UNIVERSIDADE ESTADUAL PAULISTA
"JÚLIO DE MESQUITA FILHO"
Campus de Botucatu

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MÔNICA BARTIRA DA SILVA

POTENCIAL NUTRICIONAL DA JURUBEBA (*Solanum paniculatum* L.) SUBMETIDA AO PROCESSAMENTO TÉRMICO E AO USO DE CONSERVANTES

BOTUCATU-SP
2017

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Tese apresentada à Faculdade de Ciências Agronômicas da UNESP - Câmpus de Botucatu, para obtenção do título de Doutor em Agronomia / Horticultura

BOTUCATU-SP
2017

FICHA CATALOGRÁFICA ELABORADA PELA SEÇÃO TÉCNICA DE AQUISIÇÃO E TRATAMEN-TO DA INFORMAÇÃO – DIRETORIA TÉCNICA DE BIBLIOTECA E DOCUMENTAÇÃO - UNESP – FCA – LAGEADO – BOTUCATU (SP)

Silva, Mônica Bartira, 1989-
S586p Potencial nutricional da jurubeba (Solanum paniculatum l.) submetida ao processamento térmico e ao uso de conservantes / Mônica Bartira da Silva.– Botucatu : [s.n.] 2017
81 p. : grafs., tabs.

Tese (Doutorado) - Universidade Estadual Paulista, Faculdade de Ciências Agronômicas, Botucatu, 2017
Orientador: Giuseppina Pace Pereira Lima
Inclui bibliografia

1. Compostos fenólicos. 2. Solanaceae. 3. Plantas me- dicinais – Uso terapêutico. I. Lima, Giuseppina Pace Pe- reira. II. Universidade Estadual Paulista "Júlio de Mes- quita Filho" (Câmpus de Botucatu). Faculdade de Ciências Agronômicas. III. Título.

UNIVERSIDADE ESTADUAL PAULISTA

Câmpus de Botucatu

CERTIFICADO DE APROVAÇÃO

TÍTULO: POTENCIAL NUTRICIONAL DA JURUBEBA (*Solanum paniculatum* L.) SUBMETIDA AO PROCESSAMENTO TÉRMICO E AO USO DE CONSERVANTES

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Botucatu, 11 de abril de 2017.

Aos meus pais, por não me aconselharem

a deixar de lado desenhos de jiboias abertas.

Dedico

AGRADECIMENTOS

À Faculdade de Ciências Agronômicas – UNESP, Campus de Botucatu, especialmente ao Corpo docente do Programa de Pós Graduação em Agronomia (Horticultura), pelos ensinamentos.

Ao Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) pela concessão da bolsa de doutorado (Processo CNPq 142360/2013-9).

A Fundação de Amparo a Pesquisa do Estado de São Paulo (FAPESP) pelo financiamento do projeto (2013/05644-3).

À minha família especialmente aos meus pais, Maurilio José Da Silva e Maria Gorete da Silva, por que não existem palavras que possam representar o meu agradecimento e admiração por vocês.

À Giuseppina Pace Pereira Lima, por ser uma mentora sabia, dedicada e acima de tudo ser uma grande amiga.

Agradecer é a maneira mais sensata de reconhecer o esforço que outros tiveram por nós e por isso demonstro aqui a minha gratidão por todos aqueles que dedicaram um tempo de suas vidas para me ajudar durante a condução desta tese, em especial ao Luan Fernando O. S. Rodrigues que além de ser um grande companheiro na vida, ainda me ajudou nas análises, sendo essencial em cada etapa.

Às minhas dedicadas estagiárias Ana Paula C. R. Ferraz, Talita Cardoso Rossi e Larissa Ambrósio de Andrade.

Àos meus amigos e companheiros de laboratório, Rene Campos, Marizete Cavalcante e Milena Borguini por me ensinarem as análises.

Aos meus amigos e companheiros de campo Luiz Felipe Guedes Baldini e Gean Charles Monteiro porque com vocês o serviço sempre rende.

Ao Professor e colega Igor Otávio Minatel pela disponibilidade, ajuda e concelhos.

Aos estagiários Matheus e Guilherme que embora não fossem “meus estagiários” sempre me atenderam e ajudaram.

Ao professor Santino Seabra Junior por disponibilizar os locais de coleta dos frutos.

A Professora Camila Renata Correâ pela ajuda nas análises de carotenoides.

A equipe do Laboratório de Química e Bioquímica Vegetal (LQBV) em especial aos colegas, Debora Pado, Maria Izabela, Sergio Marques, Ewerton Gasparetto, Cristine Borges, Carla Souza, Marla Diamante, Kamila Monaco, Aline Gouveia, Hector Gomez Gomez, Giovana Monnar, Andreia Dutra e Evandro Tadeu.

Aos funcionários do Departamento de Química e Bioquímica do Instituto de Biociências (IBB) em especial a Gabriela Valim.

Os meus sinceros agradecimentos

RESUMO

Foram conduzidos 3 experimentos avaliando o processamento térmico de jurubebas e seu efeito nos níveis de antioxidantes. O primeiro avaliou frutos de jurueba *in natura* e processados termicamente em diferentes tempos de cozimento (10, 20, 30, e 40 minutos), esses frutos foram preservados em óleo de soja ou vinagre de álcool e avaliados quanto as características físicas [pH, sólidos solúveis (SS), acidez titulável (AT) e a relação SS/AT], fitoquímicos (clorofilas, carotenoides, fenois totais e flavonoides totais), capacidade antioxidante (DPPH/TEAC) e poliaminas (PAs). Os dados mostram que o tratamento com cozimento por 20 minutos manteve a melhor qualidade do fruto. Posteriormente no segundo experimento os frutos de Jurubeba foram adquiridos de três formas diferentes (de plantas cultivadas, plantas espontâneas e no mercado) e estudados em relação às suas qualidades nutricionais e físico-químicas após processamento térmico e conservação. Parte destes frutos foi mantida in natura, e a outra foi submetida a cozimento por 20 min. Os frutos processados termicamente foram conservados em dois tipos de conservantes (óleo de soja e vinagre) armazenados e avaliados 1 hora após a preparação das conservas e após 30, 60 e 90 dias de prateleira quanto ao conteúdo de vitamina C, carboidratos totais, proteínas totais, lipídios totais, total flavonóides e fenóis. Tendo em consideração os flavonóides, os frutos adquiridos no mercado ou recolhidos a partir de plantas espontâneas e conservados em óleo ou vinagre são boas fontes até 90 dias. O terceiro experimento teve os mesmos tratamentos do segundo, contudo, avaliou-se o qualie quantitativamente as poliaminas. Foram detectadas variações nos conteúdos de espermina (0,02 a 3,11 mg/100 g), putrescina (18,41 a 86,48 mg/100 g), cadaverina (0,01 a 19,02 mg/100 g), espermidina (0,04 a 32,32 mg/100 g), histamina 0,01 a 8,43 mg/100g) e tiramina (0,16 a 11,74 mg/100 g) em função do local de obtenção dos frutos, assim como do tipo de conservante e do tempo de armazenamento.

Palavras-chaves: Fitoquímicos, Solanaceae, compostos fenolicos, aminas biogênicas, atividade antioxidante.

ABSTRACT

Three experiments were conducted evaluating the thermal processing of jurubebas and their effect on antioxidant levels. The first evaluated fruits of jurueba in natura and processed thermally in different cooking times (10, 20, 30, and 40 minutes), these fruits were preserved in soybean oil or alcohol vinegar and evaluated for physical characteristics [pH, solids (SSP), titratable acidity (AT) and SS / AT ratio, phytochemicals (chlorophylls, carotenoids, total phenotypes and total flavonoids), antioxidant capacity (DPPH / TEAC) and polyamines (PAs). The data show that the baking treatment for 20 minutes maintained the best fruit quality. Subsequently in the second experiment the fruits of Jurubeba were acquired in three different ways (from cultivated plants, spontaneous plants and on the market) and studied in relation to their nutritional and physical-chemical qualities after thermal processing and conservation. Part of these fruits was kept in natura, and the other was submitted to cooking for 20 min. The heat-processed fruits were stored in two types of preservatives (soybean oil and vinegar) stored and evaluated 1 hour after the preparation of the preserves and after 30, 60 and 90 days shelf life for vitamin C, total carbohydrates, total proteins , Total lipids, total Flavonoids and phenols. Taking into account flavonoids, fruits purchased on the market or collected from spontaneous plants and kept in oil or vinegar are good sources up to 90 days. The third experiment had the same treatments as the second one, however, it was evaluated quantitatively the polyamines. Changes in the contents of spermine (0.02 to 3.11 mg / 100g), putrescine (18.41 to 86.48 mg / 100g), cadaverine (0.01 to 19.02 mg / 100g), spermidine (0.04 to 32.32 mg / 100g), histamine 0.01 to 8.43 mg / 100g) and tyramine (0.16 to 11.74 mg / 100g) depending on the place of fruit picking, as well as the type of preservative and the time of storage.

Keywords: Phytochemicals, Solanaceae, phenol compounds, polyamines, aminas biogenic, antioxidanty atictivit.

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1 INTRODUÇÃO GERAL

Os alimentos são fonte de energia para o corpo humano, sendo essenciais para o desempenho das funções orgânicas. Uma alimentação saudável não necessariamente precisa ser restrita e monótona, pelo contrário, um dos pilares fundamentais para uma alimentação saudável é a diversidade de produtos, porque cada alimento contribui com um nutriente diferente e em quantidades distintas.

Não é novidade falar que frutas e hortaliças são importantes componentes de uma dieta saudável. Seu consumo, em quantidades adequadas, pode reduzir os riscos de doenças cardiovasculares e alguns tipos de cânceres (LOCK et al., 2005). Estimativas da organização mundial da saúde (OMS) apontam que a falta/baixo consumo de frutas e hortaliças estão entre os dez principais fatores de risco para a carga total de doenças em todo o mundo (WORLD HEALTH ORGANIZATION, 2002).

Ao observar os fatores associados ao consumo de frutas e hortaliças no Brasil, foi constatado que são necessárias iniciativas de promoção do consumo destes alimentos voltadas à população geral, visto que, a ingestão deles esteve aquém das recomendações atuais de no mínimo 400 g diárias. Outro ponto relativo é que deve ser dada atenção especial às cidades da região norte e nordeste, aos indivíduos jovens, ao sexo masculino e a população com baixa escolaridade (Jaime et al., 2009).

Em um levantamento realizado foi observado que o consumo brasileiro de frutas e hortaliças é equivalente a 66,8 g/dia, valor este, bastante inferior a recomendação do Food And Agriculture Organization (FAO). Contudo cabe ressaltar que no cardápio empregado nesta pesquisa foram utilizados apenas 12 alimentos (abacaxi, banana, laranja, mamão, manga, tangerina, batata, brócolis, cebola, cenoura, repolho e tomate) não sendo incluídos alimentos regionais ou hortaliças não-convencionais (Faller e Fialho 2009)

Uma importante estratégia de complementação a dieta alimentar pode ser o consumo de hortaliças não convencionais. Essas hortaliças possuem baixo custo, fácil disponibilidade e valor nutritivo, atuando como uma alternativa para a melhoria do conteúdo de alguns compostos e micronutrientes na dieta de pessoas de pouco poder

aquisitivo, substituindo alimentos de alto custo e, talvez, menor disponibilidade (MINISTERIO DA SAÚDE, 2002).

Outra importante contribuição ao incentivar o consumo e comercialização destas hortaliças é a geração de renda a agricultura familiar, previsto por lei no no Art.2° (Inciso I, II e VIII), que integra o Sistema Nacional de Segurança Alimentar e Nutricional – SISAN, instituído pela Lei nº 11.346, de 15 de setembro de 2006, e tem as seguintes finalidades: I – incentivar a agricultura familiar, promovendo a sua inclusão econômica e social, com fomento à produção com sustentabilidade, ao processamento, à industrialização de alimentos e à geração de renda; II – incentivar o consumo e a valorização dos alimentos produzidos pela agricultura familiar; VIII – promover e valorizar a biodiversidade e a produção orgânica e agroecológica de alimentos, e incentivar hábitos alimentares saudáveis em nível local e regional (BRASIL, 2006).

Dentre esses alimentos com potencial comercial e nutricional, pode-se destacar a *Solanum paniculatum* L. (Solanaceae), conhecida popularmente como jurubeba, jurupeba, juripeba, jubeba, juvena, juina ou juna (CORREIA, 1984) é uma planta amplamente utilizada na medicina popular brasileira (COIMBRA, 1958), sendo nativa das regiões norte e nordeste do Brasil (MAPA, 2010).

As formas de consumo desta hortaliça vão desde a infusão de folhas, frutos e flores, suco com as raízes e frutos até o consumo dos frutos em forma de conservas, ou cozidos junto com outros alimentos. Entretanto a jurubeba se destaca especialmente pelos seus diferentes usos medicinais, por sua distribuição ampla e, por ser um representante de *Solanum* reconhecido como fitoterápico pela Farmacopéia Brasileira, segundo a Farmacopeia dos Estados Unidos do Brasil (1959).

Solanum paniculatum é um arbusto com altura variando de 1,0 m a 1,5 m, revestido de indumento alvo-tomentoso a cinéreo, constituído basicamente de tricomas porrecto-estrelados, sésseis ou estipitados, com o raio central reduzido, unicelular. Possui raiz ramificada em crescimento secundário inicial, com xilema em estrutura hexarca.

Suas folhas são largo-ovadas a lanceoladas, com a margem lobada ou inteira, com acúleos cônicos; a epiderme da lâmina, em vista frontal, apresenta células com paredes anticlinais poligonais, retas na face adaxial e sinuosas na face abaxial; o

pecíolo, em secção transversal, exibe contorno levemente biconvexo, e o sistema vascular é formado por quatro a cinco feixes bicolaterais (NURIT et al., 2007).

A maioria das plantas do gênero *Solanum* apresenta saponinas esteroidais, glicoalcaloides e flavonoides que são importantes na defesa natural das plantas como metabolitos secundários (OLIVEIRA et al., 2006).

A avaliação do potencial terapêutico de algumas plantas medicinais e seus constituintes, tais como flavonoides, alcaloides, triterpenos, sesquiterpenos, taninos e ligninas tem sido objetivo de incessantes estudos (HAUSTEEN, 1983) e diante do potencial diversificado dessa cultura vários trabalhos foram realizados, como a viabilidade e a germinabilidade polínica de populações de jurubeba (SANTOS NETO et al., 2006)**;** avaliação da atividade antioxidante (RIBEIRO et al., 2007); a ausência de mutagenicidade (RIBEIRO et al., (2009); o potencial anti-helmintico das raiz em ovelhas do semiárido paraibano (VILELA et al., 2009); a atividade antibacteriana e prospecção fitoquímica do extrato da raiz (LOBÔ et al., 2010) dentre outros.

Ramos et al. (2012) observaram para esta cultura rendimento do óleo essencial das folhas obtidos por hidrodestilação de 0,04%, identificando o nerolidol como componente majoritário (54,3%). Além do nerolidol os autores identificaram três outros compostos: verbenono (1,1%), β-ionona (4,3%) e tricosane (38,3%) correspondentes a 98,0% da composição total.

O nerolidol é um sesquiterpeno usado como agente aromatizante pelas industrias alimentícias e como fixador natural pelas industrias de cosméticos (FRIZZO, 2000). Nogueira Neto et al. (2013) trabalhando com o potencial antioxidante in vitro do nerolidol, observaram que ele apresenta a capacidade de diminuir significativamente a produção de nitrito, a formação do radical hidroxila a produção de ácido tiobarbitúrico, demonstrando potencial atividade antioxidante protegendo as biomoléculas contra danos causados por radicais livres.

Dentre os antioxidantes existem cerca de 8.000 compostos fenólicos que são largamente distribuídos no reino vegetal, influenciando significativamente na qualidade de frutos e hortaliças por contribuírem sensorialmente e nutricionalmente (SCALZO et al., 2005). Os compostos fenólicos são agrupados em flavonoides e não flavonoides

(ácidos fenólicos e cumarinas). Segundo Reynerston et al. (2008), os polifenois de frutas são importantes componentes antioxidantes da dieta alimentar.

De maneira simplificada o termo antioxidante significa “que inibe os efeitos da oxidação”, esse processo foi primeiramente observado por Claude Berthollet em 1797 e depois esclarecido por Humphry em 1817 (HOUASIS, 2001).

A produção de radicais livres é controlada nos seres vivos por diversos compostos antioxidantes. Estes podem ser de origem endógena, ou proveniente da dieta alimentar e outras fontes. Quando ocorre uma limitação na disponibilidade de antioxidantes em humanos podem ocorrer lesões oxidativas de caráter cumulativo. Muitas evidências têm mostrado que os radicais livres e outros oxidantes são responsáveis pelo envelhecimento e pelas doenças degenerativas como câncer, doenças cardiovasculares, cataratas, disfunções cerebrais, entre outras (ATOUI et al., 2005).

Os antioxidantes podem ser classificados em: antioxidantes primários, que são compostos fenólicos capazes de remover ou inativar os radicais livres de reações (polifenois e tocofenois), antioxidantes sinergistas, que apresentam maior atividade antioxidante quando combinado com antioxidantes primários, antioxidantes removedores de oxigênio, que capturam o oxigênio presente no meio (ácido ascórbico, seus isômeros e derivados), antioxidantes biológicos, que removem o oxigênio ou compostos altamente reativos de um sistema alimentício (glicose oxidase, superóxido dismutase e catalase), agentes quelantes, que complexam íons metálicos e catalisam a oxidação lipídica (ácido cítrico e seus sais, fosfatos e sais de acido etileno diamino tetra acético) e antioxidantes mistos, onde são incluídos os compostos de plantas e animais (proteínas hidrolisadas, flavonoides) e derivados de ácido cinâmico (FOOD INGREDIENTS BRASIL, 2009).

Os extratos etanólicos das folhas de *S. paniculatum* apresentaram um fracionamento conduzindo a uma redução da atividade antioxidante, o que indica que os compostos responsáveis pela mesma não podem ser provenientes de um composto cuja atividade antioxidante é o resultado de uma ação sinérgica (Ribeiro et al., 2007)

Os mesmos autores ainda observaram que o fracionamento por solventes imiscíveis do extrato aquoso bruto de folhas de *S. paniculatum* permitiu a obtenção de

duas frações com capacidade antioxidante equivalente ao butyl-hidroxi-tolueno (BHT), contudo há poucas informações disponíveis que abordam os teores de compostos bioativos em frutos de *S. paniculatum in natura* e processados termicamente.

As hortaliças são, muitas vezes, consumidas na forma crua, porém há situações em que a cocção é necessária ou ainda preferida (CAMPOS et al., 2008). A forma como o alimento é consumido pode interferir na sua capacidade antioxidante, seja na forma *in natura* ou processado.

Kaur e Kappor (2001) consideram que o tratamento térmico é a principal causa da alteração do teor de antioxidantes naturais em alimentos. O processamento e os procedimentos para a preservação dos alimentos podem ser responsáveis tanto pelo aumento quanto pelo decréscimo da ação antioxidante, dependendo de muitos fatores, tais como: estrutura química, potencial de oxirredução, sua localização na matriz e possíveis interações com outros componentes do alimento (NICOLI et al., 1999).

No processamento térmico, o calor empregado pela cocção, pode inativar a ação da enzima peroxidase, que atuam como pró-oxidantes (TURKEN et al., 2005). Contudo o processo de cocção contribui para a formação de novos compostos, como os produtos da reação de Maillard (redutonas), que apresentam ação antioxidante, porém no estágio inicial desta reação ocorre a formação de radicais livres bastantes reativos podendo atuar como pró-oxidantes (NICOLI et al., 1999).

As informações a respeito da composição nutricional dos frutos de *S. paniculatum in natura* e submetidas a tratamentos térmicos, ainda são escassas, sendo necessário a desenvolvimento de pesquisas avaliando não só a composição nutricional destes frutos como também a resposta deles ao processamento térmico, as diferentes formas de preparo e o seu tempo de prateleira.

2 EFEITO DO TEMPO DE COZIMENTO E DE CONSERVADORES EM FRUTOS DE JURUBEBA (*Solanum paniculatum* L.)

Mônica Bartira da Silva, Luan Fernando Ormond Sobreira Rodrigues, Talita Cardoso Rossi, Marizete Cavalcante de Souza Vieira, Igor Otávio Minatel, Giuseppina Pace Pereira Lima.

RESUMO

Frutos de jurueba *in natura* e processados termicamente em diferentes tempos de cozimento (10, 20, 30, e 40 minutos), foram preservados em óleo de soja ou vinagre de álcool e avaliados quanto as características físicas [pH, sólidos solúveis (SS), acidez titulável (AT) e a relação SS/AT], fitoquímicos (clorofilas, carotenoides, fenois totais e flavonoides totais), capacidade antioxidante (DPPH/TEAC) e poliaminas (PAs). Os dados mostram que o tratamento com cozimento por 20 minutos manteve a melhor qualidade do fruto, sendo este tempo de cozimento ideal para o processamento de jurubeba. Seguindo este tratamento, nenhuma alteração no teor de clorofila ocorreu, o que é uma característica importante para esses frutos cuja a cor é verde. O pH, SS e a relação SS/AT foram elevadas em ambos os tipos de conservas usadas (óleo e vinagre). O processamento térmico não causou alterações no teor de carotenoides e flavonoides em comparação com os frutos *in natura*, mas provocou um aumento no teor de fenois. No tempo de cozimento de 10 minutos foi observada a maior atividade antioxidante. O tempo de cozimento não causou diferença significativa no teor de isorientina, rutina e ácido cafeico. O conteúdo de espermina e espermidina foram menores após 20 minutos de cozimento. Os frutos de jurubeba que foram preservados em vinagre mostraram um pH e o nível de putrescina mais baixo independente do tempo de cozimento utilizado, ao passo que o uso de óleo de soja causou aumento na atividade antioxidante e carotenoides.

Palavras-chave: Tratamento térmico, antioxidante, poliaminas, fitoquímicos, Solanaceae.

Effects of boiling time and preservatives in pickled jurubeba (*Solanum paniculatum* L) **fruits**

ABSTRACT

Jurubeba fruit raw and thermally processed in different minutes (10, 20, 30 and 40 minutes) and stored in oil and vinegar were evaluated for physical characteristics (pH, soluble solids (SS), titratable acidity (TA) and the relation SS / TA), phytochemicals (chlorophylls, carotenoids, phenolic compounds, flavonoids) antioxidant capacity (DPPH/ TEAC), and polyamines (PAs). After analyzing the data we conclude that the boiling of jurubeba for 20 minutes besides is the most widely used time in home-made processes jurubeba canning in Brazil stands by not to change the chlorophyll levels, which means no change visual appearance of the fruit. The parameters of pH and SS were high in both types of preserved adopted as well as the relationship between SS / TA; there is no change in carotenoid levels, total phenols and antioxidant activity that interferes with of the quality of the matérial. In Polyamine, the spermine and spermidine levels are lower after 20 minutes of boiling. The jurubeba in vinegar pickled lowers the pH and also the levels of putrescine, regardless of the cooking time used. Therefore, it is concluded that 20 minute cooking oil and pickled fruits jurubeba, is the combination that maintain more qualidade.

Keywords: Thermal processing, antioxidants, polyamines. phytochemicals, solanaceae

2.1 Introduction

Globally, significant improvements have been made in studies of regional dietary habits and the considerable inter- and intracountry variability (Kearney, 2010). Many important information are provided by researches focused in non conventional foods consumed along staple foods to improve taste and nutritional quality. Among these non conventional foods, we can highlight the *Solanum paniculatum* L. (Solanaceae), popularly known as “Jurubeba”, widely used in folk medicine as a tonic and antipyretic agent (Santos et al., 1988). This plant is native to northern and northeastern regions of Brazil (MAPA, 2010), and present a dark green color fruit used for culinary purpose.

S. paniculatum fruits are mainly consumed, after cooking, with rice or as pickles prepared either in oil or vinegar. Kaur and Kappor (2001) consider that the heat treatment is the main cause of change in the content of natural antioxidants in food. However, the cooking process can contribute to formation of new compounds, or promote an easier extraction of the cell matrix molecules. In fruits and vegetables, a large amount of bioactive compounds as polyphenols, carotenoids and polyamines are found in variable concentrations. The plants of the genus *Solanum* have steroidal saponins, glycoalkaloids and flavonoids, secundary metabolites which are important in natural defense of plants (Oliveira et al., 2006). An evaluation of some plants in relation to their antioxidant potential (*i.e.* the chemical constituents that may assist in free radicals scavenging), such as polyphenols, vitamins, alkaloids, triterpenes, sesquiterpenes and other molecules has been the object of several researches (Silva, et al. 2012).

The most common polyamines (PAs) in fruits and vegetables are putrescine, spermidine and spermine, compounds frequently affected by cooking process and heat treatments (Rossetto et al., 2015). Some fruits can be rich in putrescine (Lima et al., 2008), while green vegetables are richer in spermidine (Valero et al., 2002). The occurrence of polyamines in Jurubeba has not yet been described and its quantification is necessary.

Information regarding nutritional composition of *S. paniculatum* fruits, *in natura* or after cooking, are scarce or absent. Thus, the aim of this research were to evaluate the

effects of thermal processing on the quality of *S. paniculatum* fruits by assessing the vitamin C, pigments, total phenols, total flavonoid, characterization of the some polyphenols by HPLC, and total antioxidant activity in raw and heat-treated samples. In addition, the effects of preservatives as oil or vinegar, in pickled fruits were established.

2.2 Materials and methods

2.2.1 Samples

*S. paniculatum f*ruits were harvested in February 2014, from a farm located in Cáceres, Mato Grosso state, Brazil, (16º 04 '14' 'S latitude, 57 40' 44 '' W longitude and 118 m altitude), transported to the laboratory, selected and sanitized.

2.2.2 Cooking process and pickles preparation

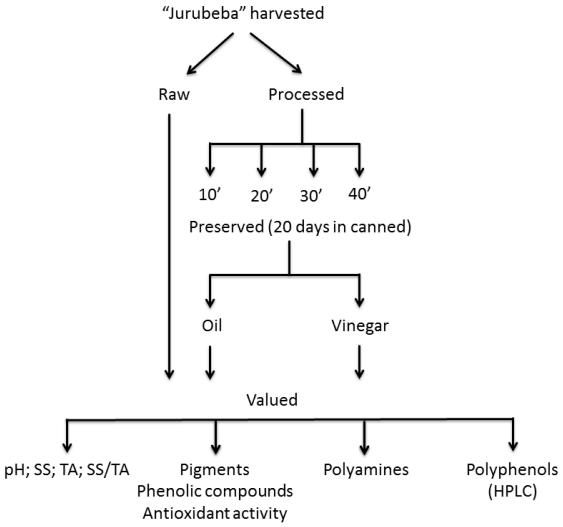
To obtain cooked samples, 150 g of fruits were placed into stainless steel pans with 1 L of boiling distilled water and cooked for 10, 20, 30 and 40 min at atmospheric pressure. After this procedure, remaining water was drained and the fruits were cooled at room temperature.

Raw and cooked fruits were pickled in sterile recipients full filled with 2,5% NaCl in two different preservatives, soybean oil and alcohol vinegar. After preparation, the pickles recipient were sealed and stored at room temperature (23 ± 2 ° C) for 20 days, as shown in the Figure 1.

2.2.3 Brix (soluble solids), pH and titratable acidity

The soluble solids was performed by digital refractometer, Atago, PAL^{-1} model. pH was determined by potentiometer (model Q Quimis -400A). Titratable acidity was determined in “g” of citric acid 100 g^{-1}, by titration, using 2 g of the ground product and 20 mL of distilled water, as described by Amerine and Ough (1987).

Figure 1 – Representative flow chart of the preparation process of pickles and analysis performed



2.2.4 Vitamin C (Ascorbic acid)

The vitamin C determination was carried out in 2 g of nitrogen grounded fruits diluted in 10 ml of oxalic acid, as described by Tillman. This method is based on the reduction of 2,6- dichlorophenolindophenol dye by ascorbic acid.

2.2.5 Carotenoids, chlorophyll, total phenols and flavonoids

Raw, cooked and pickled samples were grounded with liquid nitrogen and stored at -80 °C, until analysis.

The determination of carotenoids and chlorophyll (*a* and *b*) was carried out using the method validated by Sims and Gamon (2002). From each sample, 100 mg were homogenized in a mini-turrax (Marconi, Brazil) with 3 mL of a cold acetone / Tris-HCl (0.2M, pH 7.8, 80:20, v / v) solution for 1 min. All procedures were conducted on ice and protected from light. After, the samples were centrifuged at 2000 x g for 5 min and the supernatant was immediately used for determination of pigments in UV/VIS spectrophotometer (Amersham-Pharmacia-Biotech). Total chlorophyll was obtained by

the sum of chlorophyll a and chlorophyll b. The absorbance values were converted into ug of total carotenóids.g^{-1} based on the formulas:

Carotenoids ={A_{470}-[17.1*(Cla+Clb)]-9,479*antocianina}/119.26*
Chlorophyll *a* = 0.01373*(A_{663})-0.000897*(A_{537})-0.003046*(A_{647}).
Chlorophyll *b* = 0.02405*(A_{647})-0.004305*(A_{537})-0.005507*(A_{663}).

Total phenols were determined based on Singleton and Rossi (1965) using the Folin-Ciocalteu reagent. Fresh powdered samples (100 mg) were homogenized in mini-turrax (Marconi, Brazil) with 5 mL acetone: water (50:50 v/v). After 20 minutes in an ultrasonic bath (Eco-sonics, Ultronique), samples were centrifuged for 10 minutes at 6000 x g at 5 °C. The supernatant was removed and reserved and the process of extraction was performed once and the supernatants combined. Absorbance was read at 725 nm and the results expressed in mg of equivalent gallic acid g^{-1} fresh weight.

For flavonoid analysis, the samples were extracted in methanol. After 60 min in ultrasonic bath, the samples were centrifuged at 6,000 x g (Heitich Zentrifugen, MIKRO 220R) for 10 min and the supernatant collected. Extraction was conducted in the pellet twice and supernatants were combined and analyzed for content of total flavonoids as described by Awad et al. (2000), with adjustments made by Popova et al. (2004). The whole process was carried out in the absence of light. Results were expressed in mg quercetin g^{-1} fresh weight.

2.2.6 Trolox equivalent antioxidant capacity (TEAC) assay

The antioxidant activity was determined with the methodology proposed by Brand Williams et al. (1995), adapted by Rossetto et al. (2009). The results were expressed in uM Trolox equivalent ug / g sample^{-1} (TEAC). The extract was obtained from 100 mg of pulverized fresh samples in liquid nitrogen, homogenized in a mini-turrax (Marconi, Brazil) and kept for 15 minutes in ultrasonic bath (Eco-sonics, Ultronique) with 3 mL of ethanol. Subsequently, the samples were centrifuged for 10 minutes at 6000 x g at 5 °C

and after 30 min., the absorbance were read at 517 nm, in a UV/VIS spectrophotometer (Amersham-Pharmacia-Biotech).

2.2.7 Thin layer chromatography of polyamines (PAs)

Polyamines were analyzed as described by Flores and Galston (1982), with modifications by Lima et al. (1999). *S. paniculatum* fruits were homogenized in perchloric acid (5% v/v - cold) for 1 hour and centrifuged (10000 x g, HeitichZentrifugen, MIKRO 220R) for 30 minutes at 4 ^{o}C. 4.5 mol L^{-1} Na_2CO_3, containing 18.5 mmol L^{-1} dansyl-chloride in acetone (Sigma, 95%) was added to the supernatant. The reaction was carried out at room temperature and protected from light for 16 h. Then, 0.87 mol L^{-1} proline (Sigma, 99%) was added, and samples were maintained at room temperature for 30 min. Toluene (Sigma) was used to extract dansylated PAs. Aliquots (20 µL) were applied manually with Hamilton syringe (50 µL) onto activated (1 h at 110 ^{o}C, before use) glass plates (Adamant®Silica gel 60G, 0.25 mm, (20 x 20cm), Macherey-Nagel) and separated in a TLC developing tank, using as mobile phase chloroform:triethylamine (7.5:1). The plate was allowed to dry at room temperature (22 ± 2 °C), then dried with a hair dryer until the excess of solvent disappeared before interpretation.

Putrescine (Put) (Sigma, 98%), spermidine (Spd) (Sigma, 99%) and spermine (Spm) (Sigma, 99%), were used as standards. The entire procedure was monitored under UV light at 254nm. Free PAs were quantified by comparison against standards by fluorescence emission spectroscopy (excitation at 350 nm and emission at 495 nm), in a Video Documentation System, using the Image Master 2.0 Software (Amersham Pharmacia Biotech 1996). The calculation of quantitative analysis was done based on the area obtained in the standards and the samples. Free PAs content was expressed as nmol g^{-1} fresh weight (FW).

2.2.8 High Performance Liquid Chromatography (HPLC) analysis of flavonoids

Samples extracted as described in flavonoid analysis were filtrated (Millipore 0.22 µm filter) and used for flavonoids analyses according Escarpa et al., (1999). Briefly, 20 µL were injected into a Thermo Scientific Dionex UltiMate 3000 systems (Thermo Fisher Scientific Inc., MA, USA), coupled to a quaternary pump, Ultimate 3000RS auto sampler and diode array detector (DAD-3000RS). Flavonoids were separated on an Ace C18 (4.6 x 250 mm; 5µm) column at 25 ºC. Analysis were monitored at 280 nm and peak integration, and calibrations were performed between 210 and 350 nm with Dionex Chromeleon software,

The flow rate was 1.0 ml/min and mobile phase consisted of methanol (solvent A) and phosphoric acid 0,01M. The system was run with the following gradient elution program: 0-5 min, 0,5% A, 5-10 min, 50% A, 10-15 min 70% A, 15-20 min, 80% A, 25-30 min 100% A and to 30-35 min 5%.

2.2.9 Statistical analysis

All results are given as mean ± standard deviation. Differences between variables were tested for significance by one way ANOVA procedure, followed by Tukey, at a significance level of $p<5\%$.

2.3 Results and discussion

Cooking time influenced quality parameters (pH, soluble solids and soluble solids/titratable acidity) in *S. paniculatum* fruits conserved in oil (Table 1). The results shown a slight variation of quality parameters in fruits pickled in vegetable oil among cooking times. The highest value is found in fruits boiled for 20 minutes. In vinegar, which presents high acidity, the “jurubeba” exhibited lower pH than those preserved in oil. Besides consumer acceptance, the pH affects many chemical processes such as protein properties (denaturation), enzymatic activity and it also affects the growth of microorganisms (Stippl et al., 2004).

The effect observed in pH was reflected in titratable acidity in samples preserved in vinegar, as much as in oil. Fruits boiled for 20 min and preserved in oil showed higher levels of SS, although no difference was observed in other cooking times. In vinegar, the values found for SS after 20 min. were similar to that found at 10 min. The ratio (SS/TA) increased with the use of oil as preservative and with the cooking time in relation to raw fruits. On the other hand, in fruits preserved in vinegar, this ratio decreases proportionally to cooking time.

In our study, chlorophyll and carotenoid levels decreased after cooking when compared to raw fruits (Figures 2A and 2B). The loss of green color in many vegetables after cooking is a frequent problem affecting the quality of many canned fruits and vegetables. At room temperature, chlorophylls (*a* and *b*) exhibit stability, but when temperatures above 50 ^{o}C are used, may occur alteration of chlorophyll levels (Andrés-Bello et al., 2013).

Decreased levels of chlorophyll are promoted by heat treatment and may occur by chlorophyll conversion to pheophytins, attributed to pH change during thermal processing. The hydrogen ions change chlorophyll in pheophytins by the replacement of the Mg porphyrin ring (Minguez-Mosquera et al., 1989). Other studies also describe the loss of chlorophyll in broccoli by in 67.87% after cooking (Pellegrini et al., 2010).

After 10 and 40 minutes of boiling, “jurubebas” preserved in vinegar showed lower values of chlorophyll than “jurubebas” preserved in oil. In our study, there was a loss of chlorophyll, regardless of the types of preservatives tested. Aquino et al (2011) observed that time was a key factor in reducing the chlorophyll content in broccoli. In this study, cooking the fruits for 20 and 30 minutes presented no difference in the types of conservative used. Thus we can affirm that the cooking time of 20 minutes, common between preserve manufacturers, would be the ideal time when we compare the green color of the “jurubeba” fruit.

Table 1 - pH, soluble solids (°Brix), titratable acidity (g citric acid 100 g-1) and *ratio* (SS / TA in "Jurubeba" raw and subjected to different boiling times (10, 20, 30 and 40 minutes) and types of preservatives (oil and vinegar)

Treatment	pH		SS	
Raw	5.58 ± 0.07 abA*	5.58 ± 0.07 aA	15.33 ± 0.7 bA	15.33 ± 0.7 aA
Minute	Oil	Vinegar	Oil	Vinegar
10	5.45 ± 0.03 bA	3.94 ± 0.02 bB	18.7 ± 2.7 abA	9.9 ± 0.6 abB
20	5.63 ± 0.06 aA	3.90 ± 0.00 bB	21.3 ± 5.1 aA	9.87 ± 0.8 abB
30	5.58 ± 0.10 abA	3.92 ± 0.02 bB	19.63 ± 1.7 abA	9.03 ± 0.3 bB
40	5.52 ± 0.10 abA	3.90 ± 0.01 bB	18.73 ± 3.7 abA	9.27 ± 0.1 bB
Treatment	**TA**		**SS/TA**	
Raw	0.14± 0.01 aA	0.14± 0.01 bA	107.95 ± 11.59 cA	107.95 ± 11.59 aA
Minute	Oil	Vinegar	Oil	Vinegar
10	0.11± 0.01 abB	0.33± 0.07 aA	174.00 ± 31.43 bA	31.03 ± 8.12 bB
20	0.07 ± 0.01 bB	0.28± 0.00 aA	283.61 ±31.04 aA	35.05 ± 2.33 bB
30	0.06± 0.00 bB	0.32± 0.01 aA	295.08 ± 6.07 aA	27.47 ± 2.01 bB
40	0.10± 0.00 abB	0.32± 0.00 aA	176.59 ± 36.29 bA	28.74 ± 0.99 bB

*Average of three replicates ± standard deviation; Means followed by the same lower case letter in the column and capital on the line do not differ at 5% probability (Tukey).

The thermal processing did not cause increased levels of total carotenoids in "jurubeba" (Figure2B) compared to the raw fruits. The type of preservative influences the levels of this compound. Fruit preserved in oil contains higher levels of carotenoids when compared to those preserved in vinegar. Generally, due to the high temperature in the cooking process, matrix disruption occurs, promoting the extraction of compounds in the cell and many of these compounds migrate into the cooking water. Sa & Rodriguez-Amaya (2003) suggest an increase in carotenoid content after cooking. On the other hand, studies have demonstrated lower levels of carotenoids after thermal processing (Zhang & Hamauzu, 2004), the same results founded in our study in "jurubeba" fruits, after thermal processing.

After cooking, regardless of the preservative used (oil or vinegar), total phenol levels increased at all cooking times in relation to the raw material (Figure 2D), especially when fruits were preserved in oil and boiled for 10 minutes. This increase in total phenols after boiling has been described in eggplants (*Solanum melongena*) (Scalzo et al., 2010;. Ramirez-Anaya et al., 2015.).

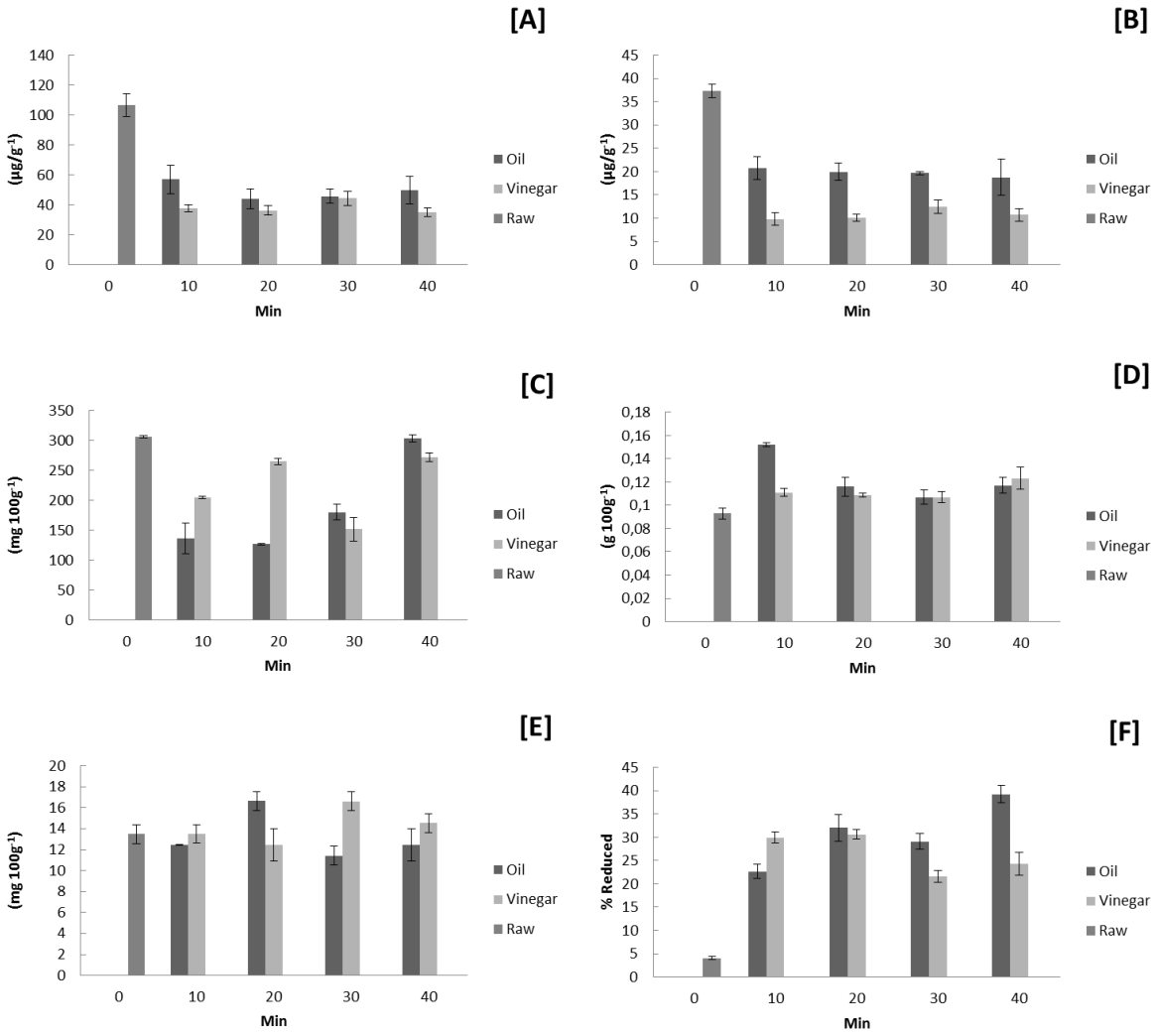
On the other hand, flavonoid levels are strongly affected by the type of preservative (Figure 2C). In vinegar, the fruits showed higher levels of these polyphenols. However, the value was lower compared to raw material, except when the jurubeba fruits were cooking for 40 minutes. This increase in the content of these compounds (phenols and flavonoids) may be due to high-temperature extraction / cooking time, which promoted denaturation of the matrix, increseasing the extractability of these compounds (Turkmen et al., 2005; Zhang & Hamauzu, 2004). Cooking promotes the softening of the cell wall and other components of cells, such as vacuoles and apoplast, releasing the phenolic compounds. Another factor that may contribute to increased of polyphenolsthe content is the decomposition of phenolic compounds linked to fibers (cellulose and pectin) (Gökmen, Serpena, & Fogliano, 2009), or even the breaking of the bonds between phenols and sugars, which have contributed to the increase in concentration of these compounds (Singleton et al., 1999).

Ascorbic acid levels (Figure 2E) in fruits boiled for 20 minutes and preserved in oil was higher than those found in raw “jurubeba” and preserved in vinegar. However, this trend disappears when cooking time increases, where fruits preserved by vinegar showed higher ascorbic acid content.

The antioxidant activity (Figure 2F) of “jurubeba” cooked and preserved in oil increased significantly in all cooking time. From the data obtained with the analysis of antioxidants in this study and the antioxidant activity, the heat treatment increases the antioxidant activity of “jurubeba”.

The type of preservative with cooking for 20 min. did not induce any difference in the antioxidant activity. “Jurubeba” in soybean oil preserves showed higher antioxidant activity after 30 and 40 minutes. Others studies with also showed that heat treatment increases the antioxidant activity (Scalzo et al., 2010). This increase can be attributed to the release of compounds with antioxidant activity of the matrix in function of the increase in temperature.

Figure 2 - [A] total chlorophyll (ug / g) [B] total carotenoids (ug / g), [C] total flavonoids (100 mg g-1), [D] total phenols (100 g g-1),[E] ascorbic acid (mg 100g-1) and [F] antioxidant activity (TEAC mmol / l,% reduced DPPH) in "Jurubeba" raw and subjected to different boiling times (10, 20, 30 and 40 minutes) and types of preservatives (oil and vinegar)



"Jurubeba" preserved in soybean oil showed no difference between the cooking times in relation to the polyamine content. In addition, the "jurubeba" preserved in vinegar presented lower values and we can observed a decrease of putrescine with the cooking time. On the other hand, spermidine and spermine levels increased with the cooking time in "jurubeba" preserved in vinegar and decreased in those preserved in oil.

The presence of the three polyamines in the preserves was expected because they occur naturally in fruits and vegetables (Figure 3). When comparing the levels of polyamines in the raw "jurubeba" (putrescine, 2.01 µmols g^{-1}, spermidine 0.10 µmols g^{-1}

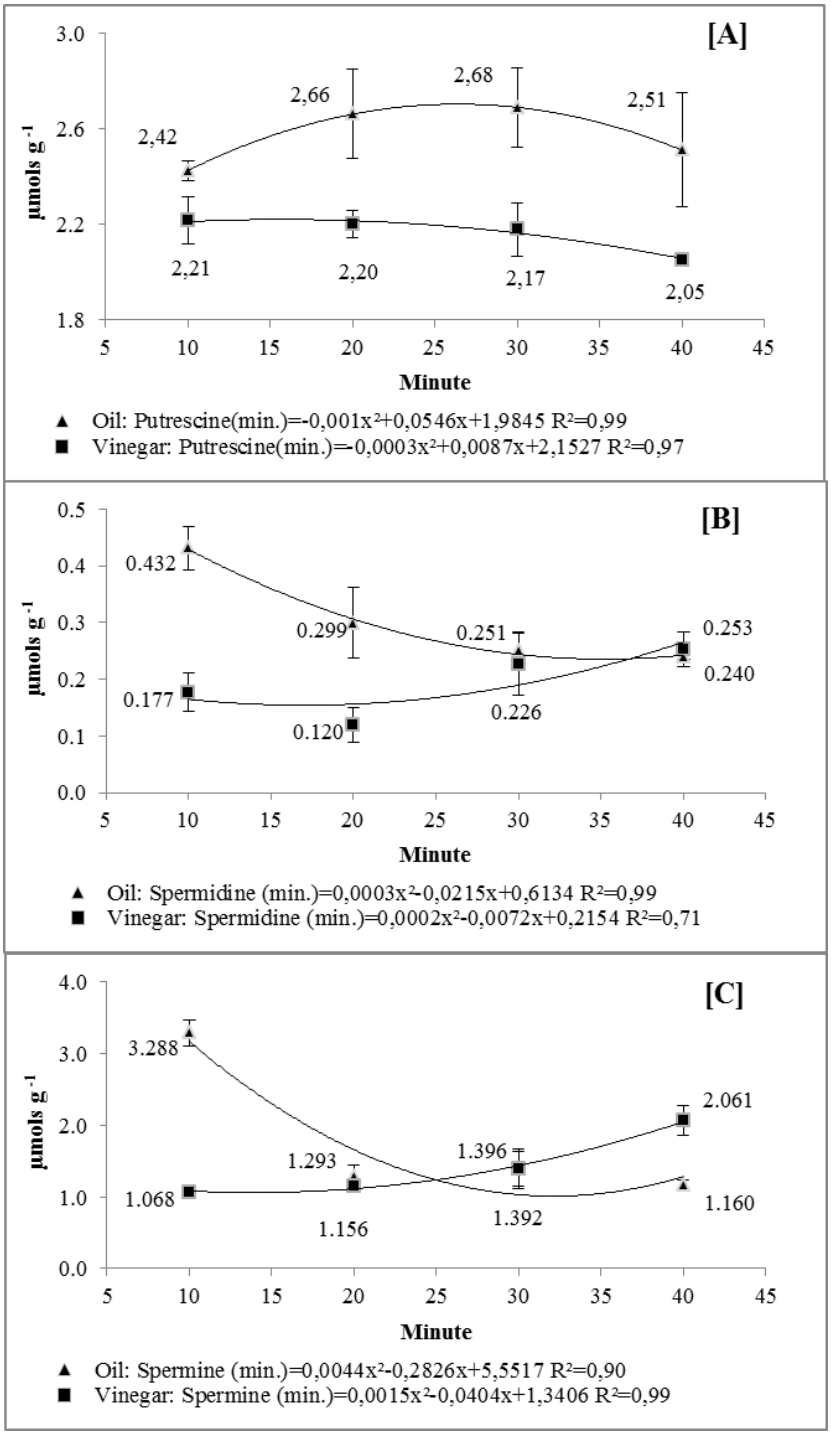
and spermine 1.77 μmols g^{-1}) and after thermal processing, regardless of the cooking time, there was an increase in the levels of putrescine and spermidine, while spermine levels had decreased when stored in vinegar and increase in fruits preserved in oil.

Some studies indicate that the cooking process can induce changes in the levels of polyamines. Rossetto et al. (2015) observed reduction of the content of putrescine, spermine and spermidine in vegetables such as carrots, broccoli, cabbage and beetroot cooked in water. In others studies, the cooking process does not alter the levels of these amines (Eliassen et al., 2002). The cooking process of “jurubeba” altered levels of putrescine, spermidine and spermine. It has been reported that polyamines can be leached in boiling water. After cooking of some vegetables, the levels of polyamines could be decrease. Some studies showed that there was putrescine loss of around 20-25% in broccoli and celery and 40% in cauliflower and asparagus. Likewise, for spermidine a 10-20% loss occurred in broccoli and celery, and 20-30% cauliflower and asparagus (Ziegler et al., 1994).

However, our results show that in addition to the changes in the levels of these substances induced by cooking in water , the type of preservative has also influenced. “Jurubeba” preserved in vinegar preserves showed lower levels of putrescine, ranging from 2.05 to 2.21 μmols / g, while for the preserved in soybean oil, the levels ranged between 2.42 and 2.68 μmols / g. Biogenic amines, especially histamine, putrescine and cadaverine have been suggested as indicators for deterioration of some types of food such as fresh fish, meat and vegetables (Riebroy et al., 2004). On vinegar preserves, the lower putrescine content could be indicative of better fruit quality.

These amines are important for the nutrion and for the health. Spermidine and spermine are directly related to the DNA and therefore with to cell division and their contents in “jurubeba” fruits cooked for 20 min. were lower compared to other cooking time. For people with certain neoplasias, the results it may be interesting, since it has low levels of these compounds, and have relatively low content of putrescine. However, spermine is important for taking part in the regulation of nitric oxide content and absorption this tetramine can contribute to the balance of excessive production of nitric oxide. This free radical (NO) can be correlated with tumor progression (Til et al., 1997) and this amine has also been linked to decreased inflammation (Moinard et al., 2005).

Figure 3 - Polyamines: [A] - putrescine in µmols g^{-1}, [B] - spermidine in µmols g^{-1}, [C] - spermine in µmols g^{-1}) in "Jurubeba" raw and subjected to different boiling times (10, 20, 30 and 40 minutes) and types of preservatives (oil and vinegar)



Polyamines occur naturally in plants. The levels of polyamines found in this study are important in relation to the consumption of these substances, because they may be related with some heart deseases and some types of cancer. Polyamines does not cause cancer, but accelerates tumor growth. Increased levels due to the synthesis of polyamines in animal tissues and to food intake can cause increased cell growth (Kalac et al., 2015).

For the analysis held in HPLC of polyphenols , we identified isoorientine, rutine and caffeic acid. All polyphenols found presented lower concentration in raw Jurubeba fruits. When this fruits were cooked, the polyphenol content incresead, but whitout no significant difference. We can observe that using 10 minutes for cook is sufficient for the polyphenol content is achieve the maxim content.

However, in the others analysis we found that 20 min. is the ideal time. In relation to the polyphenol content, the time of 20 min can be used without altering content of the analyzed polyphenols. Jurubeba fruits preserves in oil had higher content of rutin, while those stored in vinegar, had higher levels of caffeic acid. This increase release of flavonoids (rutin and isoorinetin) and the phenolic acid (caffeic acid) is a evidence that antioxidant activities of jurubeba fruits might be increased or remain unchanged after cooking and when preserved either in oil or vinegar (Jiratanan & Liu, 2004). Phenolic compounds are also water-soluble, rendering them susceptible to leaching. In our study this effect was not observed. The cooking or the canned process not influenced the isorientin, rutin or caffeic acid content. Furthermore, it has been described that occurs decline these compounds due to leaching into the brine or syrup rather than oxidation (Chaovanalikit & Wrolstad, 2004; Hong et al., 2004).

From the data obtained, the treatment using 20 minutes of cooking seems to be the one with the most interesting results. Besides being the most widely used treatment in homemade “jurubeba” preserves, it did not induce changes in chlorophyll levels, an important parameter for visual analysis. In addition, some quality parameters such as pH and SS were high in that cooking time as well as in the two types of preservatives used, which was reflected in the ratio of SS and AT. At in that same cooking time no changes in the levels of carotenoids were observed that could decrease the quality of the “jurubeba” fruits.

Table 2 - Polyphenols content (isoorientine, rutine and Acid caffeic) in raw "Jurubeba" fruits and subjected to different boiling times (10, 20, 30 and 40 minutes) and types of preservatives (oil and vinegar)

		Polyphenols (mg/100g^{-1})		
Preservative	**Minute**	**Isoorientin**	**Rutin**	**Caffeic acid**
Oil	**raw**	21.3 ± 2.16	47.90 ± 6.88	0.10 ± 0.02
	10	81.6 ± 8.67	75.38 ± 6.14	0.41 ± 0.07
	20	80.4 ± 7.81	69.38 ± 5.95	0.35 ± 0.04
	30	83.7 ± 11.70	80.94 ± 5.80	0.40 ± 0.05
	40	83.0 ± 9.91	76.45 ± 7.47	0.45 ± 0.07
Vinegar	**10**	83.0 ± 17.04	68.47 ± 4.41	0.56 ± 0.10
	20	85.6 ± 3.03	59.59 ± 4.25	0.66 ± 0.01
	30	81.0 ± 0.79	55.67 ± 4.42	0.64 ± 0.06
	40	82.2 ± 4.85	61.36 ± 3.23	0.76 ± 0.09

This same statement can be made to the contents of total phenols and antioxidant activity. The cooking time not induced significant alterations in isorientin, rutin and caffeic acid contents. Another analysis that allows choosing this cooking time is the polyamine content. Spermidine and spermine showed lower contents after 20 minutes of cooking. At that thermal process and using vinegar as a preservative, the fruits had the lowest levels of putrescine.

2.4 References

Andrés-Bello a., Barreto-Palacios V, García-Segovia P, et al. (2013) Effect of pH on Color and Texture of Food Products. *Food Engineering Reviews*, 5(3), 158–170.

Bett-Garber KL, Lea JM, Watson MA, et al. (2015) Flavor of fresh blueberry juice and the comparison to amount of sugars, acids, anthocyanidins, and physicochemical measurements. *Journal of food science*, 80(4), S818–27.

Brand-Williams W, Cuvelier ME and Berset C (1995) Use of a free radical method to evaluate antioxidant activity. *LWT - Food Science and Technology*, 28(1), 25–30.

Chaovanalikit A and Wrolstad RE (2004) Total Anthocyanins and Total Phenolics of Fresh and Processed Cherries and Their Antioxidant Properties. *Journal of Food Science*, 69(1), FCT67–FCT72.

De Sá MC and Rodriguez-Amaya DB (2003) Carotenoid composition of cooked green vegetables from restaurants. *Food Chemistry*, 83(4), 595–600.

Eliassen KA, Reistad R, Risøen U, et al. (2002) Dietary polyamines. *Food Chemistry*, 78(3), 273–280.

Escarpa A and González M. (1999) Fast separation of (poly)phenolic compounds from apples and pears by high-performance liquid chromatography with diode-array detection. *Journal of Chromatography A*, 830(2), 301–309.

Flores HE and Galston AW (1982) Analysis of polyamines in higher plants by high performance liquid chromatography. *Plant physiology*, 69(3), 701–6.

Gökmen V, Serpen A and Fogliano V (2009) Direct measurement of the total antioxidant capacity of foods: the 'QUENCHER' approach. *Trends in Food Science & Technology*, 20(6-7), 278–288.

Kaur C and Kapoor HC (2008) Antioxidants in fruits and vegetables - the millennium's health. *International Journal of Food Science & Technology*, 36(7), 703–725.

Kearney J (2010) Food consumption trends and drivers. *Philosophical transactions of the Royal Society of London. Series B, Biological sciences*, 365(1554), 2793–807.

Lima GPP, Brasil OG and Oliveira AM de (1999) POLIAMINAS E ATIVIDADE DA PEROXIDASE EM FEIJÃO (Phaseolus vulgaris L.) CULTIVADO SOB ESTRESSE SALINO. *Scientia Agricola*, Scientia Agricola, 56(1), 21–26.

Lima GPP, da Rocha SA, Takaki M, et al. (2008) Comparison of polyamine, phenol and flavonoid contents in plants grown under conventional and organic methods. *International Journal of Food Science & Technology*, 43(10), 1838–1843.

Mandal S, Mandal A and Park MH (2015) Depletion of the polyamines spermidine and spermine by overexpression of spermidine/spermine N1-acetyltransferase 1 (SAT1) leads to mitochondria-mediated apoptosis in mammalian cells. *The Biochemical journal*, 468(3), 435–47.

Minguez-Mosquera MI, Garrido-Fernandez J and Gandul-Rojas B (1989) Pigment changes in olives during fermentation and brine storage. *Journal of Agricultural and Food Chemistry*, American Chemical Society, 37(1), 8–11.

Moinard C, Cynober L and Debandt J (2005) Polyamines: metabolism and implications in human diseases. *Clinical Nutrition*, 24(2), 184–197.

Pellegrini N, Chiavaro E, Gardana C, et al. (2010) Effect of different cooking methods on color, phytochemical concentration, and antioxidant capacity of raw and frozen brassica vegetables. *Journal of agricultural and food chemistry*, American Chemical Society, 58(7), 4310–21.

Popova M, Bankova V, Butovska D, et al. Validated methods for the quantification of biologically active constituents of poplar-type propolis. *Phytochemical analysis : PCA*, 15(4), 235–40.

Ramos CS, Ramos NSM, Da Silva RR, et al. (2012) Metabolism by grasshoppers of volatile chemical constituents from Mangifera indica and Solanum paniculatum leaves. *Journal of insect physiology*, 58(12), 1663–8.

Riebroy S, Benjakul S, Visessanguan W, et al. (2004) Some characteristics of commercial Som-fug produced in Thailand. *Food Chemistry*, 88(4), 527–535.

Rossetto MRM, Vianello F, Rocha SA da, et al. (2009) Antioxidant substances and pesticide in parts of beet organic and conventional manure. *African Journal of Plant Science*, Academic Journals.

Rossetto MRM, Vianello F, Saeki MJ, et al. (2015) Polyamines in conventional and organic vegetables exposed to exogenous ethylene. *Food chemistry*, 188, 218–24.

Salerno L, Modica MN, Pittalà V, et al. (2014) Antioxidant activity and phenolic content of microwave-assisted Solanum melongena extracts. *The Scientific World Journal*, 2014, 1–6.

Santos CAM, Torres KR and Leonart R (1988) Plantas medicinais (Herbarium, flor et sciencia). . 113 pg. 1988. *Scientia et labor. Ediçoes da UFPR*, 113.

Sims DA and Gamon JA (2002) Relationships between leaf pigment content and spectral reflectance across a wide range of species, leaf structures and developmental stages. *Remote Sensing of Environment*, 81(2-3), 337–354.

Singleton VL and Rossi JA. J (1965) Colorimetry of Total Phenolics with Phosphomolybdic-Phosphotungstic Acid Reagents. *Am. J. Enol. Vitic.*, 16(3), 144–158.

Singleton VL, Orthofer R and Lamuela-Raventós RM (1999) Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. In: *Methods in Enzymology*, pp 152–178.

Stippl VM, Delgado A and Becker TM (2004) Development of a method for the optical in-situ determination of pH value during high-pressure treatment of fluid food. *Innovative Food Science & Emerging Technologies*, 5(3), 285–292.

Til HP, Falke HE, Prinsen MK, et al. (1997) Acute and subacute toxicity of tyramine, spermidine, spermine, putrescine and cadaverine in rats. *Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association*, 35(3-4), 337–48.

Valero D, Martínez-Romero D and Serrano M (2002) The role of polyamines in the improvement of the shelf life of fruit. *Trends in Food Science & Technology*, 13(6-7), 228–234.

Zhang D and Hamauzu Y (2004) Phenolics, ascorbic acid, carotenoids and antioxidant activity of broccoli and their changes during conventional and microwave cooking. *Food Chemistry*, 88(4), 503–509.

Ziegler W, Hahn M and Wallnöfer PR (1994) Changes in biogenic amine contents during processing of several plant foods. *Deutsche Lebensmittel-Rundschau*, 90, 108–112.

3 ASPECTOS NUTRICIONAIS DE JURUBEBAS SUBMETIDAS AO PROCESSAMENTO TÉRMICO E TEMPO DE ARMAZENAMENTO

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Resumo

Introdução. A hortaliça fruto, Jurubeba (*Solanum paniculatum* L.) é utilizada na medicina popular brasileira. Os frutos são comumente consumidos após o cozimento devido à adstringência e para prolongar o consumo, os alimentos em conserva podem ser feitos com óleo ou vinagre. **Material e métodos.** Os frutos de Jurubeba foram adquiridos de três formas diferentes (de plantas cultivadas, plantas espontâneas e no mercado) e estudados em relação às suas qualidades nutricionais e físico-químicas após processamento térmico e conservação. Parte destes frutos foi mantida in natura, e a outra foi submetida a cozimento por 20 min. Jurubeba processados termicamente foram conservados em dois tipos de conservantes (óleo de soja e vinagre) armazenados e avaliados 1 hora após a preparação das conservas e após 30, 60 e 90 dias de prateleira quanto ao conteúdo de vitamina C, carboidratos totais, proteínas totais, lipídios totais, total Flavonóides e fenóis. **Resultados.** Houve uma redução do teor de vitamina C após o cozimento. Observou-se um maior teor de carboidratos totais aos 90 dias de armazenamento. A Jurubeba conservada em óleo manteve o conteúdo protéico e apresentou maior teor de lipídios. Seria interessante que o consumo de jurubeba, quando o objetivo é consumo de carotenóides, seja realizado após o cozimento e conservado em vinagre e consumido dentro de 60 dias de vida útil. Tendo em consideração os flavonóides, os frutos adquiridos no mercado ou recolhidos a partir de plantas espontâneas e conservados em óleo ou vinagre são boas fontes até 90 dias. **Conclusão.** O consumo de jurubeba em conserva pode ser uma boa fonte de compostos bioativos quando preservados em óleo.

Palavras-chave: *Solanum paniculatum* (L.) / qualidades físico-químicas / vida útil / carotenóides / compostos fenólicos / flavonoides

Nutritional and bioactive aspects of jurubeba after thermal processing and storage

ABSTRACT

Introduction. Jurubeba fruit (*Solanum paniculatum* L.) is used in Brazilian folk medicine.The fruits are commonly consumed after cooking due to astringency and to prolong consumption, pickled foods can be made using oil or vinegar. **Material and methods.** Jurubeba fruit were acquired from three different ways (from cultivated plants, spontaneous plants and from market), and studied in relation to their nutritional and physicochemical qualities after thermal processing and pickling. Part of these fruit was kept in natura, and the other was submitted to cooking for 20 min. Jurubeba thermally processed were pickled in two types of preservatives (soybean oil and vinegar) and 1 hour after the prepare of the pickles and after 30, 60 and 90 shelf days the contents of vitamin C, total carbohydrates, total proteins, total lipids, total flavonoids and phenols were evaluated. **Results.** There was a reduction of the vitamin C content after cooking. A higher content of total carbohydrates was observed at 90 storage days. Jurubeba conserved in oil maintained the protein content and showed higher lipids content. It would be interesting that the jurubeba consume, in order to gather carotenoids, should be realized after the cooking and conserved in vinegar and consumed within 60 days of shelf life. Taking in consideration the flavonoids, the fruit acquired from market or collected from spontaneous plants pickled in oil or vinegar are good sources until 90 days. **Conclusion.** The consume of pickled jurubeba may be a good source of bioactive compounds when preserved in oil.

Keywords: *Solanum paniculatum* (L.) / physicochemical qualities / shelf life / carotenoids / phenolic compounds / flavonoids

Les aspects nutritionnels de jurubeba après le traitement thermique et le stockage.

RÉSUMÉ

Introduction. Des fruits de jurubeba (*Solanum paniculatum* L.) est utilisé dans la médicine populaire brésilienne. Les fruits sont populairement consommés après caisson dû à son adstringence et des conserves sont faits en utilisant de l´huile ou du vinaigre. **Matériels et méthodes**. Les fruits de jurubeba ont été acquis sous trois forms différentes (de plantes cultivées, des plantes spontanées et au marché) et etudiées à son contenu nutritionnel et les qualités physico-chimiques après traitement thermique et après la procedure de conservation. Une partie de ces fruits ont été maintenu *in nature* et l´autre partie a été soumise à une cuisson pendant 20 min. Du jurubeba traitéé thermiquement a eté immerges dans deux types de conservateurs et après une heure la preparation des conserves et après 30, 60 et 90 jours de stockage, le contenu de vitamine C, les sucres totaux, des protéines, des lipides, des flavonoïds et de phénolique totaux ont été évalues. **Résultats et discussion**. Il y a eu un réduction des niveaux de vitamine C après la caisson. Plus de sucres totaux ont été observée après 90 jours de stockage. Jurubeba conserve en huile de soja a maintenu la teneur de proteins et grand teneur des lipides. Pour l´obtention des caroténoïdes, jurubeba doit être consommé après conserve en vinaigre jusqu´à 60 jours. Jurubeba acquises dans le commerce ou récoltées des plantes spontaneés et préservés en huile ou vinaigre pendant 90 jours sont de bonnes sources de flavonoïdes. **Conclusion**. La consummation de jurubeba en conserve en huile peut être une source de composants bioactifs.

Mots clés: *Solanum paniculatum* (L.) / caractéristiques physico-chimiques / aptitude à la conservation / caroténoïdes / composés phénoliques / flavonoïdes

3.1 Introduction

With the increasing interest in functional foods, it is surprising that there is still just a limited amount of information about antioxidant properties of some non-conventional fruits and vegetables, such as jurubeba, and about the impact of post-harvest technologies. *Solanum paniculatim* L., popularly known as jurubeba, is a non-conventional fruit and is consumed by part of the population due to medicinal properties (folk medicine). Some phytotherapic jurubeba effects were already confirmed, as well as the use of its fruit (0.5 – 2 g/kg corporal weight) in the increase of the gastric acid secretion [1]. Due to the strong astringent taste, jurubeba fruit needs a prior heat treatment to be consumed, therefore its ingestion generally occurs after cooking and with other foods. According to the American Society for Testing and Materials, astringency is a complex of sensations due to shrinking, drawing or puckering of the epithelium as a result of exposure to substances such as alums or tannins. The conservation forms such as blanching and pickling in brine of maxixe, scarlet eggplants, caupi beans and guandu beans, induced the decrease of tannins and oxalates contents, which give the bitter flavor to the vegetables [2] and, in jurubeba, the thermal processing may promote the raise in some antioxidant compounds contents [3], such as carotenoids and flavonoids. However, the same process may affect other substances that present the capacity for scavenging free radicals.

The content of antioxidants in fruits and vegetables can suffer influence of biotic or abiotic factors, such as the presence of pathogens, cultivation conditions, climate, among others. Cultivated plants that receive chemical treatment usually tend to present smaller quantities of some bioactives compared to plants cultivated without agrochemicals addition [4]. In addition, the form consumption (raw or cooking) can contribute to changes in the content of molecules with the potential to eliminate free radicals. Generally, the jurubeba fruit harvest begins from 4 to 6 months after the planting (begin of the rainy season), but may be extended for up to a year due to the lack of uniformity in the flowering. As jurubeba fruit is consumed by part of the population after thermal processing, in pickles (in oil or vinegar), the bioactives quality and quantity preservation for a longer time is fundamental. Beyond the cooking and

storage factors, the fruit sources (cultivated, spontaneous and market) may also influence the levels of various antioxidant compounds. At the present time, cooking and pickling methods are important for health, but there is little information on the effects of these methods on the chemical composition and on some antioxidant activity of jurubeba fruit.

The food industry is always looking for products with high nutritional values, the presence of bioactive compounds is increasing the consumer interest and jurubeba can be an alternative between the pickled products, which has medicinal potential and is a source of antioxidants. Thus, the aim of this study was to verify whether preservatives (oil or vinegar), shelf life and acquisition methods influence some bioactive and nutritional components.

3.2 Material and methods

3.2.1 Samples

Jurubeba was harvested from cultivated and spontaneous plants and was also purchased in market. In the harvest, fruit presented uniformity in color (light green), safety, without imperfections and presented the same size (approximately 1.5 cm of diameter). The infructescences from cultivated and spontaneous plants were all harvested in the same time (same physiologic age) using a pruning shears, in the way that all fruit were transported still attached to the peduncles (Figure 4A). Fruit detached of peduncles were acquired from the local market at the same time.

Figure 1 - [A] Bunch and fruits of Jurubeba (*Solanum paniculatum* L.); [B] Separation of the fruits of the peduncles and [C] Fruits off the peduncles

3.2.2 Thermal processing and preparation of the pickles

After the selection, the fruit were thoroughly cleaned after sanitation in immersion in chlorinated water (5 °C; 100 mg L^{-1} NaClO; pH 6.5) for 10 min and rinsed with tap water (6 ± 2 °C) for 1 min. It was used 5 kg of fruit from each source (Figures 1B and 1C); one part was kept *in natura* and the other was submitted to thermal processing, with the cooking of 500 g in 1L of boiling water in a stainless steel pan with a lid, for 20 minutes [3].

After the boiling procedure, the water was drained and the fruit filled into glass jars (300 mL) (previously sterilized in boiling water for 30 minutes), containing 2 g NaCl and 150 mL of commercial soybean oil or alcohol vinegar. Jars were sealed with Parafilm®, covered with plastic lids and kept in shelfs, in a light protected room (25 ± 2 ^{o}C).

3.2.3 Shelf life study

Samples were stored at 25 ± 2 ^{o}C for three months under dark conditions. The evaluations were done in the *in natura* fruit and 1 hour, 30, 60, 90 days after the pickles prepare. Results were compared to those obtained post-processing and raw.

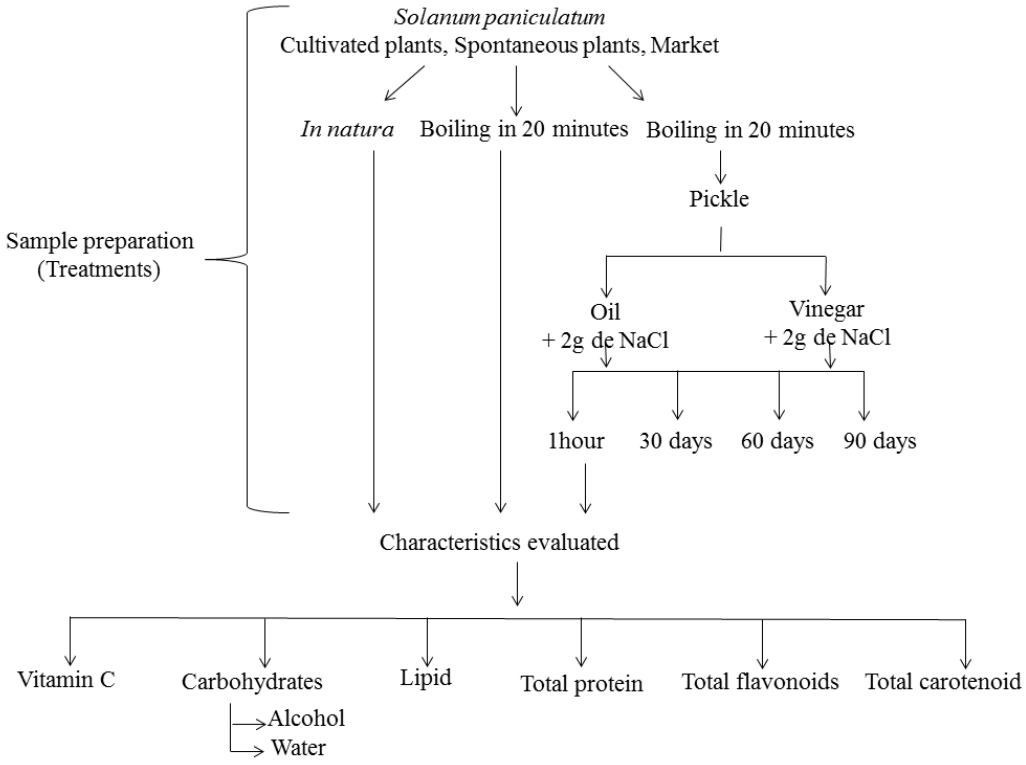
3.2.4 Physicochemical and biochemical analysis

In order to quantify the vitamin C content, fruit were ground in mini-turrax (Marconi, Brazil) until reached the aspect of a gooey paste. For the other analysis, fruit were frozen in liquid nitrogen and powdered in a cryogenic mill (Spex Sample Prep 6770 freezer/mill, MA, USA), during 5 minutes and were kept in a freezer at -80 °C (Figure 5).

3.2.5 Vitamin C

The vitamin C content was determined by using 2 g of ground fruit and homogenized in 10 mL of 1% oxalic acid, according to the methodology described [5]. The results were expressed as mg vit. C 100 $g^{-1.}$

Figure 2 - Scheme of the treatments and evaluations done in jurubeba fruit (*Solanum paniculatum* L.) *in natura*, cooked in water and pickled in two types of preservatives after 1 hour, 30, 60 and 90 of shelf days, from cultivated plants, spontaneous plants and from market



3.2.6 Total available carbohydrates

The total available (soluble) carbohydrates extraction was performed with two extractors (water and alcohol) following the method described by [6] and the results were expressed as g $100g^{-1}$.

3.2.7 Lipids

The total lipids quantification was performed according to [7] and expressed in percentage (%).

3.2.8 Total proteins

The amount of total proteins were quantified by the Kjeldahl method [5], using 6.25 as conversion factor of nitrogen to protein.

3.2.9 Total carotenoids

The extraction for the carotenoid levels determination was carried out in the fresh material powdered in liquid nitrogen, according to [8]. Briefly, 0,100 g of ground sample was homogenized in mini-turrax (Marconi, Brazil) with 3 mL of 0.2 mol L^{-1} acetone/Tris-HCl (pH 7.8, 80:20 v/v), for 1 minute in bath cold. The extraction was performed in ice and in the absence of light. Subsequently, the samples were centrifuged at 6.000 g for 5 min (MIKRO 220/220R - Hettich Lab Technology, USA) and the supernatant was immediately read in spectrophotometer UV/VIS (Amersham-Pharmacia-Biotech). The absorbance values were converted in µg total carotenoids g^{-1}.

3.2.10 Total flavonoids

In order to analyze the total flavonoids, the ground samples were extracted in methanol (P.A., HPLC) and after one hour in ultrasonic bath (Eco-sonics, Q 3.0/40, Ultronique) (homogenized at each 15 minutes in vortex) they were centrifuged for 10 min at 6.000 g (MIKRO 220/220R - Hettich Lab Technology, USA) and the supernatant was removed. Extraction was conducted in the pellet twice, and the supernatants were combined [9]. The complete process was carried out in the absence of light and the results were expressed as mg/100g.

3.2.11 Total phenol

Total phenols were determined based on the method of Singleton and Rossi (1965) [10], using the Folin-Ciocalteu reagent. Fresh powdered samples were homogenized with 50% acetone/water solution (v/v.) The precipitate was re-extracted and the supernatants were combined. The readings were performed in spectrophotometer UV/VIS (Amersham-Pharmacia-Biotech) using chlorogenic acid as standard and the results were expressed as mg/100g.

3.2.12 Statistical analysis

The experimental design was entirely randomized (DCI) with 30 treatments and 3 repetitions. To group the means, the Scott – Knott test was used at a 5% probability, using the Sisvar software 5.3 [11].

3.3 Results and discussion

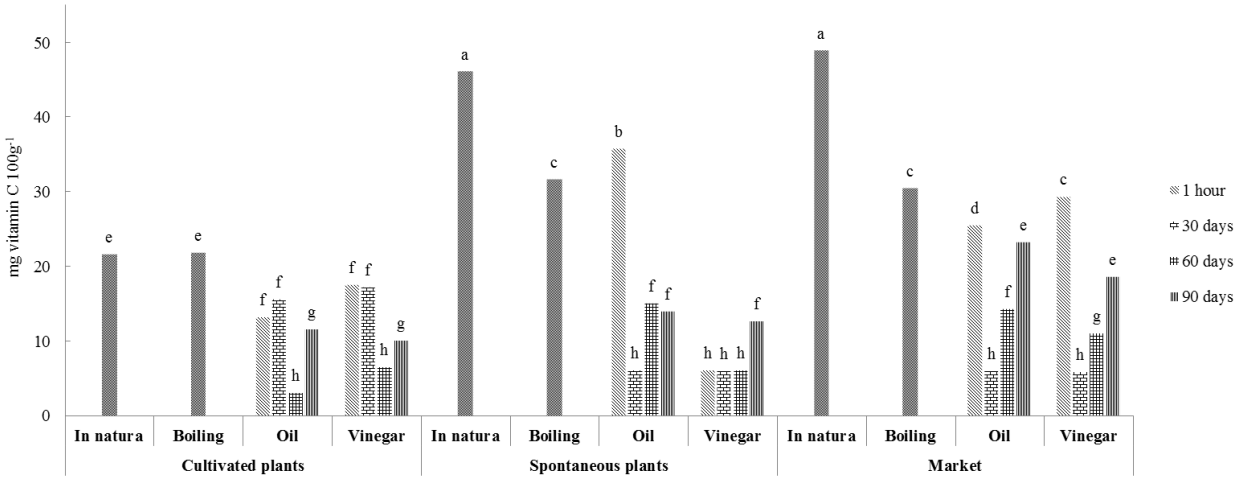
The highest vitamin C content was observed in jurubeba *in natura*. Fruit obtained from cultivated plants showed smaller vitamin C contents compared to the other forms of harvesting (Figure 6). The highest vitamin C losses after the cooking happened in the fruits from spontaneous plants and in those purchased in market. Several studies have reported vitamin C losses after cooking [12] and this effect may happen due to its thermolabile property [13].

With thermal processing and shelf time there was a reduction in the vitamin C content of 85.58% and 69.61% in the fruit pickled in oil and vinegar respectively at 60 storage days. In fruit acquired from market and collected from spontaneous plants, the highest vitamin C losses occurred after being pickled in vinegar also for 60 days. It was observed vitamin C losses of up to 86.74% in fruits from spontaneous plants conserved in vinegar for 60 days.

The vitamin C content variation found on fruit pickled in oil can be attributed to the presence of antioxidants in vegetal oils, responsible for a protective effect against oxidation [14] or this result may be attributed to the non-soluble in oil vitamin C characteristic, avoiding the possible leaching. The ascorbic acid content losses are low

in pickled fruits and vegetables (<15%) when compared to the losses that happen in fresh and frozen products [15]. Even though the highest vitamin C levels were found *in natura* fruit, the pickled jurubeba in oil can provide a source of antioxidant.

Figure 3 - Vitamin C content in jurubeba fruit (*Solanum paniculatum* L.) *in natura*, cooked in water and pickled in two preservatives (oil and vinegar) for 1 hour, 30, 60 and 90 days of shelf life, from cultivated plants, spontaneous plants and from market



In this study, we evaluated the extraction of the total carbohydrates soluble in two solvents (water and alcohol). The total carbohydrates content was the highest when water was used as an extractor (Table 3). When alcohol was used, the highest total soluble carbohydrates content occurred in the *in natura* fruit collected from spontaneous plants, followed by the content in the *in natura* fruit from cultivated plants. After thermal treatment and storage, there was a decrease in the amount of total soluble carbohydrates. The decrease of total carbohydrates within shelf time may be observed by using the aqueous extraction, except for the fruit from spontaneous plants and that were conserved for 1 hour in soybean oil.

The content variation caused by the extraction may occur due to various reasons. Heating was used during the soluble carbohydrates extraction in water, while in the alcoholic extraction this treatment did not happen. The solubility was probably increased due to the temperature effect. According to [16], the use of water is viable and the acquired contents were higher when using heated water and the contents are similar to other types of extractors, such as alcohol. Another study comparing aqueous and

alcoholic extractions showed that the soluble in alcohol sugar content is lower than the one obtained in aqueous solution [17].

Table 1 - Carbohydrates content (alcohol and water) (g/100g), total proteins (%) and total lipids (%) in jurubeba fruit (*Solanum paniculatum*L.) *in natura*, cooked in water and pickled in two types of preservatives after 1 hour, 30, 60 and 90 days of shelf life, from three different sources (cultivated plants, spontaneous plants and from market)

		Carbohydrates (alcohol) (g/100g)	Carbohydrates (water) (g/100g)	Proteins (%)	Lipids (%)
	Cultivated	1.18 ± 0.16b	1.60 ± 0.14a	1.48 ± 0.39c	0.65 ± 0.17h
In natura	Spontaneous	1.60 ± 0.14a	1.64 ± 0.13a	1.77 ± 0.10b	1.19 ± 0.06h
	Market	0.65 ± 0.12d	1.30 ± 0.03b	1.43 ± 0.19c	1.50 ± 0.34h
	Cultivated	0.73 ± 0.07d	1.88 ± 0.32a	4.26 ± 0.31a	0.84 ± 0.33h
Cooked	Spontaneous	0.91 ± 0.15c	1.83 ± 0.50a	1.47 ± 0.12c	0.83 ± 0.10h
	Market	0.65 ± 0.11d	1.29 ± 0.21b	1.32 ± 0.33c	1.75 ± 0.22h
		1 hour			
	Cultivated	0.84 ± 0.17d	1.27 ± 0.16b	2.21 ± 0.78b	5.59 ± 2.04 g
Oil	Spontaneous	0.94 ± 0.21c	1.77 ± 0.37a	1.09 ± 0.19c	7.48 ± 0.47 f
	Market	0.59 ± 0.13c	1.12 ± 0.16c	1.76 ± 0.20b	5.92 ± 1.15 g
	Cultivated	0.91 ± 0.21c	1.46 ± 0.44b	1.30 ± 0.34c	0.75 ± 0.24 h
Vinegar	Spontaneous	0.88 ± 0.13c	1.22 ± 0.20b	1.54 ± 0.19c	0.89 ± 0.12 h
	Market	0.63 ± 0.07d	0.95 ± 0.19c	1.42 ± 0.18c	0.33 ± 0.05 h
		30 days			
	Cultivated	0.79 ± 0.14c	1.07 ± 0.05c	4.43 ± 0.14a	17.33 ± 2.26a
Oil	Spontaneous	0.71 ± 0.09d	0.71 ± 0.06c	1.33 ± 0.33c	12.61 ± 2.13c
	Market	0.67 ± 0.10d	0.56 ± 0.07c	1.20 ± .037c	9.23 ± 0.55e
	Cultivated	0.93 ± 0.16c	1.16 ± 0.07b	2.49 ± 0.32b	0.57 ± 0.20h
Vinegar	Spontaneous	0.71 ± 0.09d	1.00 ± 0.20c	1.43 ± 0.50c	1.29 ± 0.22h
	Market	0.67 ± 0.09d	0.50 ± 0.03c	1.21 ± 0.19c	1.39 ± 0.39h
		60 days			
	Cultivated	1.00 ± 0.13c	1.49 ± 0.22b	2.21 ± 0.76b	14.59 ± 2.63b
Oil	Spontaneous	1.02 ± 0.07c	1.28 ± 0.38b	1.20 ± 0.19c	10.95 ± 0.90d
	Market	0.68 ± 0.08d	0.69 ± 0.12c	0.98 ± 0.33c	4.62 ± 0.54g
	Cultivated	0.71 ± 0.12d	0.95 ± 0.12c	2.22 ± 0.29b	0.53 ± 0.09h
Vinegar	Spontaneous	0.83 ± 0.04c	0.92 ± 0.15c	1.21 ± 0.19c	0.47 ± 0.08h
	Market	0.89 ± 0.06c	0.85 ± 0.08c	1.31 ± 0.33c	0.66 ± 0.08h
		90 days			
	Cultivated	0.58 ± 0.04d	0.68 ± 0.04c	1.40 ± 0.67c	17.47 ± 2.84a
Oil	Spontaneous	0.83 ± 0.14c	0.99 ± 0.21c	1.32 ± 0.00c	10.18 ± 1.09e
	Market	0.82 ± 0.03c	0.39 ± 0.03c	1.32 ± 0.33c	13.05 ± 1.05c
	Cultivated	0.60 ± 0.16d	0.87 ± 0.05c	2.84 ± 0.68b	1.00 ± 0.24h
Vinegar	Spontaneous	0.98 ± 0.11c	0.89 ± 0.13c	1.32 ± 0.57c	0.55 ± 0.09h
	Market	0.81 ± 0.14c	0.88 ± 0.18c	1.11 ± 0.18c	0.43 ± 0.10h

*Means with the same lowercase belong to the same group, according to the Scott-Knot test at 5% probability

In general, jurubeba fruit are not rich in total carbohydrates compared with *Solanum melongena* [18], regardless of the used solvent. Studies with *Solanum*

aethiopicum and *S. macrocarpon* show total available carbohydrate content of 3.60 and 6.48 g/100g, respectively [19].

The highest proteins content was found in the fruit collected from cultivated plants (Table 3). Cultivated cooked jurubeba showed high proteins content and the pickling in soybean oil seem to have been efficient to maintain the amount of this macronutrient if compared to raw fruit and the ones pickled in vinegar. The protein content may vary due to various factors, including the ripeness degree of the tissue. The values found for jurubeba varied between 0.98 and 4.43%, lower than the [20] descriptions, who found 12% of total proteins in unripen jurubeba fruit. However, the results reached by our study were close to the ones described for *Solanum melongena*, around 4% [21].

In raw or cooked jurubeba in water for 20 minutes, the total lipids varied between 0.65 and 1.75%, showing no significant difference between the fruit sources (Table 3). However, when these fruit were submitted to the preservatives (oil and vinegar), there were significant alterations. Fruit pickled in soybean oil showed higher total lipids levels when compared to fruit conserved in vinegar. The Brazilian Table of Food Composition (TACO) shows lipids contents in raw jurubeba of 3.9 g/100 g [22], which are higher than the contents described by the Agriculture Ministry [23], around 0.40 g/100g. The highest amount of lipid found in this study in jurubeba pickled in oil, was definitely due to the type of preservative used.

In the harvest day, there was no significant variation in the total carotenoids content (Table 4). Generally, cooking tends to increase the carotenoids content [24], but this result was not observed in jurubeba right after thermal treatment (20 min of cooking). When pickled in two different preservatives (oil or vinegar), fruit from cultivated plants and kept in oil for 1 hour showed higher carotenoid contents (21.30 µg/g), but smaller than the content found right after the cooking (31,99 µg/g). With shelf life, there was an increase in the content until 60 days, in the fruit purchased in market, independently of the type of preservative. The total carotenoids content observed in fruit acquired from the market and pickled in alcohol vinegar for 60 days showed the highest value (149.03 µg/g), followed by the results achieved with in soybean oil for 30 and 60 days (127.10 e 124.30 µg/g, respectively).

Table 2 - Total carotenoids (µg/g), total flavonoids (mg/100g) and total phenols (mg/100g), in jurubeba fruit (*Solanum paniculatum* L.) *in natura*, cooked in water and pickled in two types of preservatives after 1 hour, 30, 60 and 90 days of shelf life from three sources (cultivated plants, spontaneous plants and from market)

		Total carotenoids	Total flavonoids	Total phenols
		µg/g	mg/ 100g	mg/100g
In natura	Cultivated	33.44 ± 1.34e	12.74 ± 0.10g	317.81 ± 25.61i
	Spontaneous	24.85 ± 4.78e	52.90 ± 1.17e	316.74 ± 20.68i
	Market	25.17 ± 3.50e	77,54 ± 1.91d	307.44 ± 12.46i
Cooked	Cultivated	31.99 ± 1.04e	39.79 ± 3.79f	441.04 ± 29.35f
	Spontaneous	19.06 ± 0.37e	50.91 ± 1.69e	655.29 ± 15.04a
	Market	18.92 ± 0.55e	67.92 ± 1.12d	516.84 ± 12.43d
		1 hour		
Oil	Cultivated	21.30 ± 1.18e	37.81 ± 2.35f	611.89 ± 33.84b
	Spontaneous	15.33 ± 1.85f	80.00 ± 1.02d	160.86 ± 8.47j
	Market	10.39 ± 1.16f	86.98 ± 2.20d	146.95 ± 6.02j
Vinegar	Cultivated	16.68 ± 1.18f	30.65 ± 0.96f	552.02 ± 32.02c
	Spontaneous	16.67 ± 2.59f	77.54 ± 0.69d	149.74 ± 5.74j
	Market	15.22 ± 2.13f	66.56 ± 2.86d	131.74 ± 15.38k
		30 days		
Oil	Cultivated	21.31 ± 2.98e	37.48 ± 0.18f	437.41 ± 25.43f
	Spontaneous	22.71 ± 1.49e	82.63 ± 3.83d	115.75 ± 5.03l
	Market	127.10 ± 31.69b	84.27 ± 3.69d	132.78 ± 6.14k
Vinegar	Cultivated	8.89 ± 1.28f	11.52 ± 1.40g	385.17 ± 14.54h
	Spontaneous	11.36 ± 0.83f	79.18 ± 6.60d	129.63 ± 2.55k
	Market	91.39 ± 4.95c	77.81 ± 1.08d	123.32 ± 6.21l
		60 days		
Oil	Cultivated	20.17 ± 0.78e	49,08 ± 0.82e	469.37 ± 9.50e
	Spontaneous	8.96 ± 0.76f	201.65 ± 3.81c	117.61 ± 3.79l
	Market	124.30 ± 9.23b	204.14 ± 1.44c	113.90 ± 3.52l
Vinegar	Cultivated	7.70 ± 2.16f	13.98 ± 2.20g	382.35 ± 8.35h
	Spontaneous	11.78 ± 0.67f	187.42 ± 6.60c	101.74 ± 2.90l
	Market	149.03 ± 3.68a	219.98 ± 5.26b	132.56 ± 8.25k
		90 days		
Oil	Cultivated	27.22 ± 1.06e	40.41 ± 0.61f	416.27 ± 9.19g
	Spontaneous	7.47 ± 0.33f	195.25 ± 9.99c	121.30 ± 5.48l
	Market	7.39 ± 0.24f	248.25 ± 1.32a	95.12 ± 6.03l
Vinegar	Cultivated	5.94 ± 0.91f	10.97 ± 1.69g	33.82 ± 23.73i
	Spontaneous	14.81 ± 0.56f	222.91 ± 6.73b	102.97 ± 2.66l
	Market	65.69 ± 5.10d	207.10 ± 3.15c	102.21 ± 7.79l

*Means followed by the same letters do not differ statistically between themselves, according to the Scott-Knott test at 5% probability.

The highest carotenoids content in jurubeba from market may have occurred due to the deterioration state of the fruit, because they were acquired detached from the peduncles and some already showed small dark spots, which are a signal of oxidation

and deterioration. Probably, because the fruit were already in the begin of senescence, this would induce an easier extraction of this phytochemical, optimized with the heating effect [24] and with the acid medium action, promoted by the alcohol vinegar used in the preservation.

The total flavonoids levels were significantly higher at 60 and 90 days in fruit harvested from spontaneous plants and purchased in market pickled in oil or vinegar (Table 4). The content of these polyphenols was lower in the fruit obtained from cultivated plants, regardless of the preservative type used or of the shelf time. In contrast, the opposite was observed for the total phenols content at 60 days. The highest values were observed in fruit from cultivated plants cooked in water (655.29 mg/100g), followed by the cultivated fruit submitted to thermal treatment and conserved, for 1 hour, in soybean oil (611.89 mg/100g) and in alcohol vinegar (552.02 mg/100g).

The phenolic compounds are antioxidants and are subjected to oxidation during the thermal processing as well as in the storage. Some metabolic reactions may occur during the shelf life, inducing the oxidation of this compound, mainly because of the oxygen and light exposition, but in our study, all pickled jurubeba fruit (oil or vinegar) were in jars and stored in the dark. Even with these precautions, we observed darkening of the fruit with the storage time after 60 days in some treatments (from market and spontaneous).

The phenolic compounds are soluble in water and this property may induce losses by leaching [15]. The phenols losses may be increased by heating and other factors, such as pH, that can result in cell disruption. Moreover, the variations found in pickled jurubeba may be related to the lack of uniformity of the vegetal tissue. In tissues, phenols compounds are found in the cell wall, vacuoles, epidermis and sub epidermis, and in sub cell level and all factors involved in this study, such as heating, shelf life and the preservative types might have affected the fruit structure, inducing the variations found in the (poly)phenol content. However, according to our studies, pickled jurubeba could be consumed, as a phenols source, right after the cooking. Regarding the flavonoids, the fruit acquired from market or collected from spontaneous plants pickled in oil or vinegar are good sources until 90 days. Nevertheless, it is important to state that it

was observed some oxidation characteristics after 60 days, mainly in fruit acquired from market.

Thermally processed fruit showed the lowest vitamin C content, however, when the soybean oil preservative was used, there was an increased vitamin C release and lipids and proteins contents conservation, therefore, soybean oil can be considered a better preservative compared to vinegar. Fruit acquired from market showed the highest carotenoids and flavonoids contents. Decreases in these polyphenols contents occurred faster in fruit collected from both cultivated and spontaneous plants. In order to achieve the highest content of these bioactives, the jurubeba consume should be realized until 60 days of shelf life and pickled in soybean oil.

3.4 Conclusion

Thermally processed fruit showed the lowest vitamin C content, however, when the soybean oil preservative was used, there was an increased vitamin C release and lipids and proteins contents conservation, therefore, soybean oil can be considered a better preservative compared to vinegar. Fruit acquired from market showed the highest carotenoids and flavonoids contents. Decreases in these polyphenols contents occurred faster in fruit collected from both cultivated and spontaneous plants. In order to achieve the highest content of these bioactives, the jurubeba consume should be realized until 60 days of shelf life and pickled in soybean oil.

3.5 Acknowledgments

The authors are grateful to the National Council for Scientific and Technological Development (CNPq, Brazil) (142360/2013-9, 478372/2013-2, 305177/2015-0) and São Paulo Research Fundation (FAPESP) (2013/05644-3) for the financial support.

3.6 References

[1] Mesia-Vela S., Santos M.T., Souccar C., Lima-Landman M.T.R., Lapa A.J., *Solanum paniculatum* L. (Jurubeba): potent inhibitor of gastric acid secretion in mice, Phytomedicine 9 (2002) 508–514.

[2] Benevides C.M.J., Souza R.D.B., Souza M.V., Lopes M.V., Efeito do processamento sobre os teores de oxalato e tanino em maxixe (*Cucumis anuria* L.), jiló (*Solanum gilo*), feijão verde (*Vigna unguiculata* L. Walp) e feijão andu (*Cajanus cajan* L. Mill sp.), Aliment. E Nutr. – Araraquara 24 (2013) 321-327.

[3] Silva M.B., Rodrigues L.F.O.S., Rossi T.C., Vieira M.C.S., Minatel I.O., Lima G.P.P.L., Effects of boiling and oil or vinegar on pickled jurubeba (*Solanum paniculatum* L.) fruit, African J. Biotechnol. 15 (2016) 125–133.

[4] Lima G.P.P., Vianello F., Review on the main differences between organic and conventional plant-based foods, Int. J. Food Sci. Technol. 46 (2011) 1–13.

[5] Horwitz W., Latimer G.W., Official methods of analysis of AOAC International. AOAC international, Gaithersburg, Md., 2005.

[6] DuBois M., Gilles K.A., Hamilton J.K., Rebers P.A., Smith F., Colorimetric method for determination of sugars and related substances, Anal. Chem. 28 (1956) 350–356.

[7] Bligh E., Dyer W., A rapid method of total lipid extraction and purification, Can. J. Biochem. Physiol. 37 (1959) 911–917.

[8] Sims D.A., Gamon J.A., Relationships between leaf pigment content and spectral reflectance across a wide range of species, leaf structures and developmental stages, Remote Sens. Environ. 81 (2002) 337–354.

[9] Popova M., Bankova V., Butovska D., Petkov V., Nikolova-Damyanova B., Sabatini A.G., Marcazzan G.L., Bogdanov S., Validated methods for the quantification of biologically active constituents of poplar-type propolis, Phytochem. Anal. 15 (2004) 235–340.

[10] Singleton V.L., Rossi J.A.J., Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents, Am. J. Enol. Vitic. 16 (1965) 144–158.

[11] Ferreira D.F., Sisvar: a computer statistical analysis system, Ciência E Agrotecnologia 35 (2011) 1039–1042.

[12] Yamaguchi Y., Narita T., Inukai N., Wada T., Handa H., SPT genes: key players in the regulation of transcription, chromatin structure and other cellular processes., J. Biochem. 129 (2001) 185–191.

[13] Manela-Azulay M., Mandarim-de-Lacerda C.A., Perez M.A., Filgueira A.L., Cuzzi

T., Vitamina C, An. Bras. Dermatol. 78 (2003) 265–272.

[14] Horuz T.İ., Maskan M., Effect of cinnamaldehyde on oxidative stability of several fats and oils at elevated temperatures, 1 (2015) 1–8.

[15] Rickman J.C., Barrett D.M., Bruhn C.M., Nutritional comparison of fresh, frozen and canned fruits and vegetables. Part 1. Vitamins C and B and phenolic compounds, J. Sci. Food Agric. 87 (2007) 930–944.

[16] Smith D., Paulsen G.M., Raguse C.A., Extraction of total available carbohydrates from grass and legume tissue, Plant Physiol. 39 (1964) 960–962.

[17] Ayaz F.A., Torun H., Ayaz S., Correia P.J., Alaiz M., Sanz C., Grúz J., Strnad M., Determination of chemical composition of anatolian carob pod (Ceratonia siliqua L.): Sugars, amino and organic acids, minerals and phenolic compounds, J. Food Qual. 30 (2007) 1040–1055.

[18] Sims D.A., Gamon J.A., Relationships between leaf pigment content and spectral reflectance across a wide range of species, leaf structures and developmental stages, Remote Sens. Environ. 81 (2002) 337–354.

[19] Lo Scalzo R., Fibiani M., Francese G., D'Alessandro A., Rotino G.L., Conte P., Mennella G., Cooking influence on physico-chemical fruit characteristics of eggplant (*Solanum melongena* L.), Food Chem. 194 (2016) 835–842.

[20] San José R., Plazas M., Sánchez-Mata M.C., Cámara M., Prohens J., Diversity in composition of scarlet (*S. aethiopicum*) and gboma (*S. macrocarpon*) eggplants and of interspecific hybrids between *S. aethiopicum* and common eggplant (*S. melongena*), J. Food Compos. Anal. 45 (2016) 130–140.

[21] Saeedifar F., Ziarati P., Ramezan Y., Nitrate and Heavy Metal Contents In Eggplant (*Solanum melongena*) cultivated in the farmlands in the south of Tehran-Iran, Int. J. Farming Allied Sci. 3 (2014) 60–65.

[22] Taco. Tabela brasileira de composição de alimentos. NEPA - Unicamp 2011 161p.

[23] MAPA, Manual de hortaliças não-convencionais, MAPA/ACS, Brasília-DF, 2010.

[24] Colle I., Van Buggenhout S., Van Loey A., Hendrickx M., High pressure homogenization followed by thermal processing of tomato pulp: Influence on microstructure and lycopene in vitro bioaccessibility, Food Res. Int. 43 (2010) 2193–2200.

4 Aminas biogênicas em Jurubeba (*Solanum paniculatum* L.), após o processamento térmico, e tempo de armazenamento em dois tipos de conservadores

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Biogenic amines in Jurubeba (*Solanum paniculatum* L.) **after thermal processing, and storage time in two types of preservatives**

ABSTRACT

The presence of biogenic amines, such as histamine and tiramine, in canned food is usually related to health problems such as allergies. However, others bioactives amines may be present and induce some diseases. Some biogenic amines can react with nitrate and form nitrosamines, compounds harmful to human health. In this research, we evaluated qualitatively and quantitatively some biogenic amines and nitrate content in jurubeba conserved in oil or vinegar. The fruits were obtained from cultivated plants, or spontaneous plants, or purchased from market. The fruit were analysed raw and after cooking. The thermally processed fruit were preserved in soybean oil or alcohol vinegar and evaluated after 1 hour of canning and at 30, 60 and 90 days of storage, totaling 30 treatments with three replicates. Variations in the contents of spermine (0.02 to 3.11 mg / 100g), putrescine (18.41 to 86.48 mg / 100g), cadaverine (0.01 to 19.02 mg / 100g), spermidine (0.04 to 32.32 mg / 100g), histamine 0.01 to 8.43 mg / 100g) and tyramine (0.16 to 11.74 mg / 100g) were found depending on the place of fruit were picking, as well as the type of preservative and time of storage. The nitrate levels did not exceed the established limits, mainly in vinegar jurubeba, which also showed the lowest levels of biogenic amines.

Key words: cooking, polyamines, nitrate, Solanaceae, HPLC.

4.1 Introduction

The polyamines and some biogenic amines (BA) are related to many biological functions as cell duplication and differentiation with the stabilization of membranes and nucleic acids, beyond acting as secondary messengers (Larqué et al., 2007). Some biogenic amines have anti-inflammatory properties (Moulinoux e Delcros, 2005), but may present harming effect on human health. The concentration of these compounds can vary according to the type of cell, but the higher levels are generally found in tissues with a high growth rate (Moinard et al., 2005). The polyamines in low concentrations are not considered a risk to health. However, when consumed excessively, they can cause physiological damage and toxic effects (Saaid et al., 2009). Some studies evidence the possible relation between the polyamines levels and cancer (Tassoni et al., 2000).

Fresh fruits and juices are particularly rich in putrescine (Shalaby, 1996), while the green vegetables are richer in spermidine (Valero et al., 2002). According to various authors, some processes using heat can influence the polyamines content (Cirilo et al., 2007, Silva et al., 2016). Carelli et al. (2007) affirm that the alteration in the polyamines levels in canned or pickleds foods can be a result of the by the microbial decarboxylation of amino acids during the prepare or even during the storage. The consume of some biogenic amines can cause allergic reactions such as fever, hypertension, vomits, allergic processes (itching, rash), difficulty in breathing, among others (Naila et al., 2010).

Jurubeba (*Solanum paniculatum*) is a non conventional vegetable, generally consumed in canned form. This vegetable is used in the Brazilian folk medicine in the treatment of chronic hepatitis, anti-thermic, jaundice, among other gastrointestinal disorders (Mesia-Vela et al., 2002). Even though the researches related to the jurubeba production and quality have increased in the last years, little is still known about the biogenic amines concentrations in canned jurubeba. The determination of biogenic amines in foods is of great interest, not only due to its possible toxicity, but because they can be used as indicators of freshness quality and food deterioration (Silla Santos, 1996).

Some substances used as conservatives can inhibit the formation of biogenic amines. Studies with sodium nitrate (Kurt e Zorba, 2010), ascorbic acid (Bozkurt e Erkmen, 2004), glycine (Mah e Hwang, 2009), among others, have showed efficiency in inhibiting the biogenic amines content. However, only a few studies show the effects of oil or vinegar as conservatives in inhibiting the biogenic amines content.

The biogenic amines can form N-nitrosaminas in many foods, including vegetables, due to the presence of nitrate and nitrite (Yurchenko e Mölder, 2007). Nitrate naturally occurs in plants and can be transformed in nitrite in the oral cavity and in the stomach (Duncan et al., 1997), which can react with amines and form n-nitrous compounds. Furthermore, studies show that the nitrate consumption is associated with decreased risk of cardiovascular disease and blood pressure (Larsen et al., 2007).

As canned foods can contain different contents of biogenic amines and nitrate, we identified and quantified biogenic amines and nitrate in jurubeba (*Solanum paniculatum* L.) canned in oil or vinegar, during the storage.

Por outro lado, estudos demonstram que o consumo de nitrato esta relacionado com a diminuição de riscos de doenças cardiovasculares e diminuição da pressão sanguínea (Larsen et al., 2007).

4.2 Material and Methods

4.2.1 Samples

Jurubebas fruit were harvested from cultivated and spontaneous plants and were purchased in market. The fruit from cultivated plant were produced according to the recommendation for jiló (*Solanum gilo*) culture. The fruit form spontaneous plants were harvested with the same physiological age of the cultivated fruit and the ones purchased in the market were selected guaranteeing all fruit showed uniformity in color (light green), without any lesions on the surface and with the same size (approximately 1.5 cm of diameter) were randomly picked.

4.2.2 Thermal process and conserves prepare

The fruit sanitation was performed with tap water and were then immersed for 10 minutes in chlorinated water (100 mg/mL). It was used 5 kg of fruit for each acquisition form. One portion was retained raw and the other was submitted to thermal processing, through cooking of 500 g in 1L of boiling water in a stainless steel pan with a lid, and cooked for 20 minutes. The cooking time followed the recommendations by Silva et al. (2016). The samples were drained off and cooled rapidly. The cooked fruit was stored in glass jars of 300 mL, previously sterilized in boiling water for 30 minutes, containing 2 g of NaCl and 150 mL of commercial soybean oil or alcohol vinegar (acetic fermented of alcohol and water, containing 4% of acetic acid). The recipients were covered with Parafilm®, closed with plastic lids and stored on shelves at 25 ± 2 ^{o}C, protected from light. The evaluations were performed in the raw fruit, after 1 hour of cooking, 30, 60 and 90 days after the prepare of the conserves.

4.2.3 Nitrate content

The nitrate content was determined by the Nitrate Meter (C-141, **Horiba**-**Cardy**®), characterized by an interval of linearity of 0 to 9900 ppm and previously calibrated with patterns of known concentrations of nitrate (0, 500, 1000, 1500, 2000 and 2500 mg L^{1} NO^{3}). The readings were performed using the samples ground in mini-turrax without dilution.

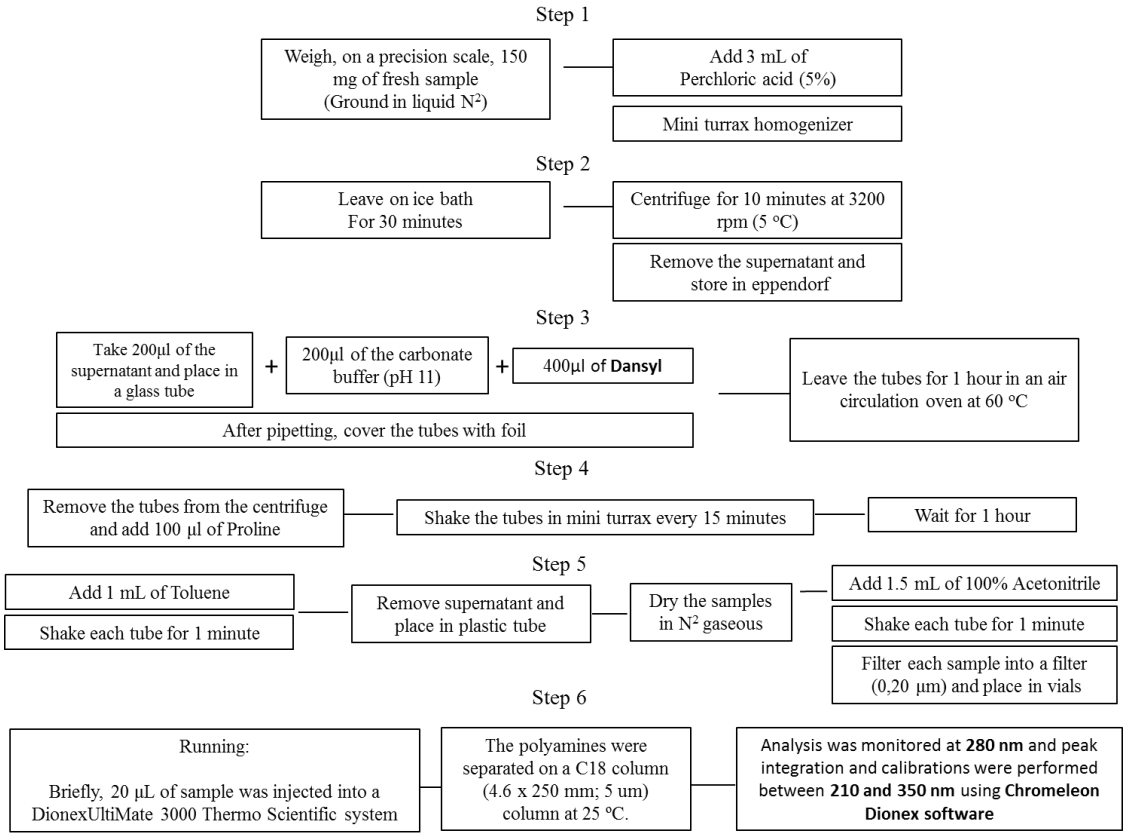
4.2.4 Extraction and quantification of polyamines

Raw and processed jurubeba fruit were ground in the cryogenic mill (Spex Sample Prep 6770, MA, USA), during 5 minutes and stored in a freezer a -80 °C, protected from light. Determination of the PAs content was performed according Dadáková et al. (2009). The polyamines and biogenic amines (spermine, putrescine, cadaverine, spermidine, histamine and tiramine) were extracted and isolated according to the procedureof Flores and Galston (1982) modified by Lima et al. (2008). Determinations of the BAs content was performed according Dadáková et al. (2009). The jurubeba samples were homogenized in perchloric acid 5% v/v during 1 hour and subsequently centrifuged (8,000 g, 30 min, 4 ^{o}C) (MIKRO 220/220R - Hettich Lab

Technology, USA). 200 µl of supernatant was put in test tubes containing Na_2CO_3 (4,5 mol L^{-1}) and it was added dansil chloride (Sigma Aldrich). The samples remained in the dark, at 60 ^{o}C for 1 hour. Then, it was added 100 µL proline (99 %) and the samples were maintained at room temperature for 30 min. All samples were homogenized at each 15 minutes using mini-turax (Marconi, Brasil) and 1000 µL of toluene were added to the tubes to extract the dansylated PAs.

Finally, sample aliquots were dried in gaseous nitrogen and resuspended in 1.5 mL of HPLC grade acetonitrile. The samples (20 µL) were injected into a UHPLC system (Ultimate 3000 BioRS, Dionex-Thermo Fisher Scientific Inc., USA), equipped with a diode array detector (DAD-3000RS), set at 225–300 nm. The flux rate was 0.7 mL / min, using n Ace 5C18 (Advanced Chromatography Technologies, UK) column (5 _m, 25 cm × 4.6 mm). The mobile phase consisted in acetonitrile 100% (solvent A) and acetonitrile 50% (solvent B). The chromatographic run gradient scheme performed was established using acetonitrile 100% (solvent A) and acetonitrile 50% (solvent B): 0–4 min, 40% A + 60% B; 4–8 min, 60% A + 40% B; 8–12 min, 65% A + 35% B; 12–15 min, 85% A + 15% B;15–21 min, 95% A + 5% B; 21–22 min, 85% A + 15% B; 22 min, 75%A + 25% B.

Figure 1 - Flowchart of the method used for the polyamines extraction



4.3 Results and Discussion

Table 1 shows the values of polyamines observed for in natura fruits obtained from cultivated plants, spontaneous plants and obtained in the market. It is possible to observe that plants of cultivated origin presented higher value of total polyamines 64.97 mg / 100g.

Table 1 - Spermine, putrescine, cadaverina, spermidine, histamine, tiramine and total polyamines (∑) (mg/100g) in jurubeba fruit (*Solanum paniculatum* L.) raw, from three forms of obtaining the fruit (cultivated plants, spontaneous plants and fruit purchased from the market)

		Spermine	Putrescine	Cadaverine	Spermidine	Histamine	Tiramine	∑
In natura	Cultivated	0,52 ± 0,10	35,54 ± 2,07	6,77 ± 0,98	13,01 ± 1,64	5,26 ± 1,79	3,87 ± 1,3	64,97
	Spontaneous	0,56 ± 0,87	18,41 ± 0,82	0,20 ± 0,08	5,09 ± 1,13	0,13 ± 0,03	1,52 ± 0,32	25,91
	Market	0,47 ± 0,21	44,63 ± 0,60	0,23 ± 0,16	4,63 ± 0,62	0,16 ± 0,03	2,52 ± 0,17	52,64

From the all analyzed amines, the highest contents were found for putrescine and there is a tendency from 30 days of storage, of higher contents occur in the fruit obtained in market (86,48 mg/100g in oil and 63,47 mg/100g in vinegar), regardless of the conservative type used. Sayem-el-Daher et al. (1984) state that the levels of putrescine, spermine, spermidine, cadaverine and tiramine are positively correlated to the temperature and to storage time of the products.

In these fruit, we found lower contents of cadaverine at 30 days, in the fruits conserved in oil (0,56 mg/100g), or in vinegar (0,72 mg/100g). Other studies show that putrescine occurs in higher quantities in some canned vegetables, as in corn (71%) (Bandeira et al., 2012). According to Alvarez and Moreno-Arribas, (2014), both putrescine and cadaverine are thermostable amines and are not activated by thermal treatments used in the foods processing and preparation.

The biogenic amines, such as putrescine and cadaverine have been correlated with the deterioration of some foods as fish, meat and vegetables (Riebrov et al., 2004). Both putrescine and cadaverine do not have toxic effects, however, they increase the adverse effects of other amines as histamine and tiramine (Landete et al., 2011), by competing for the detoxifying enzymes and by reacting with nitrites, forming carcinogenic nitrosamines (Silla Santos, 1996). There are evidences that putrescine could have a role in promoting the malignancy degree of adenomas in murine model (Ignatenko et al., 2006) and high concentrations of this diamine were also detected in gastric carcinomas caused by *Helicobacter pylori* (Shah e Swiatlo, 2008) and colorectal cancer (Wallace e Caslake, 2003).

In jurubeba fruit, the putrescine content seems to have been affected by the pH. There is a lower concentration of this amines in the fruit canned in vinegar, compared to the fruit canned in oil, except for the fruit purchased from the market. These fruit, even in vinegar, could already be in an advanced state of senescence, because they were purchased already detached from the peduncles and with an initial oxidation aspect (dark spots), which may have compromised the putrescine content. Silva et al. (2016) also observed a lower putrescine content in jurubeba fruit stored in vinegar. There is a higher difficulty of occurring microbial deterioration in low pH, one of the main causes for

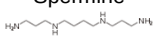
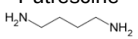
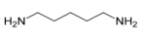
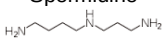
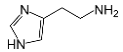
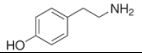
the formation of biogenic amines (Preti et al., 2016), which might have influenced the putrescine levels in canned jurubeba with vinegar.

In this way, the consumption of fruit obtained in the market and canned, both in oil and in vinegar, can provide a higher content of putrescine, which can be harmful to the organism and cause allergic reactions in sensible people. Other studies show that the accumulation of putrefactive amines, as putrescine, occurs in function of the storage time (Bandeira et al., 2012). Probably, these jurebeba fruit, could be unfit for the consumption, because they were obtained with two or more days of harvest and because they were free from the clusters and they should not be used for ingestion after 30 days of the canned making. Thus, it is recommended to use freshly harvested fruits for the manufacture of canned jurubeba fruit. Possibly, for being a Solanaceae and because the other vegetables of this group are ingested in conserves, it would be interesting that the analysis of biogenic amines are used as a form of food safety and used the food industry, as a routine analysis.

The highest spermidine content in raw fruit was found in the ones obtained from cultivated plants (13,01 mg/100 g), shortly after the thermal treatment (13,97 mg/100g) and one hour after the canning in oil (22,61 mg/100g) or in vinegar (33,32 mg/100g). With the storage time, the spermidine levels tend to decrease, presenting at 90 days, the lowest contents regarding ra*w* fruit, mainly in the ones harvested from spontaneous plants and conserved in vinegar (0,04 mg/100g). Lower spermidine contents after the processing in relation to fresh vegetables were also reported by Moret et al. (2005). Spermidine, along with spermine, are associated with various macromolecules as DNA, RNA, chromatins and proteins, promoting its stabilization and are related to the growth and development (Kusano et al., 2008). Thus, the consumption of these polyamines is important for the cells, however, the excess might not be interesting due to its relation with the carcinogenesis (Gugliucci, 2004).

The form of obtaining the fruit also showed differences regarding the spemine levels. The lowest levels were obtained in the fruit from cultivated plants, even after the thermal processing and in both of the two types of conserves (oil or vinegar), during the storage time. The levels of this polyamine increase after 30 days in conserve for the fruit harvested from spontaneous plant, as well as for the ones obtained in the market.

Table 2 - Spermine, putrescine, cadaverina, spermidine, histamine, tiramine and total polyamines (∑) (mg/100g) in jurubeba fruit (*Solanum paniculatum* L.) cooked in water and canned in two types of conservatives with 1 hour, 30, 60 and 90 shelf days, from three forms of obtaining the fruit (cultivated plants, spontaneous plants and fruit purchased from the market)

		Spermine 	Putrescine 	Cadaverine 	Spermidine 	Histamine 	Tiramine 	∑
	Cultivated	0,64 ± 0,13e	32,62 ± 2,96g	10,92 ± 0,58b	13,97 ± 1,23c	8,43 ± 1,70a	4,95 ± 0,46c	71,53d
Cooked	Spontaneous	0,60 ± 0,18e	46,63 ± 2,59e	1,08 ± 0,19f	22,47 ± 0,85b	0,37 ± 0,09d	7,77 ± 0,21b	78,92c
	Market	0,52 ± 0,33e	29,25 ± 1,85h	0,56 ± 0,10f	8,85 ± 0,62e	0,24 ± 0,02d	2,49 ± 0,94d	41,92g
				1 hour				
	Cultivated	0,57 ± 0,12e	36,70 ± 2,12g	9,06 ± 1,00c	14,83 ± 1,09c	7,99 ± 0,58a	4,80 ± 0,88c	73,95d
Óil	Spontaneous	0,52 ± 0,21e	55,34 ± 1,77d	0,94 ± 0,47f	22,61 ± 0,59b	0,76 ± 0,08d	11,74 ± 0,17a	91,92b
	Market	0,60 ± 0,11e	40,93 ± 2,69f	1,06 ± 0,12f	7,69 ± 0,83e	0,48 ± 0,02d	2,55 ± 0,52d	53,32f
	Cultivated	0,49 ± 0,19e	25,57 ± 2,91h	8,99 ± 0,32c	13,22 ± 1,32c	7,53 ± 0,94a	4,61 ± 0,47c	60,42e
Vinegar	Spontaneous	0,53 ± 0,02e	58,23 ± 0,56d	1,62 ± 0,35e	32,32 ± 1,30a	0,85 ± 0,01d	10,57 ± 2,59a	104,14a
	Market	0,68 ± 0,14e	34,68 ± 2,65g	2,60 ± 0,08e	9,57 ± 1,16e	1,42 ± 0,26c	3,39 ± 0,42d	52,35f
				30 days				
	Cultivated	0,23 ± 0,09e	32,43 ± 1,61g	6,76 ± 1,62d	11,87 ± 1,40d	7,57 ± 1,32a	3,22 ± 0,83d	62,75e
Óil	Spontaneous	2,93 ± 0,09a	35,66 ± 0,46g	19,19 ± 0,10a	6,27 ± 0,29f	0,54 ± 0,14d	9,73 ± 0,05a	74,32d
	Market	2,,66 ± 0,94b	86,48 ± 1,55a	0,56 ± 0,12f	4,78 ± 1,00f	2,45 ± 0,25c	6,28 ± 1,23c	103,04a
	Cultivated	0,38 ± 0,06e	27,37 ± 1,46h	6,35 ± 0,24d	9,21 ± 1,04e	5,09 ± 0,54b	3,22 ± 0,35d	51,63f
Vinegar	Spontaneous	1,46 ± 0,08c	23,70 ± 2,80i	18,99 ± 0,09a	3,49 ± 1,17g	0,75 ± 0,11d	10,13 ± 2,38a	58,54e
	Market	3,11 ± 0,33a	63,47 ± 4,36c	0,72 ± 0,37f	4,74 ± 0,28f	1,93 ± 0,72c	6,28 ± 1,23c	80,27c
				60 days				
	Cultivated	0,38 ± 0,09e	34,89 ± 4,34g	8,81 ± 1,32c	13,98 ± 0,85c	7,38 ± 1,31a	4,68 ± 0,36c	70,14d
Óil	Spontaneous	1,33 ± 0,26d	67,28 ± 2,48c	19,02 ± 0,12a	3,50 ± 0,52g	1,00 ± 0,08d	10,65 ± 0,65a	103,00a
	Market	2,46 ± 0,19b	56,62 ± 3,62d	0,54 ± 0,16f	4,92 ± 1,70f	1,12 ± 0,05d	4,47 ± 0,2c	70,79d
	Cultivated	0,28 ± 0,08e	23,26 ± 1,44i	6,48 ± 0,50d	9,60 ± 0,82e	5,64 ± 0,46b	3,41 ± 0,28d	48,68f
Vinegar	Spontaneous	1,73 ± 0,85c	26,39 ± 1,19h	19,07 ± 0,12a	3,08 ± 0,80g	1,42 ± 0,04c	10,19 ± 2,9a	61,89e
	Market	2,33 ± 0,65b	82,18 ± 1,63b	0,70 ± 0,10f	4,09 ± 0,46g	1,60 ± 0,17c	4,47 ± 0,2c	95,37b
				90 days				
	Cultivated	0,34 ± 0,05e	37,36 ± 2,45g	8,29 ± 1,25c	11,14 ± 0,91d	7,89 ± 1,09a	4,10 ± 0,36c	69,13d
Óil	Spontaneous	1,23 ± 0,30d	56,93 ± 3,10d	19,02 ± 0,04a	2,76 ± 0,35g	0,96 ± 0,21d	10,03 ± 0,05a	90,93b
	Market	1,54 ± 0,42c	44,58 ± 2,03e	0,57 ± 0,04f	3,15 ±0,18g	0,87 ± 0,17d	5,98 ± 0,12c	55,65f
	Cultivated	0,41 ± 0,08e	27,65 ± 2,90h	6,86 ± 0,69d	9,31 ± 0,80e	5,96 ± 0,5b	3,23 ± 0,25d	53,43f
Vinegar	Spontaneous	0,02 ± 0,01e	20,83 ± 0,26i	0,01 ± 0,0f	0,04 ± 0,01h	0,01 ± 0,00d	0,16 ± 0,01e	21,06h
	Market	2,14 ± 0,35c	53,71 ± 2,92d	0,39 ± 0,18f	3,95± 0,64g	0,84 ± 0,001d	5,98 ± 0,12c	67,02d
Average		1,11	42,99	6,63	9,45	3,00	5,99	69,18
CV (%)		29,87	5,75	8,93	9,68	20,59	17,46	4,65

According to Kalač e Krausová (2005), the levels to induce toxicity by putrescine, spermidine and spermine should be 2000, 600 and 600 mg/kg corporal weight, respectively. Our results show that jurubeba does not contain toxic levels of these three amines, because the jurubeba consumption in one meal, generally varies from 1 to 2 soup spoons, which results in about 10 grams of fruit. Thus, the amines availability would be really small and would not present toxicity to the consumer. We also noticed that the type of conservative had little influence in the indution of the total polyamines levels, except for the pH in the decreasing of putrescine.

Fruit from cultivated plants showed, in all canned treatments, high histamine contents, varying between 5.26 to 98.34 mg/100g, while in spontaneous plants, the content is lower (0.01 to 1.42 mg/100g), varying according to preservative type used and to the storage time. This biogenic amine can cause toxicity in high levels, which could be confused with allergic processes (Ladero et al., 2012). The histamine toxic levels vary between 10 to 100 mg/100g (Larqué et al., 2007) and can have its effect increased when there are high putrescine and cadaverine contents. It is described that the ingestion of 25 mg of histamine could cause intoxication symptoms (Larqué et al., 2012). The histamine content found in 100 g of jurubeba is lower than the values described causing health problems and the usual consumed quantity per person, around 10 g, is not enough to cause health problems, as allergic processes.

While the histamine content was lower in spontaneous plants, the tiramine content was higher in fruit from these plants and thermally processed, showing an increase with storage time, except for canned fruit in vinegar after 90 days. This biogenic amine can cause headaches and migraine, depending on the content in the food. According to Eerola et al. (1998), the ingestion of 10 to 100 mg of tiramine can induce toxicity. The tiramine analyses via UPLC in jurubeba show that the content did not reach the toxic or limit level.

The lowest tiramine content in jurubeba was found after processing (0.16 mg/100 g, canned in vinegar after 90 days) and the highest content occurred in fruit canned in oil after 1 hour (11.74 mg/100 g). It was possible to note that the processing and the storage promoted a raise of tiramine in jurubeba regarding the raw fruit, regardless of how the fruit were obtained, namely in jurubeba *in natura* from cultivated plants, it was

found 3.87 mg/100 g, while in fruit from spontaneous plants, the value was 1.52 mg/100 g and, when purchased in the market, the fruit showed 2.52 mg/100 g of tiramine.

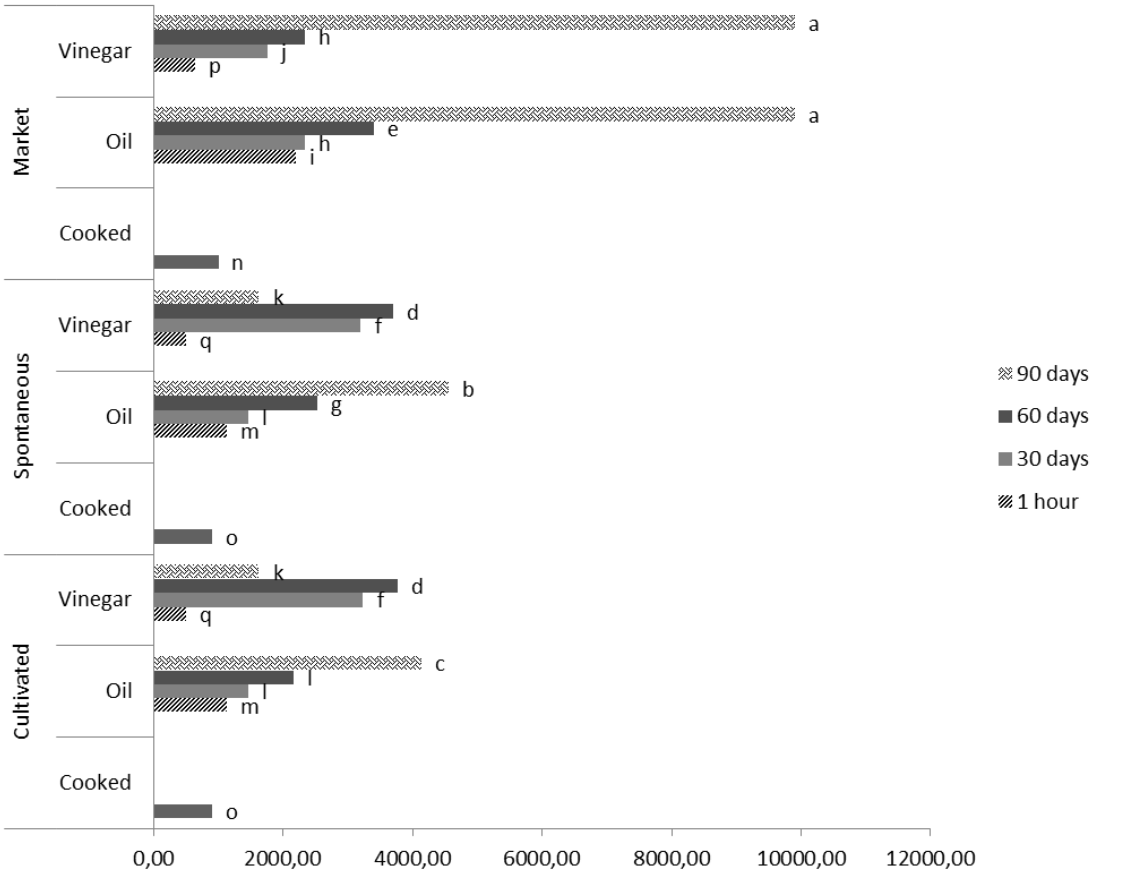
The cooking for 20 minutes did not decrease the amines content, neither when analyzed individually, neither in the total sum, as described in some studies. According to Naila et al. (2010), cooked vegetables can have its biogenic amines content decreased, due to lixiviation to the cooking water, decreasing a possible harmful effect to the consumer. According to Gonzaga et al. (2009), these compounds are stable in the heating, and after the cooking or the exposition to the heat, does not eliminate its toxic effect.

Fruits of jurubeba in natura showed 853.00 ppm of nitrate in cultivated plants, 886.00 ppm in extractivism plants and 837.00 in plantations acquired in the market. Jurubeba cooked and stored in vinegar for 1 hours showed the lowest nitrate content (Figure 2), while the highest value was observed in the fruit purchased from the market and stored for 90 days, both in soybean oil or alcohol vinegar. The FAO (2000), responsible for the worldwide agriculture and food, reports that $2.5 – 5 \times 10^3$ ppm is the maximum allowable daily intake of nitrate (Favaro-Trindade et al., 2007).

In jurubeba, the nitrate levels found are lower than the described by FAO, except for the fruit canned in soybean oil, for 90 days and purchased in market, probably due to the difficulty of lixiviation of nitrate in soybean oil. In contrast, the conservation in vinegar for 90 days induced decreased in the levels of this compound. Even showing higher levels in some treatments, the consumption in one meal, is not higher than 10 fruit, with each one weighting approximately 1 gram.

The nitrate toxicity in humans is low. From 5 to 10 % of the total NO^{3-} ingested is converted in nitrite (NO^{2-}) through oral saliva and digestive system (Boink e Speijers, 2001). When the nitrite enters the bloodstream, it can oxide the hemoglobin iron, producing methemoglobin, which is an inactive form of hemoglobin, incapable of transporting O_2 to the respiratory chain, causing methemoglobinemia, inducing anoxia in the cells (Wright e Davison, 1964).

Figure 2 - Nitrate content (ppm) in jurubeba fruit (*Solanum paniculatum* L.) cooked in water and canned in two types of conservatives with 1 hour, 30, 60 and 90 shelf days, from three forms of obtaining the fruit (cultivated plants, spontaneous plants and fruit purchased from the market)



Another problem of the nitrate presence in foods with expressive biogenic amines content, is the formation of N-nitrosamine, evidenced by the raise of nitrosamine urinary followed by the ingestion of foods containing nitrate and amines (Doyle et al., 1993). However, in canned jurubeba, the nitrate content did not exceed the value proposed by FAO and the fruit conserved in vinegar tend to present lower some amines contents, as tiramine, beyond putrescine and cadaverine, which can potentiate the histamine effect. Thus, jurubeba canned in vinegar contain low biogenic amines and nitrate levels, potential responsible for some allergies and other diseases.

4.4 Conclusion

This study showed that jurubeba used in food and popular medicine can be consumed canned. Its not demonstrating toxic effects due to the presence of biogenic amines in harmful levels to the organism. The conserves should be done using fruit from cultivated plants and in vinegar, for showing lower biogenic amines and nitrate levels. Jurubeba fruit acquired in markets, which had already been harvested a long time, can be substantial allergenic sources, which contain higher contents of putrescine and cadaverine for being in an advances state of senescence, enhancing the harmful effects caused by histamine and tiramine. The results obtained with canned jurubeba, indicates that analyses of biogenic amines are important for the health and should be performed in other fruits and vegetables from this family, when consumed in conserves.

4.5 References

Alvarez MA, Moreno-Arribas MV. The problem of biogenic amines in fermented foods and the use of potential biogenic amine-degrading microorganisms as a solution. Trends Food Sci Technol. Elsevier Ltd; 2014;39:146–155.

Bandeira CM, Evangelista WP, Gloria MBA. Bioactive amines in fresh, canned and dried sweet corn, embryo and endosperm and germinated corn. Food Chem. 2012;131:1355–1359.

Boink A, Speijers G. Health effects of nitrates and nitrites, a review. Acta Hortic. 2001;563:29–36.

Bozkurt H, Erkmen O. Effects of temperature, humidity and additives on the formation of biogenic amines in sucuk during ripening and storage periods. Food Sci Technol Int. 2004;10:21–28.

Carelli D, Centonze D, Palermo C, Quinto M, Rotunno T. An interference free amperometric biosensor for the detection of biogenic amines in food products. Biosens Bioelectron. 2007;23:640–647.

Cirilo, Marcos PG, Coelho AFS, Araújo CM, Gonçalves FRB, Nogueira FD, Glória, Beatriz A. Profile and levels of bioactive amines in instant coffee. J Food Compos Anal. 2007;20:451–457.

Dadáková E, Pelikánová T, Kalač P. Content of biogenic amines and polyamines in some species of European wild-growing edible mushrooms. Eur Food Res Technol. 2009;230:163–171.

Eerola S, Otegui I, Saari L, Rizzo A. Application of liquid chromatography-atmospheric pressure chemical ionization mass spectrometry and tandem mass spectrometry to the determination of volatile nitrosamines in dry sausages. Food Addit Contam. 1998;15:270–9.

Favaro-trindade CS, Marcatti B, Moretti TS, Petrus RR, Almeida E De, Ferraz JBS. Hidropônico e Convencional na Qualidade de Alface Lisa. 2007;111–115.

Gonzaga VE, Lescano AG, Huamán A a, Salmón-Mulanovich G, Blazes DL. Histamine levels in fish from markets in Lima, Perú. J Food Prot. 2009;72:1112–1115.

Gugliucci A. Polyamines as clinical laboratory tools. Clin Chim Acta. 2004;344:23–35.

Ignatenko NA., Besselsen DG., Roy U. KB., Stringer DE, Blohm-mangone KA., Padilla-Torres JL., Guillen-R JM., Gerner EW. A Traditional Mediterranean Diet Decreases Endogenous Estrogens in Healthy Postmenopausal Women A Traditional Mediterranean Diet Decreases Endogenous Estrogens in Healthy Postmenopausal Women. Nutr Cancer. 2006;56:253–259.

Kalač P, Krausová P. A review of dietary polyamines: Formation, implications for growth and health and occurrence in foods. Food Chem. 2005;90:219–230.

Kurt S, Zorba Ö. Effect of ripening period, nitrite level and heat treatment on the chemical characteristics of turkish dry fermented sausage (sucuk). Asian-Australasian J Anim Sci. 2010;23:1105–1111.

Kusano T, Berberich T, Tateda C, Takahashi Y. Polyamines: Essential factors for growth and survival. Planta. 2008;228:367–381.

Ladero V, Fernández M, Calles-Enríquez M, Sánchez-Llana E, Cañedo E, Martín MC, Alvarez MA. Is the production of the biogenic amines tyramine and putrescine a species-level trait in enterococci? Food Microbiol. Elsevier Ltd; 2012;30:132–138.

Landete JM, De Las Rivas B, Marcobal A, Muñoz R. PCR methods for the detection of biogenic amine-producing bacteria on wine. Ann Microbiol. 2011;61:159–166.

Larqué E, Sabater-Molina M, Zamora S. Biological significance of dietary polyamines. Nutrition. 2007. p. 87–95.

Larsen FJ, Weitzberg E, Lundberg JO, Ekblom B. Effects of dietary nitrate on oxygen cost during exercise. Acta Physiol. 2007;191:59–66.

Mah JH, Hwang HJ. Effects of food additives on biogenic amine formation in Myeolchi-jeot, a salted and fermented anchovy (Engraulis japonicus). Food Chem. Elsevier Ltd; 2009;114:168–173.

Maxa, E. and Brandes, W. 1993, Biogene Amine in frucht saften, *Mitt Klosterneuburg*. 43, 101.

Mesia-Vela S, Santos MT, Souccar C, Lima-Landman MTR, Lapa a J. Solanum paniculatum L. (Jurubeba): potent inhibitor of gastric acid secretion in mice. Phytomedicine. 2002;9:508–514.

Moinard C, Cynober L, Debandt J. Polyamines: metabolism and implications in human diseases. Clin Nutr. 2005;24:184–197.

Moret S, Smela D, Populin T, Conte LS. A survey on free biogenic amine content of fresh and preserved vegetables. Food Chem. 2005;89:355–361.

Naila A, Flint S, Fletcher G, Bremer P, Meerdink G. Control of biogenic amines in food - existing and emerging approaches. J Food Sci. 2010;75.

Preti R, Bernacchia R, Vinci G. Chemometric evaluation of biogenic amines in commercial fruit juices. Eur Food Res Technol. Springer Berlin Heidelberg; 2016;242:2031–2039.

Saaid M, Saad B, Hashim NH, Mohamed Ali AS, Saleh MI. Determination of biogenic amines in selected Malaysian food. Food Chem. Elsevier Ltd; 2009;113:1356–1362.

Sayem-el-Daher, N.; R. E. Simard & A. G. Roberge, 1984. Extraction and determination of biogenic amines in ground beef and their relation to microbial quality. Lebensm. Wiss. U. Technol., 17: 20-23.

Shah P, Swiatlo E. A multifaceted role for polyamines in bacterial pathogens. Mol Microbiol. 2008;68:4–16.

Shalaby AR. Significance of biogenic amines to food safety and human health. Food Res Int. 1996;29:675–690.

Silla Santos MH. Biogenic amines: Their importance in foods. Int J Food Microbiol. 1996;29:213–231.

Silva MB, Rodrigues LFO., Rossi T., Vieira MC., Minatel IO, Lima GPP. Effects of boiling

and oil or vinegar on pickled jurubeba (Solanum paniculatum L.) fruit. African J Biotechnol. 2016;15:125–133.

Tassoni A, Van Buuren M, Franceschetti M, Fornalè S, Bagni N. Polyamine content and metabolism in Arabidopsis thaliana and effect of spermidine on plant development. Plant Physiol Biochem. 2000;38:383–393.

Valero D, Martínez-Romero D, Serrano M. The role of polyamines in the improvement of the shelf life of fruit. Trends Food Sci Technol. 2002;13:228–234.

Wallace HM., Caslake R. Polyamines and colon cancer. Biochem Soc Trans. 2003;31:381–383.

Wright MJ, Davison KL. Nitrate accumulation. 1964; 51p.

Yurchenko S, Mölder U. The occurrence of volatile N-nitrosamines in Estonian meat products. Food Chem. 2007;100:1713–1721.

5 REFERÊNCIAS BIBLIOGRAÁFICAS

ATOUI, A. K.; MANSOURI, A.; BOSKOU, G.; KEFALAS, P.; *Food Chem*. 2005, 89p.

BRASIL. Ministério da Saúde. *Alimentos regionais brasileiros*. Brasília, DF, 2002. 140p.

BRASIL. Comissão Nacional de Normas e Padrões para Alimentos. Resolução nº 13, de maio de 1977. Estabelece características mínimas de identidade e qualidade para as hortaliças em conserva. *Diário Oficial [da] República Federativa do Brasil, Poder Executivo*, Brasília, DF, seção 1.

CAMPOS, F. M.; LIMA, A. DE. S.; MAIA, G. E. G.; PASQUI, S. C. Determinação dos teores de vitamin C em hortaliças minimamente processadas. *Alimentos e Nutrição,* Araraquara, v. 19, n. 3, p. 329-335, 2008.

COIMBRA, R. 1958. *Notas de fitoterapia*. Laboratório Clínico Silva Araújo, Rio de Janeiro.

CORREIA, P. *Dicionário das Plantas Úteis do Brasil*, Ed. Imprensa Nacional, Rio de Janeiro, v. 3, p. 545, 1984.

FALLER, A. L. K.; FIALHO E. disponibilidade de polifenóis em frutos e hortaliças consumidas no Brasil. Revista Saúde Pública. 43. (2), 211-8. 2009.

FARMACOPÉIA DOS ESTADOS UNIDOS DO BRASIL. 1959. 2. ed. São Paulo, Ed. Gráfica Siqueira. p.543-544.

FERREIRA, D. F. *Análises estatísticas por meio do Sisvar para Windows versão 5.3*. In: REUNIÃO ANUAL DA REGIÃO BRASILEIRA DA SOCIEDADE INTERNACIONAL DE BIOMETRIA, 45., 2000. São Carlos. Anais... São Carlos: UFSCar, 2000. p. 255-258.

FRIZZO C. D.; DELLACASSA, E.; SERAFINI, L. A.; CASSEL, E. Extração Supercrítica do Óleo Essencial de Planta Nativa do Gênero Baccharis. In: *Anais* do XIX Interamerican Congress of Chemical Engineering, XII Congresso Brasileiro de Engenharia Química, I Brazilian Congress of Phase Equilibrium and Fluid Properties for Chemical Process Design; 2000; Águas de São Pedro.

FOOD INGREDIENTS BRASIL. Os antioxidantes, *Revista-fi.* 16-30. 2009, Disponível em: http://www.revista-fi.com/materias/83.pdf. Acesso em 20/01/2014.

HAUSTEEN, B. 1983. Flavonoids, a class of natural products of high pharmacological potency. *Biochem. Pharm*., 32: 1141-1148.

HOUAISS, A. Dicionário Houaiss da Língua Portuguesa, 2001.

JAIME, P. C.; FIGUEIREDO, I. C. R.; MOURA, E. C. DE.; MALTAM D. C. fatores associados ao consumo de frutas e hortaliças no Brasil, 2006. *Revista Saúde pública*, 43 (Supl 2) 57-64, 2009.

KAUR, C.; KAPOR, H. C.; Antioxidants in fruits and vegetables the millennum's health. *Int J. Food Sci Technol.* n.36, v.7 p 703-725. 2001.

LÔBO, K.M.S.; ATHAYDE, A.C.R.; SILVA, A.M.A.; RODRIGUES, F.F.G.; LÔBO, I.S.; BEZERRA, D.A.C.;COSTA, J.G.M. 2010 Avaliação da atividade antibacteriana e prospecção fitoquímica de *Solanum paniculatum* Lam. e *Operculina hamiltonii* (G. Don) D. F. Austin & Staples, do semiárido paraibano. *Rev. Bras. Pl. Med*., Botucatu, v.12, n.2, p.227-233.

LOOK, K.; POMERLEAU, J.; CAUSER, L.; ALTMANN, D. R.;MCKEE, M. The global burden of disease attributable to low consumption of fruit and vegetables: implications for the global strategy on diet. *Bull World Health Organ*. 83, 2, 2005.

MAPA, MINISTERIO DA AGRICULTURA, PECUARIA E ABASTECIEMNTO. 2010. *Manual de hortaliças não-convencionais*/ Ministério de agricultura, Pecuária e Abastecimento. Secretaria de desenvolvimento Agropecuário e Cooperativismos – Brasília : MAPA/ ACS, 92p.

NICOLI, M. C.; ANESE, M.; PARPINEL, M. Influence of processing on the antioxidant properties of fruit and vegetables. *Trends Food Sci. Technol.*, v. 10, n. 3, p. 94-100, 1999.

NOGUEIRA NETO, J. D.; SOUSA, D. P. DE.; FREITAS, R. M. DE. Avaliação do potencial antioxidante *in vitro* do nerolidol. Revista de Ciências farmacêuticas Básicas e Aplicadas. 34, (1), 125-130. 2013.

NURIT, K.; AGRA, M de F.; BASILIO, I. J. L. D. 2007. Estudo farmacobotanico comparativo entre *Solanum paniculatum L. e solanum rhytidoandrum* sendth. (solanaceae) Nota Cientifica, *Revista brasileiro de Biociências*, Porto Alegre, v.5 supl. 1, p. 243-245, jul.

OLIVERIA, R. C. M.; MONTEIRO, F. S.; SILVA, J. L. V.; RIBEIRO, L. A. A.; SANTOS, R. F.; NASCIMENTO, R. J. B.; DUARTE, J. C.; AGRA, M. F.; SILVA, T. M. S.; ALMEIDA, F. R. C.; SILVA, B. A. 2006. Extratos metanólicos e acetato de etila de Solamun megalonyx Sendtn. (Solanaceaes) apresentam atividade espasmolítica em íleo isolado de cobaia: um estudo comparativo. *Revista Brasileira Farmacogn.* V.16, n.2, p.146-151.

RAMOS, C. S.; RAMOS, N. S. M.; SILVA, R. R.DA.; CÂMARA, C. A. G. DA.; ALMEIDA, A. V. Metabolism by grasshoppers of volatile chemical constituents from *Mangifera indica* and *Solanum paniculatum* leaves. *Journal of insect Phsiology* 58, 1663-1668. 2012.

REYNERTSON, K. A.; YANG, H.; JIANG, B.; BASILE, M. J.; KENNELLY, E. J. quantitative analysis of antiradical phenolic constituents from fourteen edible Myrtaceae fruts. *Food Chemistry*, Amsterdam, v.109, n.4 p. 883-890, 2008.

RIBEIRO, S. R., FORTES, C. C., OLIVEIRA, S. C. C., CASTRO, C. F. S. Avaliação da atividade antioxidante de *solanum paniculatum (solanaceae). Arq. Ciênc. Saúde Unipar, Umuarama*, v. 11, n. 3, p. 179-183, set./dez. 2007.

RIBEIRO, V.; VIEIRA, I. L. B. F.; PASSOS, D. C. S. dos.; SILVA, E. M. de.; VALE, C. R. do.; FELICIO, L. R.; FERREIRA, D. H.; VIEIRA, P. M.; CARVALHO, S de. 2009. Ausencia de mutagenicidade de solanum paniculatum L. em celulas somáticas de Drosophila melanogaster: SMART/ asa. *Rer Biol. Neotrop*. 6 (2): 27-33.

SANTOS NETO, O. D.; KARSBURG, I. V. e YOSHITOME, M. Y. Viabilidade e germinabilidade polinica de populacoes de Jurubeba (*Solanum paniculatum). Revista de Ciencias Agro-Ambientais,* Alta Floresta, v.4, n.1 p.67-74, 2006.

SCALZO, J.; POLLTI, A.; PELLEGRINI, N.; MEZZETTI, B.; BATTINO, M. Plant genotype affects total antioxidante capacity and phenolic contentes in fruit. Nutrition, London, v.21, n.2, p.207-2013, 2000.

TURKMEN, N.; SARI, F.; VELIOGLU, Y. S. The effect of cooking methods on phenolics and antioxidant activity of selected green vegetables. *Food Chem*., v. 93, n. 4, p. 713-718, 2005.

VILELA, V. L. R.; FEITOSA, T. F.; LÔBO, K; M. da. S.; BEZERRA, D. A. C.; ATHAYDE, A. C. R. Potencial anti-helmíntico da raiz de *Solanum paniculatum* L. (1762) em ovelhas do semi-árido paraibano. *Acta Veterinaria Brasilica*. v.3, n.1, p.20-24. 2009.

WORLD HEALTH OAGANIZATION. The world health report 2002. *Reducing risks, promoting healthy life*. Geneva; 2002.