Middle East Journal of Agriculture Research
ISSN 2077-4605
Volume : 04 | Issue : 02 | April-June | 2015
Pages: 373-378

Morphological and Anatomical Studies on *Jatropha curcas* L. plant

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ABSTRACT

This study was carried out at the Experimental Farm of Great Cairo Sewage Treatment Plant Station, Al-Gabal Al-Asfr, Cairo during two seasons; 2009/2010, 2010/2011 aiming to investigate the morphological and anatomical characters of the developmental root of *Jatropha curcas* L. plant. *Jatropha* seeds showed epigeal germination, development to five roots formed in early germination stage near the root tip; one central and four peripheral. Tap root length was significantly increased with a positive Relative Growth Rate per day ($RGR_{per\ day}$). The maximum length was within the ages between 5 to 9 weeks and was significantly increased from 0.0035 to 0.0211 cm, followed by relatively reduction in growth rate in the next three weeks; 0.0072 to 0.0035 cm per day. The $RGR_{per\ day}$ in length and diameter of *Jatropha* lateral roots measured on 7 days intervals throughout the first three months. Lateral root length was significantly increased with a positive $RGR_{per\ day}$. *Jatropha* lateral root length showed rapid growth within the first five weeks. Same trend was observed with regard to the average lateral root diameter. The primary root differentiated directly downward from the lower end. The four adventitious root primordia originated at the angles of an imaginary square. Endodermis showed more than one cell thick in some places. Pericycle exhibited anticlinal and periclinal divisions. A diarch primary xylem is differentiated and cells nearest the pith differentiated first while the cells nearest the pericycle matured first. In the hypocotyl, an extremely small amount of primary xylem differentiated after some secondary xylem has formed.

Key words: *Jatropha curcas* L., tap root, lateral root, secondary and primary structure.

Introduction

Jatropha curcas L.; the common name is 'jatropha plant'. It is a plant that produces seeds with high oil content. Seed is toxic and in principle non-edible (FACT, 2010). *Jatropha curcas* gained prominence because of its added features like excellent adaptability to various habitats, large fruits and seeds, high oil yield, soil conservation capabilities and thriving well as live fence.

All parts of the plant exude sticky, bitterly pungent and astringent latex, which can be used as making ink. The bark contains tanin, wax, resin and saponins for industrial purposes. The kernel, which forms 60-68% of seed weight, contains 46-58% oil of the kernel weight and 30-40% of the seed weight. Seed oil is used for illumination without smoke; substitute of diesel, kerosene, lubricants, soaps and candle manufacturing. It can be used as hair oil and livestock against sores. As an excellent source of organic manure; oil contains 3.2% nitrogen, 1.4% phosphorus and 1.2% potassium (Eric van Der, 2010 and Jimu *et al.*, 2009). Although *Jatropha* plant can grow easily from fully mature seeds, but also commercially, it can be propagated by nursery and stem cutting. In some cases *Jatropha* plant naturally forms a symbiosis with soil mycorrhiza that increases the nutrients and water plant uptake from soil. The presence of mycorrhiza increases the plant tolerance to drought and shortage of nutrients. This symbiosis occurs sometimes under natural conditions but never occurs in plantations, unless artificially introduced (Ywe *et al.,* 2010).

The main objective of this study was to investigate the morphological and anatomical root development of *Jatropha curcas* L. plants

Material and Methods

Seeds of *Jatropha* plant were secured from the Department of Oil Crops Research, Field Crops Research (FCRI), Agriculture Research Centre (ACR), Giza, Egypt according to the International Rules for Seed Testing Association (Anon., 2004). The growth rates estimates were calculated according to the methods reported by Jimu *et al*. (2009).

The microscopic study was carried out to investigate the anatomical structure of the roots during different growth stages. Plant specimens were killed and fixed in FAA solution (10ml formalin, 5ml glacial acetic acid, 50ml ethanol 95% and 35ml distilled water) for 48 hours. Then, samples were washed in 50% ethanol, dehydrated in normal butane and embedded in paraffin wax 56 – 58 °C mp. Sections were stained by crystal

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violet–erythrocin, cleared in xylene and mounted in canada balsam. The anatomical measurements were recorded (Nassar and El-Sahhar 1998).

The expermint layout was Randomized Block Design (RBD) with three rplicates. The collected data were subjected to the convinent method of statistical analysis according to the computer software (MSTAT,1986) designed for statistic analysis. The data were subjected to statistical analysis of variances procedure and the values of L.S.D were obtained (Snedecore and Cochran 1990).

Results and Discussion

a) Tap root development

Since *Jatropha* seeds showed epigeal germination, the hypocotyl showed rapid elongation as the tap root developed directly toward the apex. The most remarkable observation is the development of the five predictable root structures; one as central tap root and four as lateral roots. The five roots are formed in early germination stage near the root tip. The central root is more aggressive and thickened as compared with the other four weak perpendicular lateral roots. The average length of the central tap root was 12.6 cm measured after 21 days from seed sowing. After one month, both the tap and the four lateral roots become more branched and got the 3rd degree of roots that has fibrous texture due to extensive growth of fine roots and spread horizontally.

The relative growth rate ($RGR_{per\ day}$) in length and diameter of *Jatropha* tap root measured on 7 days intervals throughout the first three months showed in Table (1). It is evident that tap root length was significantly increased with a positive $RGR_{per\ day}$ ranged from 0.0035 to 0.0211 cm. The maximum recorded growth rate increments in tap root length were achieved within the age between 5 to 8 weeks followed by relatively reduction in growth rate in the next three weeks; 0.0072 to 0.0035 cm. Moreover, it is clear that the mean length of tap root at the 3rd week was 12.6 cm and reached 26.7 cm at the 12th week, indicating that the *Jatropha* tap root exceeded during this period to 111.9 %.

Same trend was observed with regard to the average tap root diameter. Since, data proved significant increase in tap root diameter with increasing the studied periods. The $RGR_{per\ day}$ values ranged between 0.0005 to 0.0010 mm. The maximum recorded relative growth rate increments in tap root diameter were achieved at the 3rd to the 5th week followed by relatively reduction and stable growth rate from 0.0007 to 0.0005 mm per day. It is clear that the main of tap root diameter at the 3rd weeks was 3.42 mm and reached 5.16 mm at the 12th week, indicating that the *Jatropha* tap root diameter showed 50.8% increment.

The abovementioned results of tap root development are in harmony with the findings reported by Henning (2004b), (2007) and (2008a & b). Who reported that, the root system from natural *Jatropha* plants is well developed with roots growing both laterally and vertically into deeper soil. *Jatropha* is a resilient plant that can adapted with many ecological conditions. Its survival mechanism enables it to withstand periods of stress (cold weather, severe drought and low radiation). It is able to retrieve the nutrients from its leaves and store them in the plant stem and root system.

Table 1. Average tap root length (cm), diameter (mm) and their calculated $RGR_{per\ day}$ of *Jatropha* plants during the 12 weeks (combined data over two seasons 2009/2011)

Plant age (Week)	Tap root length (cm)		Tap root diameter (mm)	
	$RGR_{/day}$	Mean±SE	$RGR_{/day}$	Mean±SE
3	0.0152	12.6±0.011	0.0010	3.42±0.006
4	0.0151	13.9±0.009	0.0010	3.66±0.004
5	0.0174	15.6±0.010	0.0008	3.87±0.007
6	0.0203	17.8±0.012	0.0006	4.03±0.002
7	0.0211	20.4±0.013	0.0006	4.20±0.003
8	0.0162	22.6±0.008	0.0007	4.41±0.009
9	0.0103	24.2±0.005	0.0006	4.59±0.003
10	0.0072	25.4±0.007	0.0007	4.82±0.005
11	0.0041	26.1±0.006	0.0005	4.99±0.004
12	0.0035	26.7±0.006	0.0006	5.16±0.004
$LSD_{5\%}$	0.91		0.0016	

b) Lateral roots development

Four lateral roots at the base of the tap root were observed during seed germination development. These roots were emerged from one specific zone. The average length of the lateral roots was 8.45 cm measured after 21 days from sowing. After one month the four lateral roots become more branched and the heavy third degree roots were observed.

Data in Table (2) proved that the relative growth rate (RGR) in length and diameter of *Jatropha* lateral roots measured on 7 days intervals throughout the first three months. The lateral root length was significantly increased with a positive $RGR_{per\ day}$ ranged from 0.0040 to 0.0241 cm. The maximum recorded $RGR_{per\ day}$ increments were achieved in the 3rd to the 7th week followed by sharp reduction in $RGR_{per\ day}$ at the 12th week to reach to 0.0040 cm. The main of lateral root length at the 3rd week was 8.45cm and reached 18.32 cm at the 12th week. Indicating that *Jatropha* lateral roots length showed rapid growth within the first five weeks. Same trend was observed with the average lateral root diameter. Since, the maximum recorded $RGR_{per\ day}$ increments in this character were achieved in the 8th week followed by sharp reduction in $RGR_{per\ day}$ at the 12th to reach 0.0093. These results are in coincidence with those mentioned by Openshaw (2000).

Table 2: Average lateral root length (cm) and diameter (cm) and their calculated $RGR_{per\ day}$ for *Jatropha* plants during the 12 weeks (combined data over two seasons 2009/2011)

Plant age (Week)	Lateral roots length (cm)		Lateral roots diameter (cm)	
	RGR_{1day}	Mean±SE	RGR_{1days}	Mean±SE
3	0.0181	8.45±0.010	0.0120	0.34±0.0010
4	0.0212	9.69±0.008	0.0131	0.38±0.0005
5	0.0241	11.29±0.009	0.0191	0.44±0.0006
6	0.0241	13.19±0.011	0.0222	0.50±0.0004
7	0.0172	14.80±0.012	0.0301	1.23±0.0008
8	0.0111	15.97±0.007	0.0872	1.32±0.0007
9	0.0083	16.84±0.005	0.0322	2.12±0.0012
10	0.0042	17.35±0.007	0.0292	2.59±0.0009
11	0.0041	17.79±0.006	0.0263	3.11±0.0013
12	0.0040	18.32±0.006	0.0093	3.68±0.0014
$LSD_{5\%}$	0.72		016	

Root anatomical structure

Main root structure

The anatomical of *Jatropha* main root was studied during two different successive developmental stages; seedling stage (7 days old) and mature tap root (4 weeks old). The primary root differentiated directly downward from the lower end of the resulting in a core of pith and a cylinder of procambium which is continuous from the hypocotyl through the primary root. The four adventitious root primordia originated at the angles of an imaginary square, the corners of which are the points of junction of the four provascular strands of the hypocotyl (Fig.1). Richard (2010) and Eric (2010) reported that the root system of the seedling consists of one primary and four adventitious roots, all five of which are similar in size, anatomy, relative position and time of origin. This is early reported by Metcalf and Chalk (1950) and Maheshwari (1998).

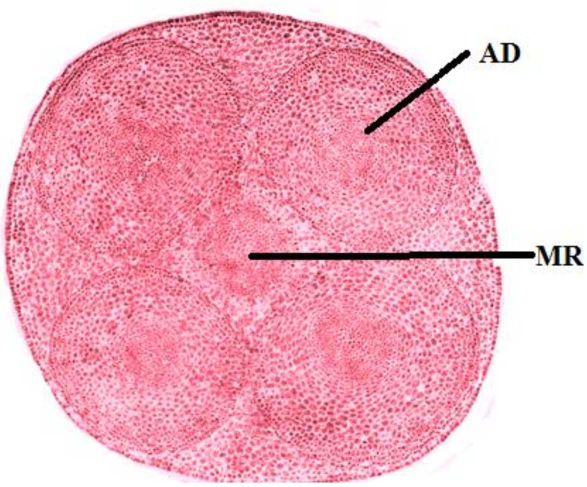

Fig. 1: Transverse section in hypocotyl of *Jatropha* showing the origin of the main root and the four adventitious roots.
Key: MR; main root – AD; adventitious root. (X 60)

At the age of 7 days the seedling root (Fig. 2) showed epidermis with thin walled cells, dense with cytoplasm and radially elongated, divided only anticlinally with enlarge in the tangential direction. Concomitantly anticlinal divisions become less frequent with very thin cuticle. Ephemeral root hairs are presented and many of these first formed root hairs showed early collapsing. At the same time the hypodermis can be distinguished from the cortical parenchyma on the basis of the cell size, shape, contents and staining properties. Although the hypodermis consists mostly of one layer of square or radially elongated cells, occasionally two and rarely more layers were observed.

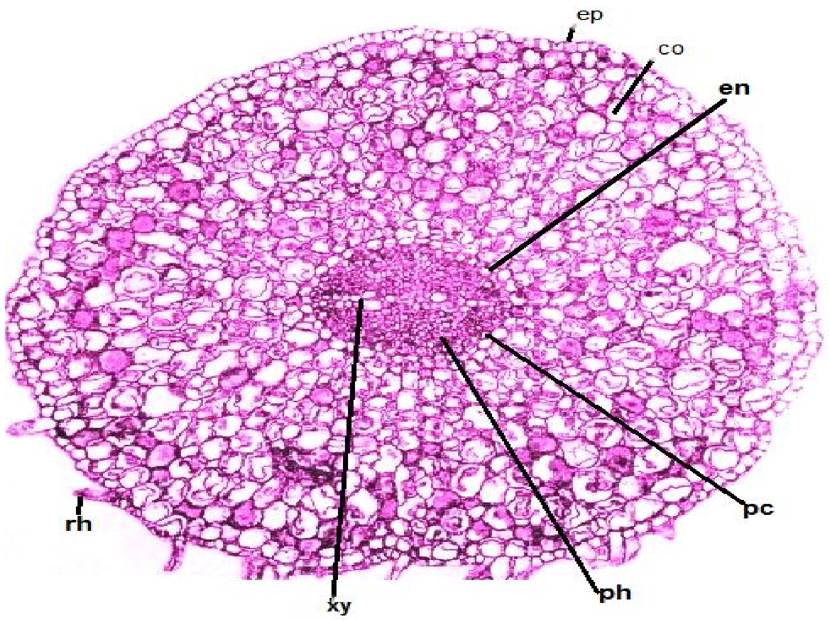

Fig. 2: Transverse section in *Jatropha* primary root at 7days.
Key: ep; epidermis, rh; root hair, co; cortex, en; endodermis, xy; xylem, ph; phloem, pc; pericycle. (X 60)

The cortical parenchyma has definite inner boundary but an approximate outer one. The number of cortical layers is twelve and remains constant until disintegration of the tissue. Periclinal as well as anticlinal divisions continue longest in those layers nearest the endodermis. Relatively large quantities of starch are present in the cells. The endodermis forms a cylinder of tangentially elongated cells lying external to the pericycle. The frequent periclinal divisions and occasional anticlinal ones contribute new cells to the cortical parenchyma and endodermis. The endodermis becomes more than one cell thick in some places. Small, intercellular spaces originated between the endodermis and the cells of the adjoining tissues. Endodermis showed simultaneously casparian strips that characterized the endodermis and become more prominent. The pericycle exhibited anticlinal and periclinal divisions contribute cells respectively to a cylinder of pericycle one cell wide and two strands of small, dividing cells appearing crescent-shape in cross section. These strands of pericyclic cells lie on each side of the central axis of the diarch root, with the convex arc adjacent to the cylinder of pericyclic cells.

Primary xylem is present in the root where primary diarch xylem is defined in the steles of the studied root. Protoxylem vessels occurred toward the periphery and the metaxylem toward the centre. The xylem cells (metaxylem) nearest the center of the root stop dividing and become greatly enlarged before divisions cease in the outer cells nearest the pericycle. Lignification and maturation of these cells begins the next day following differentiation and progress rapidly acropetally but rather slowly centripetally. Differentiation and lignification of additional vessel elements scattered among the fibers occur only occasionally, but those formed are usually extremely large. The lignified walls of these cells are reticulate or occasionally pitted. Xylem fibers become differentiated from primary cells on opposite sides of the diarch xylem axis.

The xylem fibers are similar to the pericyclic and phloem fibers. Pith is the first tissue of the root to differentiate. It is closely compacted and angular cells. Two weeks old seedling showed formation of secondary xylem and phloem as well the common production of periderm and the pericylic fibers. Since, the epidermis as well as the external portion of the cortex sloughed off. Most of the secondary tissue which originated from the vascular cambium is xylem and although the amount of it becomes constantly greater on each side of the central axis, none is formed opposite the xylem points because the vascular cambium has not yet differentiated. These gaps in the xylem, fan-shaped in cross-section and opposite the diarch xylem points, are in the meantime being filled with secondary cortex (phelloderm cells) originating from the pericyclic (cork cambium). The cells are slightly larger than those of the xylem, and many of them are tangentially elongated. The vascular cambium begins differentiating across the root (Fig. 3).

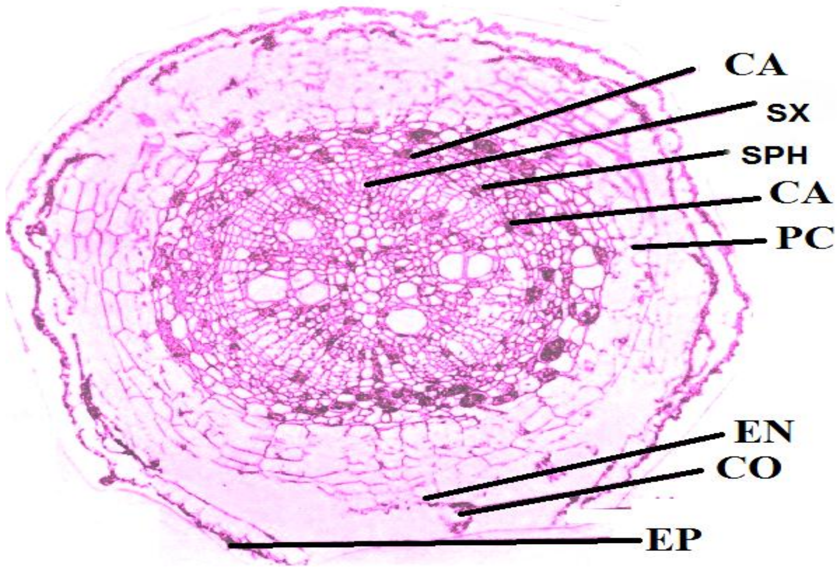

Fig. 3: Transverse section in 14 days *Jatropha* root showed secondary development.
Key: EP; epidermis, CO; cortex, EN; endodermis, PC; pericycle, CA; Cambium, SPh; secondary phloem & SX; secondary xylem. (X 60)

The differentiations of these secondary portions of the cambial cylinder proceed acropetally and by the end, they are found as complete ring surrounding the root. From four to six layers of phelloderm lying centered of the newly differentiated secondary portions of the vascular cambium differentiated to secondary xylem vessels. These vessels arranged as fan-shaped across the transverse section and accompanied with abundant amounts of ray parenchyma (Fig.4). Uni-seriate or bi-seriate rays of xylem parenchyma begin found. These large, thin-walled cells are sometimes crushed by the enlarging elements adjacent to them. The external part of the secondary portions of the vascular cambium has phloem which with pericyclic fibers are differentiated and forming a complete cylinder.

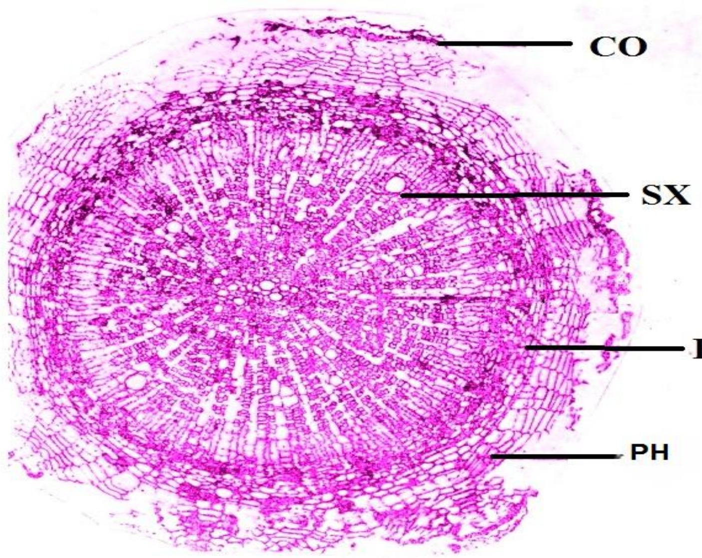

Fig. 4: Transverse section in mature *Jatropha* root showed secondary development.
Key: CO; cortex, SX; secondary xylem, P; Phloem & PH; Phelloderm. (X 60)

These results are in harmony with those mentioned by Ugborogho (1992) and Richard (2010) who reported that, in *Jatropha* seedling, the hypocotyl cannot be considered a portion of the stem occupied by the transition region but should be thought of as an organ of the plant possessing a developmental anatomy peculiar to it alone. In the root, a diarch primary xylem is differentiated. The cells nearest the pith differentiated first while the cells nearest the pericycle matured first. In the hypocotyl, an extremely small amount of primary xylem differentiated after some secondary xylem has formed. Fibers with thick, cellulose walls comprise more than half of the xylem throughout the mature plant. In the root, during the second or third week, secondary cambium differentiated across the secondary tissue opposite the xylem points and from it arise tertiary xylem and phloem.

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