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NAAMA JESSICA DE ASSIS MELO

REACTION OF PASSION FRUIT SPECIES TO *Fusarium oxysporum* f. sp. *passiflorae* AND QUALITY OF FRUIT UNDER DIFFERENT CROP SYSTEMS

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Tese apresentada ao Programa de Pós-Graduação em Fitotecnia da Universidade Federal Rural do Semi-Árido como requisito para obtenção do título de Doutor em Fitotecnia.

Linha de Pesquisa: Fitopatologia

Orientador: Prof. Dr. Rui Sales Júnior

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ABSTRACT

MELO, Naama Jessica de Assis. **Reaction of passion fruit species to *Fusarium oxysporum* f. sp. *passiflorae* and quality of fruit under different crop systems**. 2019. 68f. (Tese) Doutorado em Agronomia: Fitotecnia – Universidade Federal Rural do Semi-Árido/UFERSA, Mossoró, 2019.

Brazil is the main producer of yellow passion fruit (*Passiflora edulis*) in the world. One of the main diseases that reduce its production is fusariosis, caused by the fungus *Fusarium oxysporum* f. sp. *passiflorae* (FOP). The practice of grafting with resistant species is used to provide resistance to pests and diseases, such as fusariosis. One of these species is the *P. cincinnata*, used in much of the Northeastern region of Brazil. The objective of this work was to characterize the pathogenicity of different isolates of FOP in *P. edulis* and *P. cincinnata* in order to identify their potential for use in areas with a history of the disease and to verify postharvest quality of fruits of *P. edulis* collected inmature and mature in four areas with different forms of cultivation (conventional with and without grafting and organic with grafting). In the first experiment, thirteen isolates of the fungus were used and the inoculums produced at a concentration of 10^6 CFU mL^{-1}. The seedlings of *P. edulis* and *P. cincinnata* were produced in coconut fiber and, 45 days after sowing, the root system was then immersed for five minutes in the conidial suspension before being replanted in the 770 mL pots. In the experiment ten replicates were used, for each species, each isolate and for the control group. Seedlings were evaluated daily from the second day after inoculation (DAI) until 90 DAI. After, a second experiment was installed in a completely randomized design, in a 4 x 2 factorial scheme, with five replicates of four fruits each. The first factor was the different collection sites with their cultivation systems and the second factor was the maturity stage. The fruits were harvested, selected and submitted to physical, physical-chemical and chemical analysis, bioactive compounds and antioxidant activity. All isolates were pathogenic in both *Passiflora* species, however incidence, severity and mortality were higher in *P. edulis*. There was a statistically significant difference for the incubation period of the FOP 23 and 57 isolates, being superior for *P. edulis*. Fruit weight, length, width and firmness were superior in fruits from conventional without grafting system, however fruit yield was inferior. Soluble solids, pH, reducing sugars and total soluble sugars were superior in fruits from collection sites with the use of grafting. Bioactive compounds and antioxidant activity were similar between the fruits from different collection sites.

Keywords: Bioactive compounds; Fusariosis; *Passiflora cincinnata*; *Passiflora edulis*; Pathogenicity.

RESUMO

MELO, Naama Jessica de Assis. **Reação de espécies de maracujazeiro amarelo a *Fusarium oxysporum* f. sp. *passiflorae* e qualidade do fruto sob diferentes formas de cultivo**. 2019. 68f. (Tese) Doutorado em Agronomia: Fitotecnia – Universidade Federal Rural do Semi-Árido/UFERSA, Mossoró, 2019.

O Brasil é o principal produtor de maracujá amarelo (*Passiflora edulis*) no mundo. Uma das principais doenças é a fusariose, ocasionada pelo fungo *Fusarium oxysporum* f. sp. *passiflorae* (FOP). O uso do porta-enxerto com espécies resistentes é uma alternativa de controle para pragas e doenças. A espécie *P. cincinnata*, espécie selvagem de maracujá, tem sido bastante usada como porta-enxerto frente à FOP no Nordeste brasileiro. O objetivo deste trabalho foi caracterizar a patogenicidade de diferentes isolados de FOP em *P. edulis* e *P. cincinnata* a fim de identificar seu potencial para uso em áreas com histórico da doença, bem como verificar a qualidade pós-colheita de frutos de *P. edulis* coletados nos estádios de maturação verde e amarelo em quatro áreas com diferentes formas de cultivo (convencional com e sem enxertia e orgânico com enxertia). No primeiro experimento, treze isolados de FOP foram utilizados e os inóculos produzidos na concentração de 10^6 UFC mL^{-1}. As mudas de *P. edulis* e *P. cincinnata* foram produzidas em fibra de coco e, aos 45 dias após a semeadura, o sistema radicular foi imerso por cinco minutos em uma suspensão de conídios antes de serem replantadas em vasos de 770 mL. Foram utilizadas dez repetições, de cada espécie, para cada isolado e testemunha. As plantas foram avaliadas diariamente a partir do segundo dia após a inoculação (DAI) até o 90° DAI. Em seguida, um segundo experimento foi instalado em delineamento inteiramente casualizado com esquema fatorial 4 x 2, com cinco repetições de quatro frutos cada. O primeiro fator foi o lugar de coleta com seus respectivos sistemas de cultivo e o segundo, o estágio de maturação. Os frutos foram colhidos e submetidos à análises físicas, físico-químicas e químicas, compostos bioativos e atividade antioxidante. Todos os isolados foram patogênicos para ambas as espécies de *Passiflora*, no entanto, incidência e severidade foram maiores em *P. edulis*. Houve diferença estatisticamente significativa entre os isolados para o período de incubação dos isolados FOP 23 e 57, sendo superior para *P. edulis*. Peso do fruto, comprimento, largura e firmeza foram superiores nos frutos provenientes do sistema convencional sem enxertia, contudo o rendimento da polpa foi inferior. Sólidos solúveis, pH, açúcares redutores e açúcares solúveis totais foram superiores em frutos provenientes dos lugares de coleta com uso da enxertia. Compostos bioativos e atividade antioxidante foram similares entre os frutos de diferentes lugares de coleta.

Palavras-chave: Compostos bioativos; Fusariose; *Passiflora cincinnata*; *Passiflora edulis*; Patogenicidade.

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CHAPTER I

1 GENERAL INTRODUCTION

"Maracujá" is an indigenous name of Tupi origin that means "food in form of cuia". These are wild fruits appreciated by the natives that the first discoverers knew in the Americas (MELETTI, 2001).

The first known description was made in 1569 with the name *granadilla*, from a plant sent from America to Pope Paul V, who had it cultivated in Rome, as a divine revelation because of the individual morphology of its flower, suggesting correlation with the symbols of the Passion of Christ. A very mystical denomination of "passion flower" was originated, a popular name rarely used in Brazil, and also the scientific name of the genus: *Passiflora* (from the Latin *passio*: passion and *flos*: flower) (RUGGIERO et al., 1996).

Passion fruit belongs to the family Passifloraceae and to the genus *Passiflora*, which stands out as the most economically important. Brazil is considered the center of origin of approximately 139 known species, and also the largest center of genetic diversity of the genus (BERNACCI et al., 2013). The plant develops well in tropical and subtropical regions, where the climate is hot and humid. Altitude, temperature, relative humidity, luminosity and precipitation have an important influence on plant longevity and yield (ALMEIDA et al., 2015).

Brazil stands out in the international scene as the world's largest producer of yellow passion fruit (*Passiflora edulis* Sims f. *flavicarpa* Deg.), whose production in 2017 was in an area of 41,090 ha and a harvest of, approximately, 554,598 t of fruits. Among the producing regions, the Northeast region has the largest area of cultivation, and the State of Bahia is the largest producer of the fruit with 170,910 t in a harvested area of 16,283 ha. The State of Ceará has the second largest harvested area (5,497 ha), with a production of 94,816 t. The State of Rio Grande do Norte is in 5th place in the national ranking, with a production of 29,182 t in a harvested area of 2,551 ha (IBGE, 2017).

Passion fruit is a plant with a tropical and subtropical climate. It can be cultivated, under ideal conditions, in regions with average temperatures between 25 and 26 ºC and rainfall between 1,200 and 1,400 mm, well distributed throughout the year.

The most recommended soils for the crop are arenaceous, deep, fertile, well drained, with flat to slightly wavy topography, with pH between 5.0 and 6.5. The recommended altitude of the crop is between 100 and 1000 m, the relative humidity of the air should be low and the luminosity high. The plant needs 11 hours of light/day to flow in order to produce fruits with good appearance, taste and aroma (ALMEIDA et al., 2015).

The advance in the passion fruit culture is related to the improvement in the crops, due to the technological advance as progress in the production system, improvement in the genetics of cultivated materials that provided greater gains in productivity and agronomic performance (FALEIRO et al., 2011). One of the techniques that has gained prominence is the organic cultivation system, without the use of chemical fertilizers and pesticides (FISCHER et al., 2007).

The growth of the crop has led to an increase in the number of diseases, which affect fruit quality, productivity and longevity, reducing commercial value (FALEIRO et al., 2011), and these diseases are caused by fungi, bacteria and viruses. In addition to the diseases, the crop also suffers from summer and excess precipitation, which compromise its yield. Due to these problems, it is necessary to maintain resistant genotypes for use in breeding programs (KRAUSE et al., 2010).

Diseases caused by soil-borne pathogens are one of the most important phytosanitary problems, since there is no curative control measure that is economically feasible and environmentally acceptable. These pathogens attack the plant's root and/or vascular system, causing its sudden death. The number of plants in the area can decrease dramatically, as well as the productive period of the orchards, making economic exploitation unfeasible in highly affected areas (CHAVES, 2004).

Authors group these diseases and denominate them premature death, either attributed to an unknown causative agent or associated with soil microorganisms such as *Fusarium oxysporum* Schlecht. Emend. Snyder & Hansen f. sp. *passiflorae* (FOP), *F. solani* (Mart.) Sacc. and *Phytophthora* spp.. The lack of precision in describing the symptoms of diseases affecting the passion fruit root and/or vascular system is a major problem in the correct diagnosis of the disease. Both *F. oxysporum* f. sp. *passiflorae* and *F. solani* have been associated with premature death to passion fruit and, when occurs in isolation, has received several nomenclatures. Symptoms have been confused in disease diagnoses. *F. oxysporum* f. sp. *passiflorae* is designated in literature as causal agent of wilt in passion fruit (LIBERATO, 2002; MANICOM et al., 2003) and wilting or fusariosis (VIANA et al., 2003). *F. solani* is related to diseases of collar rot

(LIBERATO, 2002), sudden death (PLOETZ, 1991), haematonectric canker, sudden death, crown canker and base rot (MANICOM et al., 2003).

In the case of *F. solani*, the infection may start from the main root and evolve into the collar, or the reverse. With the evolution of the disease, there is the darkening of the lesion of the peel, and the tissue crumbles, detaching from the cambium. Vessel destruction occurs both in the collar region and in the roots, which causes reflex symptoms of wilting, yellowing and dryness of the foliage (VIANA et al., 2003). According to Manicom et al. (2003), the vascularity of plants attacked by *F. solani* may show reddish brown to brown discoloration, occurring associated with canker and root rot symptoms, and the lesion does not progress in the stem to a height greater than 50 cm.

Fusariosis, also known as Fusarium wilt, is a disease caused by *F. oxysporum* f. sp. *passiflorae* and is one of the main problems affecting passion fruit, leading infected plants to death without it being possible to avoid it, since there is no curative control. Its occurrence in the field occurs in turners, that is, small or large foci distributed randomly in the culture, but which spread very easily when the conditions are favorable to the pathogen. The disease has gained importance in the commercial exploitation of passion fruit because it reduces the productive period of the crop in the affected areas (JUNQUEIRA; JUNQUEIRA, 2007).

The wilt of passion fruit begins with the wilting of the pointers, which can occur at any time of the year or cycle of the plant, being more common from the first year (SANTOS FILHO; SANTOS, 2003).

The penetration of the pathogen into the roots is associated with injuries, mainly caused by nematodes or agricultural implements (LIBERATO; COSTA, 2001). Initially, the pathogen colonizes the xylem vessels, and its diffusion within the plant occurs through spores, especially microconidia, which are passively transported by the flow of transpiration (MACHARDY; BECKMAN, 1981).

This fungus causes obstruction of the passage of the sap, discoloration of the vascular bundles which can extend up to two meters above soil and wilt of the plant. The xylem vessels are reddish or darkened, and when the environment is excessively moist, there is a crack in the stem, exposing the fruiting of the fungus. In adulthood, collapse may be restricted to some branches, before total collapse. In young plants, leaves turn from bright green to pale green, and sometimes old leaves may fall. In adult plants yellowing of young leaves occurs. The green fruits wither, while those that began

to mature reach the end of the process without many changes (SANTOS FILHO; SANTOS, 2003).

This fungus survives on cultural remains, soil and infected seeds. Injuries to the root system, especially those caused by nematodes, facilitate the penetration of the fungus. The spread is made by direct contact of roots, water, seeds and infected seedlings. Temperature of 15 °C to 28 °C and high relative humidity, acidic, clayey and poorly drained soils are ideal conditions for the establishment of the disease (PICCININ et al, 2005). They present abundant microconidia, usually unicellular, ovoid, formed in simple or branched conidiophores; and macroconidia also abundant, falcate and multisept. They also produce chlamydospores (DOMSCH et al., 1993). Due to the chlamydospores (resistance spores), FOP survives in the soil for many years (LIBERATO; COSTA, 2001).

The main measures to control the disease are preventive and cannot eliminate the pathogen if it is already in the soil (RONCATTO et al., 2004). The use of resistant cultivars as rootstocks is the most practical and efficient method of control for most diseases caused by soil fungi, including the *F. oxysporum* complex (NAVAS-CORTÉS et al., 2008). In Ministério da Agricultura, Pecuária e Abastecimento, there is no resistered chemical for this crop to treat this disease.

The wild species present great potential to be used as rootstocks (JUNQUEIRA et al., 2005), as well as they can be used to improve the physical-chemical characteristics of the pulp, so that they can be commercialized as new options in the market, for fruit or to improve the functional characteristics of this genus (FALEIRO et al., 2015). In order to take advantage of the potential of these species, which are of great importance, characterization, domestication, genetic improvement, documentation, dissemination and marketing studies are necessary (FALEIRO et al., 2011). Some of the *Passiflora* species that are used as rootstocks in the production of yellow passion fruit are *P. alata* Curtis, *P. caerulea* L., *P. gibertii* N. E. Brown, *P. nitida* Kunth, *P. setacea* L. (MENEZES et al., 1994) and *P. foetida* L. in Brazilian semi-arid region (SILVA *et al*., 2017). One of the species that can be used as rootstock is *P. cincinnata* Mast., because it presents resistance to fusariosis (MENEZES et al., 1994; SILVA et al., 2013). It is known as “maracujá-mochila” or “maracujá-do-mato” and has a wide geographic distribution. It can be found in the States of Bahia, Goiás e Minas Gerais (OLIVEIRA JÚNIOR et al. 2010) and in the semi-arid region of the Northeast (CORREIA; ARAÚJO & ARAÚJO, 2011). The fruit of this species is marketed in the

off-season of the yellow passion fruit in the Northeast region, being a great option for farmers because it is native to the region (OLIVEIRA JÚNIOR et al., 2010).

In relation to the effects of rootstocks on the physical and chemical characteristics of passion fruit, Cavichioli et al. (2011) verified that there was no influence on the content of soluble solids, titratable acidity and soluble solids/titratable acidity ratio. Already, Salazar et al. (2015) observed that fruits of non-grafted plants presented higher content of soluble solids and vitamin C.

The physical-chemical characterization has great importance in the genetic improvement per verify the organoleptic properties (JUNQUEIRA et al., 2010), and is also being performed in germplasm banks to obtain information about the description and classification of the accessions, to identify individuals with adequate characters (JUNQUEIRA et al., 2005), in order that they can serve the consumer and industrial market. In addition, it assists in the correct determination of the maturation stage of the fruit, so that the harvest is carried out at the appropriate time. For this, maturation indexes that include characteristics of peel coloration or chemical changes are also used (COELHO et al., 2010).

In the case of yellow passion fruit, the indicator of the harvest point may be the color change of the peel, even before the fall, thus avoiding possible damages, dirt and contamination in the contact with the soil. The fruits are usually harvested with peel 50 to 75% yellow (green maturation stage) and consumed with peel 100% yellow (yellow maturation stage) (LIMA, CENCI & RINALDI, 2016).

Thus, the objective of this work was to characterize the pathogenicity of differents isolates of FOP in *P. edulis* e *P. cincinnata* and verify the postharvest quality of fruits of *P. edulis* mature and immature from four areas with conventional cultivation with and without grafting in resistant species and organic cultivation with grafting.

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CHAPTER II

REACTION OF PASSION FRUIT SPECIES INOCULATED WITH *Fusarium oxysporum* f. sp. *passiflorae* ISOLATES

ABSTRACT

In passion fruit culture, one of the main diseases that reduces production is fusariosis, caused by the fungus *Fusarium oxysporum* f. sp. *passiflorae* (FOP). The use of resistant rootstocks, for example the species *Passiflora cincinnata*, is one of the techniques used to control this disease. The objective of this work was to evaluate the pathogenicity of different isolates of FOP in *P. edulis* and *P. cincinnata* in order to identify its potential for use in areas with a history of the disease. Thirteen isolates of the fungus were used and the inoculums produced at a concentration of 10^6 CFU mL^{-1}. The seedlings of *P. edulis* and *P. cincinnata* were produced in coconut fiber and the root system was then immersed for five minutes in the conidial suspension before being replanted in the 770 mL pots. One assay was carried out using *P. edulis* and *P. cincinnata* seedlings. In the experiment ten replicates were used, for each species, each isolate and for the control group. Seedlings were evaluated daily from the second after inoculation (DAI) until 90 DAI. All isolates were pathogenic in both *Passiflora* species, however incidence, severity and mortality were higher in *P. edulis*. There was a statistically significant difference for the incubation period of the FOP 23 and 57 isolates, being superior for *P. edulis*.

Keywords: Fusariosis; Pathogenicity; *Passiflora cincinnata*; *Passiflora edulis*.

1 INTRODUCTION

Passion fruit belongs to the family *Passifloraceae*, widely distributed around the tropics, with more than 580 species, mostly native from tropical America (CERVI, 2006). In 2017, Brazil had a production of 554,598 tons, in an area of 41,090 hectares (IBGE, 2017), and the yellow passion fruit (*Passiflora edulis* Sims) is the most planted in the country (FAOSTAT, 2017). Despite its prominent position, Brazilian average yield is low (13.5 t/ha) (IBGE, 2017), compared to the production potential of crop, estimated at 40 to 50 t/ha (FREITAS et al., 2011). Such low average is due to expansion

of the planted area simultaneously to the appearance and, or, worsening of a significant number of diseases. Such sanitary problems have reduced the economic production period and, even making growth of this species uneconomical in certain regions (FISCHER et al., 2005).

One of the main diseases affecting passion fruit is fusarium wilt, caused by *Fusarium oxysporum* Schlecht. Emend. Snyder & Hansen f. sp. *passiflorae* (FOP). It is considered as the most complex among all diseases (RONCATTO *et al*., 2004). Fusarium wilt, also known as 'fusariosis' or 'sudden death', starts with branch yellowing and wilt, until the whole plant dries, as a consequence of root and collar rot (FISCHER et al., 2010). The disease is observed in adult plants, however, under favorable conditions, such as soils with a disease history, and high temperature and moisture, young plants can die under the pathogen attack (BENNETT; DAVIS, 2013). Disease control is done preventively since there is no effective curative measure for it. Fisher and Resende (2008) recommended avoiding planting passion fruit in areas with disease history, or in heavy and compacted soils, planting healthy seedlings, and roguing diseased plants, thus reducing inoculum sources.

An important demand for plant breeding is for genotypes resistant to fusariosis, however, maintenance of aggressive sporulating isolates of the pathogen is difficult. In this sense, vegetative propagation brings new perspectives for cultivation of passion fruit, since harvesting in orchards has shown a reduction over the years due to phytosanitary problems caused by soilborne pathogens. Passion fruit grafting is a previously described technique (ZUCARELLI et al., 2014), and the use of resistant rootstocks, associated to other integrated management techniques, has been advocated since it is an effective and economic control measure for fusariosis (CAVICHIOLLI et al., 2011).

Several wild *Passiflora* species, such as *P. alata* Curtis, *P. caerulea* L., *P. gibertii* N. E. Brown, *P. nitida* Kunth, *P. setacea* L. (MENEZES et al., 1994) and *P. foetida* L. (SILVA *et al*., 2017) have been resistant to fusariosis. The wild species *P. cincinnata* Mast. occurs frequently in several territories of Northeast and Southeastern Brazil, even in areas with FOP, and has been used as rootstock for the production of yellow passion fruit. However, the results are varied according to the grafting method, species, time of the year, isolated of the causative agent and place (SANTOS et al., 2016).

The objective of this work was to characterize the pathogenicity of different isolates of FOP in *P. edulis* and *P. cincinnata* in order to identify its potential for use in areas with a history of the disease.

2 MATERIAL AND METHODS

2.1 Plant material

Seeds of *P. edulis* and *P. cincinnata* were purchased from Dina Dinamarca farm in the State of Rio Grande do Norte, an area with history of fusariosis. The seedlings were grown in coconut fiber trays and maintained in greenhouse (Golden Mix, Amafibra®), with sprinkler irrigation. The seedlings were inoculated when they reached the three-leaves stage, 45 days after planting.

2.2 Isolates and inoculation

Thirteen isolates of *Fusarium oxysporum* f. sp. *passiflorae* (FOP) were used: eleven from Collection of Fungi Culture of Embrapa Mandioca e Fruticultura (code FOP) (SILVA et al., 2013a) and two from passion fruit plants with typical wilting symptoms from State of Ceará (Table 1). These last two isolates were identified as *Fusarium oxysporum* through of the phylogeny using fragments of the translation elongation factor *1-alpha* (*ef-1α*) using blast searches (http://www.ncbi.nlm.nih.gov/blast/). All sequences of the studied *ef-1α* of isolates were deposited in Genbank (codes MH712505 and MH712506). The isolates were deposited in the culture collection of phytopathogenic fungi "Profª. Maria Menezes" (CMM) of the Universidade Federal Rural de Pernambuco (Recife, Brazil), with the codes CMM4864 and CMM4865.

Table 1. Isolates of *Fusarium oxysporum* f. sp. *passiflorae* used on pathogenicity tests.

Isolates	Geographic origin
FOP001	Cruz das Almas (BA)
FOP003	Cruz das Almas (BA)
FOP005	Cruz das Almas (BA)
FOP008	Ubaíra (BA)
FOP0013	Ubaíra (BA)
FOP0022	Ubaíra (BA)
FOP0023	Ubaíra (BA)
FOP0028	Ubaíra (BA)
FOP0057	Ubaíra (BA)
FOP0071	Porto Seguro (BA)
FOP0072	Livramento de Nossa Senhora (BA)
CMM4864	Jaguaruana (CE)
CMM4865	Jaguaruana (CE)

To produce the inoculums, mycelial disks from each isolate were transferred to Petri dishes containing potato-dextrose-agar (PDA) media (39 g/L). The dishes were kept in BOD for seven days continuously at 25°C, under dark. The spore suspension was prepared some minutes before inoculation: 10 mL of sterile distilled water was added to the Petri dishes, and the fungal colonies were scraped off to release the spores. The resulting suspension was filtered in a double layer of sterile gauze, the spores were counted using a Neubauer chamber and spore concentration was adjusted to 10^6 macroconidia mL^{-1} (SILVA *et al*., 2013 a).

The seedlings were removed from the coconut fiber in which they were grown and their roots were washed with sterile water. The root system was then immersed for 5 min in the conidial suspension before being replanted in the 770 mL pots containing coconut fiber. Plants which roots were soaked only in sterile water were used as a control.

The action of the isolates was evaluated in the species *P. edulis* and *P. cincinnata*. For each isolate, ten replicates of one plant of each species of passion fruit were used and for the control groups (Figure 1).

Figure 1. Pathogenicity of *Fusarium oxysporum* f. sp. *passiflorae* in *Passiflora edulis* and *P. cincinnata* experiment at greenhouse.

2.3 Disease evaluation

Seedlings were evaluated daily from the second after inoculation (DAI) until 90 DAI and the occurrence of wilting or death was recorded for both experiments. Symptomatic seedlings had parts of their stem and roots sterilized and transferred to PDA media for fungus reisolation in order to complete the postulate of Koch. After five days, the colonies were evaluated for macro and microconidia to confirm the etiologic agent.

The percentage of dead plants was calculated for the species of *Passiflora* for each isolate. In addition, the average incubation period (IP) through determination of the time between inoculation and the appearance of wilt symptoms was determined for each plant and for each isolate, calculated in days. The incidence of the disease was calculated by the amount of plants that presented symptoms, represented in percentage.

Severity of disease was evaluated by scale according to Cia et al. (1977), with modifications, where: score 1 – healthy plants; score 2 – internal darkening only in the basal part of the roots and up to 35% of the leaves yellowed; score 3 – darkening above the basal part of the roots and up to 75% of the leaves yellowed; score 4 – dead plants (Figure 2).

Figure 2. Samples of *Passiflora edulis* showing the four notes of the scale of evaluation of fusariosis, according to Cia et al. (1977). a: score 1 – healthy plants; b: score 2 – internal darkening only in the basal part of the roots and up to 35% of the leaves yellowed; c: score 3 – darkening above the basal part of the roots and up to 75% of the leaves yellowed; d: score 4 – dead plants.

2.4 Statistical analysis

The experimental design was the completely randomized with two species of passion fruit (*P. cincinnata* and *P. edulis*) and thirteen isolates of FOP, with ten replicates (experimental unit being a vessel containing a plant). Two control groups (one for each passion fruit species) were installed.

The data obtained were evaluated with ASSISTAT program and the averages compared by the Mann-Whitney and the Kruskal-Wallis tests at the 5% of probability level. In the data that was not possible a statistical analysis, a descriptive analysis was performed.

3 RESULTS

In the evaluation of the pathogenicity experiments of different isolates of FOP compared to *P. edulis* and *P. cincinnata*, it was observed that there was an incidence of 80 to 100% and 10 to 90%, respectively, after 90 days (Figure 3).

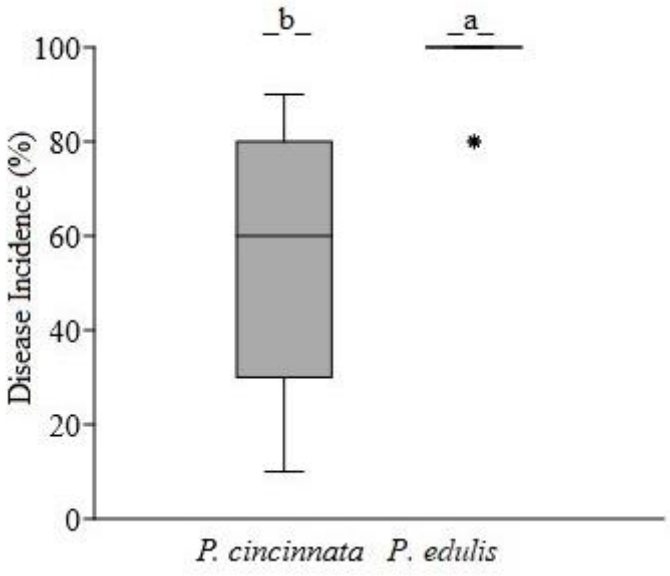

Figure 3. Incidence of fusariosis caused by all *Fusarium oxysporum* f. sp. *passiflorae* isolates in *Passiflora cincinnata* and *Passiflora edulis* at the end of 90 days after inoculation. Different lowercase letters indicate significant differences according to Mann-Whitney test ($p<0.05$).

Regarding the incidence of the disease, it was possible to observe that a larger number of *P. edulis* plants presented symptoms of fusariosis (Figure 4). The isolates FOP 1, 3, 13, 23, 28, 57, 72 and CMM4864 were highlighted in the incidence of *P. edulis*, presenting a statistically significant difference in relation to the control group, at the 5% level of significance. In *P. cincinnata*, only the isolate FOP 28 isolate showed a difference in relation to the control.

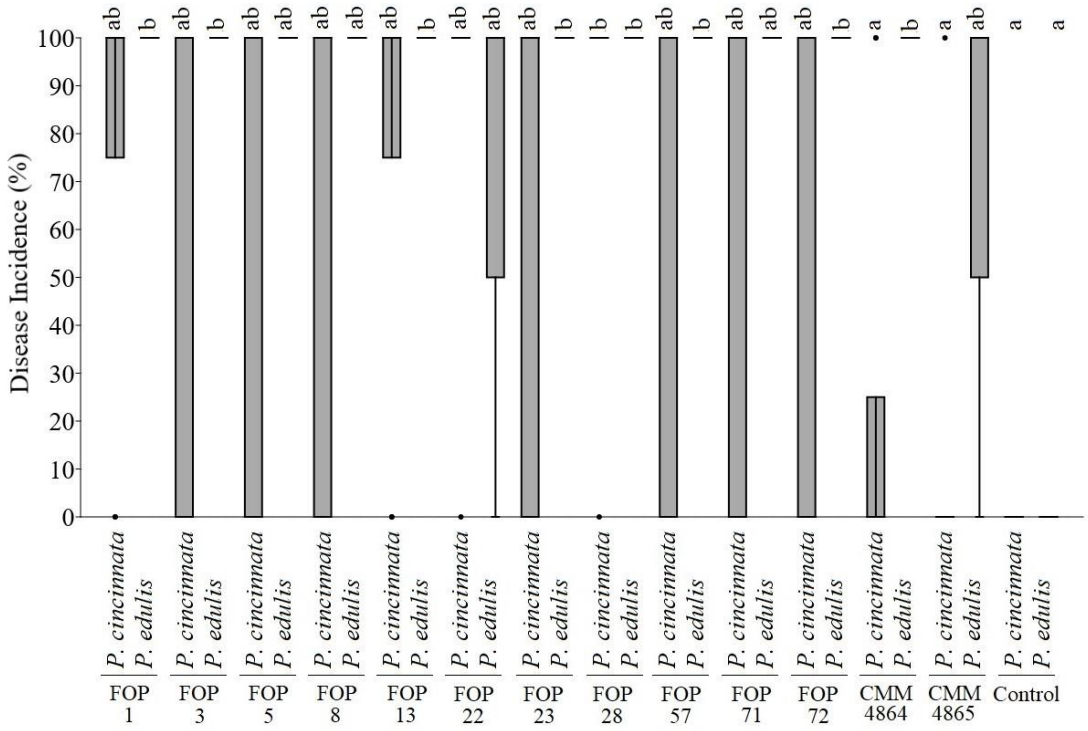

Figure 4. Incidence of fusariosis caused by each of *Fusarium oxysporum* f. sp. *passiflorae* isolate in *Passiflora cincinnata* and *Passiflora edulis* at the end of 90 days after inoculation. Different lowercase letters indicate significant differences according to Kruskall-Wallis test ($p<0.05$).

Among the isolates of FOP used in this work, FOP 28 was more virulent for both *Passiflora* species as it caused the highest number of deaths. Minor mortalities for *P. cincinnata* were found in the isolates FOP 22, CMM4864 and CMM4865. As for *P. edulis*, isolates FOP 5, FOP 22 and FOP 71 did not cause any death (Figure 5).

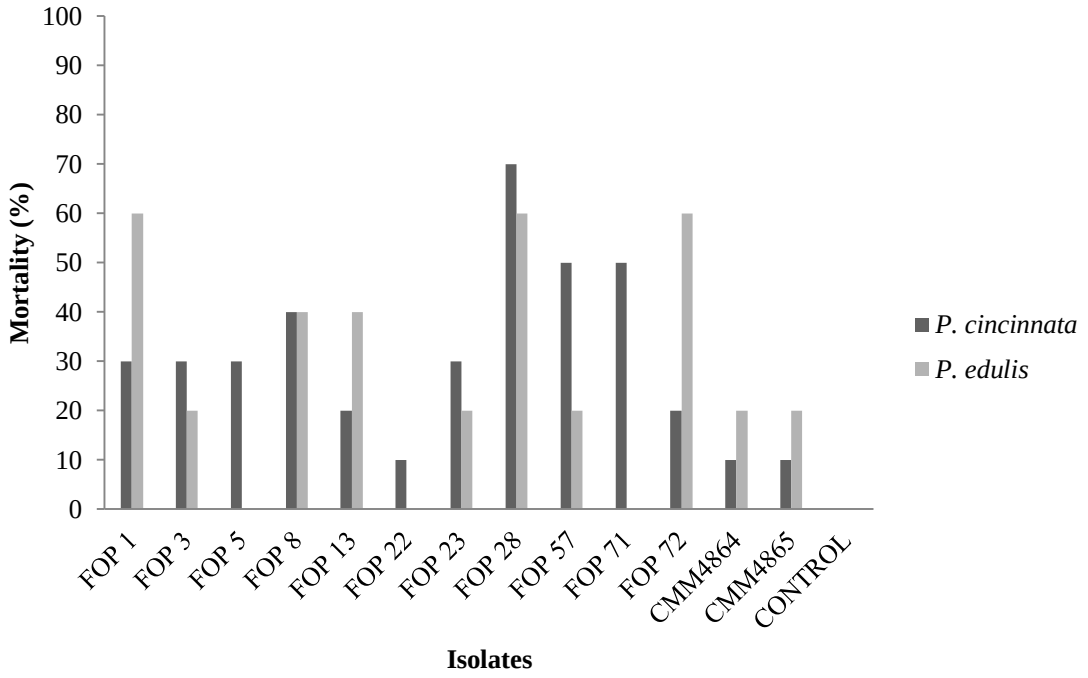

Figure 5. Mortality caused by the isolates of *Fusarium oxysporum* f. sp. *passiflorae* in *Passiflora cincinnata* and *Passiflora edulis* at the end of 90 days after inoculation.

According to the scale of scores of Cia et al. (1977), severity of fusariosis in *P. edulis* and *P. cincinnata* was more intense in the first species. Means for this evaluation were statistically different, at the 5% level of significance, indicating that the severity of the isolates of FOP was higher for *P. edulis* (Figure 6).

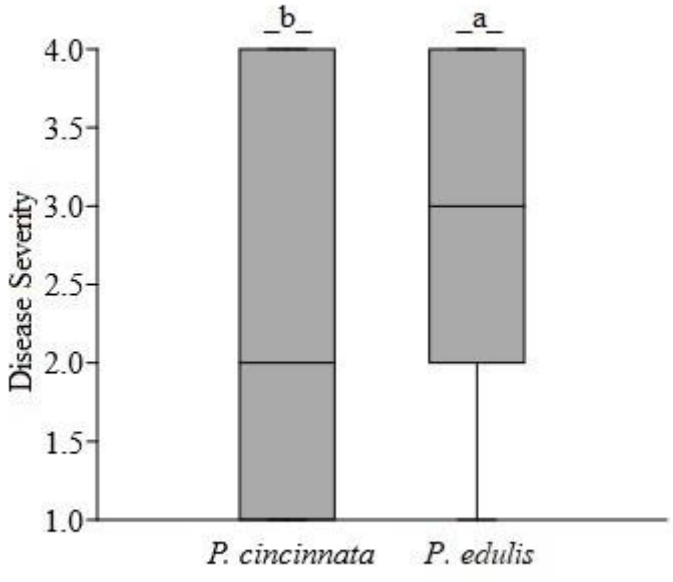

Figure 6. Fusariosis severity caused by all *Fusarium oxysporum* f. sp. *passiflorae* in *Passiflora cincinnata* and *Passiflora edulis* at the end of 90 days after inoculation. Score 1 – healthy plants; score 2 – internal darkening only in the basal part of the roots and up to 35% of the leaves yellowed; score 3 – darkening above the basal part of the roots and up to 75% of the leaves yellowed; score 4 – dead plants. Different lowercase letters indicate significant differences according to Mann-Whitney test ($p<0.05$).

In Figure 7 it is possible to observe that the two species of *Passiflor*a behaved differently when inoculated with the same isolate, been numerically dissimilar. Only the control group showed no symptoms. The isolates of FOP were more virulent for the species *P. edulis*, except for the isolated FOP 22 where it was more virulent for *P. cincinnata*.

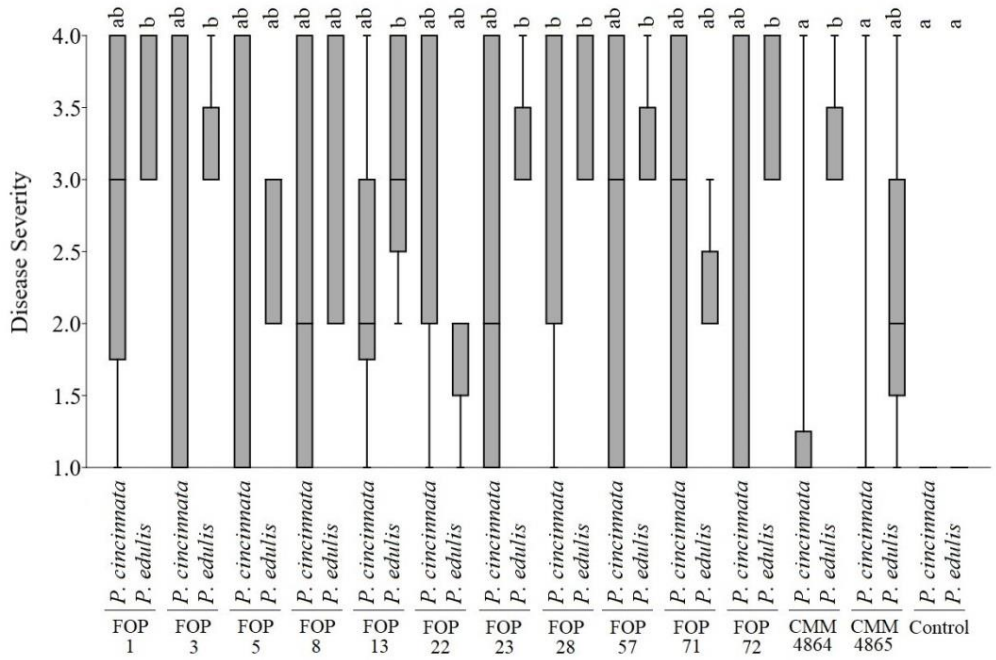

Figure 7. Severity of fusariosis caused by each *Fusarium oxysporum* f. sp. *passiflorae* isolates in *Passiflora cincinnata* and *Passiflora edulis* at the end of 90 days after inoculation. Different lowercase letters indicate significant differences according to Kruskal-Wallis test ($p<0.05$).

Pathogenicity test indicated an incubation period of 21 to 34 days for *P. cincinnata* and 17 to 52 days for *P. edulis*, presenting symptoms of mild chlorosis associated with slight to moderate wilt (Figure 8). These differences were statistically significant at the 5% probability level.

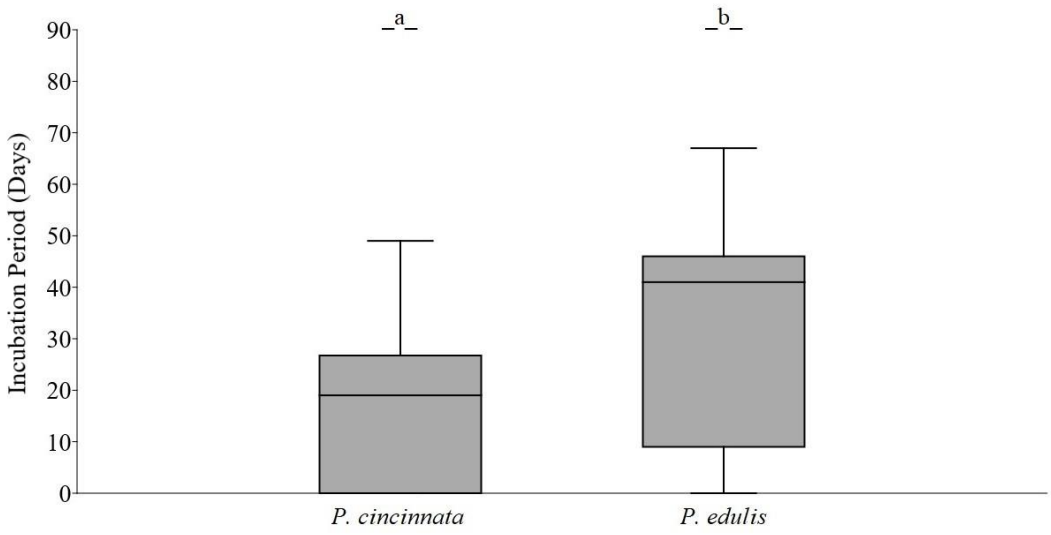

Figure 8. Incubation period of fusariosis in *Passiflora cincinnata* and *Passiflora edulis* caused by all *Fusarium oxysporum* f. sp. *passiflorae* isolates. Different lowercase letters indicate significant differences according to Mann-Whitney test ($p<0.05$).

The incubation period for each of the isolates is shown in Figure 9. Isolates FOP 28 and FOP 71 were able to cause symptoms more rapidly in *P. cincinnata* (21 days), while FOP 72 and CMM4865 isolates demonstrated a 34-day incubation period. In *P. edulis*, the shortest incubation period was found for the isolates FOP 8 and CMM4864, with 17 and 18 days, respectively. For the FOP 5 and FOP 72 isolates incubation periods of 51 and 52 days were verified, respectively. Comparing the action of the isolates between the species, FOP 23 and 57 had a higher incubation period in *P. edulis*, at a significance level of 5%.

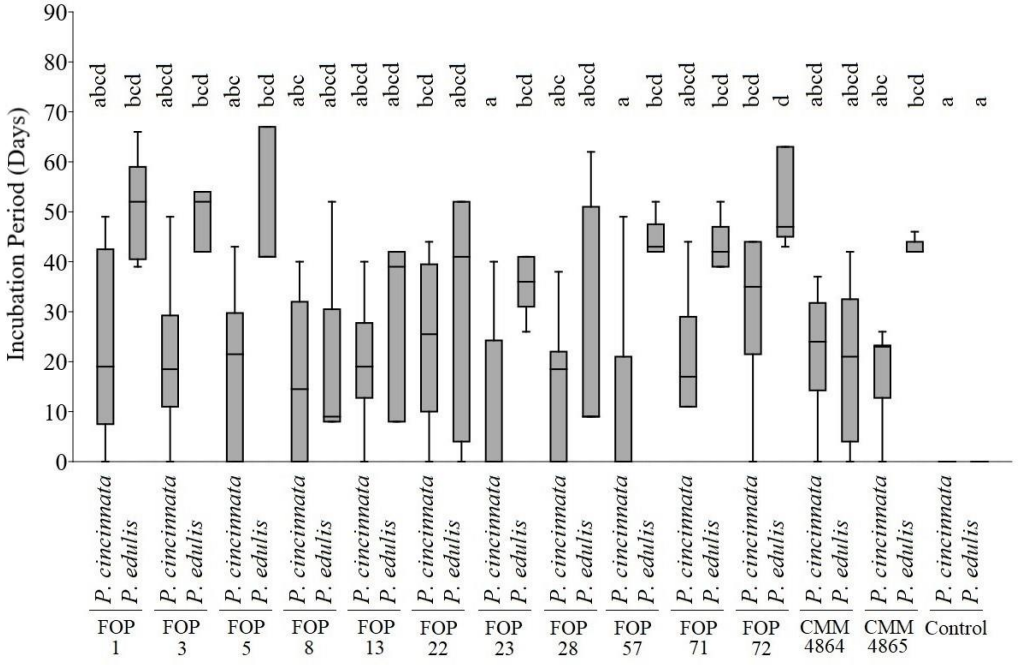

Figure 9. Incubation period of fusariosis in *Passiflora cincinnata* and *Passiflora edulis* caused by each *Fusarium oxysporum* f. sp. *passiflorae* isolates. Different lowercase letters indicate significant differences according to Kruskal-Wallis test ($p<0.05$).

4 DISCUSSION

It was possible to notice in the inoculated plants the root rot occurs due to colonization of the vascular system by the fungus. Already was observed in the control plants, a root system developed and with the emission of new roots.

Pathogenicity screening of FOP allowed detection of the corresponding isolates as virulent. All isolates evaluated demonstrated symptoms in both *Passiflora* species, with incubation periods variable, but the most virulent isolates showed during the tests higher values of incidence and severity. This suggests that these attributes are reliable

and practical for the rapid detection of pathogenic isolates (ORTIZ; HOYOS-CARVAJAL, 2016). Number of leaves are no significant at the beginning of the experiment differences, however at the end of 90 days after inoculation of the tests there was noticeable reduction in the number of sheets, which explains defoliation by the process generated by the pathogen.

The isolates of FOP tested showed variation in incidence and severity, evidencing genetic variability in this population. Similar results were found by Ferreira et al. (2015), who verified that ten isolates of this fungus in yellow passion fruit showed variability in severity and incidence, evidencing the genetic diversity of the FOP species. The same was observed by Ortiz and Hoyos-Carvajal (2016) using eight isolates of FOP. Genetic differences between FOP isolates may be associated with their pathogenicity, and these observations must be taken into consideration when the passion fruit germplasm is screened for sources of resistance and, later, in the development of resistant cultivars (SILVA et al., 2013a).

Laranjeira et al. (2005) quantified the incidence of Fusarium wilt in passion fruit plants grafted to *P. edulis*, *P. alata*, *P. gibertii* and *P. cincinnata* using a survival analysis and revealed that the *P. edulis* showed a great mortality than the others. The same occurred in the present study. Silva et al. (2017), studying the reaction of passion fruit cultivars in areas with fusariosis, verified that, according to the results obtained, the species *P. foetida* L. used as rootstock presented resistance when cultivated in soil with fusariosis in the region of Mossoró-RN. Whereas, eight cultivars of *P. edulis* were susceptible. Silva et al. (2013b) did not verify deaths in *P. cincinnata* after 120 DAI when inoculated with FOP. However, mortality of 5.8 to 100% was found for different accessions of *P. edulis*. In the present study, mortality varied for the different isolates from 10 to 70% and from 0 to 60% for *P. cincinnata* and *P. edulis*, respectively. *Passiflora cincinnata* is considered to be a strong species with wide adaptation, which are important characteristics for breeding programs. Field evaluations have shown that this species shows considerable resistance to fusariosis (LARANJEIRA et al., 2005). However, this was not observed in this work, probably due to the large genetic variability of the isolates of FOP used.

Fusarium wilt symptoms occur between the 2nd and 4th DAI for several species, but depend on the host (ALEXOPOULOS et al., 1996). The symptoms of wilt started to occur at 17 to 52 days of evaluation for *P. edulis* and 21 to 34 days for *P. cincinnata* in the present work. The initiation and continuation of symptoms varied greatly for each

isolate. For Preisigke et al. (2017), searching for fusariosis resistant *Passiflora* species and using one isolate of FOP, found a seven-day incubation period for *P. edulis* and *P. cincinnata*. The genotypes of *P. edulis* survived until 31 days of evaluation, being considered susceptible. *P. cincinnata* presented moderate resistance, surviving throughout the experiment. Silva et al. (2013b) found five accessions of *P. cincinnata* resistant to one isolate of FOP, where no symptoms were observed in the plants. For *P. edulis*, symptoms were observed at 11 days after inoculation. The differences between and within each species may be associated with the high level of heterozygosity of passion fruit, an allogamous plant with gametophytic self-incompatibility (SUASSUNA et al., 2003). The use of only one isolate of the pathogen may have also influenced the results, as the reaction of plants to different isolates can be individualized (CAVALCANTI et al., 2002). The plants used in this experiment were submitted to 13 different isolates, thus demonstrating differentiated behaviors. The incubation period found by Ortiz and Hoyos-Carvajal (2016) for eight isolates of FOP in *P. edulis* was between 17 and 19 days. A long incubation period is an important component for host plants to have a partial resistance to a given pathogen (VAN DER PLANK, 1963).

5 CONCLUSION

All the FOP isolates used were pathogenic to *P. edulis* and *P. cincinnata*. Incidence, severity and mortality were higher in *P. edulis*, demonstrating a greater degree of susceptibility as compared to *P. cincinnata*. However, period of incubation was higher in *P. edulis*.

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CHAPTER III

PHYSICAL-CHEMICAL CHARACTERIZATION OF YELLOW PASSION FRUIT PRODUCED IN DIFFERENT FORMS OF CULTIVATION

ABSTRACT

Grafting is widely used to provide resistance to pests and diseases in yellow passion fruit, such as fusariosis, caused by the fungus *Fusarium oxysporum* f. sp. *passiflorae* (FOP). However, this practice may generate some changes in the postharvest characteristics of the fruits. The objective of this work was to verify the postharvest quality of fruits of *Passiflora edulis* Sims collected inmature and mature in four areas with different forms of cultivation (conventional without grafting, two in conventional with grafting and organic with grafting). The experiment was installed in a completely randomized design, in a 4 x 2 factorial scheme, with five replicates of four fruits each. The first factor was the different collection sites with their cultivation systems and the second factor was the maturity stage. The fruits were harvested, selected and submitted to physical, physical-chemical and chemical analysis, bioactive compounds and antioxidant activity. Fruit weight, length, width and firmness were superior in fruits from conventional system, however fruit yield was inferior for this system. Soluble solids, pH, reducing sugars and total soluble sugars were superior in fruits from collection sites with the use of grafting. Bioactive compounds and antioxidant activity were similar between the fruits from different collection sites.

Keywords: Bioactive compounds; Grafting; *Passiflora edulis*; Postharvest.

1 INTRODUCTION

Passion fruit is a popular name given to several species of the genus *Passiflora* that belongs to Passifloraceae family, which there are more than 500 species distributed in regions of tropical and subtropical climate of the world (CERVI, 2006). The species *Passiflora edulis* Sims, known as passion fruit, is the most produced and marketed (IBGE, 2017). Its cultivation is primarily focusing on the juice and pulp industry, especially due to its higher acidity and pulp yield (JESUS et al., 2018).

This fruit appears to be an excellent source of nutrients as carbohydrates, vitamins, and minerals that are essential nutrients for life. The fruit has a high content in nutraceuticals, as phenolic acids, where anthocyanins and flavonoids are the majoritarian compounds of this group; carotenoids and β-carotene appear to be the principal component, with consequently increased provitamin A activity. These nutraceutical compounds have biological activities in the health, protective effect against degenerative and chronic diseases and act as mutagenesis and carcinogenesis inhibitors. Also, these compounds have been associated with antiviral, antiallergic and anti-inflammatory activities (MORAIS et al., 2016; GONZÁLEZ-GALLEGO et al., 2014). However, during the long shipping or air transportation periods of times, fruits undergo changes by accelerated ripening. They lose organoleptic quality generating economic losses for exporting companies up to 15% of the total volume shipped. Fruits are harvested immature and arrive at the consumer fully mature, but with differences in their physicochemical composition (JIMÉNEZ et al., 2011).

Grafting is widely used in fruit culture and in other perennial species to propagate superior genotypes, to control plant size, reduce juvenile period, improve the adaptation to adverse soil conditions and to provide resistance to pests and diseases (ATUCHA et al., 2014; SALAZAR et al., 2015). One of the main diseases that grafting aims to reduce the incidence is fusariosis, also known as fusarium wilt, caused by the fungus *Fusarium oxysporum* f. sp. *passiflorae* (FOP). The symptoms start with branch yellowing and wilt (FISCHER et al., 2010). Due it is a vascular disease, the chemical control of fusariosis is not efficient. In this case, preventive measures of cultural control are used, besides the use of grafted seedlings on rootstocks of wild species with potencial for resistance (CAVICHIOLI et al., 2009).

In addition to grafting, the growing demand for healthy food, valuing biological diversity and without the use of chemical fertilizers and pesticides, is a trend that favors the creation of new opportunities, especially for small farmers. The system of organic cultivation of yellow passion fruit has been adopted by producers, however, there is a lack of scientific information on the organic cultivation of the crop (FISCHER et al., 2007).

Silva et al. (2013) reported, in *Passiflora* crops, tolerance of some wild species such as *P. suberosa* L., *P. alata* Curtis and *P. cincinnata* Mast. to premature death of the plants (*Fusarium* spp.). Thus, there have been several studies evaluating the effect of rootstock and graft type on fruit quality of yellow passion fruit. Cavichioli et al. (2011)

found that the rootstocks studied did not influence the content of soluble solids (SS), titratable acidity (TA) and the SS/TA ratios. Similarly, the grafting method did not affect the diameter, length, weight of the fresh fruit, weight and thickness of the peel, and juice yield. Salazar et al. (2015) verified that fruits from ungrafted plants obtained higher values of fresh mass, diameter, peel mass, soluble solids and vitamin C. However, scarce information is available on the behavior of bioactive compounds in fruits from grafted plants and in different cropping systems (conventional and organic).

Thus, the objective of this work was to verify the postharvest quality of fruits of *P. edulis* collected inmature and mature in four areas with different forms of cultivation (conventional without grafting, conventional with grafting and organic with grafting).

2 MATERIALS AND METHODS

2.1 Sample preparation

Fruits of yellow passion fruit, *P. edulis*, were derived from four commercial orchards, located in the cities of Jaguaruana, Ceará (4°58'00.6"S and 37°47'10.3"W) and Coronel Ezequiel, Rio Grande do Norte (06°23'30.26"S and 36° 10'38.26"O), cultivated in an organic system with grafting and conventional without grafting, respectively, and in rural communities of Maisa (4°52'52.5"S and 37°26'50.0"W) and Pau Branco (4°59'30.5"S and 37°25'10.5"O) (city of Mossoró, Rio Grande do Norte), both cultivated in an conventional system with grafting, brazilian semiarid. The climate of the region, according to the Köppen classification, is "BSw", dry semiarid with low altitude and latitude, average air temperature of 26.7 °C and relative humidity of 68.9%. The average annual precipitation is about 750 mm, with a rainy period between February and June and low probability of rains between August and December. In general, they present shallow, rocky, but fertile soils (ALVARES et al., 2014).

The fruits were harvested manually, in the morning, in two maturation stages: at harvest point (peel coloration 50 to 75% yellow, commonly known as "green") and point of consumption (peel coloration 100% yellow, known as "yellow"), according to company rules. In the laboratory, the fruits went through a process of selection, being discarded those that presented damages by cuts, abrasions, attacks of insects or animals and, later, they were cleaned with the aid of a damp cloth.

The fruits were submitted to quality physical evaluations and then pulp containing the seeds was separated from the peel (epicarp) after a cross section in the fruit, manually with the help of stainless steel knives. The pulp fraction (mesocarp) was separated from the seeds with the aid of plastic sieves and stored in plastic pots and stored in a freezer at -23 ° C for further analysis.

The experiment was installed in a completely randomized design, in a 4 x 2 factorial scheme, with five replicates of four fruits each, on a bench with fruits brought from the field. The first factor was the different collection sites with their cultivation systems [C. Ezequiel (conventional cultivation without grafting), Jaguaruana (organic cultivation with grafting practice), Pau Branco (conventional cultivation with grafting practice) and Maisa (conventional cultivation with grafting)] and the second factor at maturity stage [harvest point (peel coloration 50 to 75% yellow) and point of consumption (peel coloration 100% yellow)].

2.2 Physical characteristics

For the physical analyzes of quality, the fruits were divided into five replicates of four fruits each, totaling 20 samples, for each stage of maturation of each collection site. In total, 80 yellow passion fruits were used at the harvest point and 80 fruits at the point of consumption.

The length (mm), width (mm) and peel thickness (mm) were determined using a digital pachometer (Lotus plus); fruit weight (g) and pulp weight (g) using an analytical balance (Bel Engineering); and pulp yield (%) obtained by the difference between fruit weight and pulp weight.

The color of the peel and the pulp was expressed in L (luminosity - brightness, clarity or reflectance), C* (chromaticity - saturation or intensity of color) and °H (hue angle - tonality) (Commission Internacionale de L'Eclaraige) (MINOLTA, 2007), using a digital benchtop colorimeter (CR-140, Minolta®). Readings in the peel were determined randomly at two equidistant points in the equatorial region, considering the average between them, and for the color of the pulp the readings were performed after the separation of the seeds in petri dishes.

The firmness of the fruit was determined using the Texture Analyzer®, model TA.XTExpress / TA.XTicon (Stable Micro Systems Ltd., Surrey, England) with a 10 kg load cell. The cylindrical probe of stainless steel with a diameter of 5 mm (model P/5)

was used. The pre-test, test and post-test speeds were 2 mm/s, 2 mm/s and 10 mm/s, respectively, and penetration distance of 10 mm. Two equidistant measurements were performed, one in each equatorial region of the fruit, and considered the average between them. The results were expressed in Newton (N).

2.3 Physical-chemical and chemical characteristics

The physical-chemical and chemical analyzes were performed from five replicates of four fruits each.

The hydrogenation potential (pH) was determined using a direct reading potentiometer (Model mPA-210 Tecnal®, Brazil) duly standardized with pH 7.0 and pH 4.0 buffer solutions, in analyzes made directly on the pulp. The data were expressed in real pH values (AOAC, 2002).

The titratable acidity (TA) was determined by volumetric procedure, using 1 g of pulp transferred to a 125 mL Erlenmeyer flask and the volume completed to 50 mL with distilled water. Phenolphthalein pH indicator 1% was added. Titration was performed with 0.1 N sodium hydroxide solution until the color change to slightly pink (INSTITUTO ADOLFO LUTZ, 2005), using an automatic titrator (Titrette® model Class A Precision by BRAND, USA), results were expressed as mg of citric acid/100 g of pulp.

Total soluble solids were determined with pulp juice in a digital refractometer (model PR-100, Pallete, Atago Co, LTD, Japan) (AOAC, 2002). The results were expressed as percentage (%); and the soluble solids/titratable acidity ratio (SS/TA) was determined by the ratio between the soluble solids values and the titratable acidity.

Total sugars were determined by the method of Antrona (9,10-dihydro-9-oxaanthracene) (Vetec, Brazil), according to Yemn and Willis (1954), from 1 g of the samples to obtain the extract. A 30 μL aliquot was taken to perform the spectrophotometer readings (model UV-1600, Pro-Analysis®, Brazil) at 620 nm, the results being expressed as a percentage (%). Reducing sugars by the DNS method, according to Miller (1959); the extract was obtained from the dilution of 1 g of the pulp from which 0.7 ml was taken and to this volume 0.8 ml of distilled water and 1 ml of 3,5-dinitrosalicylic acid (DNS, Vetec, Brazil) at 1%, and the reaction was carried out in a water bath at 100 ºC for 5 minutes and cooled in an ice bath. The readings were

performed in a spectrophotometer at 540 nm and the results expressed as percentage (%).

2.4 Bioactive compounds and total antioxidant activity (TAA)

Vitamin C was determined by titration with 0.02% 2,6-dichlorophenol-indophenol solution, according to the methodology proposed by Strohecker and Henning (1967). 1 g of the samples were used and diluted with 0.5% oxalic acid in a 100 mL volumetric flask. A 5 mL aliquot of this solution was diluted in distilled water to 50 mL and the titration was performed, the results being expressed as mg ascorbic acid/100 g fresh weight (FW).

Total anthocyanins and yellow flavonoids were determined according to Francis (1982). For each sample, 1 g was mixed into 50 ml of the extractive solution (85:15 ratio of ethyl alcohol-1.5 N hydrochloric acid), homogenized for 1 minute in Ultra-Turrax® (T25-IKA, Germany) and stored at 4 °C for 12 hours. The solution was filtered on filter paper for amber bottles and readings were performed in a spectrophotometer (UV-1600 model, Pró-Análise®, Brazil) with wavelength of 374 nm for flavonoids with absorption coefficient of 76.6 mol/cm and 535 nm for anthocyanins with an absorption coefficient of 98.2 mol/cm, the results being expressed in mg/100 g FW.

The procedure for the determination of total carotenoids was performed according to Higby (1962). In the dark, 5 g of each sample was weighed and 15 ml of isopropyl alcohol and 5 ml of hexane were added, following homogenization in Ultra-Turrax® (T25-IKA, Germany) for 1 min, and transfer to amber separatory funnel. Three successive washes with 100 ml of distilled water were performed, with an interval of 30 min. The top layer was filtered on cotton wool with anhydrous sodium sulfate into a 25 ml flask, washing the cotton with hexane. The reading was carried out in a spectrophotometer at 450 nm and the results expressed in mg/100 g.

β-carotene was performed according to the methodology of Nagata and Yamashito (1992), where 1 mL of each sample was taken and 10 mL of solvent hexane: acetone (3:2 ratio) was added. The contents were homogenized in Ultra-Turrax® (T25-IKA, Germany) for 1 min and centrifuged at 955 rpm for 10 min at 25 °C. The supernatant was collected for spectrophotometer reading at four absorbances: 663, 645, 505 and 453. The results were expressed as mg/100 mL juice.

2.4.1 Extract for total extractable polyphenols and total antioxidant capacity (TAA) by $ABTS^{\bullet+}$ and DPPH methods

The methodology was described by Larrauni et al. (1997), with modifications. 10 g of the samples were weighed into centrifuge tubes and 10 mL of the methanol-water extracting solution (50:50, v/v) was added at room temperature for 1 h. The tubes were centrifuged for 20 min at 10,000 rpm and the supernatant recovered. 10 mL of a second acetone-water extracting solution (70:30, v / v) was added to the residue for 1 h at room temperature, and centrifuged. The two supernatants were mixed in a volumetric flask, and added to 25 ml of distilled water. The extract was used to determine the content of total extractable polyphenols and antioxidant capacity by ABTS and DPPH method.

2.4.2 Total extractable polyphenols

Total extractable polyphenols (TEP) were determined by colorimetric assay using the Folin-Ciocalteau reagent, according to the methodology described by Obanda and Owuor (1997). The determination was performed using aliquots of 300 μL of extracts in test tubes, 700 μL of distilled water, 1 mL of Folin-Ciocalteau reagent, 2 mL of 20% sodium carbonate solution and 2 mL of distilled water. Following that the samples were agitated in a tube shaker (QL-901, Vortex®) and left at rest for at least 30 min in the dark. The readings were performed in a spectrophotometer at 700 nm, using a standard curve of gallic acid 98% (dosed at 0, 10, 20, 30, 40 and 50 μg). The results were expressed as gallic acid equivalents (GAE) mg/100g FW.

2.4.3 Total antioxidant activity - ABTS assay$^{\bullet+}$

The total antioxidant activity (TAA) by the ABTS$^{\bullet+}$ method was determined using 2,2-azinobis-3-ethylbenzthiazoline-6-sulphonic acid radical cation (ABTS$^{\bullet+}$, Sigma) (RE et al., 1999). The ABTS$^{\bullet+}$ radical was generated through the reaction of the 7 mM ABTS solution with 140 mM potassium persulfate, and leaving in the dark for 16 h at room temperature before use. The ABTS$^{\bullet+}$ radical was diluted with ethanol until an absorbance of 700 nm ± 0.05 at 734 nm. The spectrophotometer reading was performed after 6 min from the mixture of 30 μL of the extract with 3 mL of the diluted ABTS$^{\bullet+}$ radical. A calibration curve with the synthetic antioxidant Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, Sigma) at the concentration of 100 to 2000 μM

in ethanol was made and the results of the analysis were expressed in μmol of Trolox/g FW.

2.4.4 Total antioxidant activity - DPPH assay

The methodology for the determination of the antioxidant activity by the DPPH method was performed according to Sanchez-Moreno et al. (1998). From the extract of each sample, three different dilutions were prepared in 10 ml volumetric flasks. In the dark, 0.1 mL aliquot of each extract was transferred for the test tube with 3.9 mL of DPPH (0.06 mM DPPH solution dissolved in methyl alcohol) and homogenized in a tube shaker (QL-901, Vortex®). The same procedure was performed with a control solution (4 mL of 50% methyl alcohol, 4 mL of 70% acetone and 2 mL of distilled water). The rest was followed in the dark for 1 h and 10 min and the readings were carried out in a spectrophotometer at 515 nm. A calibration curve was performed with DPPH solutions in methyl alcohol varying the concentration from 10 μM to 50 μM and the results of the analysis were expressed in g of fruit/g DPPH.

2.5 Statistical analysis

Data were submitted to analysis of variance using the program SISVAR 5.6 (FERREIRA, 2014) and Tukey's test at 5% probability for comparison of means.

3 RESULTS AND DISCUSSION

3.1 Physical characteristics

There was a significant effect for the interaction of collection sites x maturation stage for all analyzes except for pulp weight (PW), peel thickness (PT) and pulp hue angle, which demonstrated significant effect considering only collection sites ($p < 0.01$) (Tables 1 and 2).

The fruits of yellow passion fruit at the green maturation stage used in the experiment had a mean weight ranging from 148.78 to 315.79 g, whereas in the yellow stage the variation was from 136.01 to 333.77 g. Regarding collection sites, the fruits with the highest weight were those harvested in the city of Coronel Ezequiel in a conventional cultivation system without grafting, in both maturation stages (Table 1),

with an increase of 128.09% in the weight of these fruits as compared with those from Maisa (with grafting). However, pulp yield was inferior. Only the weight of the fruits of this collection site was within the market preference for fresh fruits, where they must have a fresh weight of more than 200 g (AGUIAR et al., 2015). The differences observed between the weight of the fruits from grafting may have been due to the vigor of the plants, since they are materials of different species, as verified by Nogueira Filho et al. (2010), where plants grafted on *P. gibertii* presented lower vigor in the field, which may have influenced weight, length and average diameter of the fruits. Fischer et al. (2007) analyzing yellow passion fruits in conventional and organic cultivation systems verified weights of 156.13 and 175.27 g, respectively. Jesus et al. (2018) analyzing fruits of five different cultivars found a variation of 179 to 228 g. In this way, it is possible to observe that there is a great variation of fruit weight in the species *P. edulis*.

Table 1. Fruit weight, pulp yield, fruit length and fruit width of passion fruit in the green and yellow stages from different collection sites.[1]

Local	Characteristics												
		Fruit weight (g)			Pulp yield (%)			Fruit length (mm)			Fruit width (mm)		
	System[2]	Green	Yellow	Mean	Green	Yellow	Mean	Green	Yellow	Mean	Green	Yellow	Mean
Cel. Ezequiel	C	315.79 aA	333.77 aA	324.78	39.61 bA	36.25 bA	37.93	105.97 aA	108.58 aA	107.27	90.96 aA	93.92 aA	92.44
Jaguaruana	O+G	184.16 bA	178.83 bA	181.49	50.40 aA	46.66 aA	48.53	89.64 bcA	90.84 bA	90.24	78.77 bA	82.61 bA	80.69
Pau Branco	C+G	175.88 bcA	138.08 cB	156.98	50.48 aA	49.96 aA	50.22	94.61 bA	89.49 bB	92.05	85.80 aA	81.05 bB	83.42
Maisa	C+G	148.78 cA	136.01 cA	142.39	39.22 bB	50.79 aA	45.00	87.10 cA	77.88 cB	82.49	79.05 bA	71.94 cB	75.49
General Mean		201.41			45.42			93.01			83.01		
CV (%)[3]		20.47			19.17			7.64			8.27		

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

The pulp yield (Table 1) found in this study was higher than 35%, an essential characteristic for fruits destined to the juice industry (FARIAS et al., 2005), where the highest values were provided by grafted plants for presenting smaller fruits and consequently smaller length and width. Cavichioli et al. (2011) did not evidenced significant differences in pulp yield in fruits from grafted and ungrafted plants. Among the maturation stages, no differences were observed, only among the collection sites, where the fruits from Coronel Ezequiel presented lower pulp yield.

For length and width, the fruits collected in Coronel Ezequiel stood out in relation to the others due to higher values, while those from Maisa were the lowest (Table 1). In general, the fruits of grafted plants were smaller than those of ungrafted plants (weight, length and width). However, they presented higher pulp yield. Cavichioli et al. (2011), studying yellow passion fruit of grafted and ungrafted plants, evidenced greater fruits in ungrafted plants, 109.1 mm in length and 82.4 mm in width. Fruits of grafted plants obtained an average of 99.2 mm in length and 80.7 mm in width. Fruits in the green and yellow maturation stage of *P. edulis* from Pau Branco and Maisa showed statistically significant differences, where the yellow fruits were smaller than the green ones. This loss probably occurred due to the wrinkling of the fruits, depreciating the appearance. A higher rate of transpiration at room temperature associated with low relative humidity may have facilitated loss of water to the environment (CHITARRA, CHITARRA, 2005).

No statistically significant differences were observed for pulp weight and peel thickness between the two maturation stages collected, only among the collection sites was observed differences (Table 2).

Table 2. Pulp weight and peel thickness of passion fruit in the green and yellow stages from different collection sites.[1]

Maturation stage		Characteristics	
		Pulp weight (g)	Peel thickness (mm)
Green		91.60 a	9.42 a
Yellow		85.52 a	9.01 a
Local	**System**[2]		
C. Ezequiel	C	122.34 a	9.70 a
Jaguaruana	O+G	87.87 b	10.82 a
Pau Branco	C+G	79.41 bc	6.01 b
Maisa	C+G	64.62 c	10.82 a
General Mean		88.56	9.22
CV (%)[3]		28.98	28.09

[1] Means followed by the same lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

The pulp weight found for fruits of Coronel Ezequiel was the highest among all, corroborating with the data of fruit weight with a mean of 122.34 g, with an increase of 89.32%, followed by the fruits of Jaguaruana, Pau Branco and Maisa, with averages of 87.87 g, 79.41 g and 64.62 g, respectively (Table 2). These data corroborate with fruit weight. Thus, it is possible to observe that the fruits of ungrafted plants showed higher production, with fruit weight, length, width and pulp weight superior. However, pulp yield was lower. Results of Salazar et al. (2015), when evaluated fruits of *P. edulis* grafted on *P. gibertii* were similar to those of Coronel Ezequiel, with 130.46 g. However, the values of all localities were lower than those found by Cavichioli et al. (2011), which studied yellow passion fruits grafted on *P. edulis*, *P. alata* and *P. gibertii*, and ungrafted, with 218.44 g, 223.04 g, 199.68 g and 218.00 g, respectively. Statistically, these values did not differ from each other.

The peel thickness of the fruits of Coronel Ezequiel, Jaguaruana and Maisa were similar, differing from those of Pau Branco, presenting a value of 6.01 mm. Krause et al. (2012) observed, in cultivars of yellow passion fruit, peel thickness between 6.4 and 7.0 mm. Peel thickness is an important factor to be noticed because it is inversely proportional to the juice yield (FERREIRA et al., 2010), although this fact was not observed in this work, where the peel thickness did not influence the pulp yield. Fruits from grafted plants with greater thickness showed higher yield.

In the firmness of the fruits a reduction of the values was observed with the change of the stage of maturation from green to yellow, with a mean reduction of 24.41% (Table 3), however this reduction was not significant for fruits from Pau Branco. This is probably due to the action of pectinolytic enzymes present in the cell wall, which has its action intensified during maturation (LIEW, CHIN & YUSOF, 2014). Among the localities, fruits of Coronel Ezequiel, with conventional cultivation without grafting, obtained a firmness of 55.89 N in the stage of yellow maturation, being statistically different from the other fruits. In the green stage, the fruits of Coronel Ezequiel and Maisa, with conventional cultivation with grafting, stood out in relation to the others, with firmness of 73.20 N and 77.20 N, respectively. Linares, Castillo and Londoño ou Logroño (2013), studying the mechanical characteristics of passion fruit, found the value of 84.3 N for the most mature stage and 119.3 N for the green stage.

Table 3. Firmness of passion fruit in the green and yellow stages from different collection sites.[1]

Local		Firmness (N)	
	System[2]	Green	Yellow
C. Ezequiel	C	73.20 aA	55.89 aB
Jaguaruana	O+G	53.51 bA	44.07 bB
Pau Branco	C+G	54.52 bB	48.55 abB
Maisa	C+G	77.20 aA	42.13 bB
General Mean		56.13	
CV (%)[3]		23.61	

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

In peel color, there was a statistically significant difference between maturation stages and between localities (Table 4). For the values of luminosity and chromaticity, there is an increase with the advance of the maturation, according to the increment of the yellow color. Already for the values of the hue angle there was a reduction, with the change of color to a shade of darker yellow. In all localities, except Maisa in the yellow maturation stage, hue angle was located inside the second quadrant (>90°), that is, yellow color, determining greater intensity. Similar values were found by Salazar et al. (2015), with luminosity varying from 70.82 to 71.94; chromaticity from 38.88 to 43.15; and hue angle of 97.20 to 100.43. The increase in yellow color occurs due to the

degradation of chlorophyll, while yellow, orange and red pigments belonging to the group of carotenoids are revealed or synthesized. Such pigments are quite common, and their presence is a signal through the consumer and industry evaluate the maturity and quality of the fruits (FREIRE et al., 2014). The fruits of the locality of Pau Branco were highlighted with the highest values of luminosity and chromaticity and Coronel Ezequiel and Maisa with the values of hue angle.

Table 4. Luminosity (L*), chromaticity (C*) and Hue angle (°H) of the peel of passion fruit in the green and yellow stages from different collection sites.[1]

Local		Characteristics					
		Peel luminosity		Peel chromaticity		Peel °H	
	System[2]	Green	Yellow	Green	Yellow	Green	Yellow
C. Ezequiel	C	57.50 dB	73.49 bA	35.51 bB	54.41 aA	109.88 aA	92.29 aB
Jaguaruana	O+G	71.50 bB	80.46 aA	49.64 aB	55.23 aA	101.48 bA	90.29 aB
Pau Branco	C+G	75.67 aB	84.21 aA	50.25 aB	54.20 aA	99.51 bA	90.59 aB
Maisa	C+G	67.04 cB	82.11 aA	39.87 bB	53.61 aA	110.58 aA	89.47 aB
General Mean		73.99		49.09		98.08	
CV (%)[3]		6.28		12.14		5.45	

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

For pulp color, significant differences were observed only between the collection sites at the stage of green maturation (Table 5). For values of luminosity, the fruits of Maisa presented the highest value (70.25); while the fruits of Coronel Ezequiel showed the highest value of chromaticity (61.34). The results obtained by this work were superior to those found by Salazar et al. (2015), where the luminosity ranged from 42.04 to 47.66 and the chromaticity ranged from 10.00 to 13.39. Pulps of the fruits produced in the region demonstrate greater intensity of color.

Table 5. Luminosity (L*), chromaticity (C*) and Hue angle (°H) of the pulp of passion fruit in the green and yellow stages from different collection sites.[1]

Maturation stage		Characteristics				
		Pulp luminosity		Pulp chromaticity		Pulp °H
		Green	Yellow	Green	Yellow	
Green		-	-	-	-	82.12 a
Yellow		-	-	-	-	80.80 a
Local	**System**[2]					
C. Ezequiel	C	65.53 bA	64.15 aA	61.34 aA	61.98 aA	75.47 b
Jaguaruana	O+G	67.24 abA	66.80 aA	60.82 aA	62.86 aA	82.54 a
Pau Branco	C+G	62.82 bB	67.13 aA	53.71 bB	60.69 aA	83.56 a
Maisa	C+G	70.25 aA	65.94 aB	60.23 aA	59.46 aA	84.27 a
General Mean		66.36		60.14		81.46
CV (%)[3]		6.59		9.09		6.49

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

In the hue angle of the pulp, the values of all the fruits were located within the first quadrant (from zero to 90°), that is, red to yellow color, with values of 75.47 for Coronel Ezequiel and 82.54 to 84.27 for the other sites, tending to a more yellowish coloration and greater intensity of red, respectively. Fruit staining may be a parameter used as an indicator of fruit quality for industrialization, with preference for fruits having a stable yellow-gold color (MANIWARA et al., 2014). Salazar et al. (2015) found values of 84.72 to 94.45 for hue angle of the pulp in yellow passion fruits of plants grafted on wild species.

3.2 Chemical Characteristics

There was no significant difference for total soluble solids (SS) and total soluble sugars (TSS) when observed the maturation stages. However, a statistical difference was observed for the collection site for all quality variables. There was also a significant interaction between the analyzed factors (collection site and maturation stage) for titratable acidity ($p < 0.01$) and for the SS/TA ratio ($p < 0.01$) (Table 6).

Table 6. Total soluble solids (SS), hydrogenation potential (pH), reducing sugars (RS), total soluble sugars (TSS), titratable acidity (TA) and SS/TA ratio of passion fruit in the green and yellow stages from different collection sites.[1]

Maturation stage		Characteristics							
		SS (°Brix)	pH	RS (%)	TSS (%)	TA (mg/100 g)		SS/TA	
						Green	Yellow	Green	Yellow
Green		13.86 a	3.18 b	5.03 b	8.93 a	-	-	-	-
Yellow		14.17 a	3.24 a	6.13 a	8.18 a	-	-	-	-
Local	**System**[2]								
C. Ezequiel	C	11.95 c	3.05 c	2.82 c	6.66 b	4.85 aA	4.89 aA	2.30 bA	2.61 bA
Jaguaruana	O+G	14.92 b	3.26 ab	6.99 a	9.83 a	3.37 cA	3.44 bA	4.42 aA	4.38 aA
Pau Branco	C+G	12.59 c	3.20 b	4.70 b	7.54 b	3.37 cB	3.80 bA	3.86 aA	3.24 bA
Maisa	C+G	16.61 a	3.33 a	7.80 a	10.20 a	4.14 bA	3.31 bB	4.05 aB	5.11 aA
General Mean		14.01	3.21	5.58	8.56	3.90		3.75	
CV (%)[3]		8.30	1.80	22.27	16.85	7.72		13.76	

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

The pH, total soluble solids and reducing sugars increased with the advancement of the maturation stage. These facts can be attributed to the hydrolysis of starch to sugars, according to the behavior of carbohydrates during ripening (PINZON et al., 2007). Among the collection sites, the fruits from Maisa, with the conventional with grafting system, were those that presented higher values for these variables and for total soluble sugars. In relation to the production system, the fruits of the localities with grafted plants obtained a higher quality, having statistically significant differences when compared to those coming from cultivation without grafting. Lower values for soluble solids were verified by Junqueira et al. (2006), whose results were 10.8 °Brix in ungrafted passion fruit and 11.7 ° Brix in fruits of plants grafted on *Passiflora nitida*. Jiménez et al. (2011), studying yellow passion fruit in different stages of maturation, found for soluble solids the values of 13.5 and 17.4 °Brix in the stages of green and yellow maturation, respectively. For pH, they found values of 2.45 and 2.77% for the same stages.

The values of titratable acidity were higher in the fruits of Coronel Ezequiel, with conventional cultivation without grafting, with 4.85 and 4.89 mg/100g for the green and yellow stages, respectively. The titratable acidity varies due to the consumption of organic acids during fruit respiration (PINZÓN et al., 2007). Moreover, high levels of acids in the juice are important in terms of processing, since it allows greater flexibility in the addition of sugar, when preparing ready-made beverages, besides conferring conditions that hinder deterioration by microorganisms (FLORES et al., 2011). Greco et al. (2014) verified in industrialized juices of yellow passion fruit titratable acidity of 2.7 to 3.9 mg/100g, approximate values to those found in this work. Cavichioli et al. (2011) and Salazar et al. (2015) found that the titratable acidity of fruits of grafted plants did not differ statistically from those of ungrafted plants. However, in this work it was possible to observe that the fruits of Coronel Ezequiel, without grafting, presented higher acidity.

The SS/TA ratio, in the present work, varied between the collection sites and the stages of maturation. The fruits of Jaguaruana, Pau Branco and Maisa (all with grafting) stood out with the highest values, due to highest values of titratable acidity, close to those found by Cavichioli et al. (2011), ranging from 2.8 to 3.5. This relation can be influenced by environmental factors such as sunlight intensity and temperature, type and fertilizer doses (NASCIMENTO et al., 2003). The analysis of the balance between soluble solids and acids in the fruit has utility in the evaluation of the taste and in the

determination of stages of maturation, since its value tends to increase as the fruit reaches the point of harvest (CHITARRA, CHITARRA, 2005).

3.3 Bioactive compounds

According to the analysis of variance for the bioactive compounds and antioxidant activity, there was no significant difference for the factors β-carotene and total carotenoids ($p>0.05$). However, differences were observed for anthocyanins considering the collection sites ($p<0.01$) and maturation stage ($p<0.05$) and for vitamin C and TAA by ABTS only differed between the collection sites ($p<0.01$) (Table 7). There was a significant interaction between the analyzed factors (collection site and maturation stage) for TEP ($p<0.01$), flavonoids ($p<0.01$) and for TAA by DPPH ($p<0.01$) (Table 8).

Table 7. Vitamin C, β-carotene, total carotenoids, anthocyanins and TAA by ABTS method of passion fruit in the green and yellow stages from different collection sites.[1]

Maturation stage		Characteristics				
		Vitamin C	β-carotene	Total carotenoids	Anthocyanins	ABTS
		----------mg/100 g----------				(µmol Trolox/g)
Green		40.44 a	0.20 a	1.12 a	0.49 b	1.73 a
Yellow		40.71 a	0.20 a	1.20 a	0.63 a	1.63 a
Local	**System**[2]					
C. Ezequiel	C	43.02 a	0.23 a	1.31 a	0.57 ab	1.81 ab
Jaguaruana	O+G	41.66 ab	0.20 a	1.26 a	0.40 b	1.96 a
Pau Branco	C+G	37.14 c	0.18 a	0.92 a	0.67 a	1.63 ab
Maisa	C+G	40.47 ab	0.20 a	1.15 a	0.61 a	1.31 b
General Mean		40.57	0.20	1.16	0.56	1.68
CV (%)[3]		11.67	26.59	31.21	30.26	27.78

[1] Means followed by the same lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

Among the stages of green and yellow maturation, no statistically significant differences were found for the contents of vitamin C, β-carotene, total carotenoids and TAA by ABTS method. Only in anthocyanin content, in the yellow stage was 28.57% higher than green.

For vitamin C, the fruits collected in Coronel Ezequiel, Jaguaruana and Maisa, with values of 43.02; 41.66 and 40.47 mg/100g, respectively, were emphasize in relation to the fruits of Pau Branco, with a value of 37.14 mg/100g. This difference may have been due to the management used in this commercial area, since the cultivation system is similar to Maisa's. These values can be promissory, since in all the fruits contents were obtained above 20 mg/100 g, the minimum necessary for the market of yellow passion fruit (SANTOS et al., 2009). According to Ramful et al. (2011), fruits may be classified according to vitamin C content in three categories: low (30 mg/100g), medium (30-50 mg/100g) and high (>50mg/100g). In this classification, the results of this work are considered medium. Salazar et al. (2016), studying fruits from grafted and ungrafted passion fruit, found values of 29.37 to 36.06 mg/100 g, respectively. Pertuzatti et al. (2015) found in passion fruit produced under organic cultivation the value of 230 mg/100 g and 190 mg/100 g in fruits of conventional cultivation, using high-performance liquid chromatography (HPLC). The divergence with the data found in this work may be due to the different methodology used.

No statistically significant differences were found in the contents of β-carotene and total carotenoids among the fruits of the collection sites. The beta carotene content in this work ranged from 0.18 to 0.23 mg/100g. In yellow passion fruit, Reis et al. (2018) found contents of 1.33 mg/100 g. Pertuzatti et al. (2015) found the concentrations of 0.056 mg/100 g for fruits from organic cultivation and 0.077 mg/100 g from conventional cultivars. These authors attribute this variation not only to the effect of geographic distance, but also due to the maturation stage of the fruits at the moment of the analysis, crop year, variety and storage conditions. The same may have occurred in this work.

In the present study, the total carotenoid content ranged from 0.92 to 1.31 mg/100 g, with no statistically significant effect. The content of 1.78 mg/100 g was demonstrated by Reis et al. (2018) and 13.99 and 25.10 mg/100 g by Pertuzatti et al. (2015) in organic and conventional fruits, respectively. The content of total carotenoids in vegetables cannot be considered an absolute value, and may be affected by several factors. As a result of various functions or properties attributed to carotenoid production, there is a global effort to obtain reliable analytical data that provides deeper demonstration of the development of these compounds. As there are a large number of natural carotenoids, conclusive identification is hampered. Carotenoid composition in fruits is often incomplete or conflicting in the literature (MERCADANTE et al., 1998).

Among the maturation stages, the yellow fruits, with a more advanced stage of maturation, obtained in general a greater content of anthocyanins. Among the collection sites, those with conventional cultivation presented higher values, statistically significant, varying between 0.40 and 0.67 mg/100 g. Jiménez et al. (2011) found a content of 4.5 mg/100 g. Already Reis et al. (2018), with the methodology used, failed to show anthocyanins in the yellow passion fruit pulp.

The presence of phenolic compounds in yellow passion fruit makes this fruit an excellent candidate to evaluate several effects "in vivo". In Table 8, it is possible to observe that between the stages of maturation, in general, no differences were evidenced, where only in the fruits of Jaguaruana there was an expressive increase of the green to yellow stage. Similarly, among the collection sites there was no difference, except for the concentration of the yellow fruits of Pau Branco, which obtained the lowest value. Rotili et al. (2013), studying yellow passion fruit at different times of storage, found at time 0 the concentration of 20.2 mg/100 g of total extractable polyphenols. Septembre-Malaterre et al. (2016) observed the value of 286.6 mg/100 g. The phenolic composition of the fruits is determined by genetic and environmental factors, such as, for example, grafting (ROBARDS et al., 1999). However, it was not possible to observe such differences in this work.

Table 8. TEP, flavonoids and TAA by DPPH method of passion fruit in the green and yellow stages from different collection sites.[1]

Local	Caracteristics						
		TEP (mg/100 g)		Flavonoids (mg/100 g)		DPPH (g fruit/g DPPH)	
	System[2]	Green	Yellow	Green	Yellow	Green	Yellow
C. Ezequiel	C	58.98 aA	63.52 aA	5.80 aA	6.11 aA	38331.47 bA	34151.10 bA
Jaguaruana	O+G	52.82 aB	64.91 aA	2.97 bA	3.33 bA	70496.30 aB	52582.92 abA
Pau Branco	C+G	54.80 aA	46.14 bA	2.72 bB	4.05 bA	40260.84 bB	61341.31 abA
Maisa	C+G	64.88 aA	66.02 aA	3.89 bB	6.73 aA	53289.96 abA	56094.73 aA
General Mean		59.01		4.45		50818.58	
CV (%)		12.50		22.56		27.45	

[1] Means followed by the same capital letter in the line and lower case in the column do not differ, according to the Tukey test at 5% probability. [2] C = Conventional; O+G = Organic and grafted; C+G = Conventional and grafted. [3] Coefficient of variation.

The content of flavonoids in the yellow maturation stage was higher than in the green stage, where there was a yellow color development. The fruits of Coronel Ezequiel and Maisa showed the highest concentrations in both maturation stages. Zeraik

and Yariwake (2010) found the value of 0.167 mg/100 g in fruits of *P. edulis*. The concentration of 16.28 mg/100g was found by Reis et al. (2018). The results obtained in this work suggest that fruits of *P. edulis* can be compared with other sources rich in flavonoids, such as orange juice (REIS et al., 2018).

In the antioxidant activity by the ABTS method (Table 7), the fruits of Coronel Ezequiel, Jaguaruana and Pau Branco presented greater capacity to neutralize free radicals. However, superior results were found by Talcott et al. (2003), studying hydrophilic fractions containing polyphenols (14 μmol Trolox/g). In this work, only the pulp of the fruit was used. Reis et al. (2018) detected in yellow passion fruit pulp an antioxidant capacity of 121.9 μmol Trolox/g.

By the DPPH method, it was observed that with the progress of the maturation stage from green to yellow there is a reduction of the antioxidant capacity of the pulp (Table 8). However, in the fruits of Jaguaruana there was an increase, statistically significant. Regarding the sites, in the green stage, the fruits of Coronel Ezequiel and Pau Branco obtained the highest activities, while in the yellow stage this was evidenced in Coronel Ezequiel and Jaguaruana. Septembre-Malaterre et al. (2016) concluded that the highest antioxidant capacity was found in yellow passion fruit pulp (64% of reduced DPPH) when compared to other fruits, including mango, pineapple and banana. López-Vargas et al. (2013), evaluating the antioxidant capacity by the DPPH method in pulp and seed of yellow passion fruit, concluded that the seeds have higher activity. The antioxidant activity in plants is due to the action of a large variety of antioxidant compounds, which are degraded or synthesized according to the physiological state and levels of abiotic and biotic stresses experienced by the organ (ROTILI et al., 2013).

4 CONCLUSION

Fruits in the stage of yellow maturation (point of consumption) have better characteristics for the consumer market.

The use of grafted plants does not reduce the physical, chemical qualities and bioactive compounds of fruits, where pulp yield, luminosity of peel, soluble solids, pH, SS/TA ratio, total soluble sugars and reducing sugars were increased in these fruits, with higher quality.

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