GENETICS AND PLANT BREEDING

# Morpho-anatomical characterization, gene expression and protein cell wall modifications associated with natural finger drop in bananas

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**ABSTRACT.** Banana (*Musa* spp.) is one of the main fruits consumed worldwide. However, finger drop, is a physiological disorder that causes many postharvest problems, which eventually reduces market value and consumer acceptance. Therefore, the objective of the study was to evaluate the anatomical changes that occur in the pedicel rupture area (drop zone) of bananas diploids (BB França) and tetraploid (BRS Pioneira) in three ripening stages. The levels of gene expression involved in the natural ripening process and in the development of finger drop, was also investigated. The accumulation of their mRNAs and those of expansin (*EXP1*), pectate lyase (*PEL1*) and xyloglucan endotransglucosylase/hydrolase protein (*XTH4*) genes already isolated from bananas were measured by quantitative polymerase chain reaction in three ripening stages. BB França presented a higher resistance to finger drop due to the presence of some specific morphoanatomical characteristics, such as larger parenchymal cells and greater deposition of lignin. In contrast, there was degeneration of the provide parenchymal tissue of the BRS Pioneira genotype, forming large empty spaces during the ripening of the fruits, mainly in stage 6, which contributed to the finger drop. The diploid BB França is a strong candidate for use in banana breeding programs aimed at fruit drop resistance. This will certainly improve the quality of banana varieties. Moreover, *PEL1* proved to be an excellent candidate gene for functional studies of finger drop in bananas.

Keywords: Musaceae; cell wall; fruit drop; anatomical analysis; RT-qPCR.

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# Introduction

Banana and plantains are herbaceous plants belonging to the *Musaceae* family. These perennial monocotyledons are considered the most popular fruit in the world because of their attractive flavor and nutritional value (Wang et al., 2019; Busche, Pucker, Viehöver, Weisshaar, & Stracke, 2020; Dou et al., 2020). These fruits have played significant economic, social, and ecological roles in many tropical and subtropical countries as a source of food, fiber, and fruit for millions of people (Naim et al., 2018; Cenci et al., 2019; Tan et al., 2020).

Finger drop is a physiological disorder mainly associated with ripening that deteriorates the quality of bananas (Hubert, Piral, Galas, Baurens, & Mbéguié-A-Mbéguié, 2014). This phenomenon has a significant economic impact on the banana marketing sector reducing its value and acceptance by consumers, especially because bananas are sold in bunches or bouquets (Rodrigues, Amorim, Ferreira, Ledo, & Santana, 2017). In *Musa*, this disorder is not caused by the development of an abscission zone, as what occurs in most fruits, but by weakening and softening of the pedicel, leading to early detachment of individual fruits and/or their separation from the bunch (Imsabai, Saichol, & Doorn, 2006; Behera & Neog, 2020). Therefore, this condition becomes of great interest to be improved, as it is a limiting factor in the release of new banana varieties.

Studies published in the literature show that the sensitivity to fruit detachment in banana germplasm may vary according to the variety, ploidy level, and the type of genome. Hicks (1934) was the first researcher to evaluate the finger drop condition in triploid Cavendish bananas (AAA). Recent research has also demonstrated this physiological disorder in tetraploids, which are generally more susceptible, *i.e.*, "Prata

Graúda" (Pomme subgroup, genome group AAB), the triploid "Terra" (plantain, genome group AAB) that is resistant and "Prata-Anã" (Pomme subgroup, AAB) with moderate resistance to finger drop (Ruiz, Salomão, Siqueira, Rezende, & Lins, 2016). Previous studies report that the presence of the B genome confers greater resistance to fruit finger drop and may indicate that these alleles can be associated with the *Musa balbisiana* Colla species (Rodrigues et al., 2017).

In ripe banana fruits, pulp softening is considered an important condition restricting the shelf life and postharvest quality of the fruits. This process involves physiological changes in the polysaccharide structure in the cell wall matrix that is associated with the activity of cell wall modifying enzymes, such as polygalacturonase (PG), pectinmethylesterase (PME), pectate lyase (PL), xyloglucan endotransglycosylase (XET), and expansin (EXP) (Tucker et al., 2017). Previous research involving the rupture of the fruits occurring during the ripening in cultivars susceptible to finger drop may be related to the activity of a number of cell wall hydrolases (Imsabai et al., 2006; Saengpook, Ketsam, & Doorn Van, 2007).

To date, most of the studies on anatomical changes of the texture and firmness of banana fruits that occur during ripening have been focused merely on the Cavendish subgroup, AAA bananas (Amnuaysin, Seraypheap, & Kidyoo, 2012b; Brat et al., 2016; Ramírez-Sánchez, Huber, Vallejos, & Kelley, 2018). Meanwhile, the relationship between finger drop and the anatomy of the pedicel zone observed in other banana ploidies has been less frequently studied. Therefore, this study was performed to help fill this gap, seeking to better elucidate the cellular changes associated with this physiological disorder.

Gene expression studies in banana also play important roles in the regulation of banana fruit ripening and finger drop in *Musa* genotypes (Xiao et al., 2018; Yan et al., 2019; Shan et al., 2020; Song, Shan, Kuang, Chen, & Lu, 2020). Here, we mitigate the relative gene expression of cell wall-modifying genes pectate lyase 1 (*PEL1*), expansin 1 (*EXP1*), and xyloglucan endotransglucosylase/hydrolase protein 4 (*XTH4*), measured at the finger drop zone (DZ) and the control zone (CZ) in bananas with different ploidy levels and contrasting levels of finger drop resistance. To date, few studies have been carried out to investigate the behavior of banana genotypes, mainly addressing the anatomical aspects and analysis of gene expression to elucidate this physiological disorder.

Thus, the objective of the study was to evaluate the anatomical changes that occur in the pedicel rupture area (drop zone) of wild diploid banana (BB França, BB - resistant to finger drop) and the tetraploid (BRS Pioneira, AAAB - susceptible to finger drop) from three ripening stages, as well as to investigate the levels of gene expression involved in the natural ripening and in the finger drop processes. These findings provide a basis for establishing breeding strategies for the development of new banana cultivars with improved characteristics aiming the development of varieties resistant to finger drop.

# Material and methods

# **Plant material**

In our study two banana genotypes *i.e.*, BB França (wild diploid, BB), and BRS Pioneira (hybrid developed by Embrapa, AAAB), were evaluated. These were taken from the germplasm bank of Embrapa Mandioca e Fruticultura, located in the municipality of Cruz das Almas, Bahia, Brazil (12°48′19″ S, 39°06′23″ W and altitude of 225 m). These genotypes were selected for their varying responses to finger drop, as described in Table 1.

Accession name	Species or subgroup	Status	Ploidy	Finger drop*
BB França	Musa balbisiana (BB)	Wild diploid	2x = 22	Resistant
<b>BRS</b> Pioneira	AAAB	Hybrid	2x = 44	Susceptible

 Table 1. Description of the genotypes used in this study.

\*Pereira et al. (2004).

### **Anatomical studies**

Anatomical analyses were carried out at the Plant Anatomy and Histochemistry Laboratory at the *Universidade Federal do Recôncavo da Bahia*, located in Cruz das Almas, Bahia State, Brazil. For this study, segments were cut from the pedicel rupture area (drop zone - DZ) and collected in three stages of ripening: stage 4 (more yellow than green fruit), 5 (yellow fruit with green extremity), and 6 (completely yellow fruit), according Von Loesecke's (1949) scale (Figure 1A-D).



**Figure 1.** Visual aspect of BRS Pioneira banana fruits after finger drop and collected at different stages of ripening according Von Loesecke's (1949) scale. A) Pedicel region of the banana where finger drop occurs (DZ) and the control zone (CZ). B) Fruits of BRS Pioneira collected in stage 4. C) Fruits in stage 5. D) Fruits in stage 6.

The segments (n = 3) of each genotype and taken from different plants were fixed in FAA<sub>50</sub> solution (formalin, acetic acid, and 50% ethyl alcohol; 0.5: 0.5: 9 v/v) for 24 hours and then preserved in 70% ethanol, as described by Johansen (1940). After this period, the samples were dehydrated in an increasing butyl series and then mounted on histological paraffin blocks. Subsequently, serial transverse and longitudinal sections (14 µm) were cut with a Leica 2245 rotary microtome (Leica, Nussloch, Germany). Afterwards, the cuts were mounted ontoslides, and stained with 1.5 % alcoholic safranin and 1% aqueous astra blue (Gerlarch, 1969). The sections were then analyzed and photographed with an Olympus BX51 photomicroscope coupled to an Olympus A330 digital camera (Olympus, Tokyo, Japan).

The evaluated characteristics were: distribution and color of the components of the tissues from the region where the natural detachment of the fruit occurs. The tissues were digested in a solution of hydrogen peroxide and acetic acid (1: 1 v/v). After digestion, the material was washed in distilled water and preserved in 50% alcohol. The sections were stained with a 1% solution of safranin in 50% ethanol for 24 hours (Franklin, 1945). Afterwards the material was washed three times with 30% ethyl alcohol and mounted on slides with glycerin.

The obtained images were used for measurements of the cortex size (CS) in mm, fiber length (FL) in  $\mu$ m, fiber thickness (FT) in  $\mu$ m, number of fibers (NF) per mm<sup>2</sup>, number of laticifers (NL) per mm<sup>2</sup>, and fiber area (FA). The frequency of the fibers and laticifers represent the number of cells per mm<sup>2</sup> obtained using the ImageJ 1.46r software (Rasband, 1997-2016). For each characteristic, 9 sections for each pedicel segment were used, for a total of 27 units.

## Total RNA extraction and cDNA synthesis

Total RNA was isolated from a sample of tissue in the middle region (CZ) of the fruit and also of the area where the detachment of the pedicel occurs (DZ) from the banana fruits. These were collected in three ripening stages (Figure 1), according to the protocol suggested by Gambino, Perrone, and Gribaudo (2008). RNA quality and concentration were assessed by gel electrophoresis with the use of a NanoDrop ND-2000 spectrophotometer (Thermo Scientific, Waltham, MA, USA). To eliminate contaminating genomic DNA, the samples were treated with DNase (DNA TURBOfree - Ambion). cDNA synthesis was performed using total RNA treated with DNase, in accordance with the manufacturer's recommendations using the *High-Capacity RNA-to-cDNA* kit (Applied Biosystems).

## Gene expression analysis by quantitative real-time PCR

Table 2 lists the pairs of primers used in studies aimed at quantifying the expression of genes related to the ripening of fruits and the occurrence of finger drop in *Musa* spp. (Pua, Ong, Liu, & Liu, 2001; Trivedi & Nath, 2004; Mbéguié-A-Mbéguié et al., 2009), as well as the endogenous reference genes for *Musa* spp. according to Podevin, Krauss, Henry, Swennen, and Remy (2012).

Gene	Primer	Sequence (5'-3')	Annealing	Product	Reference
	name		temperature (°C)	size (bp)	
Elongation factor*	EF1-MU	F: CGGAGCGTGAAAGAGGAAT	60	185	Podevin et al. (2012)
		R: ACCAGCTTCAAAACCACCAG			
Pectate lyase	PEL1	F: TGATCATTTCTCTTCTTTCACG	60	153	Pua et al. (2001)
		R:TCCCAAGTCAAGTAGTATCAACACA			
Expansin	EXP1	F:GGTGGAGGCATTCGGTCTGGTT	60	221	Trivedi and Nath (2004)
		R:GGGAGGTGACACGATGAGAAGATG			
Xyloglucan	XTH4	F: CGACTGATGGCTGCTGGAT	60	101	Mbéguié-A-Mbéguié et
endotransferase		R:TCCATCTTTTACATACAAAACGGAACT			al. (2009)

**Table 2.** Sequences of gene-specific primers used in this study seeking to quantify the expression of genes related to the ripening of fruits and the occurrence of finger drop in banana. Each assay using the gene-specific primers amplified a single product of the expected size (bp).

\*Endogenous reference gene for Musa according to Podevin et al. (2012).

The RT-qPCR analysis was performed using the ABI 7500 Fast Real Time PCR-System (Applied Biosystems, Foster City, CA, USA) using SYBR Green. Each reaction was performed in a final volume of 9.3  $\mu$ L, containing 2  $\mu$ L (100 ng) of a pair of primers (0.4  $\mu$ L of *forward primer* + *reverse* in a concentration of 10  $\mu$ M) and 5  $\mu$ L of Platinum<sup>®</sup> SYBR<sup>®</sup> Green qPCR Super Mix-UDG w/ROX kit (Invitrogen, Carlsbad, CA, USA) according to the manufacturer's recommendations and 2.3  $\mu$ L of nuclease free water.

The following program was used for the RT-qPCR thermocycling conditions:  $50^{\circ}$ C for 2 min.,  $95^{\circ}$ C for 10 min. followed by 40 cycles of  $95^{\circ}$ C for 15 second, and  $58^{\circ}$ C for 1 min. To generate a lower cycle threshold (Ct), a test was performed to verify the cDNA concentration. A concentration test of the primers was also done to generate a lower Ct and higher  $\Delta$ Rn, with no dimers. Before evaluating the expression profile of the gene a test of the efficiency of the assay was performed from the serial dilutions with five dilutions and three replicates, and the evaluation carried out by the slope indication of the standard curve and R<sup>2</sup>. The efficiency (R<sup>2</sup>) ranges were 99-100% (0.99), 88-100% (0.98), 87-100% (0.99), and 90–102% (0.98) for elongation factor (*EF1-MU*), expansin (*EXP1*), pectate liase (*PEL1*), and xyloglucan endotransferase (*XTH4*) genes, respectively.

After the reaction, data were collected and stored in Software 7500 version 2.0.5. The results were normalized using DataAssist<sup>TM</sup> software v.3.01 (Life Technology). The *EF1-MU* gene was used as the internal control to standardize the difference between template amounts while fruit tissues taken at harvest before ripening induction was used as the calibrator. The experiments were repeated with three biological and three technical replicates for the control zone and finger drop zone. The relative fold differences in the expression of each gene among samples were determined using the  $2^{-\Delta\Delta CT}$  formula (Livak & Schmittgen, 2001).

### Statistical analyses

The anatomical variables were subjected to analysis of variance (ANOVA) by the F-test ( $p \le 0.05$  or  $p \le 0.01$ ) with subsequent comparison of the means using F test at 5% probability. All the anatomical analyses were performed with the "agricolae" package implemented in the R software (R Development Core Team, 2020). Furthermore, the ExpressionSuite software, version 1.0.3, was used for the interpretation of the gene expression analysis, which performs comparative quantification by the "Pair-Wise Fixed Reallocation Randomization Test" method (Pfaffl, Horgan, & Dempfle, 2002).

# **Results and discussion**

## Anatomical characterization of the pedicel during the finger drop at different stages of maturation

Based on the microscopic analysis of the cross sections the banana pedicel has the typical anatomical structure of monocotyledon plants presenting a wide parenchymal region with scattered vascular bundles. This structure characterizes an atactostele type of distribution, with the occurrence of non-articulated anastomosing laticifers distributed throughout the organ. Cross-sections and and longitudinal sections of the pedicel drop zone (DZ) of the bananas collected at different maturation stages showed some anatomical modifications in the cellular structure (Figure 2).

In the present study BB França, which is resistant to finger drop, presented a greater number of laticifer cells, an increase in the size of the parenchymal cells, and also a higher degree of lignification in the last stage of maturation (S6). This is due to a greater affinity of these tissues with the red dye safranin staining cell walls (Figure 2A-F).

#### Anatomical characterization and gene expression in banana

Previous studies show that the degeneration of the parenchymal tissue in the pedicel region of ripe banana fruits favors finger drop because of the large empty spaces that are formed in this region (Putra, Zacaria, Abdullah, & Saleh, 2010). These results were consistent with those obtained in our study (Figures 2G, I, K), especially because we also observed a degeneration of the pedicel parenchymal tissue of the BRS Pioneira genotype, forming large empty spaces (arrow) during the ripening of the fruits, mainly in stage 6 (Figure 2K). This justifies the high susceptibility of this genotype to finger drop.

In our study, the distribution and the types of vascular tissues varied in the pedicel region of the genotypes. Four types of vascular fibers were observed: fiber bundle, side bundles, vascular bundle with incomplete esclerenchymatic sheath, and vascular bundle with complete esclerenchymatic sheath (Table 3). The BB França genotype, resistant to finger drop, stood out from BRS Pioneira because the sheath of esclerenchyma fibers was formed by up to four layers of fibers and was heavily stained red by safranin, indicating the presence of lignin. This finding was more evident in stages 5 and 6 of maturation (Figure 2C-F).

In contrast, in the genotype BRS Pioneira, susceptible to finger drop, only the vascular bundles were stained with astra blue (Figure 2K). Here, the astra blue dye associated with safranin made it possible to distinguish cellulosic cell walls (primary) from lignified ones (secondary). Observations from other authors reveal the efficiency of the staining methods with Safranin-Astra Blue in distinguishing the primary cellulose cell walls (blue) from the lignified secondary xylem (red) (Ployet et al., 2017; Crespo-Martíne, Sobczak, Różańska, Forneck, & Griesser, 2019; Sebastian-Azcona, Hacke, & Hamann, 2020). These results highlight the fact that it is probably the variations of the vascular bundles among the two analyzed genotypes which may be related to mechanisms of resistance to finger drop. Kheng, Dinga, and Rahmanb (2011) also observed anatomical changes with fully ripe fruits, such as a reduction in peel thickness, an increase in the intercellular spaces, and less vascular bundle tissues. These features seem to be more evident in genotypes that are susceptible to finger drop.



Figure 2. Anatomical structure (cross-section and longitudinal sections) of the banana pedicel region where finger drop (DZ) occurs at different stages of ripening and stained with safranin and astra blue. A-F) BB França showing large numbers of laticifer cells, mainly in stage 6 of maturation, and also greater lignification of the parenchymatic tissues stained red by safranin. G-L) Showing greater degeneration of the parenchymatic tissue of the pedicel in BRS Pioneira, forming large empty spaces (arrow) during fruit ripening, especially in stage 6. pa: parenchyma, sc: sclerenchyma, vb: vascular bundle, la: laticifer cells, xy: xylem, ph: phloem, bd: bundle group. Bar: 200 µm (A-G, I); 500 µm (H, J-L).

Genotypes	Types of fibers	Local of occurrence	Finger drop	
BB França (BB)	Fiber bundle	Cortex		
	Collateral fibers	Peripheral region	Resistant	
	Vascular bundle with complete sclerenchyma sheath	Center		
BRS Pioneira (AAAB)	Fiber Bundle	Cortex		
	Collateral fibers	Peripheral region	Sussentible	
	Vascular bundle with incomplete sclerenchymal sheath	Center	Susceptible	
	Vascular bundle with complete sclerenchyma sheath	Center		

**Table 3.** Types of fibers and location of occurrence observed in the pedicel drop zone of banana genotypes.

From longitudinal sections, there was an abscission zone at the area of rupture in the genotypes evaluated. A probable explanation for this is that the rupture occurs due to the softening and weakening of the peel in the area where the fruit joins with the bunch (pedicel) causing the early individual detachment of the fruits. This fact corroborates the results from previous studies reported by other authors who also did not detect the formation of an abscission layer/region where the finger drop occurs (Imsabai et al., 2006; Imsabai & Ketsa, 2007; Putra et al., 2010). Here, it was possible to see that the genotypes BRS Pioneira, susceptible to finger drop, presented greater fragility of the fruit peel in the area where the break occurs.

The morphometric analysis of the pedicel drop zone from the fruits collected in stage 6 (S6) revealed significant ( $p \le 0.05$ ) responses of the banana genotypes in relation to the anatomical traits evaluated (number of lactifers, fiber area, cortex size, fiber length, fiber diameter and number of fibers), as showed in Figure 4. In this study, BB França presented the highest records for number of laticifers (5.38 mm<sup>2</sup>), fiber area (0.11 mm<sup>2</sup>), and cortex size (0.34 mm) (Figure 3A-C). In contrast, BRS Pioneira presented the highest values of fiber length (925.22 µm), fiber thickness (36.07 µm) and number of fibers (2.54 mm<sup>2</sup>) (Figure 3D-F).



**Figure 3.** Morphometric assessment of the anatomical characteristics of the pedicel drop zone of banan fruit in stage 6 of ripening. A) Number of laticifers. B) Fiber area. C) Cortex size. D) Fiber length. E) Fiber thickness. F) Number of fibers. Means followed by the same letter do not differ from each other by F test, p < 0.05.

Previous studies reported that the increase in the thickness of the fiber wall caused by the deposition of a secondary cell wall contributed to the strengthening of the peel (Amnuaysin et al., 2012b). There are numerous factors that can influence peel weakness, such as the peel thickness, fruit weight, water content of the peel, circumference of the area where the rupture occurred, and also the degradation of the pectin (Imsabai et al., 2006). However, in our study, the length, thickness and number of fibers does not seem to be related to premature fruit dropping. This is especially because BRS Pioneira presented the highest record for this variable, and is considered susceptible to this physiological disorder (Pereira et al., 2004).

#### Anatomical characterization and gene expression in banana

Therefore, based on the histological analysis of the pedicel region, the wild diploid BB França is a good candidate to be used in banana breeding crosses worldwide that are aimed at the development of banana cultivars with excellent fruit quality and resistance to fruit finger drop.

# Gene expression of the expansin (EXP1)

The expansin (*EXP1*) genes examined in our study were differentially expressed during banana fruit ripening in both the control (CZ) and drop (DZ) zones. The *EXP1* gene in BB França (resistant) was expressed less in the drop zone when compared to the control zone in all evaluated maturation stages (Figure 4A). Similar behavior was verified with BRS Pioneira (susceptible), since *EXP1* was less expressed in the DZ in the three stages of maturity (Figure 4B). This means that there was no correlation between EXP1 expression in the DZ and finger drop.

There is evidence that several ripening-associated genes in banana change their expressions during ripening (Jourda et al., 2016). For example, the expansins that are non-enzymatic cell wall proteins play an important role in fruit softening (Fan et al., 2016; Chatzopoulou et al., 2020). According to Mbéguié-A-Béguié et al. (2009), the *EXP1*, *EXP4*, and *EXP5* genes appeared to be the main candidates involved in the detachment of the cell wall that is related to finger drop in bananas, with *EXP1* as the main gene involved in this process.

### Gene expression of pectate lyase (PEL1)

The profile of the *PEL1* gene examined in our study was differentially expressed during ripening in both the CZ and DZ (Figures 4C-D). Pectate lyase (*PEL1*) catalyzes the pectin depolymerization, the main component of the cell wall. Here, the *PEL1* gene in BB França was less expressed in the DZ than in the CZ during the maturation stages (Figure 4C). Concerning the BRS Pioneira tetraploid that is susceptible to FD, we observed that the *PEL1* mRNA level was more expressed during the last two stages, especially the S5 stage (Figure 4D). This result is in accordance with the report by Amnuaysin, Jonesc, and Serayheap (2012a) who reported increased activity and gene expression levels of the pectate lyase (*PEL*) enzyme during the ripening of banana fruits. Low levels of expression and activity of PEL in the early stages of ripening, and its subsequent increase in the later stages of maturation, suggest that this gene may be associated with the degradation of pectin during fruit ripening.

Our findings of *PEL1* expression are in agreement with the degree of fruit dropping of the genotypes, i.e., all the genotypes expressed greater amounts of transcripts in comparison to the control zone, except for BB França, the resistant genotype, which confirmed its resistance due to low expression of transcripts in S6. This gene can, therefore, be an excellent candidate for functional studies regarding finger drop in bananas.



Figure 4. Relative expression of the *EXP1*, *PEL1*, and *XHT4* genes in samples of pedicel tissue from the finger drop zone (DZ) and the control zone (CZ) of BB França and BRS Pioneira genotypes at different maturity stages. A-B) *EXP1* gene. C-D) *PEL1* gene. E-F) *XTH4* gene. The measurements were normalized to the reference gene, EF1-MU. The bars indicate the standard error (±SE) evaluated from three biological replicates. The value for CZ has been set at 1.00. Means followed by the same letter do not differ from each other by F test, p ≤ 0.05.

# Gene expression of xyloglucan endotransglucosylase (XTH4)

Presented in Figure 4G-H is the profile of relative expression of *XTH4* in the tissue samples of the middle region of the peel (control area - CZ) and the area where the pedicel detachment occurs (drop zone - DZ) during the maturation stages: S4, S5, and S6 of all banana genotypes tested. In stage S6, it was observed for BB França that the *XTH4* gene was highly expressed in the drop zone compared to the control (Figure 4G). In contrast, in the genotype BRS Pioneira, there was greater expression of this gene in the DZ than in the CZ when banana fruits were collected in stage S5 (Figure 4H); nevertheless, these are expected results due to their susceptibility. Some authors have also demonstrated that the xyloglucan endotransglucosylase/hydrolase (XTH) proteins could play a role in the softening of several plants species, such as pineapple, strawberry, and banana fruits during ripening (Li et al., 2019; Yun et al., 2019; Witasari et al., 2019).

# Conclusion

The findings of the anatomical study revealed important information on the cell wall-associated metabolism occurring during banana ripening and the finger drop condition. BB França presented a higher resistance to finger drop due to the presence of some specific morphoanatomical characteristics, such as larger parenchymal cells and higher deposition of lignins. The genotype BRS Pioneira, susceptible to finger drop, showed anatomical changes such as a degeneration of parenchymal tissue, respectively, that can explain the greater fragility of the pedicel of these genotypes. The *PEL1* gene prove to be an excellent candidate gene for functional studies of banana finger drop and can be used to direct new strategies of the banana genetic breeding program aimed at producing fruits with resistance to this characteristic. This study also demonstrated that BB França was the most resistant banana accession to finger drop and therefore a strong candidate for use in breeding programs worldwide aimed at the development of bananas that are more resistant to finger drop. Our work is of great importance for subsidizing information to be used in banana genetic breeding programs that are aimed at fruit drop resistance. Consequently, it will better guide crosses from which banana genetic breeding programs globally can benefit, thereby, enabling a more sustainable banana production worldwide. Nevertheless, it is important to highlight the need to expand this study, exploring other genotypes as well as other ploidy levels, such as the Cavendish subgroup (AAA), which is commonly exported and also the Prata group (AAB), which is the variety most produced and consumed in the northeast region of Brazil.

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