ABS

Original Research

Impact of water deficit on the anatomical structure of more productive and less productive cashew trees (*Anacardium occidentale* L.) in Côte d'Ivoire

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Kouamé N'Guessan François[3]

Abstract

This study aimed to understand the mechanism of resilience of the anatomical tissues of organs of the cashew tree in water deficit in the different climatic zones of Côte d'Ivoire. To achieve this, samples of leaves and roots were taken from 55 cashew trees considered high producers (APHP) and 164 neighbouring less productive trees (non-APHP) during the dry and rainy seasons. The technique of double staining with green-carmino made it possible to obtain anatomical sections of transversely cut samples under an optical microscope. The thicknesses and surface areas of the sections were measured using Image J software. The thickness of the leaf parenchymas of APHP trees did not show any variation according to the season, while the stomatal density increased during the dry season. The thickness of the leaves cortical and medullary parenchymas of non-APHP trees did not show any variation according to the season too, but the non-APHPs' palisade parenchyma thickness increased while their lacunar parenchyma thickness decreased from 37 ± 1.5 µm to 36 ± 3.6 µm during the dry season. The thickness of leaves on both sides of the epidermis increased while the thickness of xylem bundles decreased during the dry season in both APHP and non-APHP trees. The number of the roots' xylem vessels of APHP trees increased from 11 to 27 vessels/mm^2, while those of non-APHP trees increased from 15 to 41 vessels/mm^2 during the rainy season. The surface area of the midrib medullary parenchyma of APHP trees increased from 4300 ± 837 µm^2 to 5800 ± 412 µm^2 while those of non-APHP trees increased from 3400 ± 809 µm^2 to 5200 ± 993 µm^2 during the dry season. The thicknesses of the tissues of APHPs remained greater than those of non-APHPs, regardless of the sampling season.

Keywords

Cashew tree, anatomy, adaptation, water deficit, Côte d'Ivoire

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Vpliv pomanjkanja vode na anatomsko strukturo bolj produktivnih in manj produktivnih dreves indijskega oreščka (*Anacardium occidentale* L.) v Slonokoščeni obali

Izvleček

Namen naše raziskave je bil razumeti mehanizem odpornosti anatomskih tkiv organov drevesa indijskega oreščka na pomanjkanje vode v različnih podnebnih območjih Slonokoščene obale. Za dosego tega cilja so bili med suho in deževno sezono odvzeti vzorci listov in korenin pri 55 drevesih indijskega oreščka, ki veljajo za visoko produktivna (APHP), ter pri 164 sosednjih manj produktivnih drevesih. Tehnika dvojnega barvanja z zelenim karminom je omogočila pridobitev anatomskih prerezov prečno rezanih vzorcev, ki smo jih opazovali pod optičnim mikroskopom. Debeline in površine prerezov so bile izmerjene z uporabo programske opreme Image J. Debelina parenhima listov pri drevesih APHP ni kazala sezonskih sprememb, medtem ko se je gostota rež povečala v sušni sezoni. Prav tako se debelina kortikalnega in medularnega parenhima listov dreves brez APHP ni spreminjala glede na sezono, vendar se je debelina njihovega palisadnega parenhima povečala, medtem ko se je debelina lakunarnega parenhima zmanjšala s 37 ± 1,5 µm na 36 ± 3,6 µm v sušni sezoni. Debelina epidermisa na obeh straneh listov se je povečala, medtem ko se je debelina ksilema zmanjšala v sušni sezoni pri drevesih APHP in ne-APHP. Število ksilemskih elementov v koreninah dreves APHP se je povečalo z 11 na 27 žil/mm^2, pri drevesih brez APHP pa s 15 na 41 žil/mm^2 v deževni sezoni. Površina medularnega parenhima glavne žile korenin pri drevesih APHP se je povečala s 4300 ± 837 µm^2 na 5800 ± 412 µm^2, medtem ko se je pri drevesih brez APHP povečala s 3400 ± 809 µm^2 na 5200 ± 993 µm^2 v sušni sezoni. Debeline tkiv pri drevesih APHP so ostale večje kot pri drevesih brez APHP, ne glede na sezono vzorčenja.

Ključne besede

Indijski oreščsek, anatomija, prilagoditev, pomanjkanje vode, Slonokoščena obala

Introduction

The cashew tree (*Anacardium occidentale* L.) belongs to the clades Angiospermae and Eudicotidae, the order Sapindales and the Anacardiaceae family (APG 2016). Cashew cultivation constitutes an important new source of income for African farmers, who account for more than 55% of the global cashew nut production. Côte d'Ivoire is the leading African producer and exporter of raw cashew nuts (Henry et al. 2011) and the world's third biggest producer and exporter of raw cashew nuts after India and Vietnam (FAO 2024). Its production of raw cashew nuts was estimated at 970,000 tons in 2022 (FAO 2024). Despite this performance and constant evolution, Ivorian orchard yields are between 350 to 500 kg/ha (Djaha et al. 2010, FAO 2024) compared to those obtained in India, Vietnam, Brazil and Tanzania, which are between 1,000 and 1,500 kg/ha (Kiwuso et al. 2013). Cashew nut producers encounter enormous difficulties due to several factors, such as insufficient technical supervision, the use of low-yielding varieties and unknown sources (Charahabil et al. 2017).

In Côte d'Ivoire, the mass selection technique was used to identify Potentially High-Producing APHP Cashew Trees (Konan 2016). These trees were selected on the basis of the following characteristics: good tree architecture, precocity and grouped maturity, shape and quality of nuts, nut weight ≥ 7 g and yield ≥ 10 kg of nuts/tree/year. However, some studies have pointed out the existence of a relationship between the anatomy and the production potential of fruit trees. However, abiotic factors are the trigger for stress. Plant responses to these stresses are either plastic and reversible or irreversible (Skirycz and Inze 2010, Cramer et al. 2011). Indeed, the leaf is the most adaptable organ in its response to environmental conditions (Marchi et al. 2008). Leaf structures reflect the effects of water stress more clearly than the structures of stems and/or roots, whose effects are also visible.

This work aimed to understand the resilience mechanisms of leaf and root tissues of Potentially High-Producing Cashew Trees (APHP) and their neighbours (non-APHP) in the cashew basin of Côte d'Ivoire according to different seasons.

Material and methods

Study areas

This study was carried out in the cashew basin of Côte d'Ivoire across 15 administrative regions (Figure 1) and two agro-ecological zones, characterized as follows:

- Guinean Sector, which is a transition zone between the forest zone in the south and the grassy savannah of the centre, with a dry season from January to February, a major rainy season from March to October and a small rainy season from November to December (FAO 2005, Konaté 2021). The annual rainfall is between 1,031.5 mm and 1,189.36 mm, while the temperature varies between 16 °C and 36 °C, with an average of 27 °C (FAO 2005, Ouattara et al. 2016);
- The Sudanian Sector is characterized by the north savannah vegetation, with a dry season from November to February and a long rainy season from March to October (FAO 2005, Konaté 2021). The annual rainfall is between 1,316.19 mm and 1,708.39 mm, and the temperature varies between 28 °C and 32 °C (FAO 2005, Ouattara et al. 2016).

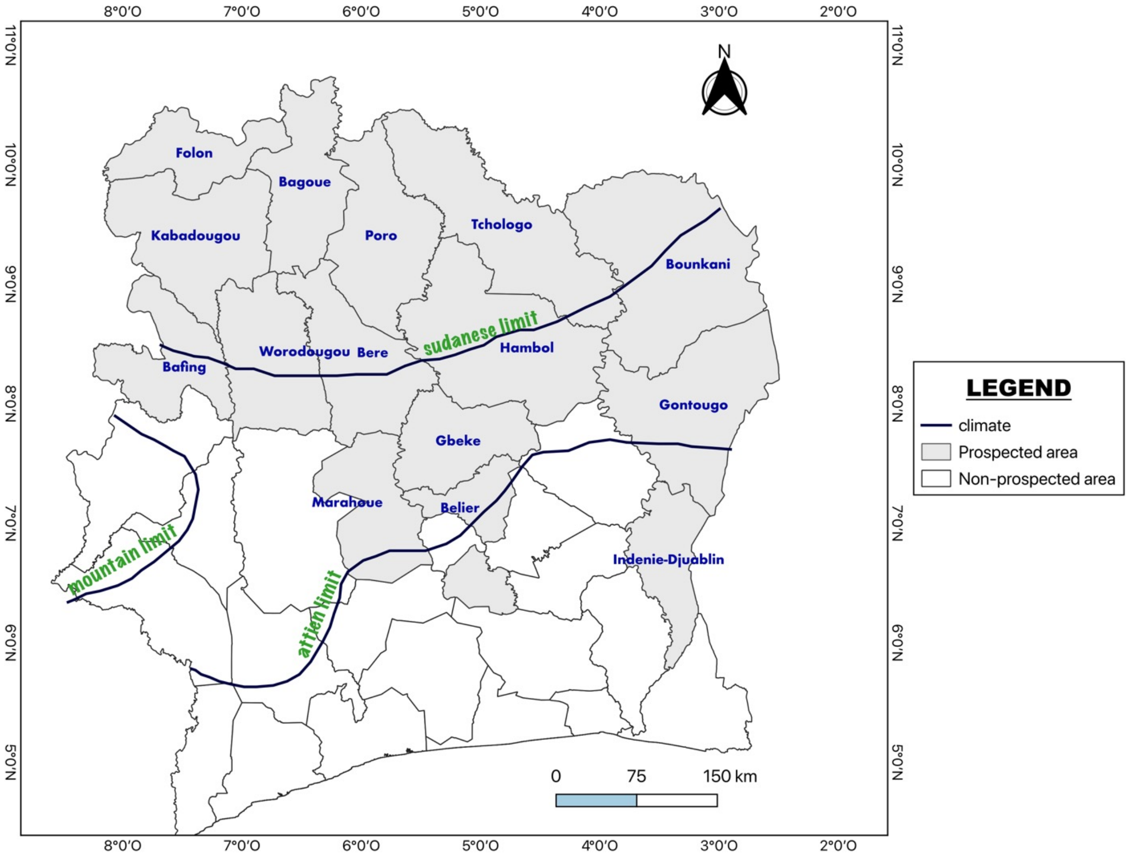

Figure 1. Map of the sample collection area.

Slika 1. Zemljevid območja zbiranja vzorcev.

Data collection

The samples of mature leaves and roots were collected according to the method of Sankharé (2018) from 219 cashew trees, including 55 that are known to be potentially more productive (APHP) and their 164 less productive immediate neighbours (non-APHP) in the four cardinal directions, over 15 administrative regions of the cashew production basin in Côte d'Ivoire (Figure 1). The leaves about 10-12 cm long and 6-8 cm wide, and the roots with a diameter of about 0.2-0.4 mm were collected during the dry season (February-March) and during the rainy season (July- August). These samples were stored in 70% alcohol before and during their preparation and observation under an optical microscope. Using a manual microtome and polystyrene, 0.1-0.2 mm microscopic sections of leaves, blade veins, midlines and roots were cut and stained using the classic green-carmino double staining technique (Ruzin 1999). To observe the stomata, a razor blade was placed on the lower surfaces of the leaves, and then the transparent epidermis located on the surfaces of the leaves was gently lifted. The fragments of the obtained transparent epidermis constituted the samples that were observed (Djinet et al. 2016). Anatomical tissues were visualized and recorded using Leica Acquire software.

Image analysis

The captured images were saved in 640×480 pixels format. Each anatomical section was measured with a micrometre graduated in millimetres (mm) and then converted to micrometres (µm) before observation under a microscope. Afterwards, the image of each anatomical section was measured in pixels at 10×, 20×, 40× and 100× magnifications. Therefore, calibration was done on each of the sections using this process to convert the photographs of the sections into µm.

Histological analysis

The anatomical tissues of leaves and roots were analyzed and measured with the Image-J 1.53t software. The parameters taken into account were:

- The thickness of the parenchymas, where the absorbed water is used for the photosynthetic activities;
- the structure of the epidermis that regulates water exchange between the interior and the exterior of the cashew aerial organs;
- the stomatal density and the surface of the opening of the ostiole of the stomata, which allow gas and water exchange between the interior and the exterior of the aerial organs of the cashew tree:

$$DS = \frac{\textit{Number of stomata}}{\text{Leaf surface}}$$

- The characteristics of the xylems that conduct the absorbed water toward the cashew organs. Regarding the leaf midrib xylem diameter, the diagonal diameters were measured as recommended (Stein et al. 2017). The diameters of the root xylem vessels were measured taking into account both the tangential diameter (DT) and the radial diameter (DR); their ratio (R) was calculated by the following expression:

$$R = \frac{\text{DT}}{\text{DR}}$$

- The number of root xylem vessels was determined with the cell counter tool on 1 area of 1×1 mm (Grishaguin 1985).

Statistical analysis

The data collected in the Excel spreadsheet were analyzed using the R software to compare thicknesses and visualize them. The ANOVA test and Student t-test were performed to compare the data during the rainy and the dry seasons at the 5% significance level ($p < 0.05$). Pearson's correlation test was performed to assess the relation between the number of xylem vessels and the ratio of the tangential and radial diameters of the vessels of the roots.

Results

Leaf tissues

Parenchymas

The average thicknesses of the lacunar parenchyma (Figure 2) of both APHPs and non-APHPs were larger during the rainy season than during the dry season (Table 1, Figure 3). The season did not show any impact (P = 0.090) on the thickness of the APHPs' lacunar parenchyma, while drought showed a high negative impact (P = 0.009) on the thickness of the non-APHPs' lacunar parenchyma, which decreased from 37 ± 1.5 µm during the rainy season to 36 ±

3.6 µm during the dry season (Table 1).

The season did not show any impact ($P > 0.105$) on the thicknesses of the APHPs' lower and upper palisade parenchyma, while drought showed a high positive impact ($P < 0.028$) on the thicknesses of the non-APHPs' palisade parenchymas (Table 1) which increased from 54 ± 4.4 µm during the rainy season to 59 ± 5.4 µm during the dry season at the upper face, and from 21 ± 10.9 µm during the rainy season to 22 ± 5.2 µm during the dry season at the lower face (Table 1, Figure 3).

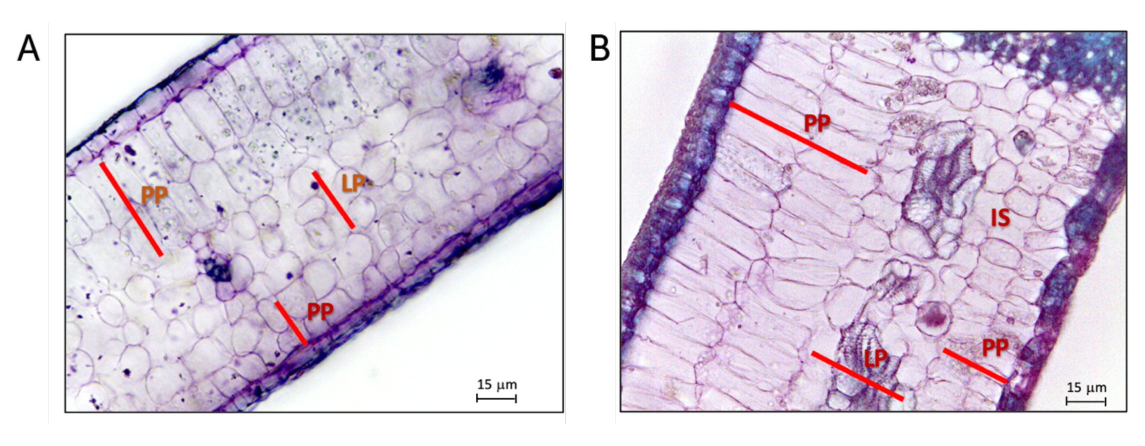

Figure 2. Pictures of the flattened blade of the cashew leaves according to the seasons (100× magnification). A: dry season; B: rainy season; PP: palisade parenchyma; LP: lacunar parenchyma; IS: intercellular space

Slika 2. Slike sploščene listne ploskve indijskega oreščka glede na letni čas (100-kratna povečava). A: suha sezona; B: deževna sezona; PP: palisadni parenhim; LP: lakunarni parenhim; IS: medcelični prostor

Table 1. Average thickness of leaf tissues according to the cashew tree categories and to the seasons. APHP: potentially higher-producing cashew trees. For the tissue, paired data with the same letter (P-value > 5%) are statistically similar, while data with different letters are different (5% > P-value > 1%) or highly different (1% > P-value > 0.1%).

Tabela 1. Povprečna debelina listnih tkiv glede na kategorije indijskih dreves in letne čase. APHP: drevesa indijskega oreščka s potencialno višjo rodnostjo. Pri tkivu so si parni podatki z isto črko (P-vrednost > 5 %) statistično podobni, medtem ko so podatki z različnimi črkami različni (5 % > P-vrednost > 1 %) ali zelo različni (1 % > P-vrednost > 0,1 %).

Tissue location		**Flattened blade tissues**					**Midrib tissues**			
Tree categories	**Season type**	**Upper palisade parenchyma (µm)**	**Lower palisade parenchyma (µm)**	**Lacunar parenchyma (µm)**	**Upper epidermis (µm)**	**Lower epidermis (µm)**	**Upper cortical parenchyma (µm)**	**Lower cortical parenchyma (µm)**	**Medullary parenchyma (µm²)**	**Xylem bundles (µm)**
APHP	Rainy season	52 ± 4.2^{a}	20 ± 2.9^{a}	40 ± 4.0^{a}	5.6 ± 0.4^{a}	5 ± 0.4^{a}	8 ± 2.3^{a}	18.5 ± 4.5^{a}	4300 ± 837^{a}	6.5± 0.7^{a}
	Dry season	52 ± 6.3^{a}	28 ± 2.1^{a}	30± 6.3^{a}	6.2 ± 1.0^{b}	5.6 ± 1.1^{b}	6.3 ± 0.8^{a}	22 ± 3.4^{a}	5800± 412^{a}	6.4± 0.5^{b}
	P-value	**0.500**	**0.105**	**0.090**	**0.032**	**0.036**	**0.075**	**0.055**	**0.094**	**0.005**
Non-APHP	Rainy season	54 ± 4.4^{a}	21 ± 10.9^{a}	37 ± 1.5^{a}	6.4 ± 0.3^{a}	5 ± 0.4^{a}	7.2 ± 1.2^{a}	15 ± 4.9^{a}	3400 ± 809^{a}	6.7 ± 0.9^{a}
	Dry season	59 ± 5.4^{b}	22± 5.2^{b}	36± 3.6^{b}	6.5 ± 1.1^{b}	5.5 ± 0.7^{b}	6.2± 0.5^{b}	20 ± 2.0^{a}	5200 ± 993^{a}	5.9 ± 0.6^{b}
	P-value	**0.028**	**0.015**	**0.009**	**0.005**	**0.030**	**0.047**	**0.090**	**0.131**	**0.040**

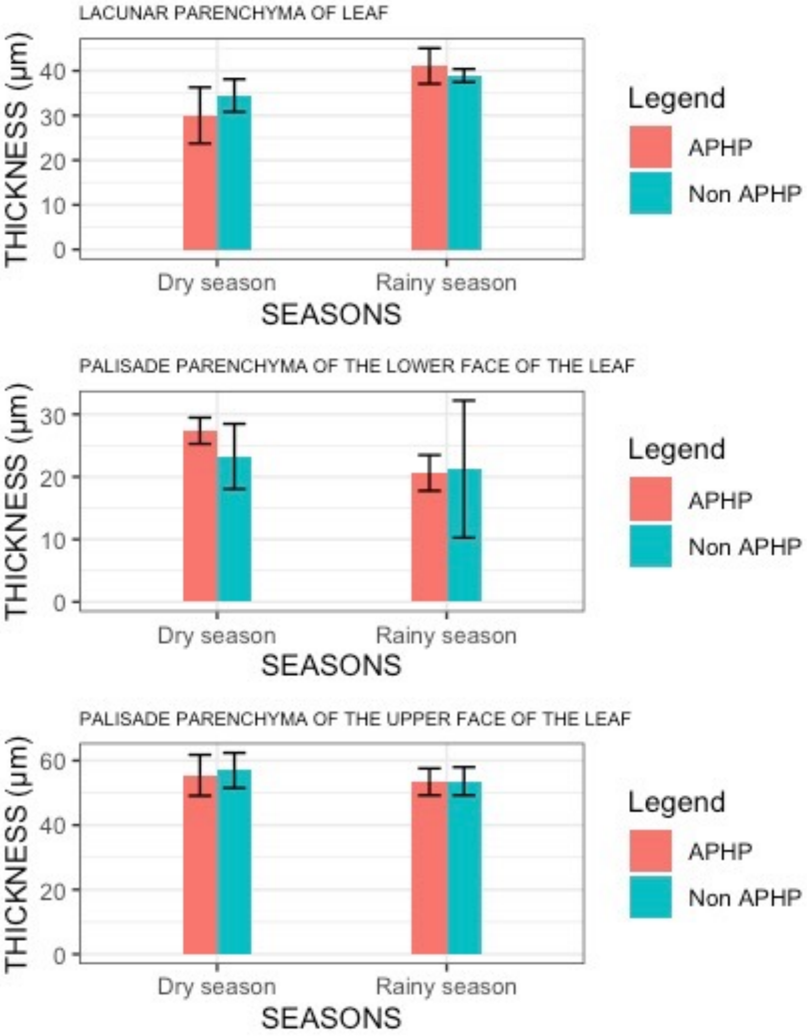

Figure 3. Thickness of the parenchyma of flattened leaf blades. IE: lower palisade parenchyma; SE: upper palisade parenchyma.

Slika 3. Debelina parenhima sploščenih listnih lopatic. IE: spodnji palisadni parenhim; SE: zgornji palisadni parenhim.

No impact ($P > 0.055$) of the season was found on the thicknesses of the APHPs' midrib in both face cortical parenchymas and on the non-APHPs' lower face cortical parenchyma thickness ($P = 0.090$), while drought showed a negative impact ($P = 0.047$) on the thickness of the non-APHPs' upper face cortical parenchyma which decreased from 7.2 ± 1.2 µm during the rainy season to 6.2 ± 0.5 µm during the dry season (Table 1, Figure 4).

The season did not show any impact ($P > 0.093$) on the area of the medullary parenchyma of both APHPs and non-APHPs, even if the average area of the medullary parenchyma of the APHPs was about 4,300 ± 837 µm^2 during the rainy season and 5,800 ± 412 µm^2 during the dry season, while that of the non-APHPs was set at 3,400 ± 809 µm^2 during the rainy season and at 5,200 ± 993 µm^2 during the dry season (Table 1, Figure 5).

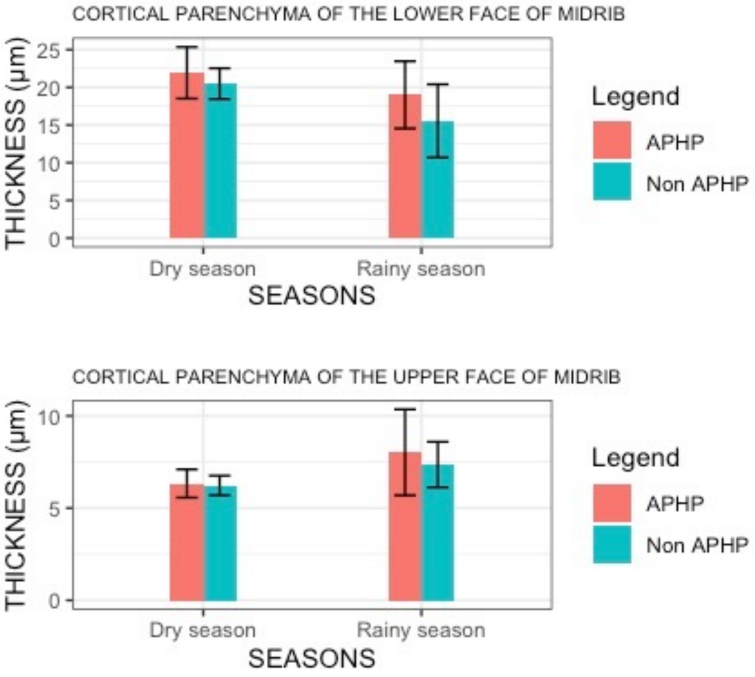

Figure 4. Thickness of cortical parenchyma.

Slika 4. Debelina kortikalnega parenhima.

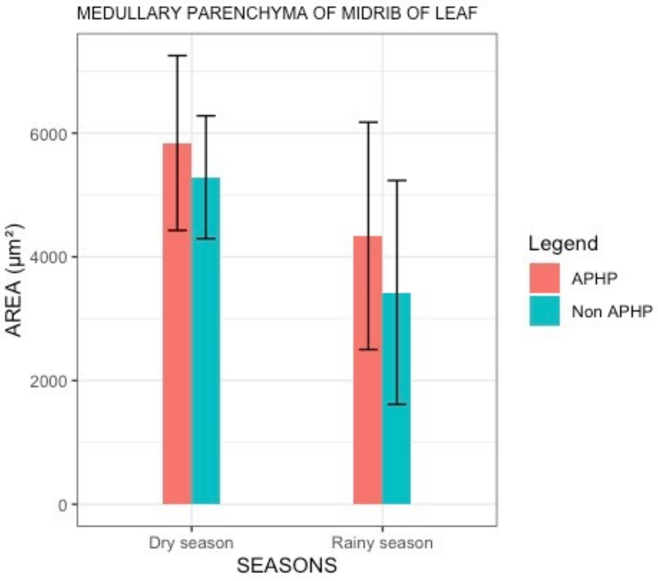

Figure 5. Area of the medullary parenchyma.

Slika 5. Območje medularnega parenhima.

Epidermis

Firstly, both APHPs and non-APHPs showed a similar thickness of the upper and the lower epidermis, and secondly, an upper epidermis thicker than the lower epidermis, independently of the seasons (Table 1, Figure 6). Moreover, both categories of cashew leaves showed an increase ($P \leq 0.036$) in the thickness of both the lower and upper epidermis from the rainy season to the dry season (Table 1, Figure 6).

Xylem

Both categories of cashew leaves showed the same thickness of xylem vessels for each season but a variation of the thickness of xylem vessels according to the seasons (Table 1, Figures 7 and 8). Indeed, there was a decrease ($P = 0.040$) in the non-APHPs' xylem vessel thickness which moved from 6.7 ± 0.9 µm during the rainy season to 5.9 ± 0.6 µm during the dry season, and a high decrease ($P = 0.004$) of the APHPs' xylem vessel thickness which changed from 6.5 ± 0.7 µm during the rainy season to 6.4 ± 0.5 µm the dry season (Table 1, Figure 7).

Stomatal density and opening surface

The average stomatal density at the abaxial faces of the leaves was the same ($P \geq 0.365$) for the APHPs and the non-APHPs during each season. However, the average stomatal density of APHPs was lower, with 155 ± 16.9 stomata/mm2 during the rainy season than those of 190.4 ±41.6 stomata/mm2 during the dry season (Figure 9). The non-APHPs also showed a lower average stomatal density of 147 ± 15.6 stomata/mm2 during the rainy season than those of 170.4 ± 20.8 stomata/mm2 during the dry season (Figure 9).

In both APHP trees and non-APHP trees, water deficit affected ($P < 0.018$) the opening width of the stomata, reducing it from 59.8 µm to 23.0 µm and from 47.3 to 22.2 µm, respectively, during the dry season (Figure 10). During each season, the stomata opening width was the same ($P > 0.552$) for the APHPs and the non-APHPs, which was 35.5 µm during the rainy season (Figure 11) and 19.2 µm during the dry season (Figure 12).

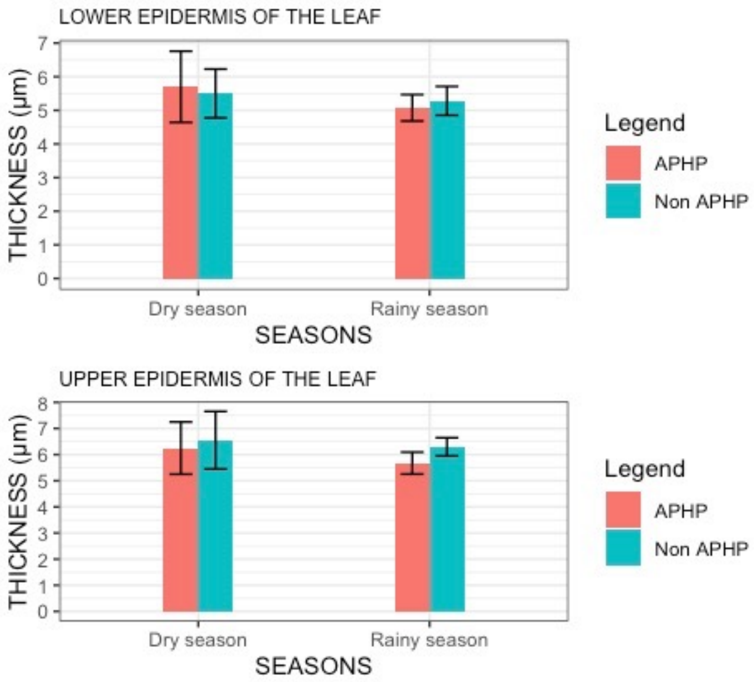

Figure 6. Thickness of the upper and lower leaf epidermis according to the season.

Slika 6. Debelina zgornje in spodnje listne povrhnjice glede na sezono.

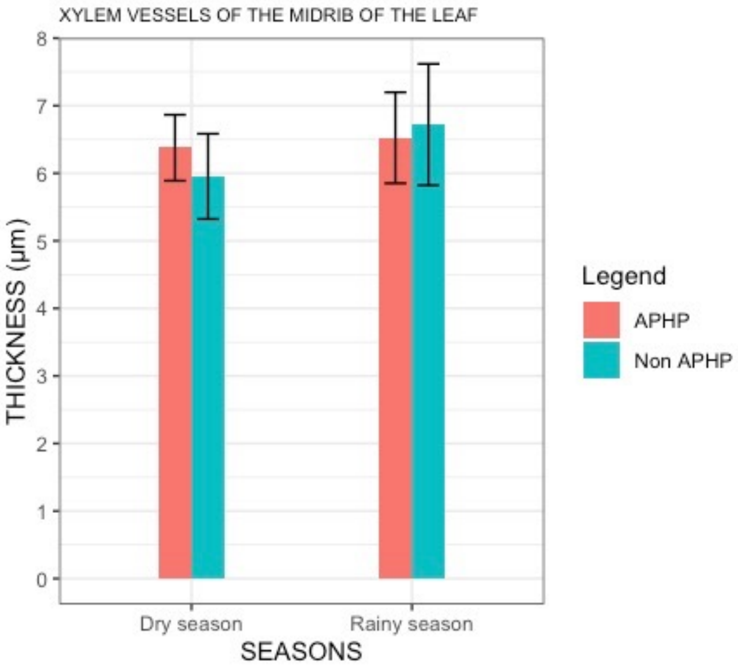

Figure 7. Xylem vessel thicknesses of the leaf midrib.

Slika 7. Debelina ksilemskih žil na sredini lista.

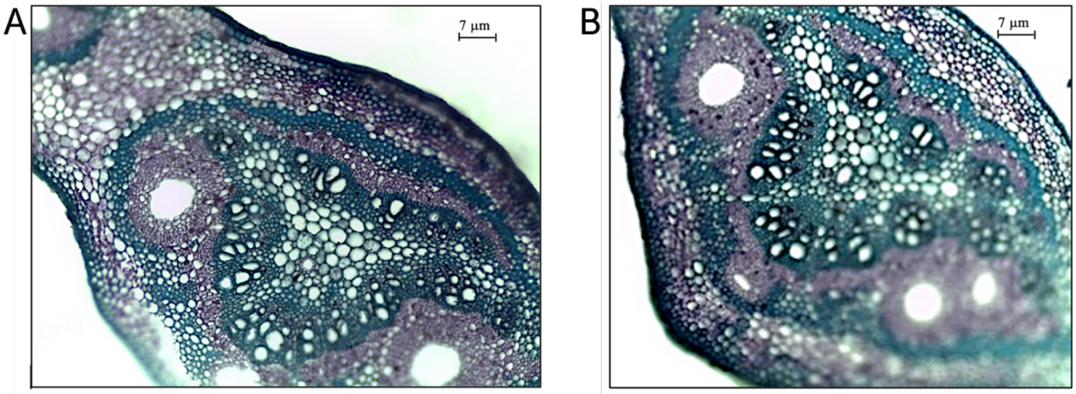

Figure 8. The anatomical structure of the main vein of cashew leaves according to the season (40× magnification). A: during the rainy season (production); B: during the dry season (after production).

Slika 8. Anatomska zgradba glavne žile listov indijskega oreščka glede na sezono (40-kratna povečava). A: v deževnem obdobju (proizvodnja); B: v suhem obdobju (po proizvodnji).

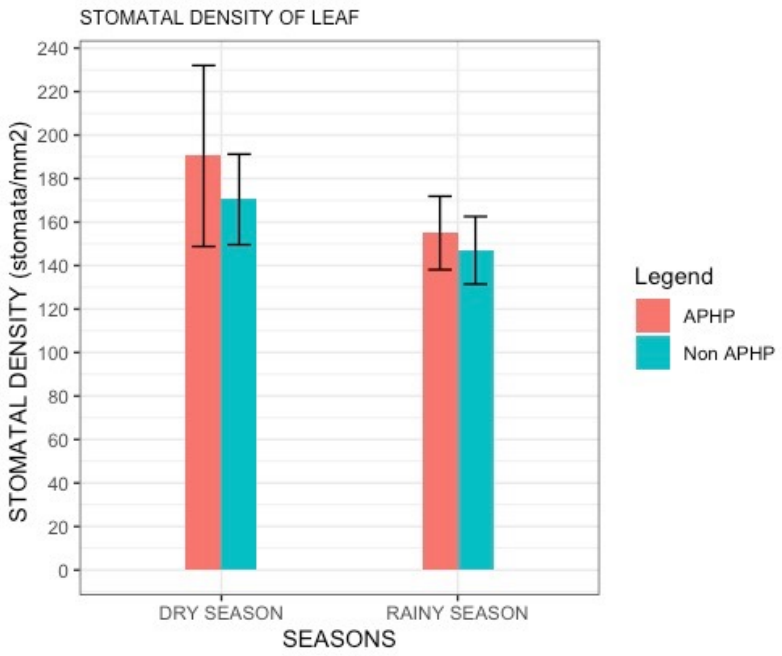

Figure 9. The stomatal density of cashew trees. For each locality of origin of the cashew trees, the letter V is used to distinguish the non-APHPs from the APHPs.

Slika 9. Stomatalna gostota indijskih dreves. Za vsako lokacijo izvora indijskih dreves je uporabljena črka V za razlikovanje med drevesi, ki niso APHP, in drevesi, ki niso APHP.

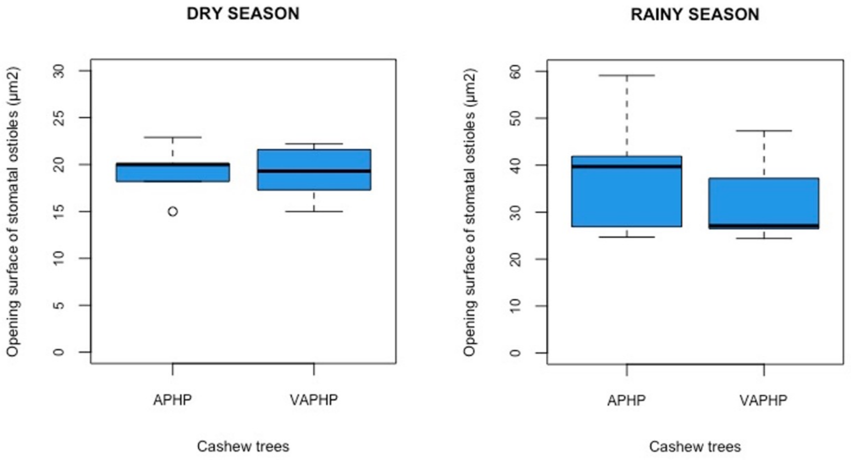

Figure 10. Variation in the surface area of cashew tree stomata ostioles depends on the seasons. For each locality of origin of the cashew trees, the letter V is used to distinguish the non-APHPs from the APHPs.

Slika 10. Razlike v površini ostiol listnih rež indijskega drevesa glede na letni čas. Za vsako lokacijo izvora indijskih dreves je uporabljena črka V za razlikovanje med drevesi, ki niso APHP, in drevesi, ki niso APHP.

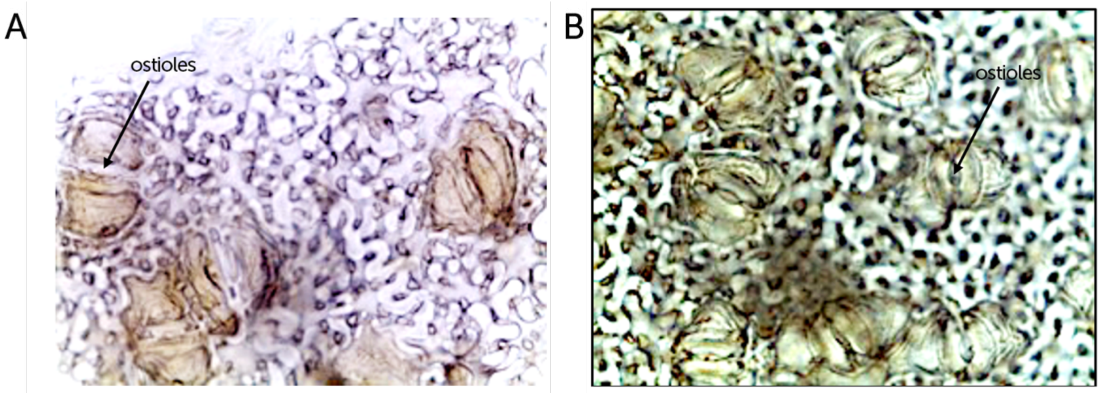

Figure 11. Surface areas of stomatal ostioles during the rainy season. A: APHP; B: non-APHP (100× magnification).

Slika 11. Površine stomatalnih ostiol v deževnem obdobju. A: APHP; B: brez APHP (100-kratna povečava).

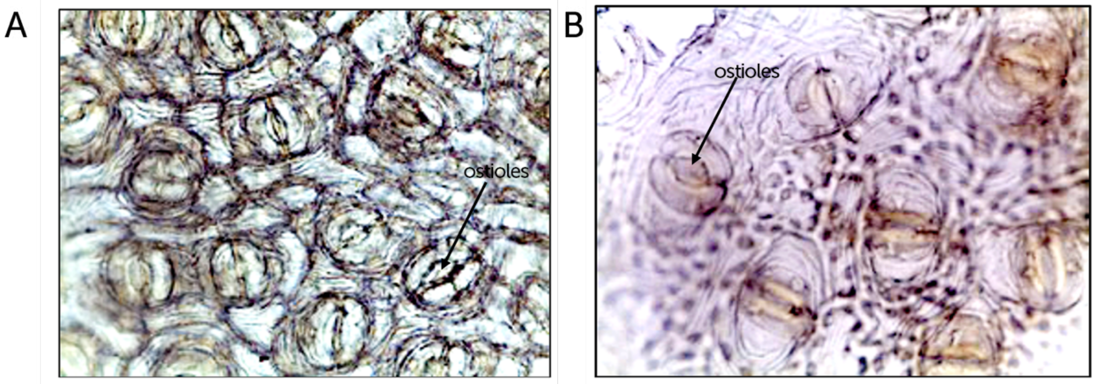

Figure 12. Surface areas of stomatal ostioles during the dry season. A: APHP; B: non-APHP (100× magnification).

Slika 12. Površina stomatalnih ostiolov v sušnem obdobju. A: APHP; B: brez APHP (povečava 100×).

Root tissues

Xylems

The root xylems were characterized by both their vessel number and diameter.

The APHPs' root xylem vessel number varied between 11 and 26 vessels/mm2, while that of the non-APHPs varied between 15 and 42 vessels/mm2 (Figure 13). ANOVA did not show (P = 0.051) any impact of the season on the number of xylem vessels in both APHP and non-APHP roots.

The largest root xylem radial diameters, set between 16.62-18.62 µm and tangential diameters set between 13.02-16.92 µm, were found during the rainy season (Figure 14), while the smallest radial diameters, about 4.88 µm and tangential diameters about 3.48 µm were found during the dry season (Figure 15). However, no impact of the season (P = 0.263) was found on the root xylem vessel diameter of both APHPs and non-APHP cashew trees.

No correlation (P > 0.380) was found between the number of xylem vessels and the ratio of the tangential and radial diameters of the vessels of the roots of both APHPs and non-APHPs.

For each locality of origin of the cashew tree, the letter V is used to distinguish the non-APHPs from the APHPs.

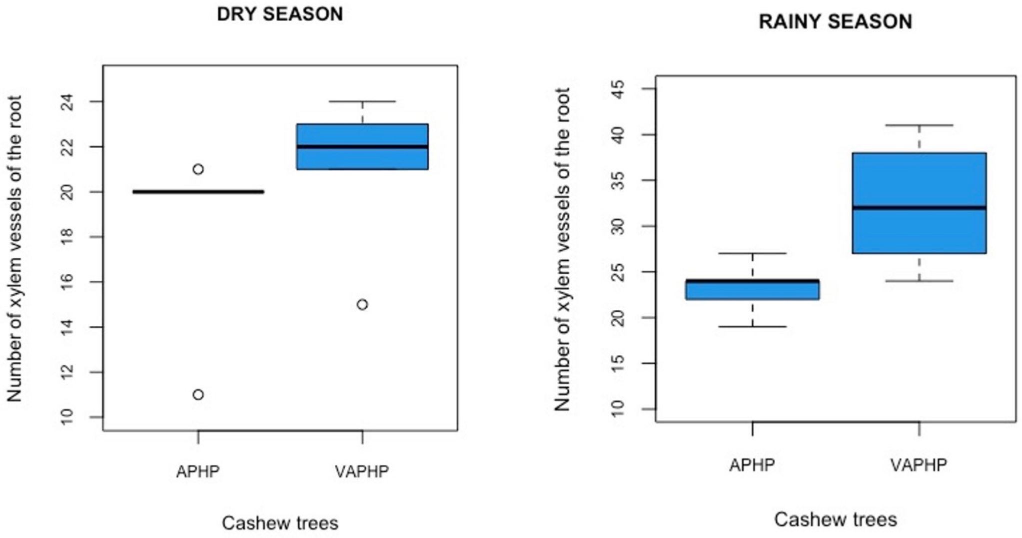

Figure 13. The number of xylem vessels in the cashew tree roots according to the cashew categories and the season.

Slika 13. Število ksilemskih žil v koreninah indijskega drevesa glede na kategorije indijskega oreščka in sezono.

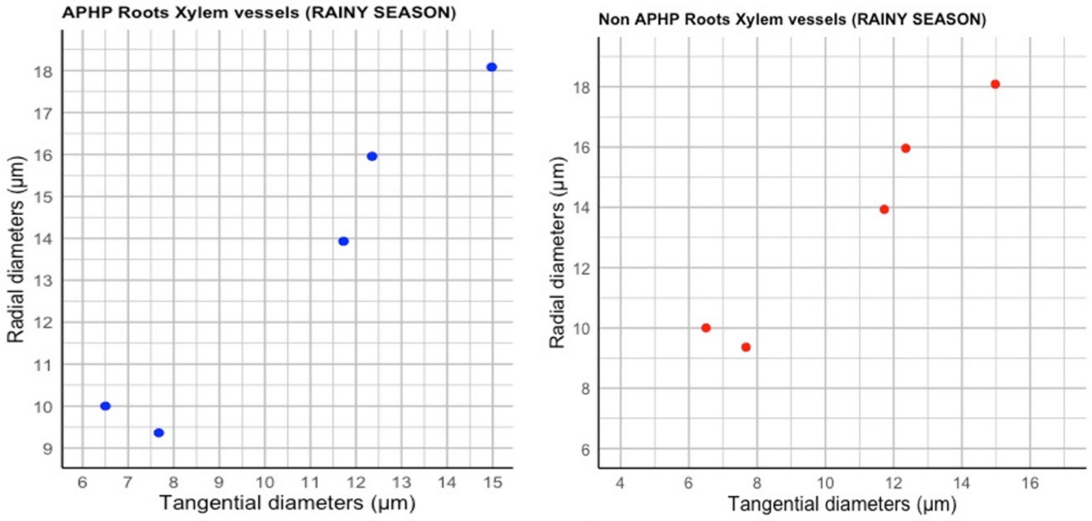

Figure 14. Tangential and radial diameters of the xylem vessels of cashew trees during the rainy season.

Slika 14. Tangencialni in radialni premeri ksilemskih žil indijskih dreves med deževno sezono.

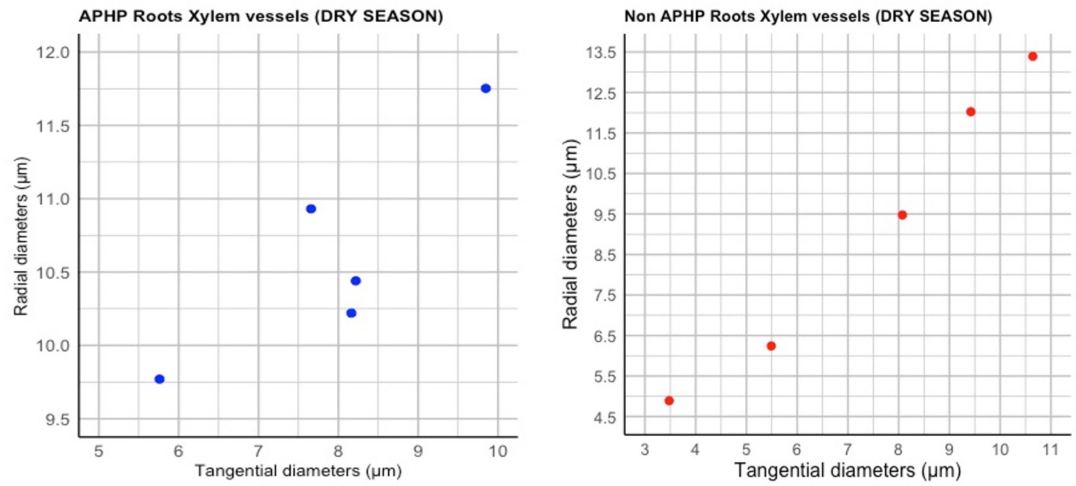

Figure 15. Tangential and radial diameters of the xylem vessels of cashew trees during the dry season.

Slika 15. Tangencialni in radialni premeri ksilemskih žil indijskih dreves v sušnem obdobju.

Medullar parenchyma

The average thickness of the roots medullary parenchyma increased with water deficit (Figure 16). The APHPs and non-APHPs experienced average medullary parenchyma thicknesses of 70 µm and 62 µm, respectively, during the dry season (Figure 16), while these values were about 44 µm for the APHPs and 20 µm for the non-APHP (Figure 17). Independently of the season, the APHPs showed a larger (P = 0.003) average medullary parenchyma thickness of roots medullar parenchyma than the non-APHPs (Figures 16 and 17).

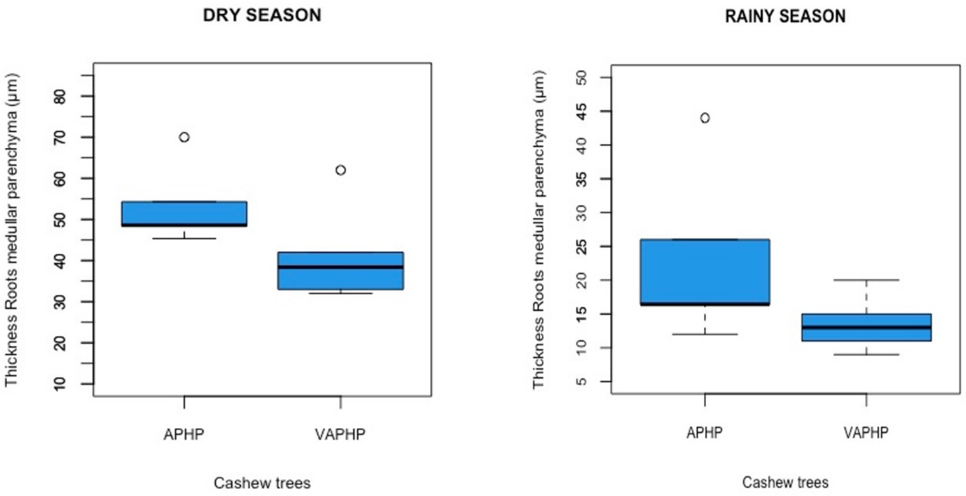

Figure 16. Thickness of the medullary parenchyma of the cashew tree roots. The letter V is used to distinguish the non-APHPs from the APHPs.

Slika 16. Debelina medularnega parenhima korenin indijskega drevesa. Črka V je uporabljena za razlikovanje med ne-APHP in APHP.

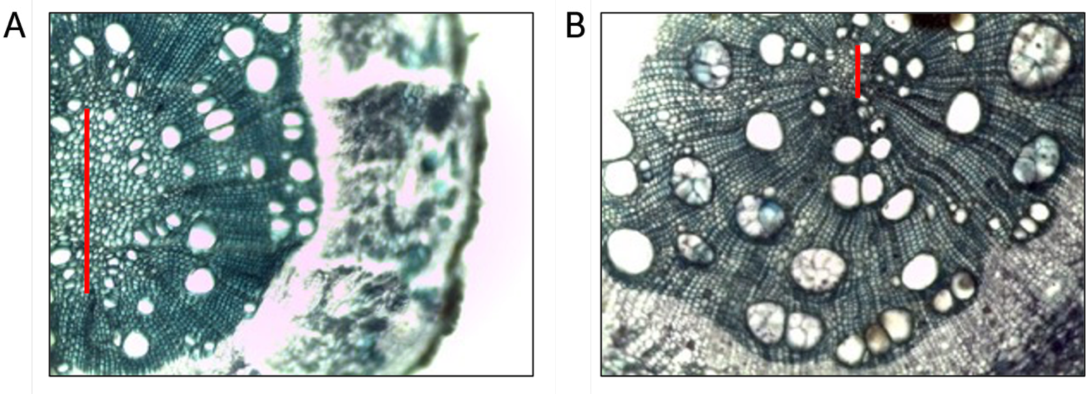

Figure 17. Roots medullary parenchyma width during the dry season (A) and the rainy season (B).

Slika 17. Širina medularnega parenhima korenin v sušnem obdobju (A) in deževnem obdobju (B).

Discussion

Plants exposed to water deficits undergo biochemical changes as well as various anatomical adaptations, enabling them to grow under stress conditions (Saeed et al. 2016). In this study, water deficit conditions during the dry season significantly increased the cortical parenchyma of the main vein from 18.5 ± 4.5 µm to 22 ± 3.4 µm. This may indicate that cashew trees adapt their anatomical structure to water-limited environments. As Boughalleb et al. (2014) demonstrated in another study on *Astragalus gombo* Coss. & Durieu ex Bunge subsp. *gomboeformis* (Pomel) Ott, various anatomical adaptations, including cortical and mesophyll parenchyma formation, occur in response to limited water availability. These features are suggested to contribute to maintaining water potential and storing energy during drought, which effectively enhances plant survival in arid environments.

A significant increase was also observed in leaf anatomical characteristics, such as the thickness of the palisade parenchyma on both surfaces (upper surface: 54 ± 4.2 µm to 59 ± 5.5 µm, lower surface: 20 µm ± 2.9 µm to 28 ± 2.1 µm) and in the thickness of the medullary parenchyma (from 4300 ± 837.7 µm^2 to 5800 ± 412.7 µm^2) of the main vein during the dry season. These findings do not align with Akram et al. (2016), who reported that water stress in radish cultivars significantly reduced the main vein parenchyma thickness. Previous studies have shown that water stress can considerably reduce leaf mesophyll thickness and main vein thickness in *Triticum aestivum* L. (Burnett et al. 2005) and *Ctenanthe setosa* (Roscoe) Eichler (Kutlu et al. 2009). Similarly, Bosabalidis and Kofidis (2002) demonstrated that severe stress negatively affects mesophyll and palisade parenchyma thickness. Our findings also revealed a decrease in cashew lacunar parenchyma, regardless of production potential (Figure 5). Similarly, Fernández et al. (1997) found that water deficit in *Olea europaea* L. reduces mesophyll intercellular spaces, causing morphological and anatomical modifications in leaves.

The reduction in epidermal cell thickness is an adaptive strategy that contributes to leaf resistance against cellular collapse due to drought and controls transpiration at the cuticle level (Bosabalidis and Kofidis 2002). However, in the cashew leaves, the epidermal cell size increased from 5.6 ± 0.4 µm during the rainy season to 6.2 ± 1.0 µm during the dry season, thus reducing water loss as stomatal density increased (Figure 9).

Ennajeh et al. (2010) also observed that the lacunar parenchyma thickness is higher under water deficit. This increased thickness during the dry season in olives is attributed to an enlargement of intercellular spaces, which enhances CO_2 diffusion.

The larger thickness of the xylem in the main leaf vein during the rainy season (6.7 ± 0.9 µm) than in the dry season (5.9 ± 0.6 µm) can likely be due to greater water availability in the soil. This water availability increases hydraulic conductivity, which increases xylem thickness and vascular bundle number. Similar results were found in roots, with both vessel number (19.7 ± 3.9 µm to 27.8 ± 7.1 µm) and vessel thickness (radial diameter: 16.6-18.6 µm to 4.9 µm; tangential diameter: 13.0-16.9 µm to 3.5 µm) increasing during the rainy season. This strategy is also adopted by grapevine cultivars, which respond to water limitations by adopting specific hydraulic strategies (Lovisolo et al. 2010; Dal Santo et al. 2016). Moreover, the formation of smaller vessels under drought conditions has been observed in the stems of various tree species, such as *Quercus* L. and *Populus* L. (Arend and Fromm 2007, Fonti et al. 2013). Small vessels are suggested to be less vulnerable to xylem embolism induced by drought and contribute to water flow regulation under water-limited growth conditions. Additionally, the anatomical response to water limitation in grapevines includes adaptation through modification of xylem traits (xylem conduit size and number). Conversely, the increase in xylem vessel number and thickness enhances cavitation tolerance, conferring drought resistance (Arend and Fromm 2007). Smaller vessel diameters facilitate hydraulic conductance, reduce cavitation, and make water and nutrient uptake more efficient (Queiroz-Voltan et al., 2014). Under water stress, smaller xylem vessels promote efficient and safer water transport, improving hydraulic conductivity (Batista et al. 2010). In contrast, larger xylem vessels are more common in areas with higher rainfall for a given species (Mauseth 1988). The olive tree, a drought-resistant species, reduces xylem vessel diameter under water stress, allowing the plant to maintain transpiration flux (Ben et al. 2007).

Our findings indicate that stomatal density in cashew trees ranges from 125 to 259 stomata per mm^2 (Figure 9). These results align with Esau (1965), who reported a density range of 100-300 stomata per mm^2 in numerous plant species. Stomata are denser during the dry season (259 stomata/mm^2) than the rainy season (176 stomata/mm^2). The stomatal aperture is smaller in the dry season (23 µm) than in the rainy season (59.3 µm), consistent with Ma (2010), who found that increased osmotic pressure in guard cell vacuoles, following high solute absorption, induces stomatal pore opening. Raven et al. (2013) observed that under hot, dry conditions, plants enter water stress, triggering the production of abscisic acid, a regulatory hormone secreted by leaves and roots that participates in various physiological regulation mechanisms. Under drought conditions, abscisic acid synthesis increases in the roots, where it is transported to the leaves to initiate prolonged stomatal closure (William 1995). Abscisic acid binds to its receptor in the plasma membrane, causing a calcium-mediated phosphorylation cascade that releases ions and leads to guard cell turgor loss, reducing stomatal opening. Thus, decreased stomatal aperture during osmotic stress is the most pronounced response (Munns and Tester 2008).

In several fruit tree species, the response to water deficit involves reducing stomatal area, as seen in pistachio trees (Abbaspour et al. 2012), other Mediterranean species (Ben Ahmed et al. 2008), and the *Olea europaea* L. as found in Guerfel et al. (2009). This mechanism limits CO_2 assimilation during water deficits. Fernández et al. (1997) also observed that reduced CO_2 assimilation may decrease leaf mesophyll conductance.

Moreover, the highest stomatal densities were observed in cashew trees during the dry season. Grisi et al. (2008) reported similar results for irrigated and non-irrigated coffee plants, with non-irrigated plants showing higher stomatal density. Similarly, Batista et al. (2010) showed that higher stomatal densities observed under water deficit in coffee plants indicate greater drought tolerance. Plants, being sessile, must adapt to various environmental factors, with stomata playing a crucial role in this function (Casson and Gray 2008). This supports the existence of drought resistance characteristics in non-irrigated coffee plants with higher stomatal density.

Conclusion

The conditions impacted leaf and root tissues. The thickness of the palisade parenchyma of non-APHPs remained constant, while that of APHPs increased considerably from the rainy season to the dry season. The two types of trees had the same reaction to water deficit, which was the increase in upper and lower epidermis thickness in order to reduce water loss during the dry season. During the dry season, the thickness of the xylem decreased, the average thickness of the cortical parenchyma of the lower midrib increased, and the average thickness of the cortical parenchyma of the lower midrib decreased, the thickness of the intercellular spaces of the lacunous parenchyma decreased, and the thickness of the medullary parenchyma of the midrib of the leaves of APHPs and their neighbours increased. The stomatal density increased during the dry season in all the cashew trees. However, the opening surface of the stomatal ostioles decreased during the dry season. These results showed the different anatomical adaptation strategies of the cashew tree to water constraints.

Authors' contributions

Conceptualization, K.A.R.F., K.M.L., and K.N.F.; methodology, K.A.R.F., K.M.L., and K.N.F.; software, K.A.R.F.; validation, K.M.L., and K.N.F.; formal analysis, K.A.R.F.; investigation, K.A.R.F. ; resources, K.A.R.F. ; data curation, K.A.R.F. ; writing—original draft preparation, K.A.R.F. ; writing—review, and editing, K.M.L., and K.N.F.; visualization, K.A.R.F. , and K.M.L.; supervision, K.N.F.; project administration, K.N.F.; funding acquisition, K.N.F.. All authors have read, and agreed to the published version of the manuscript.

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Data availability

The authors confirm that the datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Conflicts of interest

No potential conflict of interest was reported by the authors.

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