character association needs to be worked out separately for stress and non stress situation to know the contributing effect of stress and non stress conditions.

In conclusion, genotypes PCS-136, 137 and Kranthi performed better under stress in terms of yield. The genotypes PCS-136, 138, VP-1 showed their superiority in maintenance of traits related to water relations and chlorophyll status.

IN VITRO STUDIES

The present investigation was carried out with two genotypes of castor Kranthi and Aruna. The genotypes were cultured *in vivo* from 9 day old seedlings, and shoot tips were excised for culture *in vitro*. The multiple shoot induction *in vitro* under normal and PEG induced stress conditions was studied. The results obtained were summarized.

Shoot tips of Kranthi and Aruna were light green in colour at the time of inoculation. A change in colour of shoot tip to green was observed in both Kranthi and Aruna by 28 DAI.

Shoot tip explants were cultured to induce multiple shoots on MS medium supplemented with BA. A maximum 5.5 and 4.5 shoots per shoot tip were initiated in Kranthi and Aruna respectively.

Drought stress of -3 to -6 bars was imposed by fortifying the MS media containing BA 5 mg/l with polyethylene glycol (PEG). A maximum of 3.5 and 3.5 multiple shoots were recorded in Kranthi and also Aruna at -3 bars PEG. Increase in the drought stress level (-3 to -6 bars) decreased the number of multiple shoots produced per shoot tip. At -3 bars stress the initiation of a maximum of 3.5 shoots proved inhibitory. At -4 bars stress a maximum of 2.5 and 2.0 shoots were recorded by Kranthi and Aruna and at -6 bars there is still decrease in multiple shoots which recorded maximum of 1.5 and 1.0 by Kranthi and Aruna, respectively. Thus a protocol has been standardized to induce multiple shoots *in vitro* and understand the recovery ability of genotypes in terms of regenarants initiated.

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