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MORPHO-PHYSIOLOGICAL CHANGES OF *Jatropha Curcas* LEAVES CULTIVATED IN MARGINAL LAND UNDER RAIN FED CONDITION

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ABSTRACT

Jatropha (*Jatropha curcas* L.) used as a source of biofuel is well documented. The physiological parameters of Jatropha have received less attention. In the present study, research was designed to evaluate the Morpho-Physiological study of leaf quality of Jatropha. An experiment was carried out in research field of Department of Botany, University of Science and Technology, Bannu during 2012. One year old J. curcas using 1.8m and 1.8m plant to plant distance. Randomized Completed Block Design was applied and experiment was replicated three times. One year old Jatropha plants were used to collect ten leaves from each canopies. Three canopies height (branches) from the ground were selected i.e. 60-80cm (lower canopy), 100-120cm (Middle canopy) and 160-180cm (upper canopy). Lower canopy (60cm) leaves showed maximum Chlorophyll-a (2.2mg/g), Chlorophyll-b (1.933mg/g), Carotenoid (0.100mg/g), total chlorophyll content (4.133mg/g) and leaf area (946.267cm^2). Upper canopy leaves (180cm) recorded maximum relative water content (90.10%) and increased leaf relative membrane permeability (28.6%). As minimum relative membrane permeability (17.3%) was observed in middle leaves. It is concluded that lower canopy leaves were more efficient and have potential for all photosynthetic process of plant.

Keywords: Jatropha, canopy, chlorophyll content, relative membrane permeability, leaf area, relative water content.

1. INTRODUCTION

Jatropha is a potential multipurpose plant with various benefits especially for oil that can be used instead of fossil fuel and kerosene (Openshaw, 2000). *Jatropha curcas* or Physic nut is a perennial poisonous shrub belong to the *Euphorbiaceae* family. It is an uncultivated non-food wild-species. The plant, originating in Central America, and is mainly grown in Asia and Africa. It is used as a fence to protect fields and gardens from animals. It is resistant to a high degree of drought stress and as such does not compete with food crops. The seeds contain 25-40% oil that can be processed to produce a high-quality biodiesel fuel, usable in a standard diesel engine (Kongsamai *et al.*, 2014). Leaf age is closely correlated with photosynthetic capacity, such as the maximum electron transport rate (Iacono and Sommer 2000), total chlorophyll content and carotenoid composition (Yoo *et al.*, 2003). It showed that relative canopy height was significantly different among accessions only at the first four months after transplanting. Jatropha varieties had higher percentage of canopy height changes. The relative canopy height of each variety was non-significantly increased after the fourth month of transplanting (Kongsamai *et al.*, 2014).

The accumulation of osmotically active solutes in the cell and the elasticity coefficient of cell walls are mechanisms of adaptation to drought and play an important role in maintaining the growth of the plant (Pimentel *et al.*, 1999; Ghoulam *et al.*, 2002). However, an increase on ion concentration as response to decreasing on turgescence is limitanting to secondary reactions of the photosynthesis and respiration (Larcher, 2006). The frequency and intensity of water stress could affect severely the morpho-physiological mechanisms in plants. The plant survival depends on the degree of damage to cell membranes and their ability to adjust rapidly to a stress condition (Larcher, 2006).

Leaves are highly important organs of a tree, responding very sensitively to growth conditions in a stand, especially during a leaf expansion phase (Barna, 2004). Morphological alterations as reductions in root growth, leaf area and stomatal closure when the plant is acclimating to drought are the main symptoms of water deficit (Davies *et al.*, 2002). Net photosynthetic rate, stomatal conductance and transpiration rate in the upper leaves were higher than in the lower leaves of peanut (Nautiyal *et al.*, 1999). Proper prunings of the branch in the dormancy phase, when leaves are shed seem to be efficient technique to induce further branching. Jatropha curcas has deciduous leaves, with falling leaves in the dry season, which reappear soon after the first rains and is considered a xerophytic species, with strong resistance to drought (Pompelli *et al.*, 2010; Matos *et al.*, 2012).

Therefore, in order to balance vegetative and reproductive growth, appropriate pruning technique for J. curcas must be explored. One of the most important cultural practices that may affect growth and seed and oil yield of Jatropha curcas is top-cutting or pruning. However, pruning of branches requires skill or technique to achieve success. According to reviewed literature, pruning of live branches decreases the photosynthetic activity of the plant. The removal of vegetative growth reduces both stored and manufactured carbohydrates in the leaves, which could accumulate higher level of nitrogen that stimulates vegetative growth at the expense of reproductive growth (Cutler, 2000). Improper pruning could also restrict the flow of essential nutrients needed for growth, leading to defoliation and eventually death (Samsam, 2013).

The purpose of this trail was to study the effects of canopy heights on leaf parameters and proper pruning sites and avoid cutting of potential branching in canopies before spring for increased seed yield production of Jatropha curcas.

2. MATERIAL AND METHODS

2.1 Location of experiment

Bannu District is approximately 192 km (119 mi) south of Peshawar and lies within a sedimentary basin. It is flanked on all sides by the hard and dried mountain ranges of Koh-e-Safed and Koh-e-Suleiman.

Experiment was conducted in the research field of Department of Botany, Faculty of Biological Sciences, University of Science and Technology, Bannu. Jatropha was sown in April, 2012 at the depth of 15cm in soil. Cutting was sown at row to row distance of 1.8m and 1.8m plant to plant distance, experiment was replicated three times. Jatropha plants were divided into three canopy heights i.e 60-80cm, 100-120cm and 160-180cm above the ground, label them as upper, middle and lower canopy respectively. Five plants were selected in every replication and five different branches leaves were collected for data analysis.

2.2 Chlorophyll a, b, Total Chlorophyll and Carotenoids

Photosynthetic pigments were extracted that first leaves was taken and then weigh 0.1gram are place in Pistil and Mort's. Add 5ml of 95% Ethanol, a little amount of silica gel and calcium carbonate (Xiao and Wang, 2005). Then homogenize and crushed it properly in Pistil and Mort's. The extract which obtained is filter through 12.5 cm filter paper. During filtration adding ethanol to make 25 ml solution of the filtrate extract. The extract are poured in the couvets and placed it in the UV spectrophotometer at 665nm for chlorophyll-a, 649 nm for chlorophyll-b and 470nm for carotenoid. The values which taken from spectrophotometer are calculated by the following formulas. Formula for chlorophyll a: Ca = 13.95(D665) - 6.88(D649), formula for chlorophyll b: Cb = 24.96(D649) - 7.32(D665), formula for Total chlorophyll: Ct = Ca + Cb, formula for Carotenoid: cart = {1000(D470) – 2.05Ca - 114.8Cb} ÷ 245

2.3 Relative Water Content

The leaves was taken and four leaves discs of 1cm diameter of each leaves were sampled and immediately weighed fresh weight (FW). Then they were immersed in distilled water in petri dishes for 24 hours at 4C° in darkness and the turgid weight (TW) determined. The discs were dried in an Oven at 70C° for 24 hours and then the dry weight (DW) obtained (Lugojan and Ciulca, 2011). Then Relative Water Content (RWC) was calculated as give below:

RWC (%) = FW – DW ÷ TW – DW × 100

2.4 Relative Membrane Permeability

The relative membrane permeability was determine in term of ion leakage from the control and stressed following the protocol of Yang et al., 1996. Relative membrane permeability is determined as the context of ion leakage. That is three discs of each leaves cut into 1cm diameter and these freshly prepare discs placed into plastic tubes containing 10ml deionized distilled water. After Vortex the sample for 3 seconds initial electrical conductivity (EC1) of each sample are measured. The sample are then incubated at 4C° for 24 hours and electrical conductivity (EC2) measure again. The samples are then autoclaved at 120C° for 15 minutes and cooled to room temperature and electrical conductivity (EC3) measured for third time. The Relative Membrane Permeability (RMP) is calculated:

RMP = (EC2 – EC1) ÷ (EC3 – EC1) × 100

2.5 Leaf Area (Cm^2)

The leaves area is traced on the graph paper, the squares inside the perimeter of the traced leaves area are counted, both the bigger and smaller squares and leaves area calculated. Take sample of leaves from the plant and trace perimeters of the leaves on graph paper. Count the squares inside the perimeter and the smaller square in the bigger squares that are not included completely in the perimeter, if a smaller square is included more than half count it; however, ignore squares which are included less than half in the perimeter. I use the graph paper with lines draw in English system, one gig square is equal to one square inch (6.4516 cm2) and one smaller square is equal to one hundredths of a square inch (0.64516 cm2). Calculated the area as follow:

Leaves area = {(number of bigger squares) × (area of the bigger square)}
+ {(number of smaller squares) × (area of smaller square)}

2.6 Leaf Area (Cm^2)

Area of each individual leaf was determined in cm2 using leaf area meter (LI-COR, LI-3000 A, PAM 1822, USA) method adopted by Muhammad *et al.,* (2013).

2.7 Statistical Analysis

Data analysis was performed by using the M-STATC statistical software (Version 6.12). Randomized complete block design (RCBD) was used in this experiment. Mean separating were performed by least significant difference test (LSD) at 5% level. The regression analysis was plotted using the MS Excel.

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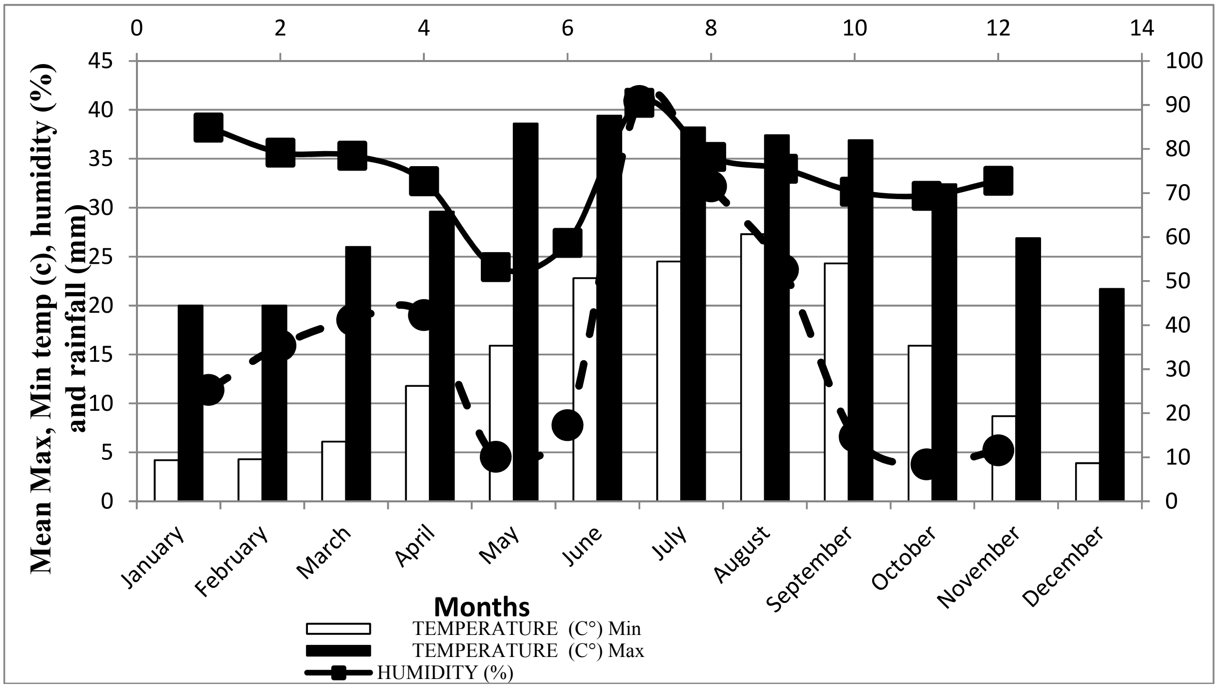

Figure-1. Mean data on monthly minimum, maximum temperature (C°), relative humidity (%) and rainfall (mm) during the growing season 2012.

3. RESULTS AND DISCUSSIONS

3.1 Chlorophyll-a (mg/g)

Chlorophyll *a* is a specific form of chlorophyll used in oxygenic photosynthesis. It absorbs most energy from wavelengths of violet-blue and orange-red light. This photosynthetic pigment is essential for photosynthesis in eukaryotes, cyanobacteria and prochlorophytes because of its role as primary electron donor in the electron transport chain. Chlorophyll-a was not significantly affected by different canopies leaves (Table-1). Lower canopy leaves showed maximum chlorophyll-a (2.23 mg g^{-1}), while upper canopies leaves showed minimum chlorophyll a (1.58 mg g^{-1}) and middle leaves showed (2.04 mg g^{-1}). The reason could be that the upper leaves are unmature leaves as compared to older leaves, which have sufficient pigments for photosynthesis. Regression analysis of chlorophyll-a showed that chlorophyll-a content gradually increased with amount of (0.32 mg g^{-1}) from younger to older leaves (Fig. 2). In C. Prucera young leaves had more chlorophyll a and b than older leaves, while there was not much differences in S. holosericea and A. Javanica (Khan, 2002).

3.2 Chlorophyll-b (mg/g)

Chlorophyll b helps in photosynthesis by absorbing light energy. Chlorophyll-b was not significantly affected by different canopies leaves. Mean data for chlorophyll-b is presented in Table-1. Lower canopy leaves showed more chlorophyll-b (1.95 mg/g), while upper leaves showed less chlorophyll-b (1.57 mg/g) and middle leaves observe as 1.31 mg/g. Regression analysis showed that chlorophyll-b content gradually increased, when the leaves collected in lower canopy with amount of (0.1884 mg/g) and have co-efficient of determination value is 0.3483 (Figure-2).

3.3 Carotenoid (mg/g)

Two main functions of Cartenoid are photo protection and light collection. Carotenoids are classified as accessory pigments in photosynthesis because they augment light harvesting in the blue spectral region by transferring the absorbed light energy to chlorophyll. Carotenoid was not significantly affected by different canopies of Jatropha leaves. Upper canopy leaves showed maximum carotenoids (0.14) as compared to lower canopy leaves (0.066) and middle leaves observe as (0.44). Regression analysis showed that carotenoids content decreased in lower canopy leaves with amount of (-0.0365) and have co-efficient of determination value is (0.0342) (Figure-2). Chlorophyll-a, chlorophyll-b and carotenoid content decreased due to salinity in young and old leaflets at all stages of Mungbean (Wahid *et al.*, 2004). The highest content of carotenoids and the greatest conversion of violaxanthin to zeaxanthin were found in the 5th leaf (middle) as compared to 3rd, 4th (younger) and 6th (older) leaf of rice (Wei *et al.*, 2002).

3.4 Total Chlorophyll (mg/g)

Total chlorophyll was not significantly affected by different stages of Jatropha leaves. Lower canopy leaves showed maximum chlorophyll (4.18) while upper leaf showed minimum chlorophyll (3.15749) and middle leaf observes as (3.36). Regression analysis showed that chlorophyll content increased when leaves were selected in lower canopies with amount of (0.5143) and have co-

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efficient of determination value is (0.8928). Accordingly, upper and lower canopy leaves at the highest altitude had approximately the same amount of chlorophylls (Rajsnerová, *et al*., 2015)

3.5 Relative Water Content (%)

Upper canopy leaves showed maximum relative water content (90.173), while lower canopy leavees showed minimum relative water content (86.498) and middle canopy leaves observe (86.947). As more photosynthetic activity in younger leaves so more water is needed as compared to older leaves. Gonçalves (2011) reported that the analysis of variance showed the RWC did not differ between treatments in all evaluation periods. The RWC remained high, around 70% even in time of more prolonged stress exposure, indicating that *Jatropha curcas* has mechanisms to minimize the loss of water in their leaf issues when submitted to drought stress. The RWC of the top leaf was higher than that of the bottom leaf (Luke, 1974). Regression analysis showed that relative water content gradually decreased when the leaf become older, with amount of (1.8375) and have co-efficient of determination value is (0.8401) (Figure-3).

3.6 Relative membrane permeability (µs/cm)

Relative membrane permeability is an indicator of plant resistance to environmental stresses (Saneoka *et al.,* 2004). Mean data for cell membrane permeability were highest in upper canopy leaves as compared to (28.636) lower canopy leaves (18.909) and middle leaves observe as (17.335). [22] Gonçalves (2011) reported that the rate of electrolyte leakage was lower than 40% for all treatment. Statistic differences, by the Tukey test at 5% probability, were observed between non stressed and stressed plants to water deficit more than 120 hours. Lower values for stressed plants after 192 hours of water restriction may indicate major membrane damage. The rate of electrolytes leakage was low in all treatments, showing that *Jatropha curcas* can keep the cell membranes intact, even after pronounced water stress and wilting visible. Minimum leaf injury was denoted in older leaves are more resistant to the environmental condition as compared to upper leaves. Besides this upper leave are soft as compared to mature leaves. Protoplasts from younger leaves were more tolerant to the toxic effect of cell wall-degrading enzymes than protoplasts from older leaves of Tobacco (Barna and Györgyi, 1992). Cell membrane permeability regression analysis showed that cell membrane permeability gradually decreased with amount of (-4.8635) and have co-efficient of determination value is (0.6313) (Figure-3).

3.7 Leaf Area (cm^2)

Leaf area was significantly affected by different stages of Jatropha leaves. Mean data for leaf area is presented in table No. 2. That lower leaf showed maximum leaf area (946.322 cm^2) while upper leaf showed minimum leaf area (443.682 cm^2) and middle leaf read as (636.129 cm^2). Amin *et al.,* (1999) reported that using compost resulted an increase in plant height, number of leaves / plant, root length; stem diameter, leaf area and upper & dry weight of all plant organs than control plants and other treatments. Compost through its content of humic substances improved soil physical, chemical and microbiological conditions, moisture content and reduced leaching of nutrients, water runoff and soil erosion. Campos *et al.*, (2012) reported that the increase in salinity level has inverse relationship with leaf area. Thus, reductions occur in the area of energy capture and fixing of CO2 per unit area. The low rates of carbon assimilation were also caused by water deficits, inherent in osmotic stress, which may cause partial stomatal closure and decreases in *g*s and *E* values. Irrigation of *J. curcas* with NaCl induced leaf abscission and increased the LT. The negative correlation between EC and leaf area and the positive one between EC and LT suggest that salt affects stomatal conductance. Regression analysis in showed that leaf area gradually increased with amount of (251.32) and have co-efficient of determination value is (0.982), the reason could be that mature leaf are fully expended while upper leaves are small size (Figure-3). Morphological, biochemical and physiological characteristics in lower leaves matured earlier than in upper leaves, moreover Leaves in the lower crown produced a large amount of photosynthate (1.35 mg C·day^{-1}) using the high light intensity of early spring (Yoshimura, 2013).

The mean leaf area of dominant trees increased from the upper layer to the lower layer crown on all the sampling plots (Barna, 2004). Leaf area is an important variable for most physiological and agronomic studies involving plant growth, light interception, photosynthetic efficiency, evapotranspiration and response to fertilizers and irrigation (Blanco and Folegatti, 2005, Peksen, 2007). Lower leaves were lower in dry matter and percentage and lighter per unit area than leaves higher on the plant. The present study showed that MDA accumulated to a greater extent in older leaves than in younger ones, indicating that the latter have a lower lipid peroxidation level whereas the former might tend to lose cell membrane integrity (Xu, *et al* 2011).

4. CONCLUSIONS

it is concluded that lower leaves recorded maximum chlorophyll-a content (2.2mg/g), chlorophyll-b content (1.933mg/g), carotenoids content (0.100mg/g), total chlorophyll content (4.133mg/g) and leaf area (946.267cm^2). Upper leaves recorded maximum relative water content (90.100%) and maximum leaf membrane permeability (28.6%) as compared to lower or mature leaves.

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REFERENCES

Amin *et al.,* 1999. Amin, G.; M.H. Mostafa; A.A. Amer and I.R. Refae (1999). Potential microorganisms for rapid and nutritionalty enriched compost. Proc. Of 1st Cong. On Recent Technology; i.e. in Agric., Cairo Univ. Faculty of Agric., Egypt, Nov. 27-29, Vol. IV: 804-822.

Barna B. and Borbála G. 1992. Resistance of young versus old tobacco leaves to necrotroph, fusaric acid, cell wall degrading enzymes and autolysis of membrane lipids. Physiological and molecular plant pathology. 40(4): 247-257.

Barna, M. 2004. Adaptation of european beech (*Fagus sylvatica* L.) To different ecological conditions: leaf size variation. Polish journal of ecology. 52 (1): 35-45.

Blanco F. F. and M. V. Folegatti. 2005. Estimation of leaf area for greenhouse cucumber by linear measurements under salinity and grafting. Sci. agric. (Piracicaba, Braz.) 62(4): 305-309.

Campos M. L. de O., Bety S. de H. João, A. de, A. G. Rafaela, M. C. Jarcilene, S. de A.C. Marcelo, F. P. 2012. Photosynthesis and antioxidant activity in Jatropha curcas L. under salt stress. Braz. J. Plant Physiol. 24(1): 55-67.

Davies W.J., Wilkinson S. and Loveys B. 2002. Stomatal control by chemical signaling and the exploitation of this mechanism to increase water use efficiency in agriculture. New Phytologist. 153, 449-460.

Ghoulam, C., Foursy, A. and Fares, K. 2002. Effects of salt stress on growth, inorganic ions and proline accumulation in relation to osmotic adjustment in five sugar beet cultivars. Environmental and Experimental Botany. 47: 39-50.

Gonçalves W.G., Alan C. C. Deucimar Pereira, L. Adinan, A. S. Clarice, A. M. 2011. Membrane permeabilty and relative water content in physic nuts plants submitted to fast water deficit. Gl. Sci. Technol. 04(1): 131-139.

Iacono F. and Sommer K. J. 2000. Response of electron transport rate of water stress-affected grapevines: influence of leaf age. VITIS. 39: 137-144.

Khan M. A. and N. Beena. 2002. Seasonal variation in water relations of desert shrubs from Karach, Pakistan. Pak. J. Bot. 34(4): 329-340.

Kongsamai B., Pojana S. and Nipaporn B. 2014. Relationship between chlorophyll content and chlorophyll fluorescence parameters on fruit yield of *jatropha curcas* l. Suranaree J. Sci. Technol. 21(4): 329-333.

Larcher W. Ecofisiologia, v. São, C. RiMa, 2006. p. 531.

Lugojan and Ciulca. 2011. Evaluation of relative water content in winter wheat. Journal of Horticulture, Forestry & Biotechnology. 15(2): 173- 177.

Luke G. J. 1974. Visual assessment of total plant relative water content, with the aid of photographic standards, as a guide to scheduling irrigation. Australian Journal of Experimental Agriculture and Animal Husbandry. 14(66): 76-79.

Matos F. S., Oliveria L. R, Freitas R. G. Evaristo A. B., Missio R. F. and Cano M. A. O. 2012. Physiological characterization of leaf senescence of *Jatropha curcas* L. populations. Biomass & Bioenerg, Oxford. 45(10): 57-64.

Muhammad A., S. K. Khalil, A. Z. Khan, Amanullah and S. Mehmood. 2013. Growth analysis of indigenous soybean land races. *Pak. J. Bot*. 45(3): 941-949.

Nautiyal P.C., Ravindra V. and Joshi Y.C. 2006. Net photosynthetic rate in peanut (*Arachis hypogaea* L.): Influence of leaf position, time of day, and reproductive-sink. Photosynthetica. 36(1): 129-138.

Openshaw K. 2000. A review of Jatropha curcas: an oil plant of unfulfilled promise. Biomass and Energy, 19(1):1-15.

Peksen E. 2007. Non-destructive leaf area estimation model for faba bean (*Vicia faba* L.). Scientia Horticulturae. 113: 322-328.

Pimentel C., Sarr B., Diouf O., Abboud A. C. S., Macauley H. R. 2002. Tolerância protoplasmática foliar à seca, em dois genótipos de caupi cultivados em campo. Revista Universidade Rural Série Ciências da Vida. 22: 07-14.

Pompelli M. F. Luis, R. B. Vitorino, H. S. Gonçalves, E. R. Rolim, E. V. Santos, M. G. 2010. Photosynthesis photoprotection and antioxidant activity of purging nut under drought deficit and recovery. Biomass & Bioenerg. 34(8): 1207-1215.

Rajsnerová P., Karel K., Petr H. Katerina, N. Kristýna V. Michaela K. Albert, R.U. Jordi, S. Michal, V. M. Josep, P. and Otmar U. 2015. Morphological, biochemical and physiological traits of upper and lower canopy leaves of European beech tend to converge with increasing altitude. Tree Physiology. 35: 47-60: doi:10.1093/treephys/tpu104.

Samsam C. l. 2013. Pruning techniques for Jatropha curcas L. to increase seed yield production. MMSU Science and Technology Journal. 3(1): 59-68.

Saneoka H., Moghaieb R. E. A, Premachandra G. S. and Fujita K. 2004. Nitrogen nutrition and water stress effects on cell membrane stability and leaf water relations in Agrostis palustris Huds. Environ Exp Bot. 52: 131-138.

Wahid A., Mansoor H. and EJAZ R. 2007. Salt-induced injury symptom, changes in nutrient and pigment composition, and yield characteristics of Mungbean. Int. J. Agric. Biol. 6(6): 1143-1152.

Wei C. Y., Peng C. L. Duan J., Lin G. Z. and Chen Y. Z. 2002. Responses of chlorophyll fluorescence and carotenoids biosynthesis to high stress in rice seedling leaves at different leaf position. Acta Botanica Sincica. 44(11): 1303-1308.

Xiao LT, Wang, SG 2005. Experimental techniques of plant physiology. China agriculture press, Beijing.

Xu Z., Guangsheng Z., Guangxuan H. and Yijun L. 2011. Photosynthetic potential and its association with lipid peroxidation in response to high temperature at different leaf ages in maize. j plant growth regul. 30: 41-50.

Yang G.D, Rhods and R.J. Joly. 1996. Effect of high temperature on membrane stability and chlorophyll florescence in glycine betaine containing maize lines. Australian J. plant physiology. 23: 437-443.

Yoo S. D., Greer D. H. Laing W. A. and McManus M. T. 2003. Changes in photosynthetic efficiency and carotenoid composition in leaves of white clover at different developmental stages. Plant Physiol Biochem. 41: 887-893.

Yoshimura K. 2013. Influences of phenological differences on leaf-level carbon budget between the upper and lower crown of *Lyonia ovalifolia*. Botany. 91(1): 25-33.

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Table-1. Chlorophyll-a, chlorophyll-b, carotenoid, total chlorophyll content, relative water content, cell membrane permeability and leaf area in different leaf ages of Jatropha curcas.

Leaf stages	Chlorophyll-a	Chlorophyll-b	Carotenoid	Total Chlorophyll	RWC	RMP	Leaf area
Upper leaf	1.533	1.533	0.100	3.100	90.1 a	28.6a	443.6 c
Middle leaf	1.967	1.300	0.400	3.333	86.9b	17.3b	636.1 b
Lower leaf	2.200	1.933	0.100	4.133	86.5c	18.8b	946.2 b
LSD value at 0.05	-	-	-	-	-	2.771	163.9

Table-2. Analyses of variance of Chlorophyll-a, chlorophyll-b, carotenoid, total chlorophyll content, relative water content, relative membrane permeability and leaf area in different leaf ages of Jatropha curcas.

S.O.V.	D.F	Chlo-a	Chlo-b	Carotenoid	Total Chlorophyll	RWC	RMP	Leaf Area
Replication	2	0.043	0.241	0.063	0.081	9.168	11.974	828.760
Canopies	2	0.343ns	0.308ns	0.090ns	0.881ns	11.548ns	112.441**	192943.522**
Error	4	0.062	0.128	0.063	0.258	1.808	1.494	5227.096
Total	8	-	-	-	-	-	-	-

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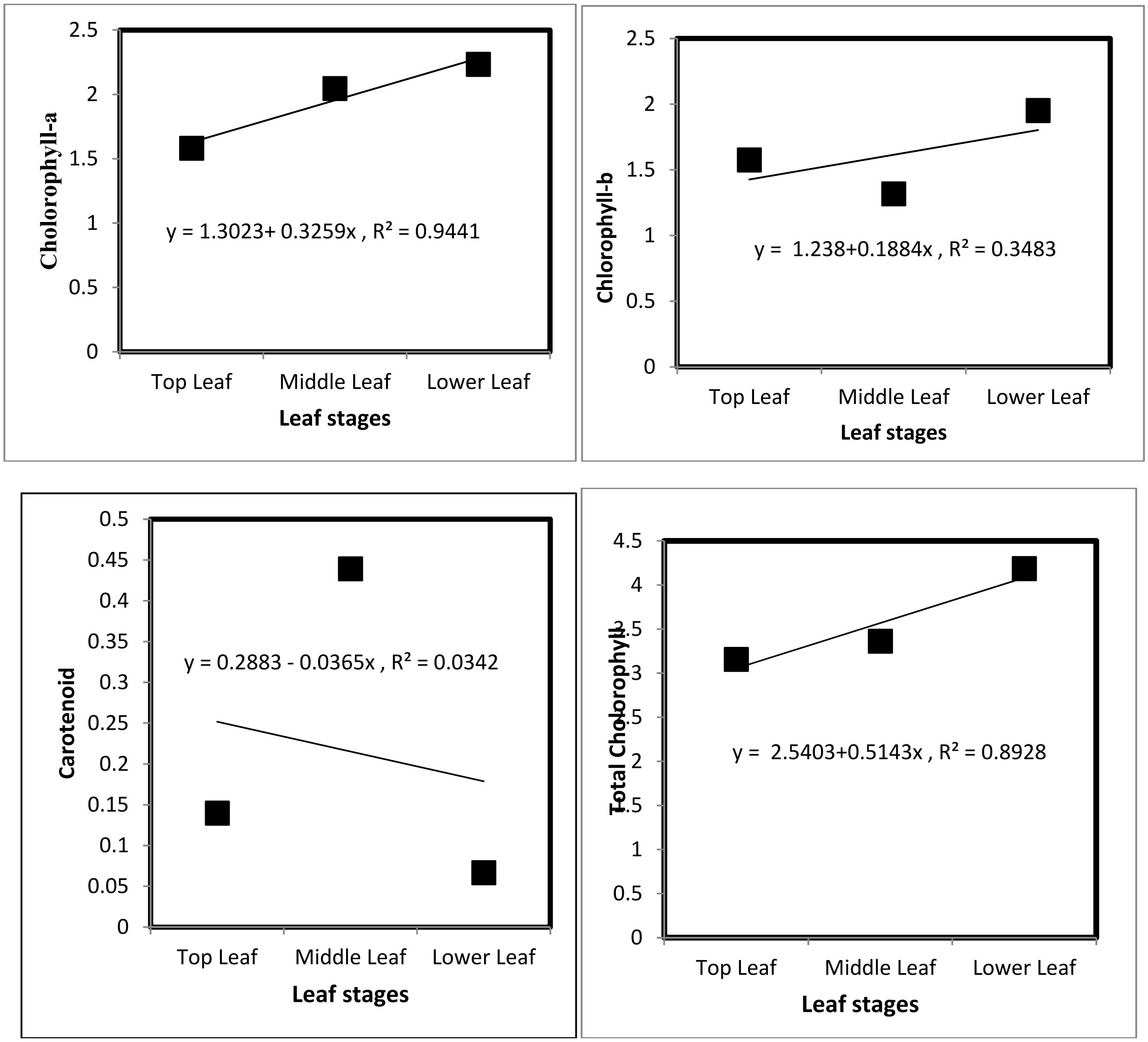

Figure-2. Chlorophyll-a, chlorophyll-b, carotenoid and total chlorophyll content different leaf ages of Jatropha.

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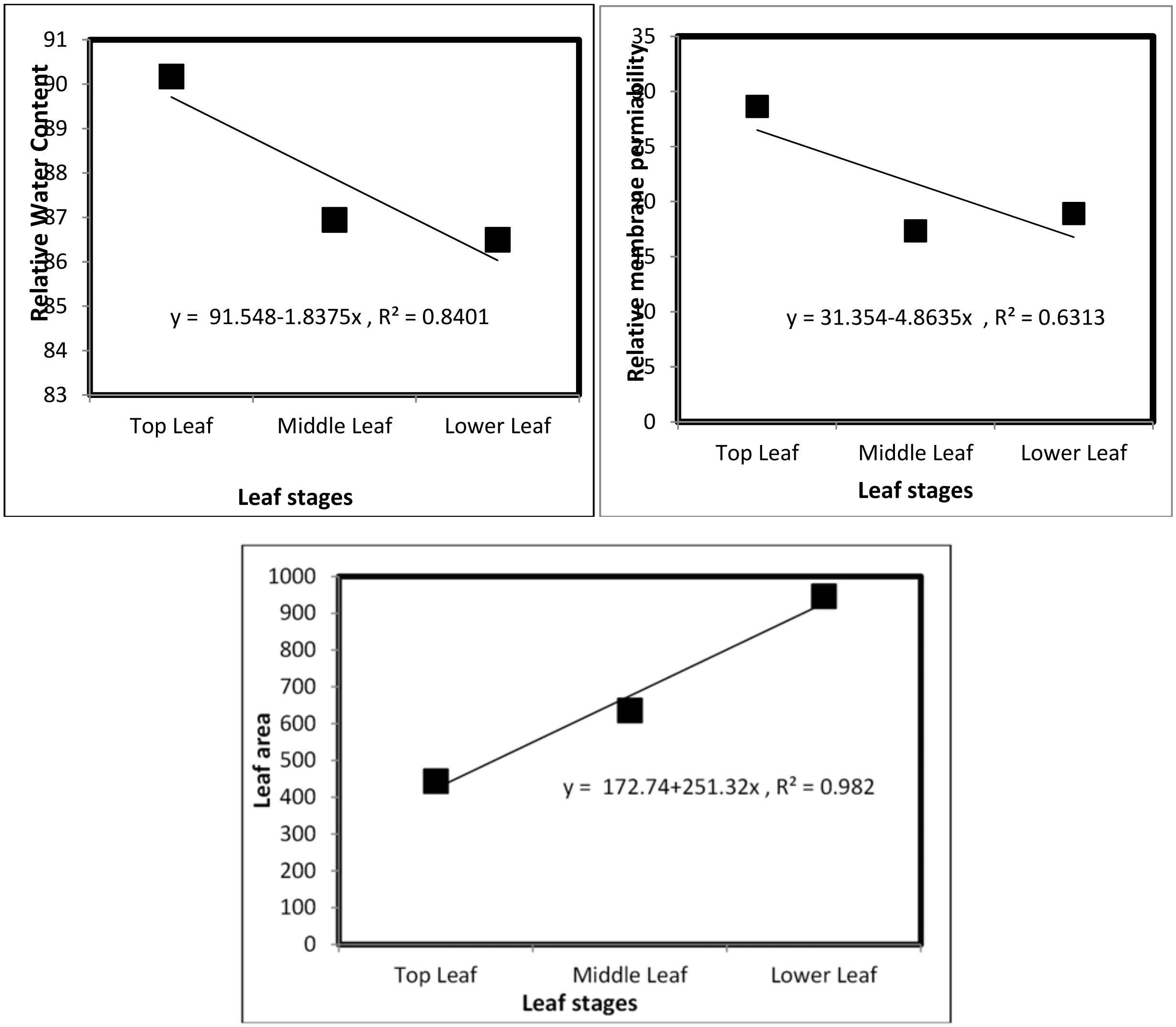

Figure-3. Relative water content, Cell membrane permeability and leaf area in different leaf ages of Jatropha.