RESEARCH ARTICLE

SABRAO Journal of Breeding and Genetics
57 (5) 1862-1869, 2025
http://doi.org/10.54910/sabrao2025.57.5.8
http://sabraojournal.org/
pISSN 1029-7073; eISSN 2224-8978

CORRELATION ANALYSIS OF THE PLANT GROWTH, LEAF CHARACTERS, AND LIPID METABOLITE MARKERS IN *JATROPHA CURCAS*

E. SETIAWAN[1], M.P. PRATAMI[2,3], I.R. KURNIYANTO[4], and M.H. FENDIYANTO[2*]

[1]Natural Resource Management Study Program, Faculty of Graduate, University of Trunojoyo Madura, Bangkalan, Indonesia
[2]Department of Biology, Indonesia Defense University, Bogor, Indonesia
[3]National Research and Innovation Agency of Indonesia (BRIN), Cibinong, Indonesia
[4]Department of Agribusiness, Faculty of Agriculture, University of Trunojoyo Madura, Bangkalan, Indonesia
*Corresponding author's email: miftahul.fendiyanto@idu.ac.id
Email addresses of co-authors: e_setiawan@trunojoyo.ac.id, mentari.putri.pratami.1@brin.go.id, Ifan_kurniyanto@trunojoyo.ac.id

SUMMARY

The physic nut (*Jatropha curcas*) plant, as a renewable alternative fuel source, has greater potential with many advantages than other plant sources. The following study sought to analyze the lipid metabolism pathway and determine its correlation with plant morphological traits. The study transpired using morphophysiological and metabolomic approaches, specifically GC-MS (gas chromatography-mass spectrometry). In the *J. curcas* fruits, metabolites detected totaled 73, which play a significant role in the fatty acid biosynthesis metabolism pathway. However, specifically in the sesquiterpenoid and triterpenoid biosynthesis pathway, only nine key metabolites gained identification in the *J. curcas* fruits, playing a vital role. The correlation values of several growth characters and marker metabolites revealed most of the traits had significant positive and negative correlations with each other. The plant height appeared considerably positively correlated with celidoniol, trans-squalene, and tetradecane. In conclusion, the growth characteristics, such as plant height and leaf traits, have a significant positive and negative correlation with marker metabolites in the formation of fatty acid biosynthesis metabolic pathways and sesquiterpenoid and triterpenoid metabolism.

Keywords: *Jatropha curcas*, plant growth, morphological traits, metabolite markers, lipid metabolism pathway, fuel source

Key findings: This research has identified the association among the plant growth and metabolite markers in *Jatropha curcas*.

Communicating Editor: Dr. Sajjad Hussain Qureshi

Manuscript received: December 31, 2024; Accepted: April 09, 2025.

Citation: Setiawan E, Pratami MP, Kurniyanto IR, Fendiyanto MH (2025). Correlation analysis of the plant growth, leaf characters, and lipid metabolite markers in *Jatropha curcas*. *SABRAO J. Breed. Genet.* 57(5): 1862-1869. http://doi.org/10.54910/sabrao2025.57.5.8.

INTRODUCTION

Jatropha curcas is a species of flowering plants in the spurge family Euphorbiaceae, which is native to the American tropics, most likely Mexico and Central America (Janick and Paull, 2008; Mishra, 2009). Its common names in English include physic nut, Barbados nut, poison nut, bubble bush, and purging nut. In Indonesia, *J. curcas* has the names 'jarak budeg,' 'jarak gundul,' and 'jarak cina.' The development of the *J. curcas* plant as a renewable alternative fuel source has huge potential because of its numerous advantages compared with other plant sources.

Jatropha curcas is a tropical plant with a wide cultivation in America, Asia, and Africa (Gubitz *et al*., 1999). The said plant's probable distribution was by Portuguese sailors through the Cape Verde Islands and Guinea-Bissau to other countries in Africa and Asia (Heller, 1996). This plant has characteristics of a woody plant with a round stem that contains much sap. It can grow up to six meters tall and can live up to 50 years. The bark is pale brown, thin, and tends to peel easily. The stem has irregular branches and grows upwards. Single leaves have alternating positions along the stem. The leaves have stalks (petioles) with a length of 2–20 cm. The leaf blades are palmate or finger-shaped with 3–5 notched sides, measuring 12.5–18 cm × 11–16 cm wide. The apex (tip) of the leaf is acuminate (Jones and Csurhes, 2008).

In addition, by having a high oil content, *J. curcas* is relatively easy to cultivate. *Jatropha curcas* grows well in dry conditions with low rainfall and on the marginal lands with low fertility, i.e., suboptimal lands. Therefore, it can support land conversion from suboptimal to optimal land in dry conditions. Moreover, *J. curcas* cultivation does not compete in land use compared with other food crops (Sharma *et al*., 2009). These characteristics provide a great opportunity for development, especially in Indonesia, because this plant still has quite extensive marginal lands that are unutilized as arable lands.

Jatropha plants do not require special growing conditions, and their planting can be widespread in tropical areas as a hedge around fields and villages (Srivastava *et al.,* 2011). *Jatropha* can easily adapt to the environment, including critical and marginal environmental conditions, with the *Jatropha* plant also used for reforestation of eroded areas (Heller, 1996). The said plant can survive well with the disperse altitude, particularly from 0 to 2000 m above sea level, with rainfall of 300–1200 mm annually and temperatures ranging from 18 °C to 30 °C. In areas with low temperatures (<18 °C), this can reduce its growth, while at high temperatures (>35 °C), the leaves and flowers may fall, and the fruit becomes dry, which eventually affects its production. However, *Jatropha* can grow in less fertile areas but must have good drainage and no flooding, with a soil pH of 5.0–6.5 (Prihandana and Hendroko, 2006). Such growing habits are preferable for its cultivation to obtain oil metabolites in *Jatropha* plants.

Studies on the use of *J. curcas* plants as a renewable alternative biofuel source remain limited, including in Indonesia (Sharma *et al*., 2009). This characteristic presents a significant opportunity for development, as Indonesia still possesses extensive marginal lands still available for agricultural utilization. Recently, no such research for metabolite markers and their relationship with growth characteristics in *J. curcas* has commenced (Yi *et al*., 2010), even though the lipid metabolism pathway has the potential for association (Syakir, 2010; Utami *et al.*, 2012). Therefore, such research on metabolite markers that play a crucial role in regulating lipid metabolism for biofuel producers is vital. Hence, the presented study aimed to analyze the lipid metabolism pathway and its correlation with plant growth characteristics, such as plant height and leaf morphology.

MATERIALS AND METHODS

Plant material and equipment

The materials used in this study were *J. curcas* plants. The tools used included a vacuum pump, Jerrycan, pH meter, Erlenmeyer flask, aerator, petri dish, pipette, Sedgewick rafter, lamp, oven, water bath, Millipore centrifuge, centrifuge tube, watch glass, and scale.

Determination of media for vegetative growth

The growth of *J. curcas* plants succeeded in polybags with 100 experimental units and various abiotic treatments. In addition, *J. curcas* from existing lands also served as specimens in this study, especially for metabolomic testing (Openshaw, 2000). The growth and development of the potential of the *J. curcas* plants progressed by following the findings of Dasumiati *et al*. (2014).

GC-MS analysis

The gas chromatography-mass spectrometry (GC-MS) analysis began by following the methods of Fendiyanto *et al*. (2020) and Pratami *et al.* (2020). An extraction of 15 g of *Jatropha* fruit using ethyl acetate continued at room temperature for 3 h (Sangwan *et al.,* 2015a). The analysis ensued using GC-MS instruments (Barqi, 2015), particularly the evaporator (Caliper-Life-Science, USA), main instrument, autosampler (Agilent Tech-Palo Alto, USA), and Mass Selective Detector (inert MSD Detector, Agilent Tech-Palo Alto, USA). Researchers used the metabolomics approach based on Kusano *et al.* (2015) and Sangwan *et al.* (2015b) as reference studies.

Statistical analysis

Statistical analysis, as performed, employed the R version 4.4.0 program (Lander, 2014) and the Agricolae package by following the methodology of Fendiyanto *et al*. (2020). The assessment included analysis of variance (ANOVA), Duncan's multiple range test (DMRT), and the T-student test (Fendiyanto *et al.,* 2019). The univariate and multivariate data's analyses also took place (Fendiyanto *et al*., 2019a, b). For statistical description, the application of R (Lander, 2014) and the Metabo Analyst R package aided this study (Chong *et al*., 2018; Chong and Xia, 2018; Chong *et al*., 2019; Pang *et al*., 2020).

RESULTS

Plant growth characters

Overall, the *J. curcas* plants showed better growth in the potted planting media. Vegetative growth displayed considerable and better plant height and leaf characteristics. Based on the study of the leafy morphology, the leaf features, such as leaf circumference, width, and length, relatively provided the same values among several replications (Figure 1).

Figure 1. Growth of *Jatropha curcas* plants at three months after cuttings. A ruler shows the length of the calibrator (1–10 cm). Plant height in the vegetative phase (A) and growth characteristics of leaves (B). Scale bar: 1 cm.

Metabolite pathway

The results revealed a total of 73 metabolites found active in the fruits of *J. curcas* plants, indicating these metabolites have significant functions in the metabolic pathways of fatty acid biosynthesis (Figure 2). According to the fatty acid biosynthesis, the main pathways occurred in the pyruvate, β-alanine, and lipoic acid metabolisms; fatty acid degradation; and mycolic acid biosynthesis pathways. The vital hexadecanoyl pathway also occurred in the fruit of the *J. curcas*. Based on the metabolite pathways, especially sesquiterpenoid and triterpenoid biosynthesis, the primary pathways that appeared in the fruit of the *J. curcas* were steroid metabolism, triterpenoids in hopene and tetrahymanol, and protosteryl type (Figure 3). Specifically in the sesquiterpenoid and triterpenoid biosynthesis pathways, only nine key metabolites, as identified, play a vital role in the fruits of the *Jatropha curcas*.

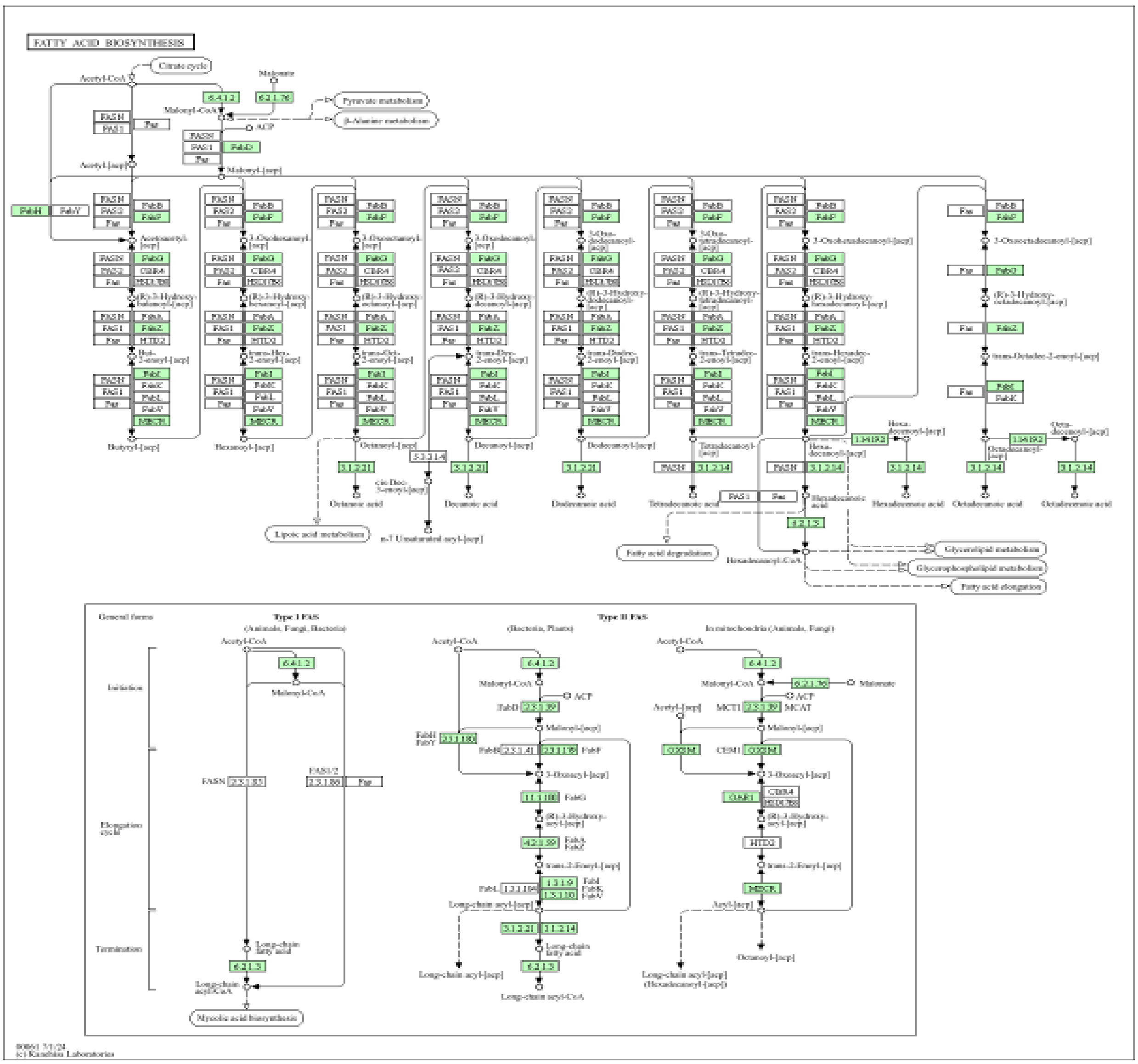

Figure 2. Metabolite pathway of fatty acid biosynthesis in *Jatropha curcas.* Metabolites were taken from the fruits of the plants.

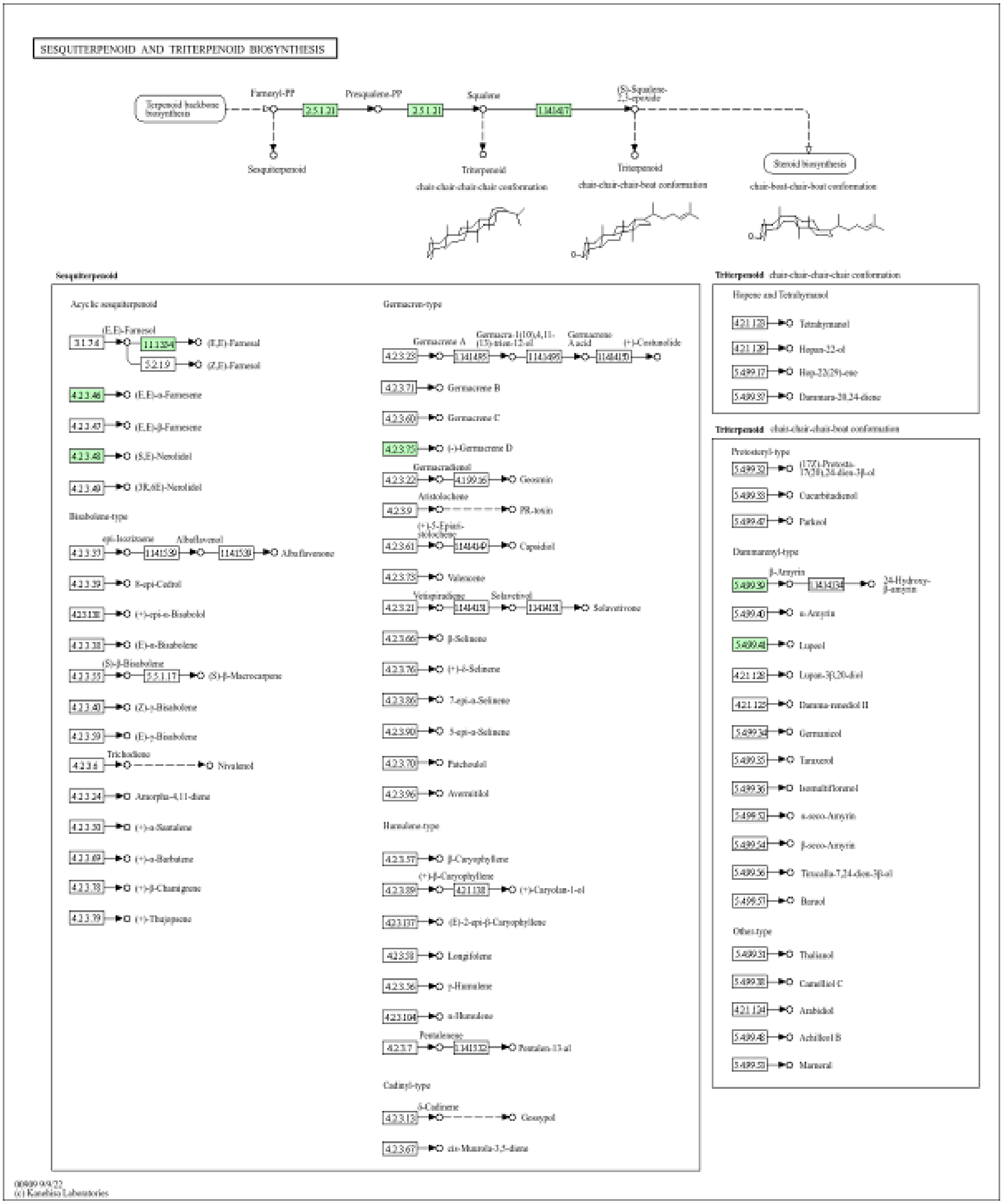

Figure 3. Metabolite pathway of sesquiterpenoid and triterpenoid biosynthesis in *Jatropha curcas*. Metabolites extracted came from the fruits of the plants.

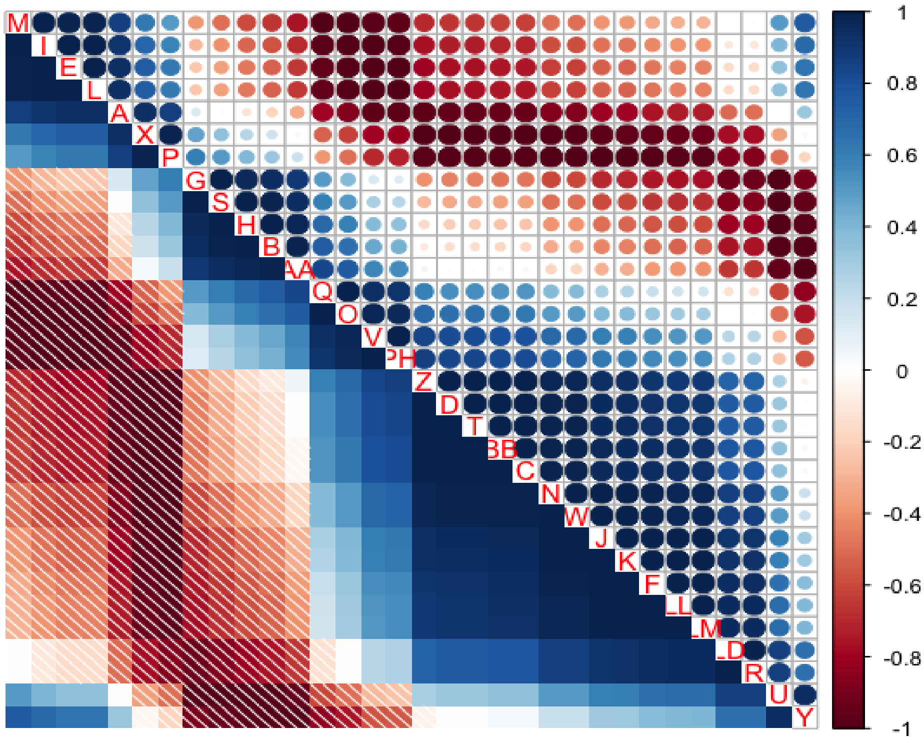

Figure 4. Correlation analysis of plant growth characters, leaf traits, and metabolites from *Jatropha curcas*. Plant height (PL), Leaf margin (LM), Leaf length (LL), Leaf diagonal (LD), Tridecane (A), 7,9-Di-tert-butyl-1-oxaspiro (B), Icosane (C), Hexadecanoic acid (D), Octadecenoic acid (E), Aminoethanethiol hydrogen sulfate (F), Heptadecene (G), Oleic acid (H), 9,12- Octadecenoic acid (I), Tetracosane (J), 9,17-Octadecadienal (K), Nonadecane (L), Heptacosane (M), Heneicosane (N), Trans-squalene (O), 14-methyl-8-Hexadecyn (P), Celidoniol (Q), Tricosane (R), 1.19-Eicosadiene (S), Hentriacontane (T), Cyclooctacosane (U), Tetradecane (V), Tetradecene acetate (W), Oxirane (X), Eicosanol (Y), gamma Sitosterol (Z), Olean (AA), and Eicosadiene (BB).

Correlation among metabolite markers and plant growth

The correlation analysis among several growth characters and metabolite markers showed that most of these traits had significant positive or negative correlations with each other (Figure 4). The plant stature exhibited considerable positive association with the metabolite markers, i.e., celidoniol, trans-squalene, and tetradecane (Figure 4).

DISCUSSION

Lipid metabolism has the potential pathways to understand how the *J. curcas* functions as a bioenergy source (Fendiyanto *et al*., 2024). The reported metabolite product related to biofuel was triacylglycerols (TAG). The TAG synthesis bears regulation from the lipid metabolism in all organisms, including in crop plants (Fendiyanto *et al*., 2024), and has different regulations with lipid-producing microalgae (Milano *et al.,* 2016). Lipid metabolism also occurs in plants; however, the said process is different in crop plants (Obata and Fernie, 2012) compared with animals. In the *J. curcas*, lipid biosynthesis investigation emerged by involving the enzymes ACC and GPAT in the triacylglyceride pathway (Maes *et al.,* 2009), beginning from the acetyl-CoA to oil bodies in the plastids and endoplasmic reticulum (Fendiyanto *et al*., 2024). Lipid biosynthesis metabolism is essential to produce the bioenergy source in the *J. curcas,* as preliminary studies show. This study performed the correlation among lipid metabolites and morphological characters (Tables 1 and 4).

The fossil fuel price in 2005 increased and had reached more than USD 70 per barrel. Various predictions estimated that the price increase was not the end of the fossil fuel increase episode but rather the initial stage that would continue to further price enhancements in the following years, and the same happened in mid-2013. The fuel price increase resulted from fuel reserves and raw materials' increasing depletion and even exhaustion. This event became an important momentum for the development of alternative energy to replace the fuels (Fendiyanto *et al*., 2024).

Biofuels are quite different from crude oil, especially in terms of their sources and the impact of their use. Biofuels require plant biomass as raw material, with more reliance on the plantation and agricultural industries, while crude oil commonly comes from the fossils formed from plants and microscopic animals over millions of years (Shu *et al.*, 2011). The basis of crude oil has more emphasis on energy farming, not on energy hunting, as carried out in crude oil processing. In this study, we found a high correlation between morphological characters and metabolites in the *J. curcas* (Tables 1–4). This study indicated that morphological attributes could be beneficial as markers to predict metabolite-related lipid metabolism in *J. curcas*.

Energy farming contains a mindset that prioritizes the collection and storage of solar energy that can be renewed by itself (self-sustainable), and of course does not damage the environment because it is pollution-free. The use of vegetable oil can also reduce greenhouse gas emissions (Achten *et al*., 2008; Sharma *et al*., 2009). In the presented study, a positive association was evident among the plant growth markers and lipid metabolism in *J. curcas*. Therefore, energy farming is the idea of cultivating energy through green plants known as green energy, such as cultivating castor oil plants for fuel raw materials.

CONCLUSIONS

In summary, this study shows growth characteristics, such as plant height and leaf features, have a strong positive and negative correlation with marker metabolites in the formation of fatty acid biosynthesis metabolic pathways, sesquiterpenoid, and triterpenoid metabolism.

ACKNOWLEDGMENTS

This research received support from the Riset Mandiri (Risman) Grant, PI: E.S, Universitas Trunojoyo Madura (UTM). The authors thank the Health Laboratory of DKI Jakarta for supporting metabolite identification of *Jatropha curcas*. MHF, MPP, and IRK wrote the manuscript, designed the experiment, and conducted metabolomics analysis. RDS edited the manuscript and conducted a morphophysiological and metabolomics study. All authors edited the manuscript.

REFERENCES

Achten WMJ, Verchot L, Franken YJ, Mathijs E, Singh VP, Aerts R, Muys B (2008). Jatropha bio-diesel production and use. *Biomass & Bioenergy* 32:1063–1084.

Barqi WS (2015). Pengambilan minyak mikroalga *Chlorella* sp. dengan metode microwave assisted extraction. JBAT 4(1): 34–41.

Chong J, Soufan O, Li C, Caraus I, Li S, Bourque G, Wishart DS, Xia J (2018). MetaboAnalyst 4.0: Towards more transparent and integrative metabolomics analysis, *Nucleic Acids Res.* 46: W486–W494. https://doi.org/10.1093/nar/gky310.

Chong J, Xia J (2018). MetaboAnalystR: An R package for flexible and reproducible analysis of metabolomics data. *Bioinformatics* 34:4313–4314. https://doi.org/10.1093/bioinformatics/bty528.

Chong J, Wishart DS, Xia J (2019). Using MetaboAnalyst 4.0 for comprehensive and integrative metabolomics data analysis. *Curr. Protoc. Bioinforma*. 68: e86.

Dasumiati D, Miftahudin M, Triadiati T, Hartana A (2014). Karakterisasi dan Peningkatan Jumlah Bunga Hermaprodit Tanaman Jarak Pagar (*Jatropha curcas*) Andromonoecious. Bogor: IPB University, Indonesia.

Gubitz GM, Mittelbach M, Trabi M (1999). Exploitation of the tropical oil seed plant *Jatropha curcas* L. *Bioresour. Technol*. 67: 73–82.
Heller J (1996). Physic nut. *Jatropha curcas* L. Promoting the conservation and use of underutilitized and neglected crop 1. International Plant Genetic Resources Institute. Rome.
Jones MH, Csurhes S (2008). Pest Plant Risk Assessment Physic Nut *(Jatropha curcas).* Bio Security Queensland Department of Primary Industries and Fisheries. Queensland.
Kusano M, Yang Z, Okazaki Y, Nakabayashi R, Fukushima A, Saito K (2015). Using metabolomic approaches to explore chemical diversity in rice. *Mol. Plant.* 8: 58–67.
Janick J, Paull RE (2008). The Encyclopedia of Fruit and Nuts. CABI. pp. 371–372. ISBN 978-0-85199-638-7.
Lander JP (2014). R for Everyone: Advanced Analytics and Graphics, Addison-Wesley USA, Boston.
Maes WH, Achten WMJ, Reubens B, Raes D, Samson R, Muys B (2009). Plant-water relationships and growth strategies of *Jatropha curcas* L. seedlings under different levels of drought stress. *J. Arid Environ*. 73: 877–884.
Milano J, Ong HC, Masjuki HH, Chong WT, Lam MK (2016). Microalgae biofuels as an alternative to fossil fuel for power generation. *Renew. Sustain. Energy Rev*. 58: 180–197.
Mishra DK (2009). Selection of candidate plus phenotypes of *Jatropha curcas* L. using method of paired comparisons. *Biomass Bioen.* 33: 542–545.
Obata T, Fernie AR (2012). The use of metabolomics to dissect plant responses to abiotic stresses. *Cell Mol. Life Sci.* 69: 3225–3243.
Openshaw K (2000). A review of *Jatropha curcas*: An oil plant of unfulfilled promise. *Biomass Bioen*. 19: 1–15.
Fendiyanto MH, Satrio RD, Darmadi D (2020). Metabolic profiling and pathway analysis in red arillus of *Salacca sumatrana* demonstrate significant pyruvate, sulfur, and fatty acid metabolisms. *Biodiversitas* 21:4361–4368. https://doi.org/10.13057/biodiv/d210955.
Fendiyanto MH, Satrio RD, Suharsono S, Tjahjoleksono A, Miftahudin M (2019). Correlation among Snpb11 markers, root growth, and physiological characters of upland rice under aluminum stress. *Biodiversitas* 20:1243–1254. https://doi.org/10.13057/biodiv/d200514.
Fendiyanto MH, Anshori MF, Pratami MP, Wasonga DO, Seleiman MF (2024). Metabolite comparative variation related lipid metabolisms among fruit, leaf, and stem of *Jatropha curcas*. *Heliyon 10*(15). e35861.
Pratami MP, Chikmawati T, Rugayah R (2020). Genetic diversity of *Cucumis* and *Mukia* (Cucurbitaceae) based on ISSR markers. *Biodiversitas* 5(1):2–10.
Pang Z, Chong J, Li S, Xia J (2020). Metaboanalystr 3.0: Toward an optimized workflow for global metabolomics. *Metabolites* 10: 1–15. https://doi.org/10.3390/metabo10050186.
Prihandana R, Hendroko R (2006) Petunjuk Budidaya Jarak Pagar. Agro Media Pustaka. Jakarta.
Sangwan NS, Tiwari P, Mishra SK, Yadav RK, Tripathi S, Kushwaha AK, Sangwan RS (2015a). Plant metabolomics: An overview of technology platforms for applications in metabolism. *Plant Omics: The Omics of Plant Science*, New Dehli, India.
Sangwan NS, Tiwari P, Mishra SK (2015b). Plant metabolomics: An overview of technology platforms for applications in metabolism. New Dehli, India.
Sharma DK, Pandey AK, Lata (2009). Use of *Jatropha curcas* hull biomass for bioactive compost production. *Biomass and Bioenergy* 33:159–162.
Shu L, Lou Q, Ma C, Ding W, Zhou J, Wu J, Feng F, Lu X, Luo L, Xu G, Mei H (2011). Genetic, proteomic and metabolic analysis of the regulation of energy storage in rice seedlings in response to drought. *Proteomics* 11: 4122–4138.
Srivastava P, Bahera SK, Gupta J, Jamil S, Singh N, Sharma YK (2011). Growth performance, variability in yield traits and oil content of selected accessions of *Jatropha curcas* L. growing in a large scale plantation site. *Biomass Bioen*. 35: 3936–3942.
Syakir M (2010). Prospek dan kendala pengembangan jarak pagar (*Jatropha curcas* L.) sebagai bahan bakar nabati di Indonesia. *Perspektif*. 9(2): 55–65.
Utami F, Hariadi A, Hariadi M (2012). Respon Morfologi dan Fisiologi Aksesi Jarak Pagar (*Jatropha curcas* Linn) dalam Kondisi Cekaman Kekeringan di Pembibitan. Bogor: IPB University, Indonesia.
Yi C, Zhang S, Liu X, Bui HTN, Hong Y (2010). Does epigenetic polymorphism contribute to phenotypic variances in *Jatropha curcas* L.?. *BMC Plant Biol*. 10:259.