Bir Bahadur · Mulpuri Sujatha Nicolas Carels *Editors*

Jatropha, Challenges for a New Energy Crop

Volume 2: Genetic Improvement and Biotechnology



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Volume 2: Genetic Improvement and Biotechnology



Editors Bir Bahadur Department of Botany Kakatiya University Warangal, India

Mulpuri Sujatha Principal Scientist Crop Improvement Sections Directorate of Oilseeds Research Hyderabad, India

Nicolas Carels Functional Genomics and Bioinformatics Fiocruz/IOC Manguinhos, Rio de Janeiro, Brazil

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Preface

Jatropha, Challenges for a New Energy Crop – Volume 2 aims to report on the state of the art of scientific investigations that were made during the past 10 years on the new crop *Jatropha curcas*. The progresses obtained on the knowledge of this abstemious, semi-wild species are already impressive and were mainly achieved in just a decade (2001–2011). This knowledge extends from basic Jatropha physiology and biological reproduction to the basic agronomic practices and systems for its productive management, but also the complete set of biotechnological tools, such as in vitro culture, genetic transformation, genome sequencing, genetic maps, and marker-assisted selection that are necessary for its selective breeding. These scientific and technological achievements paved the way for the future technological management and domestication of Jatropha as an industrial oilseed crop able to contribute to the feeding of the transport system.

In view of the importance that Jatropha demonstrated worldwide by its largescale cultivation and emerging value for energy business as a biofuel, we felt the necessity of this first comprehensive compilation by global experts. The access to objective information may be difficult to people not directly involved with Jatropha because it is scattered among science media eventually written in different languages. Thus, we gathered the information scattered worldwide in a sort of summary or general agreement of what is known on Jatropha at the moment. This form of a compilation was also necessary because the knowledge on Jatropha is shared over the tropical belt also called Jatropha Belt by different teams, in different politico-economic realities and with different technological and scientific backgrounds. A compilation was the best way to faithfully transmit the point of view of these experts with as few biases as possible. We believe and hope that this compilation will be a valuable source of inspiration for next-generation scientists investigating this new crop, for technologists invested in improving its profitability as well as for decision makers and policy implementers, and politicians, economists, environmentalists or social management who are thinking and acting for the development of a world based on sustainability.

In Volume 1, we outlined the whole *Productive Chain* of Jatropha including the worldwide economic importance of Jatropha as well as its physiology, farming, oil

processing, by-products, biodiesel, and biofuel combustion in order to provide a general picture of the Jatropha's potential to the readers. Volume 2 is presented in 4 units comprising 31 chapters covering the main aspects of its biology and reproduction, genetic diversity and domestication, germplasm, and biotechnology. It aims to give a kind of comprehensive picture on Jatropha as a *Biological System* with the purpose to understand what can be improved in Jatropha and how such improvement can be achieved.

We wish to express our gratitude to all the contributors from all over the world for readily accepting our invitations for not only sharing their knowledge, but for admirably integrating their expertise on scattered information from diverse fields in composing the chapters and enduring editorial suggestions to finally produce this venture that we hope will be a success. We greatly appreciate their commitment.

We also acknowledge the support received from many colleagues in the preparation of the manuscripts as well as thank our spouses and relatives for bearing with us, our commitment to the book.

We thank Hannah Smith, Associate Editor, Springer Science, USA, and her staff for their unstinted cooperation at every stage of the book production.

Finally, we apologize for any mistakes, omissions or failures that may subsist in this work.

Warangal, India Hyderabad, India Rio de Janeiro, Brazil Bir Bahadur Mulpuri Sujatha Nicolas Carels

About the Editors

Bir Bahadur

Graduated from Nizam College and holds a postgraduate degree from University college, Osmania University, Hyderabad, India. He obtained his Ph.D. in plant genetics from Osmania University and was closely associated with Prof. J.B.S. Haldane, FRS, the renowned geneticist of the last century. He advised, guided and encouraged Bahadur to study heterostyly and incompatibility in Indian plant species, a subject first studied by Charles Darwin in England about 160 years back. He made significant contributions in several areas of plant biology especially in incompatibility, mutagenesis, morphogenesis, tissue culture, and organism asymmetry in relation to yield and application of SEM in plant sciences. He published over 250 research papers, which are well received and quoted in national and international journals, including a number of theses and several publications on Jatropha and Castor. He served Osmania and Kakatiya Universities as lecturer, reader and professor; he also served as chairman, head of department, dean of the Faculty of Science, Kakatiya University, Warangal. He has taught genetics, biotechnology and reproduction of plants for over 40 years and accumulated research experience in these areas for about 50 years. He was a post-doctoral Fellow at the Institute of Genetics of Hungarian Academy (Budapest); recipient of the Royal Society Bursary, London; and Honorary Research Fellow at the Birmingham University (UK). He has been invited speaker of over 100 conferences including Max Planck Institute, Koln (Germany); Institute of Genetics (Budapest); Birmingham (UK); University Texas, Houston (USA); Missouri University, St Louis (USA), Sabrao Conference, Szukoba, Tokyo (Japan); Indian Science Congress; etc. He has authored/edited eight books and was editor-in-chief of both Proceedings of Andhra Pradesh Akademi of Sciences (Hyderabad, India) and Journal of Palynology (Lucknow, India). He is on the editorial boards of several journals in India. He is recipient of Best Teacher Award by Andhra Pradesh state Government and Prof. Vishwamber Puri, Gold Medal of Indian Botanical Society for his original contributions in various aspects of plant sciences. He is fellow of over a dozen professional bodies in India and abroad including the Fellow of Linnean Society, London; Fellow of Institute of Biology, and Chartered Biologist, London; and member of New York Academy of Sciences. He has been recently awarded the Bharath Jyoti Award for his sustained academic and research career at New Delhi. Presently he is on the Board of Directors of Sribiotech, Hyderabad, India, Emeritus Professor, Genetics Department, Shadan Post Graduate Centre, Osmania University, Hyderabad.

Mulpuri Sujatha

Graduated in plant sciences from the University of Hyderabad (UoH), India. She has a Ph.D. in genetics from Osmania University (OU), Hyderabad, and worked on intergeneric and interspecific affinities between Ricinus and Jatropha. She made significant contributions for the genetic improvement of oilseed crops through genetics, tissue culture and biotechnological tools. Her important achievements include development of male sterility systems in safflower, sunflower and niger and reliable and efficient tissue culture and transformation protocols for sunflower, castor, niger, safflower and Jatropha. The genetic transformation protocols developed are being used for the development of insect-resistant transgenics through the deployment of suitable cry genes in castor, development of transgenic male sterility and fertility restoration system in safflower and development of transgenics for resistance to necrosis disease in sunflower. Her experience in molecular markers resulted in mapping of downy mildew resistance gene (*Pl13*) in sunflower besides development of appropriate molecular markers for distinguishing the toxic and non-toxic accessions of *Jatropha curcas*.

Nicolas Carels

Graduated in agronomy in Belgium and did a Ph.D. in plant pathology at *Faculté* des Sciences Agronomique de Gembloux (FSAGx, Gembloux) prior to working as a scientist on the elaboration of the first genetic map of sugar beet at the end of the 1980s (ICIseed-SES, Belgium). He then moved to Paris at Institut Jacques Monod (IJM, CNRS, France) where he did a Ph.D. on the genome organization in plants. He continued his work on genomics in Italy at Stazione Zoologica 'Anton Dohrn' (SZN, Naples) and Spain at the Centro de Astrobiología of Instituto Nacional de Técnica Aeroespacial (INTA-CAB, Madrid, Torrejon de Ardoz) prior to moving to Brazil at Universidade Estadual de Santa Cruz (UESC, Ilhéus, Bahia) where he contributed to the application of bioinformatics and genomics to the improvement of the resistance of cacao and rubber tree to fungal diseases. He took Jatropha at its beginning when it was declared a strategic crop for the Brazilian economy by President Lula. His investigations covered the measure of the genome size by flow cytometry and the application of reverse genetics to detect QTLs for oil production with the purpose of breeding Jatropha for this trait. He also published an extensive review (Advanced Botanical Review - ABR) on Jatropha and more recently an overview on bioenergies (InTech) with special concern for climate change mitigation and biodiversity preservation. He is now a Federal Officer of Fundação Oswaldo Cruz (Fiocruz, Rio de Janeiro, Brazil) and is interested by the exploration of genomics, bioinformatics, and natural products for the human health benefit.

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Contributors

Lourdes Adriano-Anaya Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

Bir Bahadur Department of Botany, Kakatiya University, Warangal, India The Odyssey, Shantinagar, Hyderabad, India

Nicolas Carels Fundação Oswaldo Cruz (FIOCRUZ), Instituto Oswaldo Cruz (IOC), Laboratório de Genômica Funcional e Bioinformática, Rio de Janerio, RJ, Brazil

Carlos Augusto Colombo Centro de Pesquisa e Desenvolvimento de Recursos Genéticos Vegetais, Instituto Agronômico de Campinas (IAC), São Paulo, Brazil

E. Chamundeswari Department of Botany, Kakatiya University, Warangal, India

Rajinder Singh Chauhan Department of Biotechnology and Bioinformatics, Jaypee University of Information Technology, Waknaghat, Solan, India

Jitendra Chikara Discipline of Wasteland Research, Central Salt and Marine Chemicals Research Institute (CSIR), Bhavnagar, Gujarat, India

S.K. Datta Acharya JC Bose Biotechnology Innovation Center, Bose Institute, Kolkata, India

Paulo Cavalcanti Gomes Ferreira Laboratório de Biologia Molecular de Plantas, Instituto de Bioquímica, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

Roseli Aparecida Ferrari Centro de Ciência e Qualidade de Alimentos, Instituto de Tecnologia dos Alimentos (ITAL), Campinas-SP, Brazil

Kiichi Fukui Plant Bioengineering for Bioenergy Laboratory, Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Department of Biotechnology, Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Angharad M.R. Gatehouse School of Biology, Devonshire Building, Newcastle University, Newcastle upon Tyne, United Kingdom

Arup Ghosh Discipline of Wasteland Research, Central Salt and Marine Chemicals Research Institute (CSIR), Bhavnagar, Gujarat, India

S. Goverdhan Department of Botany, Kakatiya University, Warangal, India

Clícia Grativol Laboratório de Biologia Molecular de Plantas, Instituto de Bioquímica, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

S. Hemalatha Department of Botany and Microbiology, Acharya Nagarjuna University, Guntur (Dt.), Andhra Pradesh, India

Hideki Hirakawa Kazusa DNA Research Institute, Kisarazu, Chiba, Japan

Yan Hong JOil Pte, National University of Singapore, Singapore, Singapore

Joanna Jankowicz-Cieslak Plant Breeding and Genetics Laboratory, FAO/IAEA Agricultural and Biotechnology Laboratory, International Atomic Energy Agency, Vienna, Austria

Jochen Junker Fundação Oswaldo Cruz (FIOCRUZ), Centro de Desenvolvimento tecnológico em Saúde (CDTS), Rio de Janeiro, RJ, Brazil

S.M. Khasim Department of Botany and Microbiology, Acharya Nagarjuna University, Guntur (Dt.), Andhra Pradesh, India

Ajay Kohli Plant Molecular Biology Laboratory, Plant Breeding, Genetics and Biotechnology Division, International Rice Research Institute, Metro Manila, Philippines

School of Biology, Devonshire Building, Newcastle University, Newcastle upon Tyne, United Kingdom

K.V. Krishnamurthy Institute of Ayurveda and Integrative Medicine, FRLHT, Bangalore, India

Nitish Kumar Centre for Biotechnology, School of Earth, Biological and Environmental Sciences, Central University of Bihar, Patna, Bihar, India

Vinod Kumar National Bureau of Plant Genetic Resources Regional Station, Hyderabad, India

S. Lalitha Institute of Ayurveda and Integrative Medicine, FRLHT, Bangalore, India

Margit Laimer Plant Biotechnology Unit (PBU), Dept. Biotechnology, BOKU VIBT, Vienna, Austria

Catarina da Fonseca Lira-Medeiros Diretoria de Pesquisa, Instituto de Pesquisas Jardim Botânico do Rio de Janeiro, Rio de Janeiro, Brazil

Julyana Rosa Machado Fundação Oswaldo Cruz (FIOCRUZ), Centro de Desenvolvimento tecnológico em Saúde (CDTS), Rio de Janeiro, RJ, Brazil

Fatemeh Maghuly Plant Biotechnology Unit (PBU), Dept. Biotechnology, BOKU VIBT, Vienna, Austria

Daniela de Argollo Marques Centro de Pesquisa e Desenvolvimento de Recursos Genéticos Vegetais, Instituto Agronômico de Campinas (IAC), São Paulo, Brazil

Shaik. G. Mastan Discipline of Wasteland Research, Central Salt and Marine Chemicals Research Institute (CSIR), Bhavnagar, Gujarat, India

Sachihiro Matsunaga Department of Biotechnology, Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Department of Applied Biological Science, Tokyo University of Science, Noda, Chiba, Japan

G.V.S. Murthy Botanical Survey of India (SC), Coimbatore, India

Marcela Oliveira Nogueira Fundação Oswaldo Cruz (FIOCRUZ), Centro de Desenvolvimento tecnológico em Saúde (CDTS), Rio de Janeiro, RJ, Brazil

Isidro Ovando-Medina Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

R.K. Pandey Department of Botany, Sri Jai Narain (PG) College (KKC), Charbagh, Lucknow, India

Siam Popluechai School of Science, Mae Fah Luang University, Chiang Rai, Thailand

Aruna R. Prakash Discipline of Wasteland Research, Central Salt and Marine Chemicals Research Institute (CSIR), Bhavnagar, Gujarat, India

T. Pullaiah Department of Botany, Sri Krishnadevaraya University, Anantapur, India

Jing Qu Temasek Life Sciences Laboratory, National University of Singapore, Singapore, Singapore

Jalli Radhamani Division of Germplasm Conservation, National Bureau of Plant Genetic Resources, New Delhi, India

T.V. Ramana Rao Department of Biosciences, Sardar Patel University, Vallabh Vidyanagar, Gujarat, India

G. Ramesh Department of Botany and Microbiology, Acharya Nagarjuna University, Guntur (Dt.), Andhra Pradesh, India

Hifzur Rahman Discipline of Wasteland Research, Central Salt and Marine Chemicals Research Institute (CSIR), Bhavnagar, Gujarat, India

A.J. Solomon Raju Department of Environmental Sciences, Andhra University, Visakhapatnam, India

Manish Raorane Plant Molecular Biology Laboratory, Plant Breeding, Genetics and Biotechnology Division, International Rice Research Institute, Metro Manila, Philippines

Muppala P. Reddy Plant Stress Genomics Research Center, King Abdullah University of Science and Technology, Thuwal, Saudi Arabia

Tummala Papi Reddy Department of Genetics, Osmania University, Hyderabad, India

Manuel Rincón-Rabanales Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

Raquel Pantoja Rodrigues Fundação Oswaldo Cruz (FIOCRUZ), Centro de Desenvolvimento tecnológico em Saúde (CDTS), Rio de Janeiro, RJ, Brazil

Sonia Ruiz-González Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

Hiroe Sakai Plant Bioengineering for Bioenergy Laboratory, Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Miguel Salvador-Figueroa Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

Quézia de Sant'Anna Fundação Oswaldo Cruz (FIOCRUZ), Centro de Desenvolvimento tecnológico em Saúde (CDTS), Rio de Janeiro, RJ, Brazil

V. Sathaiah Department of Genetics, Osmania University, Hyderabad, India

Shusei Sato Kazusa DNA Research Institute, Kisarazu, Chiba, Japan

Nakako Shibagaki Bioengineering for the Interest of Environmental Sustainability (Sumitomo Electric Industries), Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Aneesha Singh Discipline of Wastelands Research, Central Salt and Marine Chemicals Research Institute (CSIR), Gijubhai Badheka Marg, Gujarat, India

Walter José Siqueira Centro de Pesquisa e Desenvolvimento de Recursos Genéticos Vegetais, Instituto Agronômico de Campinas (IAC), São Paulo, Brazil

Natarajan Sivaraj National Bureau of Plant Genetic Resources Regional Station, Rajendranagar, Hyderabad, India

Archit Sood Department of Biotechnology and Bioinformatics, Jaypee University of Information Technology, Waknaghat, Solan, India

Pamidimarri D.V.N. Sudheer School of Biotechnology, Guru Ghasidas Vishwavidyalya (A Central University) Koni, Bilaspur, Chhattisgarh, India

Mulpuri Sujatha Principal Scientist Crop Improvement Section, Directorate of Oilseeds Research, Rajendranagar, Hyderabad, India

Neelam Sunil National Bureau of Plant Genetic Resources Regional Station, Hyderabad, India

N. Rama Swamy Department of Biotechnology, Kakatiya University, Warangal, India

Satoshi Tabata Department of Applied Biological Science, Faculty of Science and Technology, Tokyo University of Science, Noda, Chiba, Japan

Satoshi Tabata Kazusa DNA Research Institute, Kisarazu, Chiba, Japan

Bradly J. Till Plant Breeding and Genetics Laboratory, FAO/IAEA Agricultural and Biotechnology Laboratory, International Atomic Energy Agency, Vienna, Austria

Suguru Tsuchimoto Plant Bioengineering for Bioenergy Laboratory, Graduate School of Engineering, Osaka University, Suita, Osaka, Japan

Kodeboyina S. Varaprasad Directorate of Oilseeds Research, Hyderabad, India

Alfredo Vázquez-Ovando Centro de Biociencias, Universidad Autónoma de Chiapas, Tapachula, Chiapas, Mexico

Padma Venkatasubramanium Institute of Ayurveda and Integrative Medicine, FRLHT, Bangalore, India

Chunming Wang Temasek Life Sciences Laboratory, National University of Singapore, Singapore

Jian Ye Temasek Life Sciences Laboratory, National University of Singapore, Singapore, Singapore

Part I Biology and Reproduction

Chapter 1 Laticifers of *Jatropha*

K.V. Krishnamurthy, Padma Venkatasubramanian, and S. Lalitha

Introduction

Laticifers are cells or series of connected cells containing a fluid called latex in suspension emulsion state. Laticifers often form a system that permeates various tissues of plants body. Latex containing plants include some 12,500 species under 900 genera and 22 families; excepting *Gnetum*, which is a gymnosperm and *Regnellidium*, which is a water fern while all the rest are angiosperms (Metcalfe 1967, 1983; Evert 2006). Among the angiosperms, Euphorbiaceae forms very important latex bearing family, containing ca. 6,300 species, under 245 genera and 37 tribes (Govaerts et al. 2000; Radcliffe-Smith 2001). *Jatropha* is an important genus of Euphorbiaceae in view of its great biodiesel potential. This paper deals with the laticifers and latex of *Jatropha*.

Types of Laticifers

Laticifers are generally classified into three broad categories: **Non-articulated**, **articulated** and **idioblastic**. The non-articulated laticifers are single cells that through continued growth develop into tube like structures. They may undergo branching to various degrees (equal to non-articulated branched type) or remain unbranched (non-articulated unbranched type). The articulated laticifers are made up of more than one cell that are placed one over the other. The number of cells forming single laticifers may vary from one laticifer to another. All the constituent cells are invariably more elongated than adjacent parenchyma cells although it is not

K.V. Krishnamurthy (🖂) • P. Venkatasubramanian • S. Lalitha

Institute of Ayurveda and Integrative Medicine (IAIM), FRLHT,

e-mail: kvkbdu@yahoo.co.in

Bangalore 560 106, India



Fig. 1.1 (a and b) Non-articulated branched and articulated unbranched laticifers respectively of *J. gossypiifolia* and *J. tanjorensis* stained with Toluidine Blue O

a rule. The cross wall between successive cells may remain intact; possess one or more pores of various sizes (perforated) or in extreme instances the cross walls may be totally dissolved to form a long duct. The articulated laticifers may remain unbranched or may get anastamosed to various extents through lateral union with similar laticifers. The third category of laticifers are usually isodiametric cells of parenchyma type filled with latex. Such cells are dispersed in the other tissues of plant organs as idioblasts. Otherwise, these idioblastic laticifers, also called as **simple laticifers**, share most other features of typical laticifers. There is often a series of integrating laticifer cell type from simple ones to very complex categories of laticiferous tubes.

The genus Jatropha is reported to possess all the three categories of laticifers (Dehgan and Craig 1978) although Pax (1884), Scott (1886) and Rao and Malaviya (1964) reported only non-articulated laticifers in the species of *Jatropha* that they have studied. The length to which non-articulated laticifers may grow varies from species to species and the growth may be straight or bent to various extents. The following species of Jatropha possess exclusively non-articulated laticifers: J. augustii and J. fremontioides (Dehgan and Craig 1978). By contrast, the following species of Jatropha exclusively have the articulated laticifers: J. multifida, J. cathartica, J. capensis, J. lobata, J. integerrima, J. hernandifolia, J. curcas, J. malacophylla, J. platyphylla, J. moranii, J. ciliata (Dehgan and Craig 1978). We have found that in J. tanjorensis also the laticifers belong to the articulated type with cross walls intact between cells (Fig. 1.1a, b). Deghan and Craig (1978) also reported that the articulated laticifers vary in length depending on the species with short or long cells arranged end to end, but all with distinct cross walls. Probably in all species of Jatropha that have articulated laticifers the cross walls remain intact. Many species of Jatropha have both articulated and non-articulated laticifers (Dehgan and Craig 1978) viz., J. gossypiifolia, J. excisa var. pubescens, J. paradoxa, J. marginata, J. fissipina, J. ferox, J. podagrica, J. trieronymii, J. unicostata, J. gallabatensis, *J. lagarinthoides, J. macvaughii, J. cordata, J. verrucosa, J. standleyi, J. canescens, J. cinerea, J. giffordiana, J. neopauciflora, J. cardiophylla, J. cuneata, J. dioica and probably J. velntina* (where non-articulated type is to be verified). Kakkar and Paliwal (1972) earlier reported articulated and non-articulated types in *J. gossypiifolia,* a fact confirmed by Dehgan and Craig (1978).

Idioblastic laticifers are also present in some species of *Jatropha* (Deghan and Craig 1978). According to these authors, this type of laticifer occurs only in the leaves, but not in petioles, stems and roots. They are of variable shapes and occur in mesophyll of the leaves often towards the leaf margins. According to these authors since the idioblastic laticifers integrate with certain other idioblasts that contain tannins, mucilage, proteinaceous material and other compounds they cannot be delimited precisely. In fact they refrain from calling them laticifers and refer to them as idioblasts, although they are convinced on evidences that these are laticiferous in nature.

Origin and Distribution of Laticifers

Laticifers in Jatropha occur both in primary and secondary bodies of the plant. It is generally believed that articulated laticifers originate both in primary and secondary tissues, whereas the non-articulated nearly and exclusively originate in primary tissues (Rudall 1987, 1994; Wurdack et al. 2005). Popham (1947) observed laticifers in the embryonic stems and root of J. cordata and these differentiate successively in cortical parenchyma, xylem and phloem. Further, a few larger, thick walled mostly empty cells that become filled with latex during the first 3–4 days after germination differentiate adjacent to phloem in the hypocotyl. They appear in the stem soon after differentiation of cortical parenchyma. In the root, laticiferous cells differentiate into xylem, phloem and tissues central of cork cambium during 2^{nd} or 3^{rd} week of germination. In the hypocotyls, laticifers differentiate in cortical parenchyma and in phloem during the 5th or 6th day of germination while in xylem between the 1st and 3rd weeks. In the stem, they are mostly formed in the cortical parenchyma. Cass (1985) made a detailed study of the origin and distribution of laticifers in J. dioica. The non-articulated laticifers of these species become recognizable when the embryo is approximately 0.3 mm long, following the initiation of cotyledons. A transection (TS) near the cotyledon node showed a ring of 5–7 laticiferous initials with large nuclei. These cells are up to 24 μ m long and are observed outside procambium. These cells extend bidirectionally along with procambium both in hypocotyl and into cotyledons. Soon the arrangement of laticifers in the hypocotyl becomes complicated by the random branching of laticiferous initials and a large ring of up to 70 laticifer branches are observed by the time the embryo matures. Finally, laticifers get themselves arranged in two rings through an inward branching of the original ring. The two rings are separated by the developing phloem. The branches of the outer ring sub-divide and extend into the cortex and no laticifiers are observed in the pith. The distribution of laticifiers in cotyledonary

and embryonic leaves differs somewhat from that in the hypocotyl. Each cotyledonary vascular trace is in close association with abaxial and adaxial laticifer extensions. Similarly, branching of cotyledonary laticifers parallels that of cotyledonary vascular bundles. A similar pattern of development of laticifers is seen in the embryonic leaf. In the mature embryo of this species, two concentric rings of laticifers extend through most of the length of hypocotyl (2.5 mm) blindly ending near the bases of four branch root primordials. Laticifer extends from the outer ring near the nodal region and enters into cotyledons and embryonic leaf. Centripedal extensions of the outer ring form a complex system of laticifers surrounding the apical meristem of the embryo. The tips of laticiferous initials remain adjacent to the apical meristematic tissue and become incorporated in the new tissues as the epicotyl elongates. During germination of embryo, mitotic divisions commence in the large nucleus of the initials to produce multinucleate and cynocytic laticifers. Thus, in this species the origin and differentiation of laticifers both temporally and spatially are similar to that observed in *Nerium oleander* (Mahlberg 1961). While Mahlberg (1961) located the origin of laticifer initial in procambium, Cass (1985) believed that it is issued from phloem parenchyma. The latter author also found a close distributional relationship between laticifers and phloem in J. dioica.

Deghan and Craig (1978) have noted that the branches of non-articulated laticifers are frequently in continuation of the tubes associated with vascular tissue, but permeate in intercellular spaces forming a network in the mesophyll. These workers found the articulated laticifers also most often on the periphery of the vascular tissue, but also occasionally within them. Non-articulated laticifers according to these authors originate in the protophloem and are in general more abundant in the phloem region in common with articulated laticifers.

We have made a detailed study on the pattern of distribution of laticifers in stems, petioles and leaves of three species of *J. tanjorensis*, *J. multifida* and *J. gossypiifolia* (Figs. 1.2, 1.3, and 1.4). We noted that articulated type predominates in the first two species and non-articulated in the third species. In all the three species, laticifers are not found in the pith of the stem and in the parenchyma inside the petiole vascularization. A careful study indicates that the first few laticifers are initiated in the peripheral cells of phloem, but perhaps through their chemical influence a larger number of laticifers differentiated in the cortical parenchyma of stem and peripheral parenchyma outside the vascularization of petioles and midribs of leaves.

It appears to be true that idioblastic laticifers are scarce/absent in stems and petioles and are restricted to lamella as Deghan and Craig (1978) have shown for some species of *Jatropha*. They may differentiate from the cells of the palisade mesophyll or spongy mesophyll depending on the species. In a few species they are specifically restricted to the mesophyll of leaf margins.

Once initiated, laticiferous initials may grow by apical intrusive growth, by symplastic growth or by both. In most if not all the cases, non-articulated laticifers growth occurs by apical intrusion. If the organ growth is unidirectional, then the apical intrusive growth of laticifers is often seen only in the direction of the organ growth. If the organ growth is bidirectional, then the apical intrusive growth of

1 Laticifers of Jatropha



Fig. 1.2 TS of portion of the stem of *J. tanjorensis* stained with Toluidine Blue O showing laticifers (*red arrows*) in the phloem and cortical regions



Fig. 1.3 TS of portion of stem of *J. multifida* stained with Toluidine Blue O showing laticifers (*darkly* stained cells) distributed in the phloem and cortical regions

laticifers can happen in both the tips. However, in embryonic organs, non-articulated laticifers associated with vascular strands often have an initial phase of symplastic growth followed by a pronounced apical intrusive growth. In most of the articulated laticifers growth is invariably symplastic. In the case of idioblastic laticifers, either there is no growth or it takes places all around the cell either



Fig. 1.4 TS of portion of petiole of *J. tanjorensis* showing laticifers (*red arrows*) in peripheral parenchyma and phloem regions

uniformly or at specific loci so that the mature idioblast may have an even or uneven outline.

Structure of Laticifers

The laticifers are generally tubular both in articulated and non-articulated categories, while it is variously shaped in case of idioblasts. Liu et al. (2006) have studied the distribution and size of laticifers in various plant parts of *J. curcas*. However, in most other respects the structure of laticifers is similar in all three categories. Milky latex is absent in species of *Jatropha* and in most species latex is not coloured and more or less sticky. In that respect, latex of this genus differs from many other genera where it is milky. The contents of non-articulated laticifers of *Jatropha* were described as generally quite granular in nature and saffranin positive (Dehgan and Craig 1978). Our studies show that the latex contains starch grains, lipids, mucilage, and predominantly phenolics. Work from other laboratories have indicated the presence of the two novel lathyrane in *J. curcas* (Naengehomnong et al. 1970), curcain, a protease (enzyme) from the latex of *J. curcas* (Nath and Dutta 1991) and curcaycline A, a novel cyclic octapeptide from the latex of *J. curcas* (van den Berg et al. 1995).

The wall of the laticifers is very thick and often made up of concentric layers of cellulose. The cellulose wall layers also contain phenolic acid, perhaps ferulic acids as judged from TBO staining. In some of the articulated laticifers, the lateral walls are frequently ridged irregularly (Dehgan and Craig 1978), a fact that we have also observed in the laticifers of *J. multifida*.

Taxonomic Considerations and Conclusions

It is already well known that at the higher taxon level laticifers are almost restricted to angiosperms (except in *Gnetum* and *Regnellidium*). It is also well known that only about 22 families of angiosperms have laticifers. Within Euphorbiaceae *sensu lato* to which *Jatropha* belongs, laticifers are restricted with one or two exceptions to subfamilies Crotonoidae and Euphorbioideae. While Acalyphoideae and Phyllanthoideae lack laticifers, at the exception of being *Dalechampia* (Hayden and Hayden 2000), *Macaranga, Kirganelia reticulata* (Balaji et al. 1996), *Dicoelia* sp. (Hayden and Hayden 2000) and *Omphalea* sp. (Wurdack et al. 2005). Thus the distribution of laticifers is a useful character that can be used in the broad classification of Euphorbiaceae.

It is generally believed that the type of laticifer is not constant to the given family and the types have evolved independently of one another. Metcalfe (1967) has shown that in Euphorbiaceae, many species have non-articulated type of laticifers, but only species like *Hevea* and *Manihot* have articulated type. However, Dehgan and Craig's (1978) work has clearly shown the occurrence of both types in the genus *Jatropha* and that many species have both types as already indicated. In the 37 species investigated by Dehgan and Craig (1978) articulated laticifers occur in two clades again emphasizing their independent origin even within a genus like *Jatropha*. But neither occurrence is in a supported position within non-articulated clade (Wurdack et al. 2005).

The work of Dehgan and Craig (1978), excluding few exceptions indicate that laticifers of leaves broadly limit taxonomic boundaries. Both articulated and nonarticulated laticifers occur in the same leaf in section Tuberosae, Sub-section Tuberosae as well as in Section Collenucia, Spinosae and Jatropha. Sections Curcas and *Mozinna* in the subgenus also show both types of laticifers. In addition, section Loureria (except J. fremonitoides) and section Mozinna show the idioblasts. J. dioica shows both articulated and non-articulated as also irregularly shaped idioblasts near leaf margin. Section Polymorphae (possibly except J. macrorhiza), Tuberosae (sub-section Capenses) in subgenus Jatropha, and section Platyphyllae in subgenus Curcas lack non-articulated laticifers and idioblasts that show the articulated type. Of the five species examined in section *Peltatae*, only *J. podagrica* and J. hieronymii show both the articulated and non-articulated types. J. multifida and J. cathartica lack the non-articulated laticifers and idioblastic laticifers, but not the articulated ones. The most striking feature of Dehgan and Craig (1978) study perhaps is the occurrence of idioblasts in the most primitive sections of the subgenus Curcas, namely Loureria and Mozinna. The origin of these idioblasts is probably independent of those found in J. augustii because in the latter, they are derived from the mesophyll cells whereas those of J. cardiophylla and others in the subgenus *Curcas* restricted to the periphery of the vascular tissue. According to these authors the presence idioblastic (laticifers) cells is assumed to be an apomorphe character.

According to Rudall (1994) many genera of Euphorbiaceae without laticifers have elongated, highly branched sclerides in the mesophyll, which may in some

instances be homologous with laticifers. However, the putative homology between laticifers and branched, foliar sclereides offers a very confused picture given the differences between these structures. In the absence of credible evidences, the suggestion of Rudall should be accepted only with reserve.

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Chapter 2 Wood Anatomy of Indian Jatrophas

Bir Bahadur, K.V. Krishnamurthy, and E. Chamundeswari

Introduction

Euphorbiaceae includes a large number of woody taxa, shrubs, trees, a few lianas and some plants with an unusual habit like the Candelabra, Euphorbias and the Ericoid Cluytia. The wood structure of these taxa shows great variation making it difficult to present a general diagnosis of the euphorbiaceous wood. Pax (1884) was the first to study wood anatomy in Euphorbiaceae and asserted that the wood is not of any systematic value. Earlier literature on wood anatomy has been reviewed by Solereder (1899) and by Metcalfe and Chalk (1950). Solereder (1899) recorded the presence of thick-walled wood fibers in the Euphorbiaceae and noted that they sometimes possess gelatinous wall, but such a character was not observed by Bamber (1974) who investigated 44 taxa of Crotonoideae and 33 taxa of Phyllanthoideae. However, Bamber (1974) distinguished two types of fibers, i.e., (1) those with moderately thick walls, small lumen and birefringence patterns, and (2) those with thick wall, without lumen cavity and birefringence pattern suggesting the absence of third secondary wall layer in Phyllanthoideae. Pax and Hoffman (1931) studied the wood anatomy of 22 taxa comprising 17 genera of the tribe Cluytieae belonging to 6 sub-tribes, i.e., Codiaeinae, Jatrophinae, Cluytinae, Galeariinae, Acidocrotoninae and Ricinodendinae. Record (1938) investigated the woods of some taxa of American Cluytieae, while Heimsch (1942) studied the LM and SEM of xylem of Euphorbiaceae and noted that both primitive and specialized woods occur in the family. Stern (1967) studied the wood anatomy of

B. Bahadur (🖂) • E. Chamundeswari

Department of Botany, Kakatiya University, Warangal 506 009, India

e-mail: birbahadur5april@gmail.com

K.V. Krishnamurthy Institute of Ayurveda and Integrative Medicine, FRLHT, Bangalore 560 106, India

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Kleinodendron and supported its inclusion in the tribe Cluytieae on the basis of the similarities with other members. Bamber (1974) and Barajas–Morales (1985; 1987) provided additional information on the wood of *J. chamelensis, J. malacophylla* and *J. platyphylla* from Mexico in respect to various characters like vessels, fibre sizes, presence of resin, crystals, etc. Ibarra (1985) made a detailed study of the woods of several Mexican Euphorbiaceae. Mennega (1987) studied extensively the wood anatomy of biovulate taxa of Phyllanthoideae (35 genera and 116 species) and distinguished two wood types on the basis of wood anatomy i.e., (a) *Aporosa* type with great number of primitive characters especially the perforation plates, (b) *Glochidion* type with absence of scalariform vessels/perforation plates.

Recently, Wiedenhoeft (2008) investigated the comparative ecological wood anatomy of several taxa of Crotineae while Ibarra (1985) made a detailed study of the woods of several Mexican Euphorbiaceae. Webster (1975) recognized five sub-families in Euphorbiaceae, i.e., Phyllanthoideae, Oldfieldoideae, Acalyphoideae, Crotonoideae and Euphorbioideae. The first two comprise the Biovulate taxa and the latter three sub-families comprise the Uniovulate taxa. According to this author, the basic pattern of wood is similar in taxa belonging to all the Uniovulate sub-families characterized by the absence of scalariform vessels, perforation plates, presence of medium to very large inter-vascular pitting and similar vessel ray pitting, presence of apotracheal, diffuse, banded parenchyma and numerous narrow heterocellular rays. Hayden and Brandt (1984) studied the wood anatomy of three specimens of *Neowawraea phyllanthoides*, a rare and endangered taxon endemic to Hawaiin Islands, and compared it with the woods of other Euphorbiaceae. Wiedenhoeft (2008) investigated the comparative ecological wood anatomy of several taxa of Crotonoideae.

It is thus obvious that though various aspects, mostly of systematic interest, have been investigated in considerable detail, the genus *Jatropha* still offers scope for further work on wood anatomy.

Wood Characteristics of Jatropha L.

The genus *Jatropha* L. is represented by about 175 species (Dehgan and Webster 1979) distributed mostly in American and African continents while in India its distribution is limited to about a dozen species. Data on wood anatomy of 10 *Jatropha* species investigated earlier by Chamundeswari et al. (2005) has been extended by a recent study of *J. heynei* from Dr. Venu Madhav (Karimnagar, Andhra Pradesh, India).

The wood characteristics described below were obtained by sectioning wood blocks in slides of 20–25 μ m thick at the Centre for Drugs Research from Osmania University (Hyderabad, India). The sections were stained, dehydrated and mounted following standard methods (Jane 1970). Free hand sections were also taken and processed similarly. Measurements (20–25) were taken for each

of the quantitative wood characters studied under a standard binocular. The terminology used is in accordance with that prescribed by the committee on Nomenclature (IAWA 1964) and the Committee on Standardization of terms of cell size (IAWA 1937, 1939). Vouchers of microtome slides of all the species are being maintained in the Department of Botany, Osmania University (Hyderabad) and Kakatiya University (Warangal, India). The wood features of the *Jatropha* species studied earlier are also described. Some species in particular *J. curcas* has been reinvestigated in addition to *J. heynei*, which was not studied earlier. By contrast, *J. maheshwarii* has not been investigated as the wood was not fully mature.

J. gossypiifolia var. gossypiifolia

J. gossypiifolia var. gossypiifolia is characterized by a diffuse and porous wood with growth rings. Pores are rarely solitary and are commonly organized in radial groups of 2–8. Radial groups are more common than solitary vessels. Solitary pores are circular to oval while pores in radial groups are flattened with tangential walls. Vessels of radial groups are long (375 µm), narrow (54-64 µm in diameter), with simple perforations mostly oblique. Intervessel pits are alternate, crowded, contiguous, angular with lenticular apertures and aligned horizontally. Parenchyma is scanty in paratracheal xylem, but abundant in apotracheal xylem; it is organized in numerous uniseriate tangential bands often alternating with fibers. Wood space is filled up more by parenchyma than by fibres, which gives it a lax structure. The mean diameter of axial parenchyma is 18.3 µm. Xylem rays are overwhelmingly uniseriate, rarely partially biseriate with 2–35 cells in length, homocellular and made of upright or vertical or square type cells forming a homogeneous ray tissue filled with starch grains. Many axial parenchyma cells also have starch although less abundant than in ray cells. On average, ray cells are 18.3 µm in diameter and 1,100 µm in length. Fibers are non-libriform, non-septate, 18.3–22 µm wide, 146.5 μ m long and 4 μ m in wall thickness (Fig. 2.1a-c).

J. gossypiifolia var. elegans

Wood diffuse, porous, pores solitary or in radial groups of 2–10, radial groups more common than solitary vessels. Solitary pores mostly oval. Vessel members long, narrow, perforations simple, oblique, pore diameter 56–73 μ m and mean vessel length 475 μ m. Intervessel pits alternate, crowded, contiguous, angular apertures lenticular, horizontally aligned, 3–7 μ m in diameter. Axial parenchyma is paratracheal and apotracheal. Paratracheal parenchyma are meager in the form of scanty vasicentric sheaths; apotracheal parenchyma abundant, in numerous uniseriate tangential rows alternating with the rows of fibres, 18.3 μ m in diameter. Some



Fig. 2.1 (a) TS of *J. gossypiifolia* var. *gossypiifolia* wood (× 500), (b) TLS of *J. gossypiifolia* var. *gossypiifolia* wood (× 70), (c) vessel of *J. gossypiifolia* var. *gossypiifolia* (× 2000), (d) TS of *J. gossypiifolia* var. *elegans* (× 600), (e) TLS of *J. gossypiifolia* var. *elegans* (× 76), (f) vessel element (enlarged) of wood of *J. gossypiifolia* var. *elegans* (× 2000)

of the axial parenchyma cells contain starch grains. Xylem rays overwhelmingly uniseriate, locally biseriate, 2–20 cells high, homocellular, of upright (vertical) or square type, ray tissue homogeneous with abundant starch grains. Ray cells 25.6 μ m wide and has a mean length of 1,000 μ m. Fibres non-libriform, angular in TS; non-septate, occupies less space than that of xylem parenchyma. Fibres 14–25 μ m across, 165 μ m long (mean length) and wall thickness is 3–5 μ m (Fig. 2.1d–f).



Fig. 2.2 (a) TS of *J. integerrima* with solitary vessels (× 700), (b) TLS of *J. integerrima* (× 80), (c) RLS of *J. integerrima* (× 500), (d) *J. integerrima* wood showing details of vessel element structure (× 1600)

J. integerrima

Wood diffuse, porous, pores predominantly solitary, rarely in radial pairs, or multiples of 3, solitary pores rounded to occasionally oval. Vessel diameter 51–73 μ m, mean length 540 μ m. Intervessel pits simple, 7.3 μ m in diameter. Perforations simple with oblique plates, intervessel pits multiseriate, crowded, angular with transverse lenticular apertures. Axial parenchyma fairly abundant, predominantly apotracheal and aligned in numerous uniseriate tangential rows alternating with rows of fibres. One or two rows of fibres present in between parenchyma rows. Diameter of xylem parenchyma is 18.3 μ m. Xylem rays overwhelmingly uniseriate, rarely partially biseriate, 3 or more than 50 cells high, rays homocellular, of upright vertical or squarish type, ray tissue homogeneous. Fibres squarish to rectangular in TS, non-libriform, non-septate, 11–15 μ m across and have a mean length of 400 μ m long and a wall thickness of 3.5 μ m (Fig. 2.2a–d).

J. panduraefolia

Wood diffuse, porous, pores solitary or in radial pairs of 2–10; radial multiples more frequent than solitary pores; solitary pores rounded to oval; vessel elements long,