RESEARCH ARTICLE

SABRAO Journal of Breeding and Genetics
55 (4) 1170-1182, 2023
http://doi.org/10.54910/sabrao2023.55.4.13
http://sabraojournal.org/
pISSN 1029-7073; eISSN 2224-8978

SEX EFFECT ON THE TELOMER LENGTH, ELEMENTAL COMPOSITION, AND PHYTOCHEMICAL CONTENT OF HIGHLAND PAPAYA (*VASCONCELLEA PUBESCENS* A.DC.) LEAVES

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SUMMARY

Highland papaya (*Vasconcellea pubescens* A.DC.) is a monoecious and dioecious plant with female and male organs. In Indonesia, three sexes of highland papaya exist with different telomere lengths, which protect chromosomes and deoxyribonucleic acid (DNA) from damage. Therefore, the purposeful study aimed to determine the telomere length, elemental composition, and phytochemical content of highland papaya leaves belonging to their different sexes. The telomere length observed and studied in said leaves (female, male, and monoecious) used the polymerase chain reaction (PCR) procedure. A scanning electron microscopy (SEM) analysis provided a more detailed material of leaves, while an energy dispersive X-Ray (EDX) helped observe elemental composition. Phytochemical content analysis ran by histochemical analysis. The results showed that the telomere lengths of young female, male, and monoecious leaves differed at 381, 391, and 396.66, respectively. According to the SEM analysis, nonsignificant differences occurred in the leaf surface of the three sexes of highland papaya leaves. Observation with EDX showed that sodium (1.98%) surfaced in female plant leaves; however, sodium and chlorine did not show in male plants. Monoecious leaves had sodium and chlorine at 0.88% and 0.28%, respectively. The histochemical analysis provided an overview of the distribution of flavonoids and tannins in young leaves of highland papaya. A discovery also noted the existence of both compounds in the adaxial and abaxial epidermis, mesophyll, xylem, phloem, sheath parenchyma, secretory cavities, and trichomes. The study concluded that sex affects telomere length, elemental composition, and the phytochemical content of highland papaya leaves. The study suggests that knowing the highland papaya's sex is vital in plant breeding and genetics and could help improve plant health and productivity.

Keywords: Highland papaya (*Vasconcellea pubescens*), monoecious and dioecious, telomere length, chromosomes, elemental composition, phytochemical content

Key findings: The telomere lengths of female, male, and monoecious highland papaya were unequal, making it possible to determine the particular sex. Sodium and chlorine did not show in male highland papaya leaves.

Citation: Laily AN, Daryono BS, Purwantoro A, Purnomo (2023). Sex effect on the telomer length, elemental composition, and phytochemical content of highland papaya (*Vasconcellea pubescens* A.DC.) leaves. *SABRAO J. Breed. Genet.* 55(4): 1170-1182. http://doi.org/10.54910/sabrao2023.55.4.13.

Communicating Editor: Prof. Zahoor Ahmed Soomro

Manuscript received: April 18, 2023; Accepted: June 23, 2023.

INTRODUCTION

Highland papaya (*Vasconcellea pubescens* A.DC.) belongs to the family Caricaceae and has limited growth in Indonesia. Its discovery in low dry mountain forest areas between 1,000 and 3,000 m had temperatures ranging from 12 °C to 18 °C (22 °C in winter at midday) (Duarte and Paull, 2015). Highland papaya describes as monoecious and dioecious and is native to Panama, Venezuela, Bolivia, Colombia, Ecuador, and Peru. It has yellowish-white flowers and produces ovate/ellipsoidal and pentagonal fruits that turn from green to yellow or yellow-orange after ripening. The leaves are broad, making a cluster as the branch ends in a dense crown, typically having five lobes with two lateral lobes on each side.

According to Duarte and Paull (2015), this species was distinct, with a layer of hairs on the underside of the leaves. Highland papaya contains various compounds, including flavonoids, polyphenols, tannins, and triterpene saponins (Minarno, 2016). Some of its biological properties included antioxidants (Indranila and Ulfah, 2015), antibacterials (Vega-Galvez *et al*., 2020), antidiabetics (Laily and Khoiri, 2016), and analgesics (Sasongko *et al*., 2016; Sudha *et al*., 2015; Sudirga *et al*., 2022). The said plant also plays a vital role in enhancing wound healing, acts as a meat tenderizer, and has various applications in the pharmacological industry (Tonaco *et al*., 2018). Furthermore, highland papaya, as an anthelmintic, has helped treat skin mycosis and verruca plana and alleviate dyspepsia (Duarte and Paull, 2015).

Telomeres appear at the non-coding ends of linear chromosomes. The telomeres not only protect the coding DNA, but also ensure the proper division of chromosomes, through a complex 3-dimensional structure. Aging is characteristic of a progressive shortening of the structure and function of telomeres (Gruber *et al*., 2021). At the end of the chromosomes, the patterns consist of TTAGGG repeat units (Bilici, 2020). The study comprising the telomere length of the dioecious plant revealed that the male willow revealed shorter than the female. Meanwhile, the female ash has a longer telomere than the male (Mu *et al*., 2014). However, a study related to the telomere length and structure in angiosperms of different sexes has limited undertakings.

Highland papaya boasts of its diverse medicinal uses, yet, only a few certified reports on the elemental (mineral) composition exist. The said plant profiles as a nutritional source of green leafy vegetables has mainly been on biochemical assays. However, investigating the elemental composition of the plant is essential, as required in the metabolic processes for the normal growth and development of the human body, and their deficiency can lead to several clinical disorders (Bharti *et al*., 2021). Therefore, determining the elemental composition of valuable medicinal plants could aid in understanding the final dosage of herbal drug preparation for treating a particular disease (Anjum *et al*., 2022).

EDX is versatile, non-destructive, and facilitates fast multiple sampling for quantification of minerals, even with a minimal quantity of samples. Its microanalysis is an approach of elemental analysis associated with electron microscopy based on the generation of characteristic X-rays that identify elements found in the specimen (Scimeca *et al*., 2018). Moreover, the EDX fitted the SEM, hence, a choice for evaluating mineral composition. Histochemical analysis of the leaves provides an overview of the spatial allocation of metabolites that affect the taste of plant organs. Detecting classes of secondary metabolites, such as, phenolics, tannins, sesquiterpenes, and triterpenes, can use specific reagents applied to fresh sections of handmade plant tissues (Vio–Michaelis *et al*., 2020). Additionally, the annotations made under a light microscope of these raw histochemical samples allowed rapid monitoring of plant organs, such as, the localization of flavonoids and tannins in young leaves of highland papaya with various sexes.

Highlights on the potential of highland papaya as a source of health-promoting compounds have materialized. Previous studies on these plants have focused on measuring their nutritional, phytochemical profile, and other bioactivity. However, a few studies have reported telomere length, elemental composition, and phytochemical content of their young leaves with different sexes. The sex effect on telomere length is important for plant reproduction. The elemental composition of a plant can affect its nutritional value and susceptibility to pests and diseases. Phytochemicals are naturally occurring compounds in plants that have beneficial health effects. The study purposely investigates the sex effect on the telomere length, elemental composition, and phytochemical content of highland papaya. The findings of this study are significant because they provide new insights into the role of sex in plant biology. They could further help develop new strategies for improving the highland papaya’s nutritional value and shelf life.

MATERIALS AND METHODS

Plants morphology

The collection of the third leaf from the highland papaya shoots came from its wild form in Batu, East Java, Indonesia (S 07 44'23.37" E 112° 32'02.93") and Banjarnegara, Central Java (S 07 10'57.36'' E 102° 53'28.04”), Indonesia. The habitus and leaves of highland papaya with different sexes appear in Figure 1. This tree species typically grow to a height of 3–6 m and often has branches. The leaves are broad and clustered at the terminal parts of the boughs, forming a dense crown. The leaves are five-lobed, with two lateral lobes on each side. The species is distinct by the coating of hairs on the underside of the leaves. Highland papaya male plants are very leafy and have an abundance of inflorescences. The male plant flowers are slender with a long, pubescent peduncle and flower tube, comprising five sepals, five yellowish-white/greenish petals, five stamens, and five sessile staminodes inserted in the corolla throat.

Figure 1. Tree habit and leaves of highland papaya in different sexes. a and d: female, b and e: male, c and f: monoecious. Bar = 10 cm in capture.

However, the female plants of highland papaya are less leafy and have larger flowers, with yellow to white petals rarely in short cymes. The flowers arise in the leaf axils with a very short peduncle and are pentamerous, with a five-lobed stigma. The fruit is obovoid/ellipsoidal, five-sided, and changes from green to yellow or orange-yellow after ripening (Duarte and Paull, 2015). The telomere analysis consisted of elemental composition and histochemical analysis of nine young leaf samples collected from different individuals of highland papaya (three each young leaves from female, male, and monoecious).

Telomer length by PCR

Collecting samples of choice young leaves of highland papaya served as experimental material. The telomere length determination used PCR with the following process: leaf DNA extraction by cetyl trimethyl ammonium bromide (CTAB) method -leaves cut and weighed to 1 g, adding liquid nitrogen then crushed with a mortar, subsequently adding 700 µl of CTAB buffer solution and 1 µl RNAase (Promega, Beijing - China) to the mixture. The CTAB solution consisted of 2% CTAB, 1.26 M NaCl, 100 mM Tris-HCl pH 8, 20 mM ethylenediaminetetraacetic acid (EDTA), 1% PVP-40, and sterile aquadest. The mixture underwent incubation in a water bath at 66 °C for 10 min to ensure homogeneity. After incubation, mixing each sample continued with 500 µl of a blend of 24 Chloroform: 1 isoamyl alcohol (CIAA), then vortexed for 5 min.

The mixture gained centrifugation for 15 min at a rate of 13,000 rpm, with the resulting supernatant carefully removed and transferred to a new microtube, recording its volume. Isopropanol addition to 2/3 of the total volume followed, mixing the content well by flipping the tube, then centrifuging the mixture at a rate of 13,000 rpm for 10 min, and removing the supernatant after. The precipitated DNA, washed with 500 µl cold ethanol at 70%, ran into a centrifuge for another 5 min at a rate of 13,000 rpm. Finally, discarding the supernatant, the DNA precipitate attained aerating before storing in the refrigerator at 4 °C.

Dilutions progressed to obtain the required DNA concentration in the amplification protocol by PCR. The optimized primer annealing temperature was from 50 °C to 69 °C. Using the telomere primer of *Arabidopsis thaliana* (CCCTAAACCC-TAAACCCTAAACCCTAAACC) helped the DNA amplification by PCR to multiply the sequences. The stages of the DNA amplification reaction are in Table 1. The result attained electrophoresis at 100 volts for 55 min using 1.5% agarose in an electrophoresis tank containing pH 8 Tris/Borate EDTA (TBE) buffers, completely dissolved by heating in the microwave and then supplemented with 4 µl DNA staining. Agarose removal from the electrophoresis tank followed, then visualization using UV light, and photographed with a digital camera. Finally, telomere length measurements ran by the bands of amplification.

Elemental composition by SEM-EDX

The pieces of young leaves of highland papaya with various sexes served as samples for studying the elemental composition. Before scanning with the SEM-EDX machine, several preparatory steps included a) preparation of the samples, b) cutting the sample to the

Table 1. DNA amplification stages.

No.	Stages	Temperature (°C)	Time (minutes)
1	Pre-heating	94	5
2	Denaturation	94	0.5
3	Annealing	65	1.5
4	Elongation	72	0.5
5	Final elongation	72	7

appropriate size on an SEM-EDX machine, c) sanding the samples, and d) applying a coating of gold to the surface of the sample. Furthermore, performing microanalysis was at 20 kV.

Histochemical analysis

Preparation of fresh transverse sections of young leaves from different physiological stages for histochemical analysis comprised each segment, including female, male, and monoecious plant leaves, treated with a 5% NaOH solution (Sigma – Aldrich, Germany) and examined under a microscope for the presence of flavonoid compounds, identified with yellow color in the tissues (Etika and Iryani, 2019). Further soaking of each section in 1% $FeCl_3$ followed, then examined under a light microscope (Olympus, Tokyo - Japan) for tannin presence, indicated by the brown/black color (Trimanto *et al*., 2018).

Statistical analysis

For all the studied traits, the mean values determined came from three replications, with the quantitative data statistically analyzed by one-way analysis of variance with significant differences set at $p < 0.05$ using Microsoft Excel®. The analysis of variance also continued with a Tukey Test when the observed means for various treatments were significantly different.

Limitations

The study had a small sample size and ran in a controlled environment. It limits the generalizability of the findings to a larger population of plants and to a more natural environment. In future research, a larger sample size needs usage, with the study requiring a more natural environment for experimentation. It will help to ensure that the findings are universal to a wider population of plants.

RESULTS

Telomere length

The telomere length based on different sex of highland papaya young leaves had the length measurements of the bands amplified by female, male, and monoecious plants revealed by PCR (Figure 2). The female, male, and monoecious leaves of highland papaya exhibited 381, 391, and 396.67 base pairs, respectively (Table 2). PCR is a technique that amplifies DNA sequences. It can aid in measuring telomere length by amplifying a telomere repeat sequence and then measuring the amount of amplified DNA. The telomere length calculation comprised the amount of amplified DNA divided by the amount of input DNA.

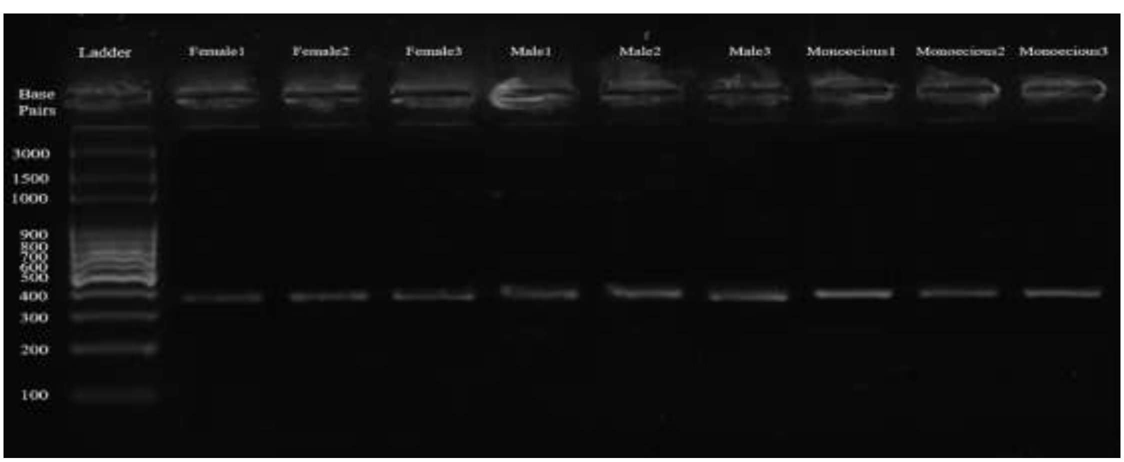

Figure 2. DNA amplification on different sexes of highland papaya, 1–3 = sample number.

Table 2. Telomere length of highland papaya leaves in different sexes.

Highland Papaya	Telomere Length (base pairs)
Female	381 a
Male	391 ab
Monoecious	396.67 b

Remarks: The means in one column and row followed by the same letter were not significantly different, according to Tukey (α 1%).

The observed differences in telomere length and elemental composition between female and male highland papaya leaves may have implications for the health-promoting properties of these leaves. Monoecious leaves have longer telomeres and a higher elemental composition than male leaves. It suggests that monoecious leaves may be more resistant to cell damage and may have a reduced risk of developing cancer and heart disease. They may also be more nutritious and less susceptible to pests and diseases.

Elemental composition

Based on the different sex of highland papaya for young leaves of female plants, the SEM analysis showed the absence of any material on the surface of plant leaves. A thin cuticle covers the epidermal layers, and one stoma on the upper epidermis. The EDX spectrum of female highland papaya leaves showed six absorbance peaks (Figure 3). The young leaves of highland papaya contain carbon, oxygen, magnesium, and sodium elements. Among these elements, carbon had the highest peak, while sodium had the lowest. Additionally, two weak absorbance peaks for the sodium element emerged.

For male highland papaya, the SEM observations also exhibited the absence of any material on the surface of the leaves. A covering of thin cuticles on epidermal layers spreads, and one stoma on the upper epidermis. The EDX spectrum of male highland papaya leaves showed five absorbance peaks (Figure 4). The young leaves of highland papaya hold carbon, oxygen, magnesium, and sodium elements. Among these elements, carbon peaked at the highest, while sodium was the lowest. Moreover, the sodium element displayed two weak absorbance peaks.

For monoecious highland papaya, the SEM interpretations disclosed the absence of any material on the plant leaves' surface. The epidermal layers have a thin cuticle covering and one stoma on the upper epidermis. The EDX spectrum of monoecious highland papaya leaves provided eight absorbance peaks (Figure 5). Also, they contain carbon, oxygen, sodium, magnesium, chlorine, and potassium elements. Carbon showed the highest peak, while Chlorine had the lowest. Furthermore, two weak absorbance peaks came out for the sodium and chlorine elements.

The highland papaya plant leaves for all three sexes had a thin cuticle/epicuticular wax covering, and a limited number of stomata appeared on the upper epidermis. In the observed elements, there were variations, where carbon and oxygen ranged from 48.24 to 50.77 and 45.73 to 46.76, respectively. The highest and lowest carbon content discovered were on the surface of male and female plant leaves. However, the highest oxygen content resulted in female plant leaves, whereas the lowest value was in crystals on the surface of male plant leaves. Magnesium and sodium elements ranged from 0.27 to 0.85 and 2.59 to 3.52, respectively. The lowest and highest magnesium values detected showed from monoecious and male plant leaves, respectively. The sodium element was minimum in female plant leaves, whereas the highest was in their monoecious counterpart. On the other hand, limited chlorine arose on the surface of monoecious plant leaves (Table 3).

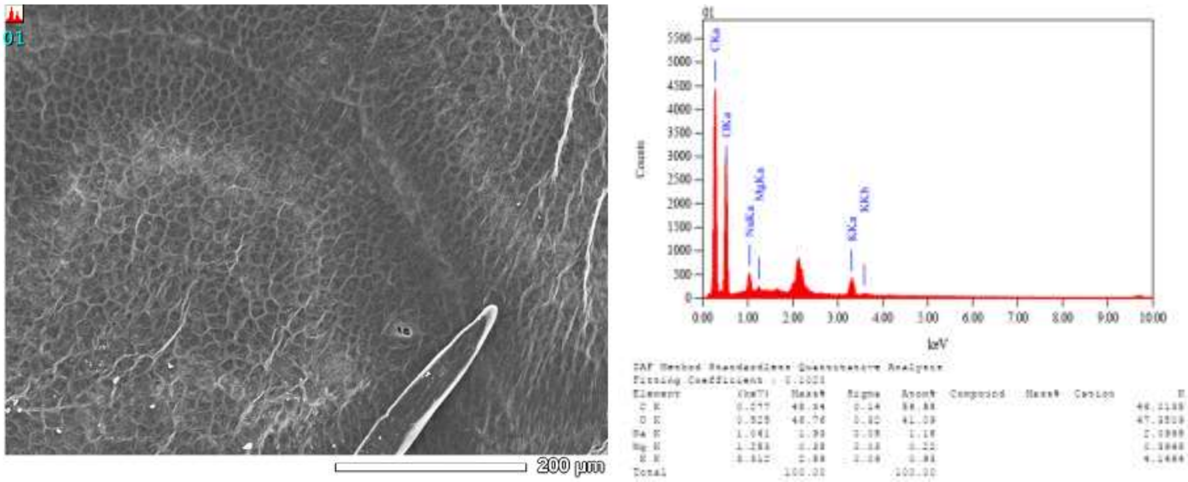

Figure 3. SEM-EDX analysis on the female leaf of highland papaya.

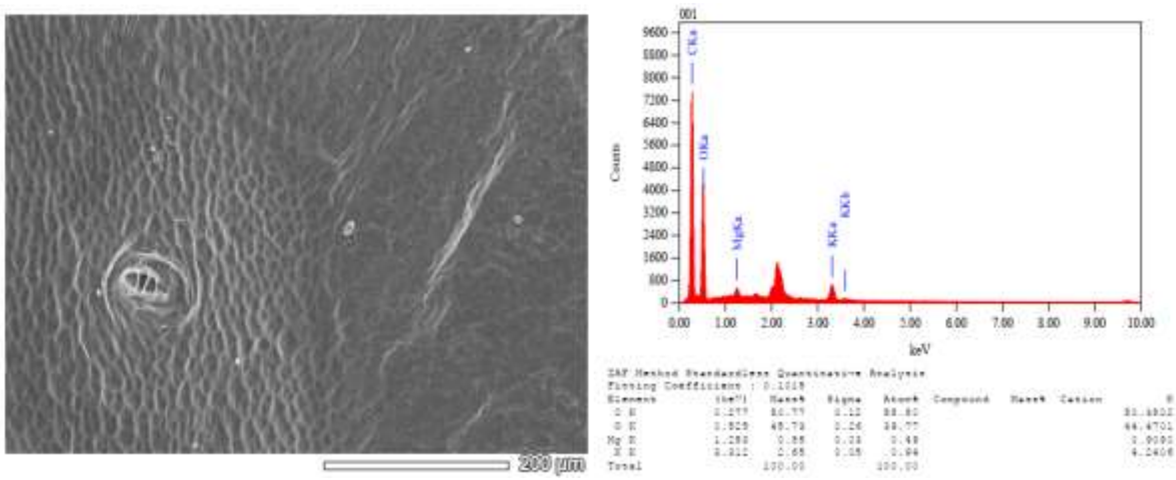

Figure 4. SEM-EDX analysis on the male leaf of highland papaya.

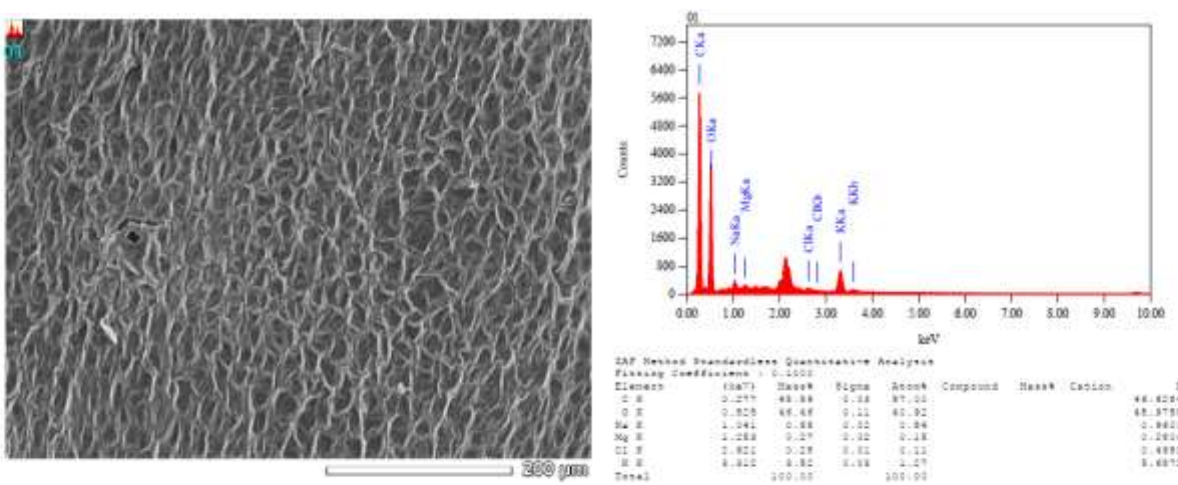

Figure 5. SEM-EDX analysis on the monoecious leaf of highland papaya.

Table 3. Elemental composition (mass %) of highland papaya leaves in different sexes.

No.	Element	Female leaf (%)	Male leaf (%)	Monoecious leaf (%)
1	Carbon	48.24	50.77	48.59
2	Oxygen	46.76	45.73	46.46
3	Magnesium	0.38	0.85	0.27
4	Potassium	2.59	2.65	3.52
5	Sodium	1.93	0	0.88
6	Chlorine	0	0	0.28

Histochemical analysis

Based on the different sex of the highland papaya plants' young leaves, histochemical analysis showed the presence of compounds with medicinal properties, such as, flavonoids and tannins, distributed throughout the tissues of their organs. The results indicated the presence of these two compounds in the leaves of highland papaya (Figure 6). These components, detected in specific areas, spread across the plant tissues. The NaOH staining of flavonoids produced a yellow color, while the $FeCl_3$ staining of tannins produced a black color. Flavonoid content recurred in the adaxial and abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, and trichome of the highland papaya leaves for the female, male, and monoecious plants studied (Table 4). Similarly, tannins surfaced in the adaxial and abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, and trichome in the female, male, and monoecious plant leaves (Figure 6).

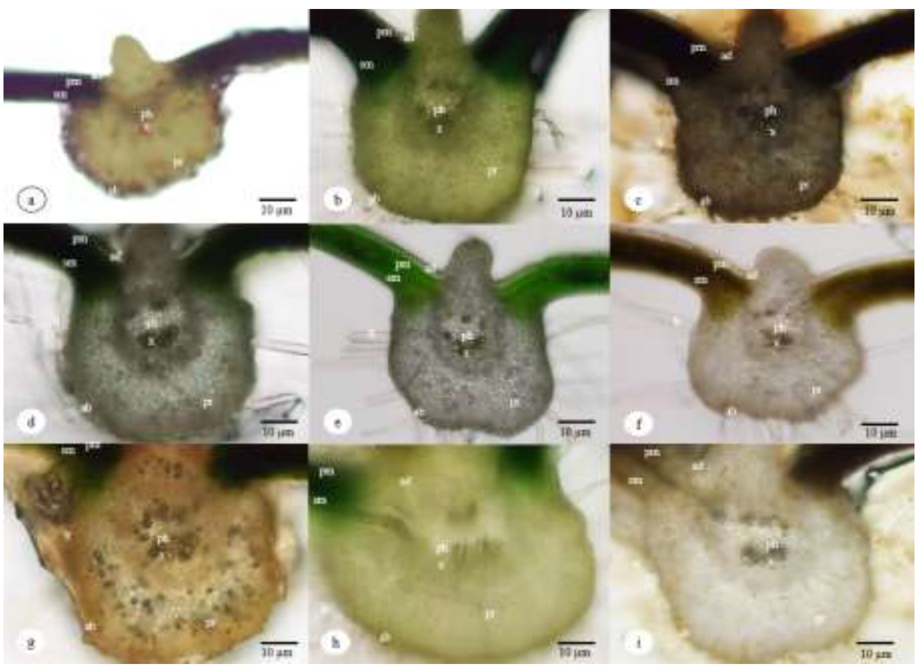

Figure 6. Transverse sections of histochemically stained female, male, and monoecious leaves of highland papaya. a: Freehand of a female leaf without staining, b: the positive reaction of 5% NaOH, c: the positive reaction of 1% $FeCl_3$, d: Freehand of a male leaf without staining, e: the positive reaction of 5% NaOH, f: the positive reaction of 1% $FeCl_3$, g: Freehand of a monoecious leaf without staining, h: the positive reaction of 5% NaOH, i: the positive reaction of 1% $FeCl_3$, palisade mesophyll (pm), sponge mesophyll (sm), xylem (x), phloem (ph), parenchyma (pr), trichomes (tr), the abaxial epidermis (ab), and adaxial epidermis (ad) Bars: a–i: 10 µm.

Table 4. Distribution of phytochemical content of highland papaya leaves in different sexes.

Phytochemical Content	Reagent Test	Positive color	Female leaves	Male leaves	Monoecious leaves
Flavonoid	NaOH 5%	Yellow	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome
Tannin	$FeCl_3$ 1%	Black	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome	adaxial and the abaxial epidermis, mesophyll, xylem, phloem, parenchyma midrib, secretory cavity, trichome

DISCUSSION

The longer telomeres in crop plants may be more adaptive with faster developmental rates and early flowering (Choi *et al*., 2021). The female plants' ash had shorter telomeres than the males, while the female willow had longer (Mu *et al*., 2014). However, further study needs execution to examine the length in females, males, and monoecious highland papaya plants. The telomere length of the female and male *Salacca zalacca* plants was equal, making it impossible to determine the particular sex (Indhirawati and Purwantoro, 2018). Developing early methods for determining sex types in dioecious crops could help alleviate the difficulties faced by farmers. It occurs particularly when all the superior parental selections have an unknown composition of commercially undesirable sex types. Moreover, it aids the farming community in saving field space, time, and other valuable resources that would otherwise go to waste in maintaining undesirable plants until flowering (Heikrujam *et al*., 2015).

Research by Liang *et al*. (2015) on *Panax ginseng* has suggested a complex relationship between telomere length and plant growth. It could be due to influences by environmental factors and telomerase activity. Telomere length variations might occur because of the length and the enzyme activities' relationships (Mu *et al*., 2014). The enzymes shown in several plant species have the highest activity observed in meristems and reproductive organs, while little action emerged in endosperms, leaves, and stems (Liang *et al*., 2015). Studies have shown that telomeres' shortening may result from the early stages of apoptosis (Rescalvo-Morales *et al*., 2016). As the activity and relative length are complex phenomena, further studies require an undertaking to examine the molecular mechanisms of telomere-related proteins (Yun *et al*., 2022). The average telomere length of female highland papaya leaves (381 base pairs) is similar to the female leaves' average telomere length of other plant species, such as, spinach (380 base pairs) and kale (385 base pairs), according to Dekker *et al*. (1994). However, the average telomere length of male highland papaya leaves (391 base pairs) is slightly longer than the male leaves' average telomere length of other plant species, such as, spinach (385 base pairs) and kale (380 base pairs). The average telomere length of monoecious highland papaya leaves (396.67 base pairs) is the longest of all three sexes and significantly longer than the average telomere length of male and female leaves of other plant species.

Numerous studies have shown that oxidative stress correlates with accelerated telomere shortening and dysfunction (Barnes *et al*., 2019). Scientific evidence supports that oxidative stress can contribute to telomeric attrition and plays a vital role in developing certain age-related diseases (Gavia-García and Rosado-Pérez, 2021). Earlier studies have associated telomere shortening with functional decline, and alongside oxidative stress markers, it has links with higher mortality risks and decreased survival (Bernabeu-Wittel *et al*., 2020). Standard growth conditions also showed telomeres accumulating 8-oxoG at approximately 100-fold greater than the rest of the genome. This accumulation was inversely proportional to the length, supporting the hypothesis that telomeres were hotspots of 8-oxoG and may function as sentinels of oxidative stress in crop plants (Castillo-González *et al*., 2022). The observed differences in telomere length and elemental composition between female, male, and monoecious highland papaya plants may be due to the different hormonal and metabolic pathways that are active in each sex, as well as, the varied nutrient requirements of each sex. More research needs confirmation of these findings to determine the specific hormonal and metabolic pathways responsible for the differences in telomere length and elemental composition between female, male, and monoecious highland papaya plants.

According to Rashmi and Sanjay (2017), the strong carbon signal in the EDX spectral analysis was due to the grid used, while the strong oxygen peaks resulted from sample oxidation. The result showed that the carbon content increased significantly, while the other elements, such as, oxygen, magnesium, silicon, and calcium, decreased during the activation in *Carica papaya* L. leaves (Agustina *et al*., 2016). It was attributable to the decomposition of volatile compounds and degradation of organic substances during wet activation, leaving behind carbon with high purity (Bello *et al*., 2016). The effectiveness of the sample as an adsorbent was said to increase with a higher amount of carbon (Agustina *et al*., 2016). In the presented study, the observed elements contributed to the therapeutic action of the plant (Yashvanth *et al*., 2015). The elemental composition of highland papaya leaves is similar to other plant species' elemental composition of leaves. The most abundant elements in upland papaya leaves are carbon, hydrogen, oxygen, and nitrogen. These elements are essential for plant growth and development.

Results of this study showed that all tissues examined in the leaves of highland papaya contained flavonoids. The distribution of flavonoids in the different tissues suggests that they could play varied roles in the plant, including protection from UV radiation, antioxidant activity, signal transduction, and plant defense. Past studies revealed flavonoids emerging in the *Acalypha indica* L. (Purwestri *et al*., 2016). Flavonoids are common in most crop plant species yet distinctive to many other categories. The flavonoids are relatively straightforward to recognize and can serve as chemical markers in taxonomic classification. De-Oliveira *et al*. (2017) also reported that flavonoids can benefit as a taxonomic characteristic in the genus *Chromolaena*. Highland papaya leaves contain a variety of phytochemicals, including flavonoids and alkaloids. These phytochemicals have a variety of health benefits, including antioxidant, anti-inflammatory, and anticancer properties. Monoecious highland papaya leaves have longer telomeres and a more diverse range of phytochemicals than male or female leaves; however, the composition of elements is almost similar in the three sexes.

The research findings on telomere length, elemental composition, and phytochemical properties of highland papaya leaves are consistent with outcomes from previous studies on other plant species. These findings suggest that upland papaya leaves could have similar health benefits to other plant species' leaves. However, more research requires validating these findings and determination of the specific health benefits of highland papaya leaves.

Several areas can further enhance and improve the quality of this research, namely, 1) sample size: larger sample sizes can increase the statistical power of a study and improve the universality of the findings, and 2) control for confounding variables that may affect the outcome of the study to ensure that the observed effects are due to the independent variable. It can consider using appropriate microscopy techniques, such as, light or fluorescence microscopy, to visualize the stained plant tissues and accurately identify the spatial allocation of flavonoid compounds. In addition, it can utilize other analytical techniques in quantifying flavonoid compounds' contents in plant tissues, such as, high-performance liquid chromatography (HPLC) or mass spectrometry (MS), to provide measurements of the flavonoid content.

CONCLUSIONS

The study showed that highland papaya exhibited significantly different telomere lengths in its female, male, and monoecious plant leaves. The female plants have the shortest telomeres, at 381 base pairs. The male plants have the longest telomeres, at 391 base pairs. The monoecious plants have telomeres that are intermediate in length, at 396.67 base pairs. The results of the PCR test suggest that telomere length may be a useful sex marker in highland papaya plants. Elemental and chemical composition analyses indicated the presence of sodium in female plant leaves, while the absence of sodium and chlorine in male plant leaves, but present in monoecious plant leaves. It implies that highland papaya leaves are a greater source of health-promoting compounds. Phytochemical studies also verified the presence of pharmaceutically active secondary metabolites, primarily flavonoids, and tannins. Further studies need exploration of the therapeutic potential of these compounds and their effects on human health.

ACKNOWLEDGMENTS

The authors thank the RTA Program, with Grant Number 5722/UN1.P.III/Dit-Lit/PT.01.05/2022, as well as, the farmers in Dieng, Batu, and Bromo, Indonesia, for their tremendous assistance and great support.

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