

KAJIAN ANATOMIS, FITOKIMIA, DAN MOLEKULER BUNGA DAN BUAH CIPLUKAN (*Physalis peruviana* L.) DALAM BERBAGAI TAHAP PERKEMBANGAN

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INTISARI

Familia Solanaceae mencakup spesies bernilai ekonomis, salah satunya adalah ciplukan (*Physalis peruviana* L.). *Physalis* memiliki ciri khas yang membedakan dengan anggota Solanaceae lainnya yaitu *calyx* yang ikut berkembang selama perkembangan buah. Beberapa penelitian telah mendukung pemanfaatan tanaman ini dalam bidang kesehatan, akan tetapi penelitian mengenai perubahan morfologis, struktur anatomis, kandungan metabolit, serta kajian molekuler dalam berbagai tahap perkembangan bunga dan buah pada *P. Peruviana* masih sangat terbatas. Penelitian ini bertujuan mengkaji morfologi dan anatomi buah dalam berbagai tahap perkembangan; membandingkan profil metabolit bunga dan buah dalam berbagai tahap perkembangan; serta mempelajari pengaturan molekuler perkembangan bunga menjadi buah pada *P. peruviana*. Sampel untuk analisis morfologis dan anatomis bunga diambil pada 7 tahap perkembangan (12-13 Hari Menuju Antesis (HMA), 10-11 HMA, 8-9 HMA, 6-7 HMA, 4-5 HMA, 2-3 HMA), dan 0 HMA (antesis), buah terdiri dari 5 tahap perkembangan (bunga Antesis, 5 HSA (Hari Setelah Antesis), 10 HSA, 15 HSA 20 HSA, 25 HSA). Untuk analisis profil metabolit, pengambilan sampel terdiri dari empat tahap perkembangan, yaitu 13 HMA, 7 HMA, 0 HMA, dan buah (5 HSA), sedangkan sampel untuk analisis molekuler adalah bunga (antesis/ 0 HMA) dan buah (5 HSA). Sediaan anatomis disiapkan menggunakan metode penyelubungan parafin. Analisis anatomis organ penyusun bunga didasarkan pada deskripsi jaringan penyusun organ, tahapan mikrosporogenesis, megasporogenesis dan megagametogenesis. Analisis perkembangan buah didasarkan pada deskripsi jaringan penyusun buah, diameter buah, serta deskripsi anatomis perkembangan buah. Analisis profil metabolit diawali dengan ekstraksi sampel menggunakan pelarut etanol kemudian dilanjutkan dengan analisis menggunakan *Liquid Chromatography-High-Resolution Mass Spectrometry* (LC- HRMS). Data hasil LC-HRMS dianalisis secara multivariat dengan *Metaboanalyst 6.0*. Analisis transkriptomik diawali dengan ekstraksi RNA dilanjutkan dengan *RNA-Seq* menggunakan *Illumina sequencing*. Hasil *RNA-Seq* dianalisis *differentially expressed genes* (DEGs), *Gene Ontology* (GO), dan *Kyoto Encyclopedia of Genes and Genomes* (KEGG). Hasil analisis morfologis menunjukkan *P. peruviana* mempunyai bunga yang lengkap dan sempurna, tumbuh secara aksiler, termasuk bunga tunggal dengan *calyx* terdiri dari lima sepal, saling menempel dan berwarna hijau dengan semburat ungu. *Corolla* dengan lima petal berwarna kuning dengan semburat ungu dan saling berlekatan. Benang sari terdiri

dari lima kepala sari berwarna kuning dan filamen berwarna ungu. Epidermis sepal dan petal bagian adaksial dan abaksial bersifat isodiametrik pada saat kuncup, kemudian epidermis berdiferensiasi menjadi papila saat antesis. Trikoma glanduler ditemukan pada epidermis adaksial sepal. Kepala sari mempunyai dua lobus atau dua teka (*ditheca*) dengan empat mikrosporangia (*tetrasporangiate*). Dinding antera terdiri dari empat macam jaringan pada tahap I-V, dan dua macam jaringan pada tahap VI dan VII. Mikrospora isobilateral berbentuk tetrad. Ovarium terdiri dari dua karpel. Ovulum dengan tipe anatropus dan bitegmik. Kajian profil metabolit pada bunga dan buah *P. peruviana* mampu mendeteksi dan mengidentifikasi 48 senyawa pada berbagai tahap perkembangan. Senyawa yang terdeteksi didominasi oleh asam organik sebanyak 38%, asam lemak 29%, dan karbohidrat 10%. Sebanyak 33% sisanya adalah metabolit sekunder yang terdiri dari terpenoid 10%, flavonoid 3%, alkaloid 2%, dan fenol 2%. Senyawa spesifik pada genus *Physalis*, yaitu *calotropin* yang termasuk golongan senyawa *withanolides* turut terdeteksi. Berdasarkan analisis DEG diketahui bahwa jumlah total gen yang terdeteksi pada bunga dan buah adalah sekitar 42.000 gen. Sejumlah 18.000 gen terdeteksi mengalami *downregulation* dan 24.000 gen *upregulation*. Gen yang terekspresi tinggi pada bunga adalah *Pollen_Ole_e_1* dan *PMR5N*, sedangkan gen yang terekspresi tinggi pada buah adalah *X8*, *Sugar_tr*, *Pyridoxal_deC*, *Pectate_lyase_3*, *Fasciclin*, dan *DUF761*. Hasil analisis GO menunjukkan proses biologi fase bunga mekar melibatkan sejumlah gen yang berperan pada proses organisasi enkapsulasi struktur eksternal, organisasi dinding sel, dan proses metabolisme karbohidrat sebagai persiapan pembentukan buah, sedangkan pada komponen seluler, peningkatan ekspresi gen bagian ekstraseluler menunjukkan perubahan bentuk bunga menjadi buah. Analisis GO pada fungsi molekuler menunjukkan sebagian besar gen berperan pada pengaturan aktivitas enzim hidrolase dan transporter transmembran. Hasil analisis KEGG menunjukkan sejumlah besar gen terdapat di ribosom dan banyak gen dalam dataset *RNA-seq* yang terlibat dalam sintesis senyawa metabolit sekunder pada bunga. Terdapat hubungan antara aspek morfologis, anatomis, metabolit, dan molekuler bunga dan buah *P. Peruviana* dalam berbagai tahap perkembangan.

Kata kunci : anatomis, buah, bunga, metabolomik, *Physalis peruviana* L., transkriptomik

ANATOMICAL, PHYTOCHEMICAL, AND MOLECULAR STUDY OF FLOWER AND FRUIT OF CAPE GOOSEBERRY (*Physalis peruviana* L.) IN VARIOUS STAGES OF DEVELOPMENT

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ABSTRACT

The Solanaceae family includes economically valuable species, one of which is *Physalis peruviana* L. *Physalis* has a distinctive characteristic that distinguishes it from other members of the Solanaceae, namely, flower petals that develop during fruit development. Several studies have supported the use of this plant in the health sector. However, research on morphological changes, anatomical structure, metabolite content, and molecular studies in various stages of flower and fruit development in ciplukan (*Physalis peruviana* L.) is still minimal. This study aims to examine the morphology and anatomy of the fruit at various stages of development, compare the metabolite profiles of flowers and fruits at different stages of development, and investigate the molecular regulation of flower and fruit development in *P. peruviana*. Samples for morphological and anatomical analysis of flowers were taken at seven stages of development (12-13 Days Before Anthesis (DBA), 10-11 DBA, 8-9 DBA, 6-7 DBA, 4-5 DBA, 2-3 DBA), and 0 DBA (anthesis), fruit consists of five stages of development (flower Anthesis, 5 Days After Anthesis (DAA), 10 DAA, 15 DAA, 20 DAA, 25 DAA). For phytochemical analysis, sampling consisted of four stages of development: 13 DBA, 7 DBA, anthesis, and fruit (5 DAA). Samples for molecular analysis included flowers (anthesis) and fruit (5 DAA). Sampling was carried out at 9-10 WIB, with three replications at each stage of development. Anatomical preparations were prepared using the paraffin embedding method. Anatomical analysis of flower organs is based on the description of the organ tissue, stages of microsporogenesis, megasporogenesis, and megagametogenesis. Fruit development analysis is based on the description of the fruit tissue, fruit diameter, and anatomical description of fruit development. Metabolite profile analysis begins with sample extraction using ethanol solvent, followed by analysis using Liquid Chromatography-High-Resolution Mass Spectrometry (LC-HRMS). LC-HRMS data were analyzed multivariately with Metaboanalyst 6.0. Transcriptomic analysis begins with RNA extraction followed by RNA-Seq using Illumina sequencing. RNA-Seq results were analyzed for differentially expressed genes (DEGs), Gene Ontology (GO), and Kyoto Encyclopedia of Genes and Genomes (KEGG). The results of morphological analysis show that *P. peruviana* has complete and perfect flowers, which grow axially, including single flowers with petals consisting of five sepals, attached, and green with a purple tinge. The crown with five petals is yellow with a purple tinge and is attached. The stamens consist of five yellow anthers and purple filaments.

The epidermis of the adaxial and abaxial sepals and petals is isodiametric at the time of budding, then the epidermis differentiates into papillae at anthesis. Glandular trichomes are found on the adaxial epidermis of the sepals. The anthers have two lobes or two thecas (ditheous) with four microsporangia (tetrasporangiate). The anther wall consists of four types of tissues at stages I through V, and two types of tissues at stages VI and VII. Isolateral microspores are tetrad-shaped. The ovary consists of two carpels. The ovules are anatropous and bitegmic. The study of metabolite profiles in *P. peruviana* flowers and fruits detected and identified 48 compounds at various stages of development. The compounds detected were dominated by organic acids at 38%, fatty acids at 29%, and carbohydrates at 10%. The remaining 33% consisted of secondary metabolites, including terpenoids (10%), flavonoids (3%), alkaloids (2%), and phenols (2%). Specific compounds in the *Physalis* genus, namely calotropin, which belongs to the withanolides compound group, were also detected. Based on DEG analysis, it is known that the total number of genes detected in flowers and fruits is approximately 42,000. A total of 18,000 genes were detected as being downregulated, and 24,000 genes were found to be upregulated. The genes that are highly expressed in flowers are *Pollen_Ole_e_1* and *PMR5N*, while the genes that are highly expressed in fruits are *X8*, *Sugar_tr*, *Pyridoxal_deC*, *Pectate_lyase_3*, *Fasciclin*, and *DUF761*. The results of GO analysis show that the biological process of the blooming phase involves many genes that play a role in the process of organizing external structure encapsulation, cell wall organization, and carbohydrate metabolism in preparation for fruit formation, while in cellular components, increased expression of extracellular genes indicates a change in the shape of flowers into fruit. GO analysis of molecular function shows that most genes regulate the activity of hydrolase enzymes and transmembrane transporters. The KEGG analysis results revealed that many genes are located in the ribosome, and a significant number of genes in the RNA-seq dataset are involved in synthesizing secondary metabolite compounds in flowers. There is a correlation between the morphological, anatomical, metabolite, and molecular aspects of *P. Peruviana* flowers and fruits at various stages of development.

Keywords: anatomical, flowers, fruit, molecular, *Physalis peruviana* L., phytochemical