DANEYSA LAHIS KALSCHNE

TRATAMENTO COM VAPOR EM GRÃOS PVA DE *Coffea canephora*: EFEITO NA COMPOSIÇÃO E CARACTERÍSTICAS SENSORIAIS

Londrina
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Orientadora: Profa. Dra. Marta de Toledo Benassi.
Co-orientadora: Profa. Dra. Marinês Paula Corso.

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Londrina, 19 de setembro de 2017.

DEDICATÓRIA

Aos meus pais Loreno e Nelda,

Ao meu noivo Everton,

A vocês dedico.

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“Uma criança, um professor, um livro e uma caneta podem mudar o mundo.”

Malala Yousafzai

KALSCHNE, Daneysa Lahis. **Tratamento com vapor em grãos PVA de** *Coffea canephora***:** efeito na composição e características sensoriais. 120 f. Tese (Doutorado em Ciência de Alimentos) – Universidade Estadual de Londrina, Londrina. 2016.

RESUMO

O Brasil é o maior produtor e o líder em exportação de café do mundo. Dos frutos maduros/cereja obtêm-se bebidas com maior qualidade sensorial comparativamente às de grãos defeituosos pretos, verdes/imaturos e ardidos (PVA). Para selecionar apenas frutos cereja, é necessário realizar várias colheitas manuais, envolvendo altos custos. No Brasil esse tipo de colheita é pouco empregado, logo as indústrias beneficiadoras de café recebem entre 15% e 20% de grãos defeituosos. A espécie *Coffea canephora* (café canéfora) é empregada na indústria do solúvel e em *blends* de café torrado e moído. Por apresentar menor qualidade sensorial que a *Coffea arabica* (café arábica) tem menor valor comercial, e pode ser mais depreciada pela presença de defeitos. O tratamento com vapor antes do processo de torra tem sido reportado como alternativa para melhorar a qualidade da bebida de café canéfora, mas não existem propostas para uso em PVA. Objetivou-se estudar o efeito do tratamento com vapor (condições de pressão de 2 a 8 bar e tempo de 3 a 29 min) na composição e características de defeitos PVA de café canéfora torrado, bem como propor um processo para obtenção de produto para ser empregado em *blends* de café torrado e moído. Após o tratamento com vapor os cafés foram secos e torrados para padronização da cor. As condições de tratamento foram estudadas visando o aumento nos voláteis de impacto positivo e redução nos de impacto negativo e manuteção dos teores de compostos bioativos e de interesse para a qualidade da bebida e atividade antioxidante. Os tratamentos mais promissores foram submetidos a testes sensoriais em *blends* com cafés sem defeito. O tratamento com vapor afetou o teor dos bioativos cafeína, trigonelina, ácidos clorogênicos, melanoidinas, cafestol e 16-O-metilcafestol. Tratamentos com tempos longos e baixas pressões devem ser evitados para melhor preservação dos compostos bioativos e atividade antioxidante. O tratamento com vapor modificou também o perfil de voláteis do PVA torrado; a condição 5 bar/16 min foi a mais promissora pelo aumento de compostos com impacto positivo (acetoína, álcool benzílico, maltol, benzaldeído, 2,6- dimetilpirazina, 2-furfurilthiol, 5-metilfurfural, 2-acetilpiridina e 2-acetil-3,5-dimetilpirazina) e descréscimo nos de impacto negativo (4-etilguaiacol, ácido isovalérico, metional, 2,3-dietil-5-metilpirazina e 2-metoxi-3-metilpirazina). Foi possível adicionar 30% de defeito tratado (5 bar/16 min) ao café arábica sem que houvesse percepção do defeito ou rejeição da bebida e com aceitação maior que a do café controle (sem tratamento). *Blends* de café arábica e canéfora com adição de 50% de defeito tratado (5 bar/16 min) foram descritos como apresentando características sensoriais similares as bebidas puras, mas com menor intensidade em alguns atributos. Bebidas de café arábica (puro ou *blend*) foram descritas como de cor marrom, aroma frutado/de erva/de grão verde, e sabor de café/residual de café, e bebidas de canéfora, como de cor preta, viscosas, com maior presença de espuma, sabor amargo, e com aromas e sabores associados ao processo de torra. A espécie de café teve maior relevância na diferenciação sensorial das bebidas do que a adição do defeito tratado. As bebidas dos *blends* com tratamento com vapor 5 bar/16 min foram aceitas, e indicadas como preferidas e com maior intenção de compra pelos consumidores. O tratamento dos grãos PVA com vapor na condição 5 bar/16 min mostrou melhora no perfil volátil, boa retenção de bioativos e atividade antioxidante, sendo proposto para uso em *blends* com cafés arábica e canéfora sem prejuízo nas características sensoriais e aceitação das bebidas.

Palavras-chave: Preto. Ardido. Imaturo. Compostos voláteis. Compostos bioativos. Análise sensorial. Cromatografia gasosa. Cromatografia líquida. Limiar de detecção. Limiar de rejeição. Aceitação. Mapa de preferência interno.

KALSCHNE, Daneysa Lahis. **Steam treatment of** *Coffea canephora* **PVA beans:** effect on the composition and sensory characteristics. 120 p. Thesis (Doctoral Degree in Food Science) – Universidade Estadual de Londrina, Londrina. 2016.

ABSTRACT

Brazil is the world's largest coffee producer and exporter. The ripe/cherry fruits provide high sensory quality coffee brews compared to the defective beans: black, green/immature and sour ones (PVA). It is necessary to perform several manual harvest process, involving high costs, in order to harvest only cherry coffee fruits. In Brazil this type of harvesting is little employed, thus coffee industries receive between 15% and 20% of defective beans. The *Coffea canephora* species (robusta coffee) is employed in the instant coffee industry and blends of roasted and ground coffee. It presents lower commercial value and sensory quality than *C. arabica* (arabica coffee), thus the presence of defects could depreciate it even more. The steam treatment has been reported as an alternative to improve robusta coffee quality, but there are no proposals for the use of this treatment on PVA beans. The aim of this research was to study the steam treatment effect (steam pressure from 2 to 8 bar and contact time from 3 to 29 min) over the composition and characteristics of roasted PVA beans, as well as propose a process condition to obtain a product that can be used in blends of roasted and ground coffee. After treatment, the coffees were dried and roasted to standardize the color. The steam treatment conditions were studied aiming to increase the impact of positive volatile compounds and reduce negative volatile ones. The effect of treatment on the composition - bioactive compounds and compounds of interest for the coffee brew quality - and antioxidant activity was also studied. The most promissing treated coffees were applied in blends with non-defective ones and sensory analyzed. The steam treatment significantly affects the contents of the bioactives caffeine, trigonelline, chlorogenic acids, melanoidins, cafestol and 16-O-methylcafestol; treatments with longer times and low pressures should be avoided to prevent the loss of bioactive compounds and antioxidant acitvity. The steam treatment also modified the volatile profile of the roasted PVA; the most promising condition was 5 bar/16 min. On this condition, positive impact volatiles (acetoin, benzyl alcohol, maltol, benzaldehyde, 2,6-dimethylpyrazine, 2- furfurylthiol, 5-methylfurfural, 2-acetylpyridine and 2-acetyl-3,5-dimethylpyilrazine) were increased and negative impact ones (4-ethylguaiacol, isovaleric acid, methional, 2,3- diethyl-5-methylpyrazine and 2-methoxy-3-methylpyrazine) were decreased. Up to 30% of trated defect (5 bar/16 min) was added to arabica coffee without the perception or rejection of the coffee brew and this blend was more accepted than the control (without treatment). Blends of arabica or robusta coffee with 50% of treated defect (5 bar/16 min) were described as presenting sensory characteristics similar to pure coffees, but with the perception of least intensity. Arabica coffee brews (pure or blend) were described as having a brown color, fruity/herb/green bean aroma and coffee/residual coffee flavor. Robusta coffee brews were characterized by a black color, greater viscosity and presence of foam, bitter taste, and with aromas and flavors associated with the roasting process. The coffee species had more relevance towards differentiating sensory characteristics than the addition of the steam treated defect. Coffee brews with treated defect (5 bar/16 min) were sensory accepted, indicated as preferred and with greater purchase intent by consumers. The steam treatment performed at 5 bar/16 min on PVA beans improved the volatile profile and allowed good retention of bioatives compounds and antioxidant activity. It could be proposed to be used in blends with non-defective arabica and robusta coffees with no decrease in positive sensory charcateristics and acceptance.

Keywords: Black. Sour. Immature. Volatile compounds. Bioactive compounds. Sensory analyses. Gas chromatography. Liquid chromatography. Detection threshold. Rejection threshold. Acceptance. Internal preference mapping.

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LISTA DE ABREVIATURAS E SIGLAS

5-ACQ/5-CQA	Ácido-5-cafeoilquínico/5-caffeoylquinic acid
AA	Antioxidant activity
AB	*C. arabica* brew
ABIC	Associação Brasileira da Indústria do Café
ABTS	2,2-azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) radical scavenging activity
AC	*C. arabica*
ACG/CGA	Ácidos clorogênicos/chlorogenic acids
AISI	American Iron and Steel Institute
ANOVA	Análise de variância
ASDB	Blend with 50% of SDC and *C. arabica*
BET	Best estimates threshold
C	Curvature
CB	*C. canephora* brew
CC	*C. canephora*
CONAB	Companhia Nacional de Abastecimento
CRT	Consumer Rejection Threshold
CSDB	Blend with 50% of SDC and *C. canephora*
CV%	Coeficient of variation in percent
DT	Detection Threshold
GC-MS	Gas chromatography with mass spectrometry
HS-SPME	Headspace Solid Phase Microextraction
L*	Luminosidade
PVA	Preto, verde e ardido
SC	Stemead coffee
SDC	Stemead defective coffee
TEAC	Trolox equivalent antioxidant capacity
UHPLC	Ultra-High Performance Liquid Chromatography
UV-Visible	Regiões do espectro ultravioleta e visível
NY 2	Café com qualidade de bebida tipo 2

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CAPÍTULO 1

INTRODUÇÃO, REVISÃO BIBLIOGRÁFICA E OBJETIVOS

1.1 INTRODUÇÃO

O Brasil é reconhecidamente o maior produtor e o líder em exportação de café do mundo (ABIC, 2016a), destacando-se ainda por produzir cafés das duas principais espécies comerciais: *Coffea arabica* (café arábica) e *Coffea canephora* (café canéfora). Na safra 2017, a produção de café arábica, espécie em que o Brasil lidera a produção global e as exportações, está estimada em 35,4 milhões de sacas de 60 kg, enquanto que a de canéfora deve chegar a 10,1 milhões (CONAB, 2017).

O café canéfora é reconhecido pela maior rusticidade e vigor que o café arábica, adaptado a temperaturas elevadas e com resistência a deficiências hídricas prolongadas (CLARKE; MACRAE, 1985; EMBRAPA, 2004). Apesar das chuvas abaixo da média em duas safras consecutivas no estado do Espírito Santo (responsável por 63% da produção de café canéfora no país), principalmente nas épocas do florescimento, formação e enchimento de grãos, além da falta de águas nos mananciais para irrigação; a produção de café canéfora deve apresentar um crescimento de 26,9% em relação à 2016, justificado pela recuperação da produtividade nos estados de Rondônia e Bahia, segundo e terceiro maiores produtores nacionais, respectivamente (CONAB, 2017). Logo, ressalta-se a importância do café canéfora como alternativa para manutenção da produtividade do agronegócio de café no Brasil.

Em relação ao consumo de café, no Brasil, segundo maior consumidor mundial, existe uma perspectiva de crescimento moderado, que deve alcançar 906 mil toneladas de café torrado e moído em 2019. Em 2015 o consumo *per capita* de café torrado e moído foi de 81 L, equivalente a 819 mil toneladas (ABIC, 2015; 2016b).

Os cafés mais valorizados no mercado são provenientes dos frutos maduros ou cereja, enquanto que, cafés oriundos dos frutos imaturos e dos frutos que já passaram do ponto de maturação e secam na planta, proporcionam bebidas de qualidade inferior. Para colher apenas frutos maduros, é necessário realizar várias colheitas manuais, selecionando somente os frutos cereja, envolvendo altos custos. No Brasil esse tipo de colheita é pouco empregado, assim as indústrias beneficiadoras e produtoras de café recebem grande quantidade de grãos defeituosos, destacando-se grãos pretos, verdes (imaturos) e ardidos (usualmente denominados PVA). Apesar dos grãos PVA apresentarem poucas diferenças na composição em relação ao café sem defeitos a qualidade da bebida é bastante prejudicada.

O emprego de vapor no tratamento de grãos de café canéfora tem demonstrado resultados interessantes na qualidade do perfil de compostos voláteis observados após o processo de torra, característica importante para a qualidade da bebida de café torrado. Na literatura as informações relativas a composição de grãos defeituosos de café canéfora são escassas, e não se observou pesquisas associadas ao efeito do tratamento com vapor em café canéfora PVA. Baseado nisso, a presente tese visou estudar o impacto do tratamento com vapor em grãos PVA de café canéfora buscando alternativas para obtenção de melhor qualidade sensorial na bebida do café torrado produzido com essa matriz.

O capítulo 1 da tese apresenta uma introdução geral, uma revisão bibliográfica sobre o tema, e a descrição dos objetivos do trabalho. Nesse estudo, defeitos PVA de café canéfora crus foram tratados com vapor empregando um planejamento fatorial (condições de pressão de 2 a 8 bar e tempo de 3 a 29 min), e após secagem e torra em condições padronizadas foi estudado o impacto do tratamento nas características do produto considerando vários enfoques, resultando nos capítulos 2, 3, e 4.

O capítulo 2 apresenta uma avaliação do impacto do tratamento com vapor no perfil de voláteis do PVA tratado, descrevendo as condições que permitem maximizar compostos de impacto positivo e minimizar compostos de impacto negativo. Foram ainda avaliados o limiar de rejeição do consumidor e o limiar de detecção, para estimar possíveis porcentagens de adição do PVA tratado na melhor condição de estudo em *blends.*

No capítulo 3, descreve-se o impacto dos tratamentos com vapor nos teores de alguns compostos bioativos e de importância para a qualidade de bebida (cafeína, trigonelina, ácidos clorogênicos, melanoidinas e diterpenos) e na atividade antioxidante das bebidas dos cafés PVA tratados torrados, indicando as condições de tratamento que permitem melhor retenção desses compostos.

No capítulo 4 está apresentado um perfil sensorial e aceitação das bebidas de *blends* de café PVA tratados com cafés arábica e canéfora, mostrando o impacto sensorial do emprego do PVA tratado na melhor condição de estudo pela comparação com bebidas de cafés puros (arábica e canéfora).

A avaliação conjunta dos resultados permite propor, nas conclusões finais, produtos (*blends* de PVA tratado adicionados a café sem defeito) com boas características de bebida e aceitação sensorial para uso no mercado brasileiro de café torrado e moído.

1.2 REVISÃO BIBLIOGRÁFICA

Coffea canephora Pierre é uma espécie de café nativa das florestas baixas da África Equatorial, na bacia do rio Congo, cultivada em alguns países da África Central e Ocidental, no sudeste da Ásia e na América do Sul (EMBRAPA, 2004). No Brasil, predomina nas lavouras do Espírito Santo e Rondônia e pode ser encontrado em parte da Bahia e de Minas Gerais (MAPA, 2017).

A duas variedades de *C. canephora* mais importantes são a Conilon e Robusta (RONCHI, 2009) sendo que a primeira representa quase a totalidade das lavouras de café (BRAGANÇA et al., 2001). As denominações café conilon e robusta são usuais, nesse trabalho será empregada a denominação café canéfora, de maneira a agrupar todas as variedades da espécie *Coffea canephora*. Algumas características gerais dessa espécie são apresentadas na Tabela 1.1.

Tabela 1.1 - Principais características da planta e do café *Coffea canephora*.

Características	*Coffea canephora*
Origem	Bacia do Congo
Altitude preferencial	Abaixo de 500 m
Temperatura preferencial	24 a 28 ºC
Déficit hídrico	Até 150 mm/ano
Rusticidade	Maior
Fecundação	Alógama, autoincompatível
Ploidia	Diplóide, com 22 cromossomos
Porte	Mais alto
Propagação	Semente e estaca
Período de maturação	Em média 300 dias
Cor do fruto	Mais escuro que o do café arábica
Grãos maduros	Permanecem na planta
Mucilagem	Menos que o café arábica
Bebida	Sabor diferenciado
Preço	Menor que café arábica

Fonte: Embrapa (2004).

O café canéfora é empregado no preparo de *blends* de café torrado e moído, nos quais é misturado ao café arábica, podendo compor até 30% do produto final (EMBRAPA, 2004; RONCHI, 2009). Por possuir maior teor de sólidos solúveis, apresentar maior rendimento após o processo de torra e pelo menor valor comercial que o café arábica, o café canéfora é a principal matéria prima da indústria do café solúvel (EMBRAPA, 2004). No geral, o café canéfora apresenta qualidade sensorial inferior ao café arábica, sendo

descrito como uma bebida de sabor único (CLARKE; MACRAE, 1985; MAPA, 2017), e pode ser ainda mais depreciado pela presença de defeitos.

Os grãos de café podem ser classificados quanto à presença de defeitos intrínsecos, decorrentes de grãos alterados devido a imperfeição de processos agrícolas, modificações fisiológicas ou genéticas do fruto, ou por práticas de beneficiamento inadequadas; ou extrínsecos, decorrentes de impurezas incorporadas ao café durante a colheita, como pedras, gravetos, cascas, cocos entre outros.

A Associação Brasileira da Indústria de Café (ABIC) preconiza um critério de classificação da qualidade de café baseado nos defeitos (Tabela 1.2) em que se estabelece um impacto para cada tipo de defeito (BRASIL, 2003).

Tabela 1.2 - Critérios de classificação do grão cru de café beneficiado quanto a equivalência de defeitos e impurezas (amostras de 300 g).

	Defeitos	Quantidade	Equivalência
Intrínsecos (defeitos)	Grão Preto	1	1
	Grãos Ardidos	2	1
	Conchas	3	1
	Grãos Verdes	5	1
	Grãos Quebrados	5	1
	Grãos Brocados	2 a 5	1
	Grãos Mal Granados ou Chochos	5	1
Extrínsecos (impurezas)	Coco	1	1
	Marinheiros	2	1
	Pau, Pedra, Torrão grande	1	5
	Pau, Pedra, Torrão regular	1	2
	Pau, Pedra, Torrão pequeno	1	1
	Casca grande	1	1
	Casca pequena	2	3

Fonte: Brasil (2003).

As bebidas por sua vez, podem ser classificadas no Brasil como de tipo 2 até tipo 8, segundo a classificação indicada na Tabela 1.3, o que corresponde a classificação internacional NY 2 a NY 8 (FRANCA et al., 2005). Assim uma bebida tipo 2 pode conter apenas 6 defeitos em 300 g de amostra. A totalização de 6 defeitos poderia, por exemplo, ser devido a presença de 1 grão preto (1 defeito), 8 ardidos (4 defeitos) e 5 verdes (1 defeito).

Os grãos defeituosos mais comuns e de grande significância para a qualidade da bebida do café correspondem aos defeitos intrínsecos grãos preto, verde (imaturo) e ardido

(denominados usualmente pela sigla PVA) (BANDEIRA et al., 2009; FARAH, SANTOS, 2015).

Tabela 1.3– Descrição da classificação dos tipos de bebida do café.

Café tipo (NY)*	Quantidade máxima de defeitos permitidos em 300 g
2	6
3	13
4	30
5	60
6	120
7	240
8	450

* O termo tipo é usado na classificação no Brasil, e NY na classificação internacional. Fonte: Franca et al. (2005).

Os grãos pretos são definidos como grãos ou pedaços de grão de coloração preta opaca; os grãos ardidos são classificados como grãos ou pedaços de grão que apresentam coloração marrom, em diversos tons, devido à ação de processos fermentativos; e os grãos verdes/imaturos são caracterizados como grãos imaturos, com película prateada aderida, com sulco ventral fechado e de coloração verde em tons diversos (BRASIL, 2003; FRANCA, OLIVEIRA, 2015).

Os cafés mais valorizados no mercado são provenientes dos frutos maduros ou cereja, que propiciam a obtenção das melhores bebidas, no entanto para colher apenas frutos maduros, é necessário realizar várias colheitas manuais, selecionando somente os frutos cereja, envolvendo altos custos (SOARES et al., 2010). No Brasil esse tipo de colheita é pouco empregado, assim as indústrias beneficiadoras e produtoras de café recebem grande quantidade de grãos PVA.

De um modo geral, estima-se que entre 15 e 20% do total de grãos de café produzidos no Brasil são defeituosos (FRANCA; OLIVEIRA, 2015; OLIVEIRA et al., 2006; TOLEDO et al., 2016; WEI; TANOKURA, 2015). Oliveira et al. (2006) reportaram que um café arábica, proveniente de Minas Gerais, rejeitado por uma selecionadora automática continha 68,8% de grãos defeituosos (40,5% ardido; 21,1% imaturo; 3,2% preto e 4,0% referente à outros defeitos) enquanto restaram 31,2% de grãos não defeituosos. Em amostras similares de café arábica de Minas Gerais rejeitado pela selecionadora automática, Vasconcelos et al. (2007) quantificaram 59% de grãos defeituosos (34% ardido; 18% imaturo e 7% preto) e 41% de grãos não defeituosos.

Na literatura, as informações sobre a composição e características sensoriais do café contendo defeitos PVA são escassas, especialmente para o café canéfora.

Oliveira et al. (2006) avaliaram a composição de grãos pretos, imaturos, ardidos e grãos não defeituosos de café arábica torrado proveniente de Minas Gerais (Tabela 1.4). O teor de água e cinzas não diferiu, mas os grãos pretos tiveram o maior conteúdo de proteína, e os grãos imaturos o menor teor de lipídios. Morais et al. (2007) relataram um maior conteúdo de proteínas e menor teor de óleo essencial em grãos PVA de café arábica em comparação aos grãos sem defeito.

Tabela 1.4 - Composição centesimal de grãos de café torrado (g 100 g^{-1} bs).

	Mistura PVA	Pretos	Imaturos	Ardidos	Não defeituosos
Água	$1,5^{a} \pm 0,2$	$1,4^{a} \pm 0,2$	$1,6^{a} \pm 0,1$	$1,4^{a} \pm 0,0$	$1,3^{a} \pm 0,1$
Proteína	$14,4^{b} \pm 0,2$	$16,1^{a} \pm 0,2$	$14,5^{b} \pm 0,2$	$14,6^{b} \pm 0,1$	$14,0^{c} \pm 0,3$
Lipídios	$10,3^{ab} \pm 0,2$	$10,2^{b} \pm 0,1$	$9,0^{c} \pm 0,1$	$10,3^{ab} \pm 0,4$	$10,3^{a} \pm 0,1$
Carboidratos	67,3	65,9	67,6	66,5	67,7
Cinzas	$4,2^{a} \pm 0,6$	$4,5^{a} \pm 1,0$	$4,7^{a} \pm 0,6$	$4,9^{a} \pm 0,0$	$4,0^{a} \pm 0,0$

Médias com a mesma letra na linha não diferiram pelo teste de Duncan ($p > 0,05$); teor de carboidratos estimado por deiferença. Fonte: Oliveira et al. (2006).

Vasconcelos et al. (2007) avaliaram a atividade de água (Aw), acidez e pH de grãos pretos, imaturos, ardidos e não defeituosos de café arábica cultivado em Minas Gerais com grau de torra média. Não houve diferença na Aw e acidez. O maior pH foi observado para grãos pretos, seguido dos ardidos, imaturos e grãos não defeituosos (Tabela 1.5). Em contrapartida, Morais et al. (2007) relataram menor pH para grãos PVA de café canéfora em comparação ao não defeituoso, justificado pela fermentação sofrida pelos grãos PVA.

Tabela 1.5 – Características de Aw, acidez e pH de grãos de café com torra média.

	Pretos	Imaturos	Ardidos	Não defeituosos
Aw	$0,16^{a} \pm 0,01$	$0,17^{a} \pm 0,02$	$0,20^{a} \pm 0,02$	$0,18^{a} \pm 0,04$
Acidez (mL NaOH. g café$^{-1}$)	$108^{a} \pm 13,51$	$97,4^{a} \pm 5,08$	$99,2^{a} \pm 5,36$	$109^{a} \pm 10,02$
pH	$6,87^{a} \pm 0,01$	$6,28^{b} \pm 0,02$	$6,47^{c} \pm 0,03$	$6,08^{d} \pm 0,04$

Médias com a mesma letra na linha não diferiram pelo teste de Duncan ($p > 0,05$). Fonte: Vasconcelos et al. (2007).

Vasconcelos et al. (2007) ainda relataram, nos grãos crus antes da torra, a presença de cadaverina em grãos pretos e histamina em grãos pretos, imaturos e ardidos.

Esses parâmetros de diferenciação, no entanto, não podem ser empregados em cafés torrados, uma vez que essas aminas são reduzidas drasticamente com o processo de torra.

A matriz do café é extremamente complexa e o processo de torra dá origem a uma grande quantidade de compostos voláteis responsáveis pelo aroma desse produto (DE MARIA; MOREIRA; TRUGO, 1999). Vários autores têm estudado a possibilidade de identificar a presença de defeitos pelos compostos voláteis, mostrando que o perfil volátil de grãos defeituosos é diferenciado do observado para cafés sem defeito (MORAIS et al., 2007; TOCI; FARAH, 2008; BANDEIRA et a., 2009; TOCI; FARAH, 2014; TOLEDO et al., 2016).

Bandeira et al. (2009) analisaram compostos voláteis de grãos crus e torrados de cafés arábica e canéfora sem defeitos (controles) e com defeitos (verde, preto e mistura de defeitos PVA do café arábica; e defeito preto do café canéfora). Os compostos 2-metil-propanal, hexanal, decanoato de etila e dodecanoato de metila foram encontrados somente nos grãos defeituosos de café arábica torrados, enquanto 1-hidroxi-2-propanona e pirrol foram encontrado em defeitos de café arábica e canéfora. Nenhum dos compostos mencionados esteve presente nos grãos controle. Alguns compostos foram exclusivos para um defeito, como 2-pentanona que foi encontrada somente em grãos imaturos (arábica), e pentanal e 1-pentanol em grãos pretos (arábica). O etanal e 1-hidroxi-2-propanona foram identificados somente em cafés arábica e canéfora não defeituosos. A Tabela 1.6 mostra um resumo dos voláteis identificados por Bandeira et al. (2009).

Toci e Farah (2008) estudando um conjunto de defeitos PVA de café arábica com grau de torra de claro a médio identificaram alguns compostos voláteis como potenciais marcadores de defeitos, como pirazina; 2,3-butanodiol *meso*, 2-metil-5-(1-propenil) pirazina, ácido hexanóico, 2-metoxi-4-etilfenol (4-etil-guaiacol) e 1-metil-4-[(1-metil)tio]benzeno (sulfito de *iso*propil *p*-cresol).

Em grãos PVA de café arábica comparados com grãos não defeituosos, com torras clara, média e escura, Morais et al. (2007) relataram a presença de maiores teores dos compostos voláteis 2-metilpropanal, 3-metilbutanal, 2-metilbutanal, acetato de furfurila e 1-metil-2-pirrolcarboxaaldeído e, de menores teores de 2-metiltetraidrofuran-3-ona, 2-metilpirazina, 2,6-dimetilpirazina, 2-etilpirazina e 2-etil-6-metilpirazina. Os compostos voláteis 2-heptanona, 1-acetoxibutan-2-ona mostram-se exclusivos dos grãos não defeituosos e os ácidos 2-propenilbutanóico e 2-hidroximetilbenzóico foram exclusivos dos grãos PVA.

Tabela 1.6 - Compostos voláteis identificados em café arábica e canéfora defeituosos e não defeituosos com grau de torra moderadamente escura.

Coffea arabica			
Não defeituosos	**Imaturos**	**Pretos**	**PVA**
Voláteis exclusivos			
etanal; 1-hidroxi-2-propanona;	2-pentanona	pentanal; 1-pentanol;	
Voláteis não exclusivos			
2-metilbutanal; 3-metilbutanal; 2-propanona; 2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; metilpirrol; pirazina, 2-metilpirazina, piridina, ácido etanóico; acetato de 2-furfurilmetano	2-metilbutanal; 3-metilbutanal; 2-propanona; 2-butanona; 1-hidroxi-2-butanona; 3-hidroxi-2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; pirrol; metilpirrol; pirazina; 2-metilpirazina; 2,5-dimetilpirazina; 2,6-dimetilpirazina; 2-etilpirazina; piridina; ácido etanóico; decanoato de etila; dodecanoato de metila; acetato de 2-furilmetila	2-metilpropanal; 2-metilbutanal; hexanal; 2-propanona; 2-butanona; 3-hidroxi-2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; pirrol; pirazina; 2-metilpirazina; 2,5-dimetilpirazina; 2,6-dimetilpirazina; 2-etilpirazina; piridina; ácido etanóico; decanoato de etila; dodecanoato de metila; acetato de 2-furilmetila	2-metilbutanal; 3-metilbutanal; 2-propanona; 2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; metilpirrol; pirazina; 2-metilpirazina; 2-etilpirazina; piridina, ácido etanóico; acetato de 2-furilmetila
Coffea canephora			
Não defeituosos		**Pretos**	
Voláteis exclusivos			
etanal; 1-hidroxi-2-propanona;			
Voláteis não exclusivos			
2-metilbutanal; 3-metilbutanal; 2-propanona; 2-butanona; 3-hidroxi-2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; pirazina; 2-metilpirazina; 2,5-dimetilpirazina; 2,6-dimetilpirazina; 2-etilpirazina; piridina; acetato de 2-furilmetila		2-metilbutanal; 3-metilbutanal; 2-propanona; 2-butanona; 1-hidroxi-2-butanona; 3-hidroxi-2-butanona; 2,3-butanodiona; 2,3-pentanodiona; butirolactona; etanol; 2-metilfurano; 2-furilmetanol; metilpirrol; pirazina; 2-metilpirazina; 2,5-dimetilpirazina; 2,6-dimetilpirazina; 2-etilpirazina; piridina; ácido etanóico; acetato de 2-furilmetila	

Fonte: Bandeira et al. (2009).

O composto volátil 2-metilisoborneol foi relacionado com a impressão sensorial de “mofo”, “bolor” e “terra” no café (BLANK; GROSCH, 2002). Encontra-se presente no café canéfora em quantidade superior ao arábica (BADE-WEGNER; HOLSCHER; VITZTHUM, 1993). Foram reportados teores de 80 a 420 ng kg^{-1} no café arábica torrado em comparação a variações de 740 a 1280 ng kg^{-1} no café canéfora (GROSCH; SEMMELROCH; MASANETZ, 1993). O limiar de detecção ortonasal do 2-metisoborneol

em água foi descrito como sendo observado em concentrações entre 20 e 30 ng L^{-1} (BLANK; GROSCH, 2002).

O café possui também diversos compostos bioativos e de importância para a qualidade de bebida. Benefícios à saúde relacionados com o consumo moderado e regular de café têm sido reportados na literatura, descrevendo-se efeitos cognitivos e na memória, redução dos níveis de glicose sanguínea, atividades anti-inflamatórias e neuroprotetoras, efeitos citoprotetivos contra o estresse oxidativo e prevenção na incidência de doenças crônico degenerativas como diabetes tipo 2, Parkinson, Alzheimer, alterações da função hepática (cirrose) e alguns tipos de câncer (AMER; MAZEN; MOHAMED, 2017; BEDOYA-RAMÍREZ et al., 2017; BOROTA et al., 2014; BUTT; SULTAN, 2011; FROST-MEYER; LOGOMARSINO, 2012; HIGDON; FREI, 2006; MARTINI et al., 2016; NKONDJOCK, 2009; PREEDY, 2015). Em estudos de grande amplitude observou-se associação inversa entre o consumo de café e óbitos devido a doenças cardíacas, respiratórias, acidente vascular cerebral, lesões e acidentes, diabetes e infecções, mesmo considerando indivíduos fumantes (FREEDMAN et al. 2012) e do risco de mortalidade total (DING et al., 2015).

Os benefícios à saúde com o consumo regular de café têm sido correlacionados a presença de vários compostos bioativos, mas existe na literatura um menor número de informações sobre a composição para cafés canéfora, comparado a que se observa para cafés arábica.

A cafeína (1,3,7-trimetilxantina) é o principal alcaloide encontrado no café e um de seus componentes mais estudados (MOREIRA; TRUGO; DE MARIA, 2000). É reconhecida pelo efeito estimulante ao sistema nervoso central e do músculo cardíaco e na diminuição do sono (NEHLIG, 1999; MAZZAFERA, 2012). A cafeína também tem sido investigada por sua ação antioxidante e propriedades anticarcinogênicas, atribuída a atividade sequestrante contra espécies reativas de oxigênio (hidroxil, peroxil e oxigênio singleto) (SHI; DALAL; JAIN, 1991; GEORGE; RAMALAKSHMI; RAO, 2008). Mais recentemente, a ingestão de cafeína tem sido ainda inversamente associada ao risco do desenvolvimento da doença de Parkinson, Alzheimer e diabetes tipo 2 (MEJIA; RAMIREZ-MARES, 2014), diminuição do estresse oxidativo e biomarcadores inflamatórios do fígado (AMER; MAZEN; MOHAMED, 2017), e efeito hepatoprotetor (SALOMONE; GALVANO; VOLTI, 2017, 2017).

A cafeína, assim como outros alcaloides, possui gosto amargo característico que é transferido para a bebida do café (HOMMA, 2001; LEY, 2008). A solubilidade desse biotivo em água aumenta com o acréscimo de temperatura, chegando a 40% a 100 °C (URGEL, 2010). A cafeína é estável ao processo de torra, mas seu teor no café é altamente dependente da espécie, sendo mais alto no café canéfora (VIGNOLI et al., 2014). Souza e Benassi (2012), estudando cafés torrados e moídos, reportaram teores de cafeína de 1,98 a 2,04 g $100g^{-1}$ para café canéfora e de 1,10 a 1,29 g $100g^{-1}$ para arábica. Franca et al. (2005), estudando cafés arábica crus e torrados, reportaram maiores teores de cafeína nos grãos pretos e ardidos, comparativamente a grãos imaturos e não defeituosos (Tabela 1.7).

Tabela 1.7 - Cafeína, trigonelina e ácido-5-cafeoilquínico (5-ACQ) (g 100 g^{-1} bs) em grãos de café arábica defeituosos e não defeituosos torrados.

	PVA	Pretos	Imaturos	Ardidos	Não defeituosos
Cafeína	$0,79^{b} \pm 0,04$	$1,38^{a} \pm 0,06$	$0,64^{c} \pm 0,04$	$1,26^{a} \pm 0,00$	$0,70^{c} \pm 0,08$
Trigonelina	$0,19^{c} \pm 0,00$	$0,33^{a} \pm 0,01$	$0,14^{d} \pm 0,01$	$0,26^{b} \pm 0,00$	$0,18^{c} \pm 0,01$
5-ACQ	$0,13^{c} \pm 0,01$	$0,34^{a} \pm 0,02$	$0,14^{c} \pm 0,01$	$0,24^{b} \pm 0,00$	$0,18^{bc} \pm 0,05$

Médias com a mesma letra na linha não diferiram pelo teste de Duncan ($p > 0,05$). Fonte: Franca et al. (2005).

Além do interesse de saúde e qualidade de bebida, a importância da quantificação da cafeína especialmente nos cafés tratados com pressão de vapor se deve à similaridade com o processo de descafeinização do café. Nesse o grão verde é tratado com vapor e depois a extração da cafeína é feita por lavagem com água quente (URGEL, 2010; HEILMANN, 2001). Sob condições de tratamento com vapor podem ser esperadas perdas, devido a solubilidade desse composto em água.

A trigonelina (1-metil piridina-3-carboxílico) é uma N-metil betaína, alcaloide importante para o sabor e aroma do café (MONTEIRO; TRUGO, 2005). Os produtos da degradação da trigonelina, derivados de pirrol, são precursores de aroma e gosto amargo e isso se reflete nas características de bebida (VIANI; HORMAN, 1974). A trigonelina é precursora do ácido nicotínico (vitamina B_3), o qual é formado durante o processo de torra pela remoção do grupo metila da trigonelina (DE MARIA; MOREIRA, 2011). Estudos relacionados com a trigonelina têm demonstrado melhoras na tolerância à glicose (YOSHINARI; IGARASHI, 2010), relação inversa com a incidência de diabetes tipo 2 e controle dos níveis de lipídios séricos (YOSHINARI; IGARASHI, 2015), propriedades

antimicrobianas (ALMEIDA et al., 2005), atividade antioxidante (LIANG; KITTS, 2014) e efeitos neuroprotetores na prevenção de Alzheimer (MAKOWSKA et al., 2014).

A trigonelina é hidrossolúvel, possui baixa estabilidade ao processo de torra, e seu teor é dependente da espécie: café canéfora apresenta menor teor de trigonelina que o arábica. Alves et al. (2006) reportaram 0,68 g trigonelina 100 g^{-1} de café canéfora com grau de torra médio. Vignoli et al. (2014), comparando cafés com grau de torra de muito clara a muito escura, relataram redução no teor de trigonelina de 3,3 para 1,4 g 100 g^{-1} em café arábica e de 2,2 para 0,2 g 100 g^{-1} em café canéfora, com um decréscimo de 90% com o aumento do grau de torra. Com relação ao teor de trigonelina em cafés com defeito, Franca et al. (2005) reportou para café arábica torrado, maiores teores nos grãos pretos e ardidos comparativamente a café imaturo e não defeituoso (Tabela 1.6).

Os ácidos clorogênicos (ACG) são os principais componentes da fração fenólica dos grãos de café e apresentam propriedades benéficas à saúde, devido à sua atividade antioxidante, hepatoprotetora, hipoglicemiante, antiviral, anti-inflamatória, neuroprotetiva e de redução do risco de desenvolvimento de diabetes tipo 2 (FARAH; DONANGELO, 2006; BEDOYA-RAMÍREZ et al., 2017; BODE; DONG, 2015; FROST-MEYER; LOGOMARSINO, 2012; LUDWIG et al., 2014; VAN DIJK et al., 2009). O consumo regular de café, uma bebida rica em polifenóis, demonstrou exercer efeito protetivo contra fatores de risco cardiovascular (MIRANDA et al., 2017).

Os principais grupos de ACG encontrados no café cru incluem os ácidos cafeoilquínicos, dicafeoilquínicos, feruloilquínicos, p-cumaroilquínicos e ácidos cafeoilferuloilquínicos (MONTEIRO; TRUGO, 2005; FARAH; DONANGELO, 2006; PERRONE et al., 2007). O ácido-5-cafeoilquínico (5-ACQ) é o composto mais abundante, representando mais de 30% do total de ACG em bebidas de café torrado (PERRONE; FARAH; DONANGELO, 2012). Zanin et al. (2016), avaliando 32 cafés arábica brasileiros procedentes de concursos de qualidade, reportaram que o 5-ACQ representou de 38 a 50% do total de ácidos clorogênicos em cafés arábica. Mori (2016) descreve que, para bebidas de café canéfora de diferentes cultivares, o 5-ACQ representou de 31% a 40% do total de ácidos clorogênicos totais da bebida.

Os ACG são compostos hidrossolúveis, com baixa estabilidade a torra. Uma redução no conteúdo de ACG totais de 56 a 99% foi observada comparando-se bebidas de café verde com bebidas de café torrado por 6 e 15 minutos, respectivamente (PERRONE; FARAH; DONANGELO, 2012). Um decréscimo no conteúdo de 5-ACQ de 5,96 g 100 g^{-1}

para 0,22 g 100 g^{-1} para o café arábica e de 6,19 g 100 g^{-1} para 0,13 g 100 g^{-1} no café canéfora foi observado com o aumento do grau de torra, de muito claro a muito escura (VIGNOLI et al., 2014). Apesar do café canéfora cru possuir maior teor de 5-ACQ, a literatura descreve que ocorre uma degradação mais acentuada desse bioativo nessa matriz em comparação ao café arábica (VIGNOLI et al., 2014, DIAS, BENASSI, 2015). Franca et al. (2005) relataram um maior teor de 5-ACQ para os grãos de café preto, comparativamente aos grãos imaturo, ardido e não defeituoso (Tabela 1.6).

Melanoidinas são compostos de alta massa molecular que possuem uma estrutura heterogênea contendo o átomo de nitrogênio, e são formados nas etapas finais da reação de Maillard (MORALES; SOMOZA; FOGLIANE, 2012). Tem sido atribuída a esses compostos diversas propriedades biológicas como atividade antioxidante (PASTORIZA; RUFIAN-HENARES, 2014; BEDOYA-RAMÍREZ et al., 2017) e atividade antimicrobiana (RUFIAN-HENARES; DE LA CUEVA, 2009), ambas influenciadas pelo poder de quelar metais (RUFIÁN-HENARES, PASTORIZA, 2015), além da atuação como fibra alimentar na modulação do intestino (MORALES; SOMOZA; FOGLIANO, 2012). Esses compostos são predominantemente responsáveis pela coloração marrom observada em produtos de café torrado (LINDENMEIER; FAIST; HOFMANN, 2002). Teores de melanoidinas entre 18,50 a 27,30 g 100 g^{-1} foram relatados para o café canéfora com diferentes graus de torra (L* de 14 a 33) (VIGNOLI et al., 2014). Para bebidas de café canéfora (15 genótipos de 3 cultivares de cafés canéfora brasileiros produzidos em dois locais de cultivo) foram reportados teores de melanoidinas entre 5,9 e 9,7 mg mL^{-1} da bebida (MORI, 2016).

A fração lipídica do café é composta principalmente de triacilgliceróis, esteróis e tocoferóis, presentes em todo óleo vegetal comestível comum, entretanto o óleo de café também contém diterpenos da família dos kaurenos, em proporção de até 20% dos lipídeos totais (SPEER; KÖLLING-SPEER, 2006). Os dois principais diterpenos do café são caveol e cafestol. O aumento dos níveis séricos do colesterol com alto consumo de café, principalmente em bebidas de café fervido são atribuídos a atividade hipercolesterolêmica do cafestol (URGERT et al., 1995), mas a literatura mais recente tem ressaltado os efeitos positivos como a atividade anti-inflamatória, antioxidante e anticarcinogênica, destacando principalmente a redução do risco de incidência de doenças hepáticas (CAVIN et al., 2002; HIGDON; FREI, 2006; CHU, 2012; WANG et al., 2012; GAASCHT; DICATO; DIEDERICH, 2015; MARTINI et al., 2016).

Os diterpenos são insolúveis em água e relativamente estáveis ao processo de torra. Dias et al. (2014) relataram para cafés arábica e canéfora, que ao longo da torra ocorre formação de produtos de degradação de diterpenos (dehidrocaveol e dehidrocafestol), no entanto, devido ao aumento proporcional da concentração dos lipídios (pela degradação de outros componentes da matriz) os teores de caveol e cafestol mantêm-se estáveis ou podem ter pequenos aumentos na concentração mesmo em torras mais intensas.

O cafestol e caveol estão presentes em café arábica e canéfora (PACETTI et al., 2012; MORI et al., 2016), enquanto o derivado 16-O-metilcafestol está presente somente em café canéfora (SPEER; KOLLING-SPEER, 2006). Pacetti et al. (2012) reportaram teor de caveol variando de 0,7 a 2,3 mg 100 g^{-1} em café canéfora. Campanha, Dias e Benassi (2010) reportaram para cafés canéfora brasileiros com diferentes graus de torra, ausência de caveol e teores de cafestol de 163 a 250 mg 100 g^{-1} para amostras com poucos defeitos e de 188 a 275 mg 100 g^{-1} para amostras com muito defeito, concluindo que o perfil de diterpenos é pouco afetado pela presença de defeitos. Mori et al. (2016) analisaram 15 genótipos de 3 cultivares de cafés canéfora brasileiros produzidos em dois locais de cultivo e detectaram teores de caveol entre 0 e 14,1 mg 100 g^{-1}, de cafestol entre 151,7 e 359,7 mg 100 g^{-1} e de 16-O-metilcafestol entre 26,3 e 132,1 mg 100 g^{-1}.

No geral se relata maior atividade antioxidante para produtos de café canéfora, comparativamente ao arábica, principalmente em função de seu maior teor de cafeína (PERRONE; FARAH; DONANGELO, 2012; VIGNOLI; BASSOLI, BENASSI, 2011; VIGNOLI et al., 2014).

A atividade antioxidante (AA) é definida como a capacidade de um composto de inibir degradação oxidativa. Em alimentos, envolve pelo menos duas questões: o potencial antioxidativo determinado pela composição e propriedades antioxidantes dos constituintes e os efeitos biológicos que dependem, entre outras coisas, da biodisponibilidade do antioxidante (ROGINSKY; LISSI, 2005; LIANG; KITTS, 2014).

Diversas metodologias já foram empregadas para determinação da AA para produtos de café (LUDWING et al., 2012; VIGNOLI; BASSOLI; BENASSI, 2012; LIANG; KITTS, 2014; POKORNÁ et al., 2015, MARCUCCI et al., 2017), dentre os métodos indiretos mais empregados destaca-se o ABTS, ensaio que se baseia na habilidade de sequestro do cátion radical de longa vida ABTS•+. Termodinamicamente, um composto pode reduzir ABTS•+ quando tem um potencial redox menor que o ABTS (0,68 V), o que

acontece com muitos compostos fenólicos (AWIKA et al., 2003). No café torrado, a AA é usualmente expressa em mmol equivalente de Trolox g^{-1} ou g de Trolox 100 g^{-1}.

Vignoli et al. (2014), estudando cafés arábica e canéfora em diferentes graus de torra, verificaram que as mudanças na composição de bioativos foram muito mais expressivas que as observadas na AA. Os autores reportaram maior AA para café canéfora e arábica com menor grau de torra, pelo maior teor de ACG. Observou-se, no entanto, que a AA do café torrado é mais dependente da espécie de café do que do processo, destacando-se que o café canéfora, pelo alto teor de cafeína, apresentou AA sempre superior ao café arábica. Para cafés arábica de diferentes variedades e grau de torra médio Kitzberger, Scholz e Benassi (2014) reportaram AA variando de 3,75 a 5,42 g Trolox 100 g^{-1}. Almeida e Benassi (2011) analisaram a AA de cafés torrados e moídos comerciais, provavelmente *blends* de arábica e canéfora, e observaram a variação de 2,28 a 6,76 g Trolox 100 g^{-1}. Para café solúveis comerciais regulares e descafeinados procedentes do mercado brasileiro, Marcucci et al. (2017) reportaram altos valores de AA (entre 20,4 e 37,0 g Trolox 100 g^{-1}), atribuídos a presença de café canéfora como principal matéria-prima. Mori (2016) relatou para bebidas de café canéfora de diferentes cultivares brasileiras, valores médios de 6,78 a 8,80 mg de Trolox mL^{-1} da bebida.

O tratamento do café com vapor se desenvolveu na Alemanha, com intuito de fazer um café mais "aceitável" para certos consumidores que relataram vários graus de desconforto no estômago quando do consumo de café regular, já que o procedimento permite que os grãos passem por mudanças químicas e físicas (ITC, 2014). O emprego de vapor no tratamento de grãos de café tem demonstrado resultados interessantes no balanço de compostos voláteis observados após o processo de torra, característica importante para a qualidade da bebida de café torrado (BAGGENSTOSS et al., 2008; BECKER; SCHLABS; WEISEMANN, 1991; DE CONTI, 2013).

Baggenstoss et al. (2008) compararam a composição de voláteis em café arábica não tratado e tratado com vapor, e submetidos posteriormente a processo de torra clara (Tabela 1.8). O tratamento com vapor proporcionou um aumento do composto volátil de impacto positivo/negativo 2-furfuriltiol, enquanto propiciou uma redução nos compostos voláteis 2-metil-butanal e 4-vinilguaiacol, ambos de impacto negativo na bebida (Tabela 1.8).

Tabela 1.8 – Compostos voláteis (μg kg^{-1} bs) identificados em café arábica torra média tratado e não tratado com vapor antes do processo de torra média.

Composto aromático	Não tratado	Tratado com vapor
2-furfuriltiol	$0,39^{b} \pm 0,03$	$0,91^{a} \pm 0,15$
2-metil-butanal	$21,9^{a} \pm 0,5$	$20,2^{b} \pm 0,7$
3-metil-butanal	$13,0^{a} \pm 0,3$	$12,7^{a} \pm 0,6$
Hexanal	$3,9^{a} \pm 0,5$	$2,97^{a} \pm 0,06$
2,3-butanodiona	$22,1^{a} \pm 0,6$	$23,2^{a} \pm 0,2$
2,3-pentanodiona	$13,2^{a} \pm 0,3$	$13,2^{a} \pm 0,4$
Piridina	$30,1^{a} \pm 1,5$	$24,8^{b} \pm 0,3$
4-vinil-guaiacol	$27,8^{a} \pm 1,1$	$23,7^{b} \pm 0,4$
2,3,5-trimetil-pirazina	$4,7^{a} \pm 0,3$	$4,4^{a} \pm 0,4$

Médias com a mesma letra na linha não diferiram pelo teste de *t* de Student ($p > 0,05$). Fonte: Adaptado de Baggenstoss et al. (2008).

O tratamento com vapor tem sido proposto com mais frequência para café canéfora, pois verificou-se que no café tratado com vapor poderia ser percebida uma maior "acidez". Um café canéfora com qualidade incrementada é especialmente interessante nos períodos de preços altos do café arábica, no preparo de *blends* (ITC, 2014).

Algumas patentes mencionam a possibilidade do emprego de tratamento com vapor em grãos para melhora na qualidade da bebida de café, mas a literatura científica tem pouca informação sobre o efeito do processo na composição e qualidade sensorial.

Becker, Schlabs e Weisemann (1991) trataram grãos de café canéfora com umidade de 30-45% por injeção de vapor a 135-140 °C, com pressão de 3-4 bar por 60-120 min, obtendo melhora na qualidade da bebida, justificada pela eliminação do composto 2-metilisoborneol, responsável pelos sabores de “mofo” e “terra”.

Manfred (1997) propôs um método de tratamento de café cru para remoção de compostos que prejudicam o aroma e sabor, especialmente aqueles que surgem durante a maturação, colheita ou processamento. A patente prevê que o café cru seja tratado com vapor a uma pressão elevada e uma temperatura acima de 100 °C, até que quantidade suficiente dos elementos indesejáveis sejam removidos.

De Conti (2013) utilizou processo combinando de tratamento com vapor e ácido em café canéfora. Primeiramente, os grãos de café cru foram imersos durante 10 min em soluções com concentrações de ácido acético variando de 0 a 6,9%. Para o tratamento posterior, foi empregada pressão de vapor entre 0,66 e 2,34 bar, tempo de contato entre grãos e vapor de 6,6 a 23,4 min. O tratamento conjunto com vapor e ácido reduziu a acidez

titulável, teor de cafeína e de compostos voláteis de impacto negativo na bebida (furfural, 4-vinilguaiacol e ácido acético), enquanto os compostos voláteis de impacto positivo (2,3-pentanodiona, 3-metil-butanal, maltol, benzaldeído e vanilina) foram maximizados. O autor relatou ainda que as variáveis pressão de vapor e tempo de contato com vapor se compensam, logo sob pressões menores e tempos maiores de contato é possível obter resultados similares na composição volátil quando aplicado pressões maiores em tempos de contato mais curtos.

Nesse mesmo estudo, De Conti (2013) utilizou, para avaliar o efeito do tratamento em café canéfora torrado, uma classificação dos compostos voláteis que interferem positiva ou negativamente na qualidade da bebida proposta por Bassoli (2006), que separou diversos compostos presentes em café solúvel produzido com café arábica e/ou canéfora considerando os grupos olfatométricos e sua influência na bebida (Tabela 1.9). No café canéfora contendo 8,18% de grãos ardidos, 7,1% quebrados, 2,9% brocados, 3,67% verdes, 2,21% pretos, 2,1% chochos ou mal granados, 0,32% de coco e 0,8% de cascas pequenas, o autor ressaltou como voláteis de impacto positivo na bebida a 2,3-pentanodiona, 3-metil-butanal, maltol, benzaldeído e vanilina, enquanto ressaltou como voláteis de impacto negativo na bebida o furfural, 4-vinil-guaiacol e ácido acético (DE CONTI, 2013).

Tabela 1.9 - Grupos olfatométricos propostos para vários compostos odoríferos de importância para a qualidade do café.

Grupo olfatométrico/descritor	Compostos voláteis
	Qualidade positiva para a bebida
Doce, caramelo	2,3-butanodiona; 2,3-pentanodiona; 3-hidroxi-2-butanona; 2-furfuriltiol; 3-metil-2-buten-1-ol; 2,5-dimetil-3-etil-pirazina; álcool benzílico; maltol; furaneol; 2-pirrolidona
Floral	acetato de metila; 3-hexen-2-ona; benzaldeído; acetato de 2-furanometanoila
Frutal	acetato de etila, 3-metil-butanal, 2,4-dimetil-3-pentanona, limoneno, 1-hidroxi-2-propanona, 2,5-dimetilpirazina, etil-pirazina; 2,3-dimetil-pirazina; 2-etil-3-metil-pirazina; propanoato de 2-furanometanoila
Nozes	1-metil-piperidina; 2,5-dimetil-pirazina; 2,3-dimetil-pirazina; 2,5-dimetil-3-etil-pirazina; 5H-5-metil-6,7-diidrociclopentapirazina, butirolactona
Queimado, torrado, cereal	propanal; 2,3-diidro-4-metil-furano; etil-benzeno; 2,3-dimetil-isoxazol; piridina; 1-etil-3-metil-benzeno; metil-pirazina; 4-metil-tiazol; 2,5-dimetil-pirazina; 2,6-dimetil-pirazina; 2,3-dimetil-pirazina; 4,5-dimetil-tiazol; 3-etil-piridina; 2-etil-6-metil-pirazina; 2-etil-5-metil-pirazina; 2-etil-3-metil-pirazina; 2,6-dietil-pirazina; 2-furfuriltiol; 2,5-dimetil-3-etil-pirazina; 1-(furanil)-etanona; benzaldeído; 5-metil-2-furanocarboxialdeído; 1-(2-piridinil)-etanona; acetil-pirazina; 2-furilmetanol; 3,4-dimetil-2,5-furanodiona; guaiacol; álcool fenil-etenílico; 4-etil-guaiacol; ácido nonanóico; 4-vinil-guaiacol

	Qualidade negativa para a bebida
Defumado, fenólico	1-hidroxi-2-propanona; 2-metil-2-ciclopenten-1-ona; 2-etil-6-metil-pirazina; (1-metil-etenil)-pirazina; 2-furilmetanol; 2-hidroxi-3-metil-2-ciclopenten-1-ona; 2-hidroxi-3-etil-2-ciclopenten-1-ona; 1-(2-furanil-metil)-1H-pirrol; guaiacol; álcool fenil-etenílico; 4-etil-guaiacol; ácido-nonanóico; 4-vinil-guaiacol; *cis*-isoeugenol
Fermentado, azedo	2-butanona; ácido isovalérico; 2-(2-furanil-metil)-5-metil-furano
Mofado, terra	4,5-dimetil-oxazol; pirazina; metil-pirazina; 4-metil-tiazol; 2,5-dimetil-pirazina; etil-pirazina; 2,3-dimetil-pirazina; 2,6-dietil-pirazina; 2-etil-1-hexanol; 2-hidroxi-3-etil-2-ciclopenten-1-ona
Pungente, pútrido	metanotiol; etanal; butanal; 2-metil-butanal; decano; dissulfeto de dimetila
Químico, etérico, solvente	acetona; 2-metil-furano; 2-butanona; pirazina; 2-metoxi-metil-furano; 2,6,11-trimetil-dodecano; 2,6-dimetil-pirazina; 2-etil-5-metil-pirazina; 2-etil-3-metil-pirazina; 2-furfuriltiol; 3-metil-2-buten-1-ol; ácido acético; guaiacol; 2,4-tert-butil-fenol
Tempero, cozido, sulfuroso	1,2,3,6-tetraidro-1-metil-piridina; 1-(2-metil-1-propenil)-pirrolidona; 4-metil-tiazol; 2,6-dimetil-pirazina; 2,3,5-trimetil-pirazina; metional; 2,3-dietil-5-metil-pirazina; 1,5-dimetil-1H-pirrol-2-carbonitrilo
Vegetal, herbáceo	hexanal; 3-penten-2-ona; 2-metoxi-metil-furano; 2(n-propril)-pirazina; 2,6-dietil-pirazina; 2,5-dimetil-3-etil-pirazina; furfural; linalol; ácido propanoico

Fonte: Bassoli (2006).

Apesar da informação de que o tratamento com vapor pode afetar o perfil de voláteis dos cafés, com redução e eliminação de compostos voláteis indesejáveis, não há estimativa do emprego desse tipo de tratamento e do uso comercial de cafés tratados com vapor pela indústria (ITC, 2014).

A análise sensorial é uma disciplina científica usada para evocar, medir, analisar e interpretar as reações das características dos alimentos e materiais como são percebidas pelos sentidos da visão, olfato, gosto, tato e audição (ABNT, 1993). É realizada em função das respostas transmitidas pelos indivíduos às várias sensações que se originam de reações fisiológicas e são resultantes de certos estímulos, gerando a interpretação das propriedades intrínsecas aos produtos (IAL, 2005).

Os métodos sensoriais são classificados nas categorias de testes afetivos, discriminativos ou descritivos. Entre os métodos afetivos é bastante usual o emprego da escala hedônica para avaliar a aceitação. O julgador expressa o grau de gostar ou desgostar de um determinado produto, de forma global ou em relação a um atributo específico. Os métodos discriminativos têm como objetivo indicar se há diferenças perceptíveis na comparação de amostras, enquanto os métodos descritivos tem por finalidade caracterizar uma amostra quanto aos atributos sensoriais (DUTCOSKY, 2013; IAL, 2005; LAWLESS, HEYMANN, 2010).

Além dos métodos sensoriais mais tradicionais, novas abordagens tem sido propostas combinando métodos e procedimentos já usuais para permitir a obtenção de informações sensoriais mais específicas, facilitar avaliação por um julgador não treinado ou consumidor e/ou maior agilidade na realização dos testes.

A escala hedônica híbrida é uma escala linear resultante da combinação das escalas estruturada e não estruturada. Villanueva, Petenate e Silva (2005) descrevem que para ficar mais próximo do consumidor do que a escala não estruturada, a escala hedônica híbrida possui âncoras com rótulos afetivos verbais nas regiões média ("não gostei/nem desgostei") e extremos da escala ("desgostei extremamente" e "gostei extremamente"). Os autores relatam que essa escala pode apresentar algumas vantagens pela sua facilidade de uso, reduzindo os efeitos numéricos e contextuais e favorecendo seu emprego em estudos interculturais.

Prescott et al. (2005) propuseram uma metodologia que combina o teste pareado preferência e o teste triangular para determinar o limiar de rejeição do consumidor (*consumer rejection threshold*) e o limiar de detecção do consumidor (*consumer detection threshold*) em relação a um constituinte presente na amostra. O método permite determinar o ponto a partir do qual começa a ocorrer rejeição e percepção sensorial do constituinte no alimento, pela adição em diferentes proporções e comparado com um padrão. Já foram reportadas aplicações da metodologia citada em trabalhos que incluem a rejeição/identificação em diferentes tipo de vinhos de vários compostos como 2,4,6-tricloroanisol (PRESCOTT et al., 2005, TEIXEIRA et al., 2006), 1,8-cineol (eucaliptol) (SALIBA; BULLOCK; HARDIE, 2009), 1,1,6-trimetil-1,2-di-hidronaftaleno (ROSS et al., 2014), fenil acetato de etila e ácido fenilacético (CAMPO et al., 2012), bem como da presença de chá verde e extrato de semente de uva em vinho branco e bordô (YOO et al., 2012). Foi descrita a aplicação do método quanto a identificação da adição de componentes e/ou uso de processos diversos em produtos de chocolate (HARWOOD; ZIEGLER; HAYESET, 2012a; HARWOOD; ZIEGLER; HAYESET, 2012b; HARWOOD et al., 2013), frutas e derivados (LIMA FILHO et al., 2014; METHVEN et al., 2016; PINELI et al., 2016) e produtos lácteos (BAKKE; SHEHAN; HAYES, 2016). Entretanto, para produtos de café não foram localizadas aplicações da metodologia até o momento.

Com relação a novas metodologias descritivas, Varela e Ares (2012) destacam o interesse de ter alternativas confiáveis, simples e rápidas para caracterização sensorial, proporcionando mapas sensoriais próximos a uma análise descritiva clássica com

avaliadores altamente treinados. Os autores ressaltam que essas metodologias não devem ser consideradas como substitutas das análises descritivas clássicas mas sim como técnicas complementares.

O Perfil Flash é uma análise descritiva, que combina o levantamento de atributos do Perfil Livre com a avaliação dos atributos empregando um procedimento de ordenação, baseado na apresentação simultânea de todas as amostras a serem avaliadas, proporcionando uma descrição e discriminação rápida de um conjunto de produtos (DAIROU; SIEFFERMANN, 2002). Os participantes são convidados a gerar descritores sensoriais que diferenciam ou são comuns ao conjunto de amostras. Na sequência, após desenvolver uma lista individual de atributos e definições, as amostras são apresentadas simultaneamente para avaliação (LAWLESS; HEYMANN, 2010; DELARUE; SIEFFERMANN, 2004).

No procedimento original proposto por Dairou e Sieffermann (2002), o Perfil Flash é realizado em duas sessões. Na primeira, os avaliadores avaliam um conjunto de amostras e geram os atributos que considerarem adequados para discriminá-las. Após a geração dos atributos, é permitido ao avaliador consultar as listas de atributos/definições dos demais membros da equipe, para, se necessário, adicionar ou retirar descritores de sua lista. Na segunda sessão, os avaliadores recebem novamente o conjunto de amostras e ordenam os produtos em relação à intensidade dos atributos. Os autores reportam que o método poderia ser empregado por avaliadores não treinados, mas também por experts ou avaliadores com experiência anterior com o produto.

Para permitir a aplicação em uma única sessão, Terhaag e Benassi (2010) propuseram uma modificação no método, retirando a etapa onde o avaliador é informado sobre a lista de atributos dos outros membros da equipe (que não é permitida no Perfil Livre). Dessa forma, após a discussão e montagem da ficha individual de avaliação, o avaliador retorna a cabine para avaliação do conjunto de amostras.

Perfil Flash já foi empregado em diferentes matrizes alimentares como vinho, soja, geleia, maçã, salsicha, pera e pão (LIU et al., 2016; TERHAAG; BENASSI, 2010; BLANCHER et al., 2007; DAIROU; SIEFFERMANN, 2002; JAROS et al., 2009; RASON et al., 2006; TAREA; CUVELIER; SIEFFERMANN, 2007; POINOT et al., 2007), sendo encontrado apenas um estudo com produtos de café (KOBAYASHI; BENASSI, 2012).

Apesar da importância da análise sensorial na avaliação das características e qualidade das bebidas de café, a literatura reporta poucas informações sobre produtos de café canéfora em comparação aos de arábica. Ressalta-se também que para cafés, tanto canéfora como arábica, não foram localizados na literatura estudos que envolvem análise sensorial de cafés defeituosos.

1.3 OBJETIVO GERAL

Avaliar o efeito do tratamento com vapor na composição e características de defeitos preto, verde e ardido (PVA) de café canéfora torrado, bem como propor um processo (condições de pressão de vapor e tempo) para obtenção de um produto que possa ser empregado em *blends* com obtenção de bebidas de café torrado com boa qualidade.

1.4 OBJETIVOS ESPECÍFICOS

O trabalho teve como objetivos específicos:

- Caracterizar a matéria prima, café canéfora, em relação à proporção de cada defeito que compõe o conjunto de grãos PVA e tratar os grãos em diferentes condições de pressão de vapor e tempo, utilizando um planejamento fatorial completo, compararando as características dos cafés PVA tratados com um café PVA controle (não tratado) após processo de torra padronizado;

- Avaliar o impacto dos tratamentos nos teores de alguns compostos bioativos e de importância para a qualidade de bebida (cafeína, trigonelina, ácidos clorogênicos, melanoidinas e diterpenos) e atividade antioxidante (pela atividade sequestrante do radical ABTS), verificando as condições de tratamento que permitem melhor retenção desses compostos;

- Avaliar o impacto dos tratamentos no perfil de voláteis, de impacto positivo e negativo na qualidade de bebida, verificando as condições que permitem maximizar compostos de impacto positivo e minimizar compostos de impacto negativo;

- Selecionar os tratamentos mais promissores para avaliação sensorial, determinar o limiar de rejeição do consumidor e o limiar de detecção de bebidas com PVA tratado adicionado ao café arábica sem defeito, definindo possíveis porcentagens de adição em *blends* e avaliar a aceitação sensorial dos *blends*;

- Estudar a viabilidade do uso do café PVA tratado na melhor condição em blends com cafés arábica e canéfora, comparando as características sensoriais (avaliadas por Perfil Flash) e aceitação das bebidas de blends as obtidas com cafés puros (arábica e canéfora).

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CAPÍTULO 2

STEAM PRESSURE TREATMENT OF DEFECTIVE *Coffea canephora* BEANS IMPROVES THE VOLATILE PROFILE AND SENSORY ACCEPTANCE OF ROASTED COFFEE BLENDS.

(Artigo submetido para avaliação na revista Food Research International).

Steam pressure treatment of defective *Coffea canephora* beans improves the volatile profile and sensory acceptance of roasted coffee blends.

Abstract

Between 15 and 20% of Brazilian coffee production corresponds to defective beans (PVA), which decreases the quality of the coffee brew. Steam treatment has been reported as an alternative to improve the volatile profile and cup quality of coffee. The aim of this study was to propose a steam treatment of defective *Coffea canephora* beans to improve the volatile profile of the roasted coffee. The sensory impacts of adding steamed coffee (SC) in *Coffea arabica* blends were evaluated. The steam treatments studied modified the volatile profile of roasted SCs, increasing the contents of acetoin, benzyl alcohol, maltol, 2,6-dimethylpyrazine, 2-furfurylthiol, and 5-methylfurfural and decreasing the contents of 4-ethylguaiacol, isovaleric acid, methional, 2,3-diethyl-5-methylpyrazine, and 3-methoxy-3-methylpyrazine. Among the evaluated parameters, the best condition to maximized the content of the volatiles with a positive impact and minimize those with a negative impact was 5 bar/16 min (SC 5). The thresholds of consumer rejection and of detection indicate that up to 30% SC 5 can be added to a high cup quality *C. arabica* coffee without perception or rejection of the coffee brew. A blend of 30/70% SC 5/*C. arabica* was well accepted.

Keywords: roasted and ground coffee, coffee brew, PVA beans, black, immature, sour.

2.1 Introduction

Brazil is the world's largest coffee producer and exporter, producing the following two main commercial species: *Coffea arabica* L. (arabica coffee) and *Coffea canephora* Pierre (robusta coffee).

The ripe fruits or cherry coffees provide high quality brews, and therefore, they are more valued in the market. Defective coffee beans are usually present in Brazilian coffee due to the strip-picking harvesting and processing practices used by the coffee producers. Approximately 20% of all coffee produced in Brazil is defective beans, which are considered inappropriate for export and are usually incorporated into the internal market. In addition to the differences in their composition compared to non-defective coffee

(Craig, Franca, & Oliveira, 2012), defective beans markedly decrease cup quality (Bandeira, Toci, Trugo & Farah, 2009; Toci & Farah, 2014).

The main defective beans related to coffee cup quality are the intrinsically defective ones known as PVA, from the Portuguese “preto” (black beans), “verde” (green or immature beans) and “ardido” (sour beans). Immature beans originate mainly from immature fruits; sour beans can be generated through abnormal fermentation; and black beans often originate from over-ripened cherries that fall and remain in contact with the soil, which favors fermentation during post-harvesting (Mendonça, Franca, & Oliveira, 2009; Mendonça, Franca, Oliveira, & Nunes, 2008).

Some volatile compounds were cited as potential drivers for defective coffee. Compared to non-defective coffees, higher differences in the volatile profile were observed for black and sour beans (probably due to fermentation) (Franca & Oliveira, 2008) than for immature beans (Toci & Farah, 2014). The compound 2,3,5,6-tetramethylpyrazine was reported for medium roasted black and sour beans (Toci & Farah, 2014), while 2,3-butanedione, 2-methylbutanal, 3-methylbutanal, 4-ethylguaiacol, 4-methylthiazole were reported for roasted PVA mixture (Toledo, Pezza, Pezza, & Toci, 2016). Meanwhile volatiles as 2,5-dimethylpyrazine, 2,6-dimethylpyrazine, 2-acetyl-3,5-dimethylpyrazine, 2,3-dimethylpyrazine and pyridine were detected in all beans (black, sour, immature and non-defective coffees) (Toci & Farah, 2014).

C. canephora is the main raw material for the instant coffee industry. However, another of its relevant uses is roasted and ground coffee, blended with *C. arabica*. The total production of *C. canephora* in Brazil reached 11.19 million 60 kg bags in 2015 (Companhia Nacional de Abastecimento (CONAB), 2016). *C. canephora* presents a higher content of soluble solids and a higher yield after the roasting process compared to *C. arabica;* on the other hand, it also presents lower commercial value and sensory quality. *C. canephora* can have objectionable tarry, earthy, and bitter flavor notes associated with undesirable volatiles (Ponzoni, Protomastro & Stefanucci, 1973), and its sensory quality can be further downgraded due to the presence of defects.

Steam processing has been applied to coffee as an industrial pretreatment (PIETSCH, 2017). One of the goals is to reduce unwanted taste components in *C. canephora*, mainly by increasing the pressure during treatment. The process improves the volatile profile generated by the roasting process, an important characteristic for the quality of the roasted coffee brew. Early research in this area includes mainly patents (Becker,

Schlabs & Ag, 1991; Dar, Bruckmann & Kelly, 1985; Manfred 1997). Regarding the impact of volatile profile on the characteristics and the cup quality of a coffee brew, several compounds present in roasted coffees, which were studied in this research, have been classified into positive or negative active odors groups considering their sensory characteristics (Table 2.1).

Table 2.1 – Active odor, aroma description and odor thresholds of volatile compounds identified in coffee.

Volatile compounds	Active odor[a]	Aroma description	Threshold (water) ($\mu g\ kg^{-1}$)
2,3-Butanedione	(+) Sweet, caramel	Oily, buttery[c, h]	4.40[h], 15[c]
2,3-Pentanedione	(+) Sweet, caramel	Oily, buttery[c, h]	20[d], 30[c], 30.19[h]
Acetoin	(+) Sweet, caramel	Flowery, wet[e]	8000[d]
Benzyl alcohol	(+) Sweet, caramel	Sweet, flowerly[f]	2546.21[f]
Maltol	(+) Sweet, caramel	Caramel-like[b]	2500[d], 9000[b]
Furaneol	(+) Sweet, caramel	Sweet, caramel-like, candy cotton[c, e]	30[b], 60[d], 100[c]
Benzaldehyde	(+) Floral (+) Burned, toasted, cereal	Bitter almond[f], fruity[j]	350[b,d], 750.89[f]
Furfuryl acetate	(+) Floral	Flowery (very soft), fruity (sweet)[h]	
Limonene	(+) Fruity	Citrus-like[b]	10[b]
3-Methylbutanal	(+) Fruity	Fruity (peach), chocolate, burned matter, malty[b, c, h]	0.2[b, d, g], 0.35[h]
2,4-Dimethyl-3-pentanone	(+) Fruity		
2,5-Dimethylpyrazine	(+) Fruity (+) Nut (+) Burned, toasted, cereal (-) Moldy, earth	Coffee (nuts, roasted)[j]	1700[d], 1800[g, i]
2,3-Dimethylpyrazine	(+) Fruity (+) Nut (+) Burned, toasted, cereal (-) Moldy, earth	Linseed oil, moldy, chocolate, walnuts[h]	2500[d, g]
Pyridine	(+) Burned, toasted, cereal	Rotten fish, burned matter, smoky[h]	2000[d]
2,6-Dimethylpyrazine	(+) Burned, toasted, cereal (-) Chemical, ethereal, solvent (-) Spicy, cooked, sulfur	Cocoa, toast nuts, roasted meat[h]	1500[d, g]
4,5-Dimethylthiazole	(+) Burned, toasted, cereal		
2-Ethyl-3-methylpyrazine	(+) Fruity (+) Burned, toasted, cereal (-) Chemical, ethereal, solvent	Roasted matter[h]	130[g]
2-Furfurylthiol	(+) Sweet, caramel (+) Burned, toasted, cereal (-) Chemical, ethereal, solvent	Burned matter, caramel, roasty, coffee-like[c,b, h]	0.01[h], 0.012[b]
5-Methylfurfural	(+) Burned, toasted, cereal	Caramel, burned sugar, aromatic, sweet[h]	500[d]
2-Acetylpyridine	(+) Burned, toasted, cereal	White-bread crust, roasted barley, popcorn[b, h]	19[b]
2-Acetyl-3,5-dimethylpyrazine	(+) Sweet	Sweet[j]	
Vanillin	(+) Sweet	Sweet, vanilla-like[b, c]	20[b], 58[d]
Phenylethyl alcohol	(+) Burned, toasted, cereal (-) Smoked, phenolic	Honey, rosy[f]	564.23[f]
4-Ethylguaiacol	(+) Burned, toasted, cereal (-) Smoked, phenolic	Spicy, clove[c, f]	50[h], 89.25[f]
4-Vinylguaiacol	(+) Burned, toasted, cereal (-) Smoked, phenolic	Spicy, woody, amber[c, f]	3[d], 5[b], 12.02[f], 20[h]
Cis-isoeugenol	(-) Smoked, phenolic		

Isovaleric acid	(-) Fermented, sour	Fruity, rancid[e], cheese, herbaceous[h]	4.50[h]
Pyrazine	(-) Moldy, earth (-) Chemical, ethereal, solvent	Lightly ammonia and sweetened [h]	18000[d]
4-Methylthiazole	(+) Burned, toasted, cereal (-) Moldy, earth	Fruity, meat, vegetables[h]	
Methanethiol	(-) Pungent, putrid	Sulfurous, putrid[b]	0.02[b]
Dimethyldisulfide	(-) Pungent, putrid	cabbage-like[b, f]	1.10[f], 7.6[b]
Acetic acid	(-) Chemical, ethereal, solvent	Vinegar-like[b], acid, fatty[e]	22000[b]
2,3,5-Trimethylpyrazine	(-) Spicy, cooked, sulfur	Nutty, grass, burned matter, raw potato, roasty, earthy[c, b, h]	23[d], 90[b]
Propanoic acid	(-) Vegetable, herbaceous		
Acetaldehyde (ethanal)	(-) Pungent, putrid	Ether, pungent, acrid[h]	10[b], 25.1[f]
Guaiacol	(+) Burned, toasted, cereal (-) Smoked, phenolic (-) Chemical, ethereal, solvent	Phenolic, chemical, spicy, smoky, burning, sweet[b, c, e]	1[b], 3[d], 12[h]
Methional	(-) Spicy, cooked, sulfur	Potato-like, boiled, sweet[c,j]	0.2[d, g], 0.45[f]
2,3-Diethyl-5-methylpyrazine	(-) Spicy, cooked, sulfur	Mouldy, burned matter, nutty, potato-like, hazelnut, earthy, roasty[c, h]	0.09[h]
Furfural	(-) Vegetable, herbaceous	Bread, almond, nutty, caramel-like (burned notes), grass[h]	3000[b, d, g]
Linalool	(-) Vegetable, herbaceous	Flowery, fruity, citric [b, c, e]	6[b]
5-Hydroxymethylfurfural	(-) Chemical	Chemical	
2-Methoxy-3-methylpyrazine	(+)	Roasted peanuts, hazelnuts, almond[k]	3[k], 4[i]
2,3,5,6-Tetramethylpyrazine	(-)	Fermented soy[f]	2525.02[f]
2-Isobutyl-3-methylpyrazine	(+)	Grass, caramel, sugar burned[h]	35[g]
2-Isobutyl-3-methoxypyrazine	(-)	Grass, earthy, green pepper, pea-like, hot paprika (red pepper)[b, h]	0.002[b]

(+) positive impact; (-) negative impact.

References: [a] Bassoli (2006); [b] Belitz et al. (2009); [c] Blank et al. (1991); [d] Buttery et al. (1999); [e] Ferreira et al. (2002); [f] Giri et al. (2010); [g] Guadagni et al. (1972); [h] Nascimento et al. (2007); [i] Seifert et al. (1970); [j] Misharina et al. (2009); [k] Leffingwell & Associates (2017).

Literature about the volatile profile of *C. canephora* defective beans is scarce (Bandeira *et al*. 2009; Mendonça, Franca, & Oliveira, 2009; Mendonça, Franca, Oliveira, & Nunes, 2008; De Conti 2013). Additionally, there is no research on the effect of steam treatment on *C. canephora* PVA beans. It is generally reported that the addition of up to 20% of *C. canephora* in *C. arabica* blends does not affect sensory characteristics and is not perceived by consumers (Santos, Deliza, Freitas & Corrêa, 2013). However, there is no information regarding the impact of the use of *C. canephora* defective beans in coffee blends.

The aim of this study was to propose a steam pressure treatment for *C. canephora* defective beans to improve the volatile profile of the roasted coffee. The sensory effect of the process was evaluated in blends of *C. arabica* with *C. canephora* steamed coffees through the threshold of rejection/detection and an acceptance test.

2.2 Material and Methods

2.2.1 Coffees

To obtain defective beans for the study, *C. canephora* raw coffee from the 2014 crop from Rondônia State (Brazil) was subjected to selection on an electronic sorter until 20 kg of rejected beans was obtained. The raw material was characterized as 34.9% black, 8.9% immature, 37.9% sour, 15.7% non-defective beans, and 2.6% other (including shells and sticks, excluding stones). This material was used for the steamed coffee preparation.

A high cup quality *C. arabica* coffee NY 2 (corresponding to 6 defects per 300 g of sample – more details on classification see Franca, Oliveira, Mendonça, & Silva (2005) - from the 2015 crop from the Mogiana region (São Paulo, Brazil) was used in the sensory tests.

2.2.2 Preparation of steamed coffee

Batches of 1.5kg of the rejected beans (81.7% PVA beans) were submitted to steam treatment following a full factorial design 2^2 with 3 repetitions on the central point (Table 2.2). An untreated sample was maintained as the control.

The steam treatments were performed in an equipment based on the one used by De Conti (2013). The basket consists of a cylindrical stainless steel container AISI standard, with 1200 mm of height and 60 mm of diameter. The container surface has holes of 3.0 mm in diameter, resulting in a 20% area opened for steam passage. The container was placed inside a pressure vessel where the steam was injected from biomass boiler. The inlet pressure in the vessel was adjusted through a controlling valve installed in the steam pipe and a manometer recorded the applied pressure (Figure 2.1).

Table 2.2 - Full factorial design (2^2) matrix with coded and real values for the variables.

Run/SC*	Steam pressure (x_1) (bar)	Time (x_2) (min)
SC 1	-1 (2)	-1 (3)
SC 2	+1 (8)	-1 (3)
SC 3	-1 (2)	+1 (29)
SC 4	+1 (8)	+1 (29)
SC 5	0 (5)	0 (16)
SC 6	0 (5)	0 (16)
SC 7	0 (5)	0 (16)

SC: steamed coffee.

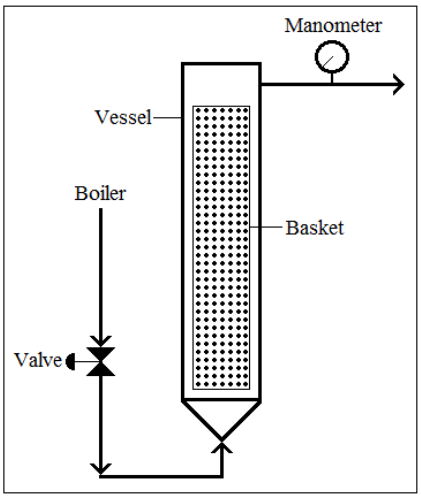

Figure 2.1 - Scheme of equipment used for steam treatment.

Before the roasting process, the steamed coffee beans (SC) were previously dried in vacuum oven (Q819V2, Quimis, São Paulo, Brazil) at 70 °C until moisture reached 7.4 to 9.5 g 100g^{-1} (8.7 ± 0.8 g 100 g^{-1}).

2.2.3 Standardization of the roasting process for steamed coffees and characterization of samples

Subsequently, for moisture standardization, each dry steamed coffee was subjected to a medium roasting process (Rod-Bel, São Paulo, Brazil) at approximately 230 °C, suggested as optimal for *C. canephora* (Mendes, De Menezes & Silva, 2001). The roasting process time was standardized based on L* value, as described in the Results. The weight loss was determined by the difference between the mass before and after roasting.

The roasted coffees were middle ground (0.7% retained in sieve size 1.18 mm; 73% retained in sieve size 0.60 mm, and 20% passing a sieve size 0.60 mm) in a Burr Grinder GVX2 (Krups, Solingen, Germany). The roasted coffees were ground just before each analysis and coffee brews preparation, whenever necessary it was stored in plastic flasks at −18 °C until use.

Color measurements were performed in triplicate on roasted and ground coffee samples placed into a CR A50 accessory of a colorimeter (CR-410, Konica Minolta, Tokyo, Japan). The analyses were recorded under the conditions of standard illuminant C and a 10 degree observer and data were reported on CIELAB color scale.

Regarding the *C. arabica* used in the sensory analysis, the coffee was roasted to medium degree (roasting time of 19 min, L* of 24.5, moisture of 1.9 g 100 g^{-1}).

Moisture content of roasted and ground coffees was measured by infrared moisture analyzer (MB200, Ohaus, Mumbai, India) at 105 ºC for 7 min (triplicate analysis) (Corso, Vignoli & Benassi, 2016).

2.2.4 Analysis of volatile compounds

Volatile compounds analysis was performed through isolation using solid-phase micro extraction (SPME) followed by quantification using an Agilent 6890 N gas chromatograph (California, USA) equipped with an Agilent 5973 mass spectrometric detector and MSD Chemstation software. Sample preparation and chromatograph conditions were applied as described by Viegas and Bassoli (2007).

For headspace analysis, the roasted and ground coffee samples were weighed (2.8 g) in 20 mL vial (Agilent, California, USA) immediately sealed with a silicone septum and kept in water bath (70 °C). After 10 min, the septum was perforated and a Divinylbenzene/Carboxen/Polydimethylsiloxane fiber (film thickness of 50/30 μm) (Sigma Aldrich, St. Louis, USA) was exposed to the headspace for 30 min. The fiber was injected into the gas chromatograph; compounds were thermally desorbed (desorption time of 10 min) and transferred to an Innowax column (60 m x 0.32 mm x 0.25 μm) (Agilent, California, USA).

Carrier gas helium at 1.3 mL min^{-1} flow rate and injector temperature of 250 °C were employed. The heating profile started at 40 °C, kept 5 min, up to 60 °C at 4 °C min^{-1} rate, maintained at 60 °C for 5 min, and up to 250 °C at 8 °C min^{-1} rate, held for 3 min. The mass spectrometer operated at 280 °C interface temperature, ion source temperature of 230 °C, quadrupole temperature of 150 °C to scan a range m/z of 35-400 atomic mass units.

The following standards (Sigma Aldrich, St. Louis, USA) were used: 2-3 dimethylpyrazine, 2-ethyl-3-methylpyrazine, pyrazine, 4-methylthiazole, 2-isobutyl-3-methylpyrazine, 2,3-butanedione, 2,3-pentanedione, acetoin, benzyl alcohol, maltol, furaneol, benzaldehyde, furfuryl acetate, limonene, 3-methylbutanal, 2,4-dimethyl-3-pentanone, 2,5-dimethylpyrazine, pyridine, 2,6-dimethylpyrazine, 4,5-dimethylthiazole, 2-furfurylthiol, 5-methylfurfural, 2-acetylpyridine, vanillin, phenylethyl alcohol, 4-ethylguaiacol, 4-vinylguaiacol, *cis*-isoeugenol, isovaleric acid, methanethiol, dimethyldisulfide, acetic acid, 2,3,5-trimethylpyrazine, propanoic acid, acetaldehyde, guaiacol, methional, 2,3-diethyl-5-methylpyrazine, furfural, linalool, 5-

hydroxymethylfurfural, 2-methoxy-3-methylpyrazine, 2,3,5,6-tetramethylpyrazine, 2-isobutyl-3-methoxypyrazine and 2-acetyl-3 5-dimethylpyrazine. The standards were injected into the GC-MS using the same extraction technique (HS-SPME) applied to the volatile compounds. A 10000 μg kg^{-1} stoke standard solution was prepared for each compound, with the exception of methanethiol (which is a pressurized gas) that was injected with gas tight syringe. The working standard solution range varied between 0.16 and 1000 μg kg^{-1} depending on the compound. The identification was based on mass spectra and on the linear retention index as previously described by Viegas and Bassoli (2007). The quantification was performed by external standardization using calibration curves with six concentration levels each. Recoveries above 80% were obtained for all compounds (data not shown).

The contents of the volatiles compounds (μg kg^{-1}) on the roasted samples obtained through the factorial design (Table 2.2) were analyzed by Experimental Design procedure of Statistica 8.0 software. The adequacy of the models generated by the factorial design was assessed; volatiles with significant effects ($p \leq 0.05$) and $R^2 \geq 0.75$ (used as cutting limit) were considered (Supporting Table S2.1). In order to have a simultaneous optimization of all the responses of the factorial design, the global desirability function was determined using the Response Desirability Profiling procedure. The goal was to maximize the content of the volatiles with positive impact and minimize those with negative impact. The overall desirability value varied from 0, outside an acceptable region, to 1, where the response is at its goal or target. The higher value indicated the more desirable treatment, which is considered as the optimal solution.

2.2.5 Sensory analysis of the blends

Sensory analyses were carried out in lab-scale tests. The participants were university students, teachers and employees, all regular coffee consumers. Before the tests, participants were informed about each analysis and answered a self-administered questionnaire on socio-demographic data and consumption habits. This study was authorized by the Ethics Committee of Universidade Estadual de Londrina (Certificate of Ethical Evaluation Presentation 36840214.0.0000.5231).

A pure *C. arabica* roasted coffee, with high cup quality, was used as a base coffee in order to obtain the blends with selected steamed coffees (SC) and control (untreated) roasted coffees.

For sensory tests, coffee brews were prepared through percolation with Melitta 102 paper filters (Guaíba, Brazil) following the proportion of 50 g of roasted and medium-ground coffee to 500 mL of mineral water (Ouro Fino, Campo Largo, Brazil) at 92 °C. No sugar was added to coffee brews for the threshold tests; for the acceptance test, 12.5% sucrose (suggested as the ideal sweetness by Moraes & Bolini, 2010) was added. After the preparation, the samples were stored in thermal bottles and kept for up to 2 h until being served at 70 °C (Corso, Vignoli & Benassi, 2016).

Tests were conducted in a sensory analysis laboratory, in individual booths, under white light (preference/acceptance tests) or red light (difference test). Coffee brews, approximately 20 mL, were served at 70 °C in 50 mL transparent plastic cups codified with three randomized digits. Consumers were required to clean their palate with water before and between samples.

2.2.6 Consumer rejection threshold and detection threshold

The consumer rejection threshold (CRT) and the detection threshold (DT) were determined according to Prescott, Norris, Kunst and Kim (2005). Seventy-one regular coffee consumers (42 females and 29 males) were recruited. The participants presented diversity in the age (48% aged between 18 to 25, 30% between 26 to 35 and 22% above 36 years old) and in the educational level (45% completed high school, 38% undergraduate and 17% graduate). All participants were coffee consumers: 70% consumed daily, 14% at least 3 times a week, and 16% occasionally. They consumed mostly filtered brew (43%), followed by instant coffee (25%), cappuccino (12%), espresso (11%) and others (9% - including single-dose capsule, decaffeinated and gourmet). The majority of the panel declared themselves as consumers of sweetened coffee (57% with sugar and 12% with sweetening) and 31% without sweeten.

Based on the desirability results, sample SC 5 (5 bar/16 min, central point) was chosen for the threshold test. Considering that up to 20% regular *C. canephora* can be added to *C. arabica* in a blend without reduction of the cup quality (Santos *et al.* 2013), preliminary tests were performed, and the range of SC was defined. SC 5 was added to *C. arabica* coffee in order to achieve 15, 30 and 45% (w/w) blends. The base coffee (pure *C. arabica*) was compared to each blend to establish the thresholds (rejection and detection) of the SC 5.

In a first session, CRT was measured using a series of four paired comparison tests. Each pair consisted of the base coffee (*C. arabica*) and one of the three SC blends or the own base coffee itself. In each test, consumers were required to taste both samples and indicate on a score sheet which sample of the pair they preferred.

In a second session (one week later), DT was measured using a series of three triangle tests, in which one of the three samples was an SC blend. Consumers were asked to identify the different sample.

In both tests, steamed coffee proportion were presented in ascending order, and the order of the SC blend within each pair/trio order was randomized across each of the series. During the sessions, each pair/trio of coded samples was offered sequentially.

Criteria for significant rejection or detection as a function of SC 5 proportion were based on binomial distribution tables for paired-preference and triangle tests ($p \leq 0.05$) (Roessler, Pangborn, Sidel & Stone, 1978). The best estimates thresholds (BETs) were calculated as the geometric mean group (Lawless & Heymann 2010).

2.2.7 Acceptance of steamed coffee blends

The sensory acceptance of the coffee brews was evaluated by 124 regular coffee consumers (65 females and 59 males). The consumers were young (85% aged between 18 and 25, 11% between 26 and 36 and 4% above 36 years old), and the panel was diversified in terms of educational level (85% completed high school, 8% undergraduate and 7% graduate). All participants were coffee consumers; 69% consumed daily, 23% at least 3 times a week, and 8% occasionally. They consumed mostly filtered brew (41%), followed by instant coffee (23%), cappuccino (15%), espresso (11%) and other (10%). The panel was composed mainly of sweetened coffee consumers (81% with sugar and 6% with sweetener).

The proportion of steamed coffees on the blends (30% - w/w) was defined considering the results of the CRT and DT. In addition to SC 5, the acceptance of other SC blends (SC 1 and SC 4) was also studied. SC 1 also presented good results in the desirability study (Figure 2); SC 4 was chosen based on its wide reduction of volatiles with negative impact. Four blends with *C. arabica* were prepared: three with the steamed coffees (SCs 1, 4 and 5) and one with the control (untreated) sample.

The sensory acceptance was measured using a series of four hedonic scale tests. The samples were offered in a monadic way and sequentially, and the presentation order

was randomized across each of the series. A 10-cm hybrid hedonic scale anchored with verbal terms (0=disliked extremely, 5=neither liked, nor disliked, 10=like extremely) (Villanueva, Petenate & Silva, 2005) was used to evaluate the global impression of the samples. The acceptance data were evaluated by main effects ANOVA (considering the samples and consumers as sources of variation and Tukey test ($p \leq 0.05$) using the software Statistica 8.0.

2.3 Results and discussions

2.3.1 Characterization of roasting process

After the steam pressure treatment, all SCs presented high moisture content ranging from 32.0 to 49.8 g 100 g^{-1}. The control sample had a moisture content of 8.6 ± 0.1 g 100 g^{-1}. Therefore, the samples were vacuum dried at 70 °C until reaching similar moisture content levels (8.8 ± 0.8 g 100 g^{-1}).

The roasting process was standardized based on color; an average L* of 26.4 ±1.1 and an average h_0 of 55.7 ± 2.7 were obtained (Table 2.3). The colors of the roasted coffee samples are in accordance with L* range values (from 19 to 27) reported by Vignoli, Viegas, Bassoli and Benassi (2014) for medium roasted *Coffea canephora* coffees. As a result of the roasting process, the weight loss ranged between 15.4 and 17.7%, while the moisture content ranged from 0.8 to 1.3 g 100 g^{-1} (Table 2.3). Wei and Tanokura (2015) described for medium roasted coffee L* of 24.2 and weight loss from 14.0 to 17.0%. The exception was SC 4, which was submitted to the most drastic steam treatment. On this treatment, the previous alteration by the heat was more intense, and therefore, SC 4 underwent a less aggressive roasting process, showing 5.0% weight loss and a moisture content of 4.7 g 100 g^{-1}. Similarly, De Souza and Benassi (2012) estimated an average weight loss of 15.9% in 38 Brazilian commercial coffees. The characterization of the roasting process reinforces that the SCs produced were similar to commercial coffee.

Table 2.3 – Weight loss (%) during the roasting process, and color (lightness and hue) and moisture (g 100 g^{-1}) of roasted samples*.

Parameters / Samples**		SC1	SC2	SC3	SC4	SC5	SC6	SC7	Control
Weight loss		17.7	16.9	17.2	5.0	16.0	16.1	16.4	15.4
Roasted and ground coffees**	Lightness (L*)	27.7 (0.9)	26.7 (0.7)	26.9 (1.1)	26.7 (1.6)	24.3 (0.7)	25.3 (0.5)	25.9 (0.5)	27.6 (0.2)
	Hue (h_0)	59.0 (0.3)	55.3 (0.2)	56.3 (0.3)	57.7 (2.1)	50.7 (0.9)	53.7 (1.2)	55.3 (0.5)	57.9 (0.4)
	Moisture	1.1 (3.4)	0.9 (1.8)	1.0 (2.5)	4.7 (7.6)	1.3 (3.0)	1.0 (2.2)	1.1 (2.8)	0.8 (5.4)

*Mean of triplicate of analysis; values between parentheses represent the coefficient of variation (CV%). **Steamed coffees (SC 1 = 2 bar/3 min; SC 2 = 8 bar/3 min; SC 3 = 2 bar/29 min; SC 4 = 8 bar/29 min; SC 5, SC 6 and SC 7 = 5 bar/16 min) and control (untreated) sample.

2.3.2 Volatile compounds

Thirty-nine volatile compounds were quantified on SC and/or control samples (Table 2.4). Other six compounds (2-3-dimethylpyrazine, 2-ethyl-3-methylpyrazine, pyrazine, 2,3,5-trimethylpyrazine, 4-methylthiazole and 2-isobutyl-3-methylpyrazine) were identified, but they were below the quantification limit.

Table 2.4 – Content (μg kg^{-1}) of volatile compounds in the roasted samples.

Volatile compounds (μg kg^{-1})	Samples*							
	SC 1	SC 2	SC 3	SC 4	SC 5	SC 6	SC 7	Control
Positive impact volatiles								
2,3-Butanedione	0.00	0.00	0.00	0.00	5.43	6.10	0.00	3.59
2,3-Pentanedione	0.00	6.57	6.92	0.00	7.19	3.43	4.59	6.47
Acetoin	32.92	39.83	37.76	46.53	47.97	48.34	50.01	26.40
Benzyl alcohol	0.63	1.91	1.58	0.63	1.46	2.08	2.01	0.76
Maltol	334.49	404.48	394.64	194.52	701.89	575.56	472.39	313.33
Furaneol	3.11	3.76	2.15	1.11	3.93	0.48	2.73	3.12
Benzaldehyde	5.40	3.48	4.20	4.33	3.89	4.93	4.62	3.53
Furfuryl acetate	104.68	124.65	112.00	37.73	124.98	118.14	126.19	127.14
Limonene	0.10	0.00	0.00	0.00	0.00	0.00	0.00	0.00
3-Methylbutanal	3.65	6.54	10.78	8.25	9.86	5.86	7.65	7.78
2,4-Dimethyl-3-pentanone	0.03	0.00	0.05	0.00	0.09	0.00	0.00	0.00
2,5-Dimethylpyrazine	109.43	116.81	128.78	65.05	89.93	116.30	114.36	96.83
2,3-Dimethylpyrazine	-	-	-	-	-	-	-	-
Pyridine	9.55	105.55	100.50	28.45	122.09	98.59	108.04	114.22
2,6-Dimethylpyrazine	17.34	16.41	11.72	0.00	8.36	11.02	2.47	0.00
4,5-Dimethylthiazole	1.18	1.54	1.60	0.38	1.02	1.12	1.06	1.15
2-Ethyl-3-methylpyrazine	-	-	-	-	-	-	-	-
2-Furfurylthiol	10.82	17.21	14.53	8.68	17.25	17.50	18.25	10.06
5-Methylfurfural	216.33	264.03	247.79	211.72	267.79	330.53	335.27	184.54
2-Acetylpyridine	5.86	5.73	5.42	3.38	6.82	6.10	6.23	5.35
2-Acetyl-3,5-dimethylpyrazine	9.92	9.52	10.05	1.17	6.78	8.00	8.07	9.45
Vanillin	0.14	0.00	0.18	0.27	0.23	0.14	0.14	0.14
Negative impact volatiles								
Phenylethyl alcohol	0.16	0.39	0.30	0.16	0.46	0.33	0.46	0.24
4-Ethylguaiacol	100.73	106.27	90.80	34.23	118.20	89.27	93.60	107.95
4-Vinylguaiacol	519.23	533.49	482.61	480.33	547.01	477.12	504.04	421.76
Cis-isoeugenol	5.63	6.56	7.53	7.67	7.24	7.13	7.56	5.70
Isovaleric acid	54.30	54.21	37.11	49.46	51.29	50.30	57.05	58.96
Pyrazine	-	-	-	-	-	-	-	-
4-Methylthiazole	-	-	-	-	-	-	-	-
Methanethiol	0.03	0.03	0.03	0.01	0.04	0.03	0.03	0.02
Dimethyldisulfide	0.01	0.71	1.02	0.31	0.55	0.75	0.81	0.77
Acetic acid	674.53	643.07	506.55	1622.61	742.51	738.41	767.20	496.12
2,3,5-Trimethylpyrazine	-	-	-	-	-	-	-	-
Propanoic acid	15.19	16.88	12.30	41.27	21.49	20.77	19.03	10.24
Acetaldehyde (ethanal)	392.89	510.45	363.36	327.37	575.93	450.86	428.93	344.85
Guaiacol	127.33	143.97	118.47	71.89	151.49	121.98	127.49	143.10

Methional	0.10	0.00	0.00	0.00	0.00	0.00	0.00	0.14
2,3-Diethyl-5-methylpyrazine	5.60	6.06	8.49	3.16	3.99	5.25	5.63	6.77
Furfural	199.50	221.09	237.95	543.43	252.99	300.48	295.63	155.82
Linalool	1.58	1.40	1.03	1.29	1.16	0.38	1.70	0.02
5-Hydroxymethylfurfural	3.95	0.50	1.32	46.41	3.62	0.39	0.00	0.00
2-Methoxy-3-methylpyrazine	0.30	0.03	0.15	0.00	0.26	0.00	0.48	0.60
2,3,5,6-Tetramethylpyrazine	0.00	0.00	0.00	0.86	0.00	0.00	0.00	0.00
2-Isobutyl-3-methylpyrazine	-	-	-	-	-	-	-	-
2-Isobutyl-3-methoxypyrazine	2.17	0.27	2.54	1.41	2.92	2.47	0.41	1.65

- Below the quantification limit.

*Steamed coffees (SC 1 = 2 bar/3 min; SC 2 = 8 bar/3 min; SC 3 = 2 bar/29 min; SC 4 = 8 bar/29 min; SC 5, SC 6 and SC 7 = 5 bar/16 min) and control (untreated) sample.

The steam treatments provided extensive modification to the SC volatile profiles. Literature described the mechanisms of production and degradation for each chemical class groups of volatiles compounds (pyrazines, aldehydes, alcohols, ketones, esters, carboxylic acids, sulfur compounds, thiazoles and pyridines) with heat treatments (Toci & Farah, 2014). In the range studied, eleven volatile compounds with positive impacts and eight with negative impacts were significantly influenced by the steam treatment. The linear models are presented in Table 2.5 (more information in Supporting Table S2.1).

Table 2.5 - Linear models of the factorial design for the prediction of volatile compounds.

Volatile compounds	Model	R^2
	Positive impact on brew	
2,3-Pentanedione	$Y= 4.10 - 3.37x_1x_2$	0.79
Acetoin	$y= 39.26 + 3.92x_1 + 2.89 x_2 + 9.51C$	0.99
Benzyl alcohol	$y= 1.19 - 0.56x_1x_2 + 0.66C$	0.88
Furfuryl acetate	$y= 94.77 - 13.58x_1 - 19.90x_2 - 23.56x_1x_2 + 28.34C$	0.99
3-Methylbutanal	$y= 7.51 + 2.21x_2 - 1.36x_1x_2$	0.76
2,5-Dimethyl- pyrazine	$y= 105.81 - 14.09x_1 - 17.78x_1x_2$	0.75
Pyridine	$y= 61.01 - 42.01x_1x_2 + 48.56C$	0.96
4,5-Dimethyl- thiazole	$y= 1.13 - 0.22x_1 - 0.19x_2 - 0.40x_1x_2$	0.97
2-Furfurylthiol	$y= 12.81 - 1.21x_2 - 3.06x_1x_2 - 4.86C$	0.99
2-Acetyl-3,5-dimethylpyrazine	$y= 7.64 - 2.32x_1 - 2.06x_2 -2.12x_1x_2$	0.98
Vanillin	$y= 0.16 + 0.08x_1 + 0.06x_1x_2$	0.84
	Negative impact on brew	
Phenylethyl alcohol	$y= 0.25 - 0.09x_1x_2 + 0.16C$	0.84
Cis-isoeugenol	$y= 7.05 + 0.27x_1 + 0.75 x_2$	0.80
Dimethyldisulfide	$y= 0.59 + 0.15x_1 - 0.35x_1x_2$	0.86
Acetic acid	$y= 861.69 + 271.15x_1 + 202.89x_2 + 286.88x_1x_2 - 112.32C$	0.99
Propanoic acid	$y= 20.99 + 7.67x_1 + 5.38x_2 + 6.82x_1x_2$	0.99
2,3-Diethyl-5- methylpyrazine	$y= 5.45 - 1.22x_1 - 1.45 x_1x_2$	0.84
Furfural	$Y= 293.01 + 81.77x_1 + 90.20x_2 + 70.97x_1x_2$	0.98
5-Hydroxymethyl- furfural	$y= 13.05 + 10.41x_1 + 10.82x_2 + 2.14x_1x_2 - 11.71C$	0.99

x_1 = steam pressure; x_2 = time; C = curvature (C = 1 on central point, for x_1 and x_2 = 0; C = 0 on the other points) $p \leq 0.05$.

Considering the volatiles with a positive impact, such as acetoin, benzyl alcohol, furfuryl acetate, pyridine, and 2-furfurylthiol, for which the intent is to maximize their concentration, it was found that the maximum concentration was located in the region near the central point (positive curvature). The pressure increase had a negative effect on the levels of 2,5-dimethylpyrazine, which were further reduced with an increase in processing time. For 4-5-dimethylthiazole and 2-acetyl-3,5-dimethylpyrazine, the increase in pressure and/or time reduced volatile contents, and the best results were obtained using lower pressures for longer process times. For 2,3-pentanedione, the highest contents were observed at low pressures with longer processing time or at high pressures for shorter times. The increase in the processing time, associated with the use of low pressures, increased the content of 3-methylbutanal. The most notable behavior was observed for vanillin, wherein the increase in processing time associated with a high pressure increased the volatile content (Tables 2.4, 2.5 and Supporting Table S2.1).

Additionally, two volatiles with positive impact, 3-methylbutanal and 2-furfurylthiol, were highlighted for being detected in all samples (Table 2.4) above the thresholds of 0.2 μg kg^{-1} and of 0.012 μg kg^{-1} (Belitz, Grosch & Schieberle, 2009) (Table 2.1), respectively. Contents of 35.19 and 61.55 μg kg^{-1} have been reported for 3-methylbutanal in *C. canephora* defective beans treated by immersion in diluted acetic acid, followed by steam treatment at 1 bar/10 min and 2 bar/20 min, respectively (De Conti 2013). Similarly, an increase from 390 to 910 μg kg^{-1} of the concentration of 2-furfurylthiol was reported by steam treatment (2 bar/6 min) in light roasted *C. arabica* (Baggenstoss, Poisson, Kaegi, Perren & Escher, 2008).

Some volatile compounds with positive impact showed no change in concentration in the range studied. However, they were affected by the steam treatment, as observed when comparing their contents in SC coffees with their concentration in the control sample (Table 2.4). 2,6-Dimethylpyrazine (from 2.47 to 17.34 μg kg^{-1}) was found in treated samples (except for SC 4, the most aggressive treatment), but this volatile was not present in the control (Table 2.4). For 5-methylfurfural, higher levels were observed in SCs (from 211.72 to 335.27 μg kg^{-1}) than in the control (184.54 μg kg^{-1}) (Table 2.4).

Considering the volatiles with negative impact, for which the intent is to minimize the concentration, the contents of 5-hydroxymethylfurfural and acetic acid were reduced in the region near the central point (negative curvature). Lower contents of *cis*-isoeugenol, dimethyldisulfide, propanoic acid and furfural were observed in mild process conditions

(lower times and pressures). The level of 2,3-diethyl-5-methylpyrazine was reduced in the higher pressure processes associated with longer times. The most notable behavior was observed for phenylethyl alcohol, for which decreases in concentration were observed under both mild (shorter time and low pressure) and aggressive (long time and high pressure) conditions (Tables 2.4, 2.5, and Supporting Table S2.1).

Additionally, two negative impact volatiles, guaiacol and 2,3-diethyl-5-methylpyrazine, showed no change in concentration in the range studied, but they were affected by the steam treatment. Lower levels of both volatiles were present in the SCs, notably so in SC 4 (most drastic treatment), compared to the control (Table 2.4).

It is important to note that several of the undesirable volatiles studied, such as 4-ethylguaiacol, methanethiol, isovaleric acid, acetaldehyde, guaiacol, 2,3-dimethyl-5-methylpyrazine and 2-isobutyl-3-methoxypyrazine, were quantified (Table 2.4) above their odor threshold values in water (respectively, 50 $\mu g\ kg^{-1}$, 3 $\mu g\ kg^{-1}$, 4,5 $\mu g\ kg^{-1}$, 10 $\mu g\ kg^{-1}$, 1 $\mu g\ kg^{-1}$, 0.09 $\mu g\ kg^{-1}$, and 0.002 $\mu g\ kg^{-1}$) (Belitz *et al.* 2009; Nascimento, Aquino, Nascimento, Chang & Morais, 2007; Buttery, Orts, Takeoka & Nam, 1999) (Table 2.1). Therefore, these compounds should negatively impact on volatile profile of the coffee, 4-ethylguaiacol and guaiacol were also previously described as potential drivers for defective beans and low quality indicators (Toci, Farah, 2014).

There is little literature information regarding steam treatment effects, but a few authors have reported a change in the volatile profile of steam pressure-treated coffees. Baggenstoss *et al.* (2008) reported an increase in volatiles with positive (pyridine, 4-vinylguaiacol) and negative impacts (2,3,5-trimethylpyrazine) and a decrease in dimethyldisulfide (undesirable) for *C. arabica* beans steamed at 2 bar/6 min compared to an untreated coffee (both dark roasted). Additionally, in the condition studied, the same authors observed no effect of the steam treatment on the content of other desirable volatile compounds (2,3-butanedione, 2,3-pentanedione, 3-methylbutanal and 2-furfurylthiol). De Conti (2013), studying a steam treatment of *C. canephora* previously submitted to an acid treatment, reported an increase in some volatiles with positive impact (2,3-pentanedione, 3-methylbutanal, maltol, benzaldehyde, and vanillin), even using a milder steam treatment (2 bar/20 min) than the condition applied in this research.

Considering that compounds with positive impact should be maximized and those with negative impact should be minimized, the desirability procedure indicates the central point (5 bar/16 min) as the best condition (overall desirability value $\cong 0.6$), with a major

content of positive impact volatiles and a minor content of negative impact volatiles, followed by the least drastic treatment SC 1 (2 bar/3 min) (Figure 2.2).

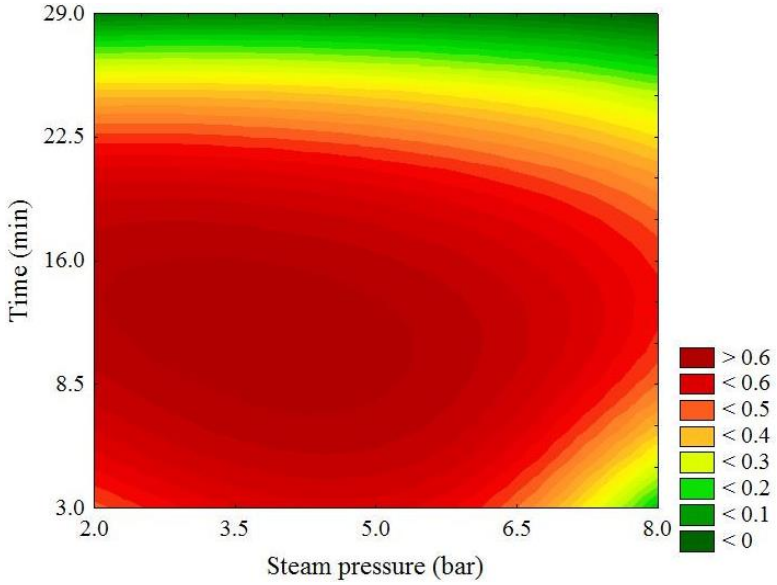

Figure 2.2 - Desirability graph of positive and negative impact volatile compounds

It is still interesting to observe the desirabily results for the steam treatment at 8 bar/29 min. For the SC 4 sample, a wide reduction in the content of some negative impact volatiles was observed (Table 2.5), especially for the compounds that were observed in other SC samples above the threshold (4-ethylguaiacol and methanethiol). However, because of the aggressive condition applied, this sample also presented a decrease in the positive impact volatile contents (Tables 2.4 and 2.5) justifying the worst results in the global desirability (Figure 2.2).

2.3.3 Sensory analysis

Figure 2.3 shows the results for CRT (a) and DT (b), with the proportion of consumers that rejected or identified the SC 5 blends, respectively.

For the consumer rejection threshold (CRT), the proportion of consumers that preferred the base *C. arabica* coffee (0.46) was below the criterion of significant value (0.50). On paired preference tests, no difference ($p > 0.05$) was observed between the base *C. arabica* coffee and the blends with SC 5 (15, 30 and 45%). Similarly, Gonçalves (2006) reported no difference ($p > 0.05$) on the preference for coffee blends containing 5, 10, 20, 30 and 40% defective beans blended with non-defective beans (both *C. arabica*). However,

considering the BET approach, it was estimated that above 31.0% steamed coffee addition, consumers tend to prefer the base coffee without defective beans (Figure 2.3).

For the detection threshold (DT), the proportion of consumers that chose blends with defective coffee beans (0.38) was above the criterion of significant value (0.33), and there was a significant difference ($p \leq 0.05$) between samples above the DT (Figure 3). It was interesting to observe that the DT estimated through BET (34.0%) was near but slightly above the CRT (31.0%). Deliza, Marques, Santos and Farah (2007) reported that some consumers did not find defective beans unpleasant, even when they were able to detect them; a similar situation was described by (Prescott *et al*. 2005) for 2,4,6-trichloroanisole in white wine. Gonçalves (2006) reported a considerably lower DT (16%) for defective beans in *C. arabica* blends, stressing that the steam treatment significantly reduces the perception of the defective beans presence in a coffee brew.

In Brazil, the addition of up to 30% of *C. canephora* in blends with *C. arabica* is common (Souza, Santos, Costa & Santos, 2004). Based on this information and on the CRT and DT obtained, for the acceptance tests, a proportion of 30% of SC or control (untreated) *C. canephora* to 70% of *C. arabica* was defined. Three SCs were chosen for the acceptance evaluation: SC 5 and SC 1, based on their good results in the desirability study, and SC 4, considering its reduction of volatiles with negative impact.

The SC 5 blend was significantly more accepted ($p \leq 0.05$) than the blends with control (untreated) and SC 4 (Table 2.6).

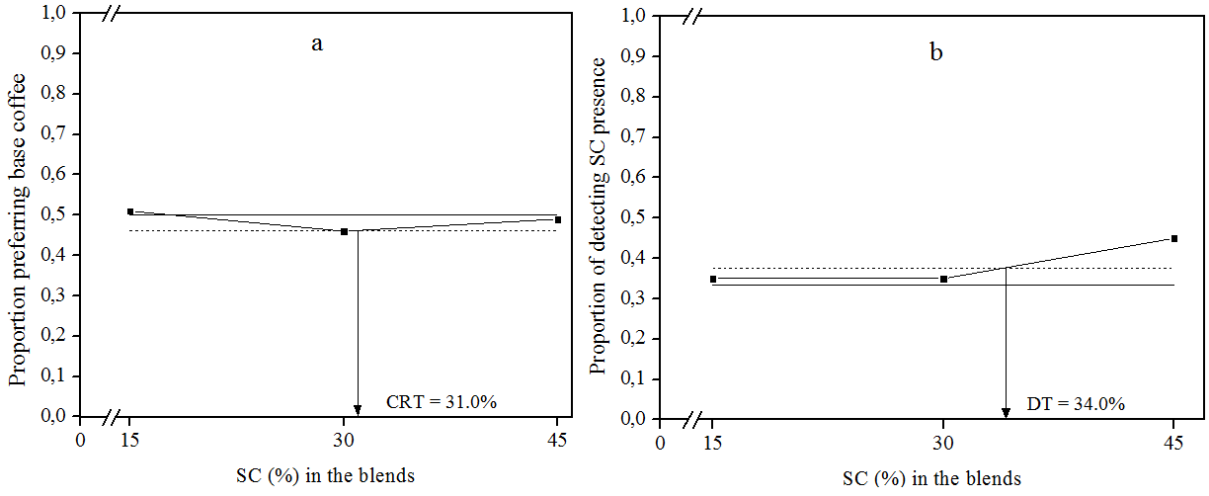

Figure 2.3 - Proportion of consumers that: (a) choose a coffee brew without SC; (b) identify the sc adition .

The solid line indicates no preference (a) or chance responding (b), while the dotted line represents the 5%

criteria considering the binomial distribuition for paired preference test (a) or triangle test (b) (N = 71).

Table 2.6 - Sensory acceptance of coffee blends brew.

Blends (30%)*	Acceptance**
SC 1	$6.7^{ab} \pm 1.9$
SC 4	$6.3^{b} \pm 2.1$
SC 5	$7.1^{a} \pm 1.8$
Control	$6.6^{b} \pm 2.1$

* Blends 30/70% of *C. canephora* steamed coffees/*C. arabica*.

**Mean ± standard deviation; different letters in the column indicate significant differences by Tukey test ($p \leq 0.05$).

The good acceptance of the SC 5 blend reinforces the results of desirability, showing that the changes in the volatile profile produced after the steam treatment of 5 bar/16 min (central point) (Figure 2.2) improved the sensory acceptance. It is also relevant to emphasize the importance of both effects of the steam treatment: an increase in the content of positive impact volatiles and a decrease in the content of negative impact volatiles. The blend with SC 1, which also has a good balance of volatile compounds, did not differ from the SC 5 blend. However, the blend with SC 4, which had the greatest reduction of negative impact volatiles, presented a lower sensory acceptance compared to SC 5.

Considering all findings, the best results were observed for a steam pressure of 5 bar for 16 min of treatment (SC 5 coffee). However, the treatment of 2 bar/3 min (SC 1) also exhibited promising results with a less aggressive and faster treatment and could be considered for further studies.

2.4 Conclusions

The steam pressure treatment provides an extensive modification of the volatile profile of defective *C. canephora* roasted coffee. The most desirable profile was obtained at conditions of 5 bar/16 min (SC 5 coffee). In this condition, the contents of volatiles with a positive impact, such as acetoin, benzyl alcohol, maltol, benzaldehyde, 2,6-dimethylpyrazine, 2-furfurylthiol, 5-methylfurfural, 2-acetylpyridine and 2-acetyl-3,5-dimethylpyrazine, were increased, and the contents of volatiles with a negative impact, such as 4-ethylguaiacol, isovaleric acid, methional, 2,3-diethyl-5-methylpyrazine and 2-methoxy-3-methylpyrazine, were decreased.

Based on the consumer rejection threshold and on the detection threshold, a percentage up to 30% of SC 5 can be added to a high cup quality *C. arabica* coffee without perception or rejection of the coffee brew. An increase in the acceptance of an SC 5 blend (30%) compared to control (untreated) reinforces the importance of a balanced volatile profile and confirms the positive sensory impact of the steam treatment (5 bar/16 min) on the cup quality of the roasted coffee brew.

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Supplementary Material

Table 2.1S - Effects of the factors studied on FFD for the response volatile compounds.

Volatile compounds	Factor	Effect ($\mu g\ kg^{-1}$)	Standard error	t(3)	p-Value	$F_{calculated}$	$F_{tabulated}$	R^2
			Positive impact on brew					
2,3-Pentanedione	Mean	4.10	0.60	6.88	0.001*	21.98	5.99	0.79
	x_1 by x_2	-6.75	1.58	-4.28	0.008*			
Acetoin	Mean	39.26	0.52	75.70	<0.000*	103.16	6.59	0.99
	Curvature	19.03	1.58	12.01	0.001*			
	x_1	7.84	1.04	7.56	0.005*			
	x_2	5.77	1.04	5.56	0.011*			
Benzyl alcohol	Mean	1.188	0.13	8.90	0.001*	17.16	5.79	0.88
	Curvature	1.33	0.41	3.25	0.031*			
	x_1 by x_2	-1.12	0.27	-4.18	0.014*			
Furfuryl acetate	Mean	94.77	2.17	43.66	0.001*	117.80	9.12	0.99
	Curvature	56.68	6.63	8.55	0.013*			
	x_1	-27.15	4.34	-6.25	0.025*			
	x_2	-39.80	4.34	-9.17	0.012*			
	x_1 by x_2	-47.12	4.34	-10.86	0.008*			
3-Methylbutanal	Mean	7.51	0.55	13.66	<0.000*	7.93	5.79	0.76
	x_2	4.42	1.45	3.04	0.038*			
	x_1 by x_2	-2.71	1.45	-1.86	0.136			
2,5-Dimethylpyrazine	Mean	105.81	5.00	21.16	<0.000*	7.35	5.79	0.75
	x_1	-28.18	13.23	-2.13	0.100			
	x_1 by x_2	-35.56	13.23	-2.69	0.055*			
Pyridine	Mean	61.01	5.43	11.25	<0.000*	58.93	5.79	0.96
	Curvature	97.12	16.58	5.86	0.004*			
	x_1 by x_2	-84.03	10.85	-7.74	0.001*			
4,5-Dimethylthiazole	Mean	1.13	0.03	32.59	<0.000*	50.45	6.59	0.97
	x_1	-0.43	0.09	-4.69	0.018*			
	x_2	-0.37	0.09	-4.04	0.027*			
	x_1 by x_2	-0.79	0.09	-8.62	0.003*			
2-Furfurylthiol	Mean	12.81	0.23	56.61	<0.000*	182.95	6.59	0.99
	Curvature	9.71	0.69	14.05	0.001*			
	x_2	-2.41	0.45	-5.32	0.013*			
	x_1 by x_2	-6.12	0.45	-13.52	0.001*			
2-Acetyl-3 5-dimethylpyrazine	Mean	7.64	0.22	34.08	<0.000*	70.94	6.59	0.98
	x_1	-4.64	0.59	-7.82	0.004*			
	x_2	-4.11	0.59	-6.93	0.006*			
	x_1 by x_2	-4.24	0.59	-7.14	0.006*			
Vanillin	Mean	0.16	0.02	10.02	0.001*	13.51	5.79	0.84
	x_2	0.16	0.04	3.73	0.020*			
	x_1 by x_2	0.12	0.04	2.77	0.050*			
			Negative impact on brew					
Phenylethyl alcohol	Mean	0.25	0.03	8.16	0.001*	13.13	5.79	0.84
	Curvature	0.33	0.09	3.47	0.026*			
	x_1 by x_2	-0.19	0.06	-2.99	0.040*			
Cis-isoeugenol	Mean	7.05	0.15	47.25	<0.000*	10.32	5.79	0.80
	x_1	0.54	0.39	1.36	0.247			
	x_2	1.51	0.39	3.81	0.019*			
Dimethyldisulfide	Mean	0.59	0.06	9.97	0.001*	14.82	5.79	0.86
	x_2	0.31	0.16	1.93	0.125			
	x_1 by x_2	-0.71	0.16	-4.47	0.011*			
Acetic acid	Mean	861.69	7.79	110.66	<0.000*	1251.66	9.12	0.99
	Curvature	-224.63	23.79	-9.44	0.011*			
	x_1	542.30	15.57	34.82	0.001*			

	x_2	405.78	15.57	26.06	0.001*			
	x_1 by x_2	573.76	15.57	36.8412	0.001*			
Propanoic acid	Mean	20.99	0.48	43.70	<0.000*	147.52	6.59	0.99
	x_1	15.33	1.27	12.06	0.001*			
	x_2	10.75	1.27	8.46	0.003*			
	x_1 by x_2	13.64	1.27	10.73	0.002*			
2,3-Diethyl-5-methylpyrazine	Mean	5.45	0.31	17.33	<0.000*	12.92	5.79	0.84
	x_1	-2.44	0.83	-2.92	0.043*			
	x_1 by x_2	-2.90	0.83	-3.48	0.025*			
Furfural	Mean	293.01	9.48	30.90	<0.000*	56.09	6.59	0.98
	x_1	163.54	25.09	6.52	0.007*			
	x_2	180.40	25.09	7.19	0.006*			
	x_1 by x_2	141.95	25.09	5.66	0.011*			
5-Hydroxy methylfurfural	Mean	13.05	0.99	13.13	0.006*	163.92	9.12	0.99
	Curvature	-23.42	3.04	-7.71	0.016*			
	x_1	20.82	1.99	10.48	0.009*			
	x_2	21.64	1.99	10.89	0.008*			
	x_1 by x_2	24.27	1.99	12.21	0.007*			

* $p \leq 0.05$.

CAPÍTULO 3

STEAM PRESSURE TREATMENT OF *Coffea canephora* DEFECTIVE BEANS: EFFECT ON THE BIOACTIVE COMPOUNDS PROFILE AND ANTIOXIDANT ACTIVITY OF ROASTED COFFEE

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Steam pressure treatment of *Coffea canephora* defective beans: effect on the bioactive compounds profile and antioxidant activity of roasted coffee

Abstract

Steam pressure treatment has been reported as an alternative to improve the volatile profile and sensory quality of *Coffea canephora* beans. However the use of steam process can also affect the bioactive composition profile, by solubilization or thermal degradation of some components. The aim of this article was to evaluate the influence of steam treatment on *Coffea canephora* defective beans regarding the bioactive compounds profile and the antioxidant activity of roasted coffee. Defective crude beans were treated following a full factorial design 2^2 with the variables steam pressure (2 to 8 bar) and process time (3 to 29 min). The steamed coffees were dried in a vacuum oven, subjected to a medium roasting process (standardized by color) and ground. The steam treatment significantly affects ($p < 0.05$) the contents of caffeine, trigonelline, chlorogenic acids, melanoidins, cafestol and 16-O-methylcafestol on the roasted coffee. In contrast, kahweol content and antioxidant activity were not influenced by steam treatment conditions ($p > 0.05$). SC roasted coffee compounds profile depended on both steam treatment and the subsequent roasting process. In general, the bioactive compounds content in roasted steamed coffee were higher in conditions that combined a more drastic steam treatment (8 bar/29 min) with a shorter roasting process, or a mild steam treatment (2 bar/3 min) with a longer roasting process. Steam treatments with longer times and low pressures should be avoided to prevent the loss of bioactive compounds.

Keywords

Caffeine, chlorogenic acids, diterpenes, trigonelline, liquid chromatography, melanoidins.

3.1 Introduction

A ripe fruit or cherry bean provides a better quality coffee brew. Coffee fruits do not ripen at the same time in a plant, so the harvesting step begins when the majority of fruits are ripe. Harvesting can be performed by picking cherry fruits by hand one by one, in a labor-intensive, time-consuming and expensive process. Coffee can also be harvest by stripping the twigs and collecting all fruits including the ripe, immature, and overripe ones. In addition, fruits that oxidize or ferment after falling on the ground are frequently

collected at the end of harvesting season. Intrinsic defects as black beans, green (immature) and sour coffees - called PVA (from the Portuguese *preto*, *verde* and *ardido*) - usually represents about 15 to 20% of the coffee production on a weight basis - and their presence reduces the cup coffee quality (Farah & Santos, 2015; Franca & Oliveira, 2015; Toledo, Pezza, Pezza, & Toci, 2016; Wei & Tanokura, 2015). Defective coffee beans are usually present in Brazilian coffee due to the strip-picking harvesting and processing practices adopted by the coffee producers.

Brazil is the world leader in the production and exportation of coffee being a heavy producer of both species *Coffea arabica* L. (arabica coffee) and *Coffea canephora* Pierre (robusta coffee). In 2017, it is estimated that Brazil will produce up to 10.1 million of 60 kg bags of *C. canephora* (Companhia Nacional de Abastecimento - CONAB, 2017). *C. canephora* is widely accepted in the market due to its low cost being the main raw material for instant coffee products and is also used in roasted and ground coffees, blended with *C. arabica*. Conversely, *C. canephora* presents lower sensory acceptance compared to *C. arabica*, and the presence of defects can further reduce the cup quality.

Many health benefits associated with moderate and regular consumption of coffee have been related to positive cognitive effects and in memory, antimutagenic action, decrease in bloodstream glucose levels, cardiovascular benefits, neuroprotective and anti-inflammatory activities and cytoprotective effect against oxidative stress, as well as in the reduction of incidence of chronic degenerative diseases as type 2 diabetes, Parkinson's, Alzheimer's, liver impairment (cirrhosis) and some cancers (Amer, Mazen, & Mohamed, 2017; Bedoya-Ramírez, Cilla, Contreras-Calderón, & Alegría-Torán, 2017; Borota et al., 2014; Butt & Sultan, 2011; Frost-Meyer & Logomarsino, 2012; Higdon & Frei, 2006; Martini et al., 2016; Nkondjock, 2009; Preedy, 2015). An inverse association was observed between regular coffee consumption and death due to cardiac disease, respiratory, stroke, injuries and accidents, diabetes and infections (Freedman, Park, Abnet, Hollenbeck, & Sinha, 2012) and with the total risk of mortality (Ding et al., 2015). The benefits promoted on health by coffee intake have been correlated with bioactive compounds such as caffeine, trigonelline, chlorogenic acids, melanoidins and diterpenes. The same compounds have been cited as the main responsible for the antioxidant activity of roasted coffee (Daglia et al., 2004; Kitzberger, Scholz, & Benassi, 2014; Lee, Choi, & Jeong, 2007; Martini et al., 2016; Perrone, Farah, & Donangelo, 2012; Preedy, 2015; Rodrigues, Benassi, & Bragagnolo, 2014; Rosa, Freitas-Silva, Godoy, & Rezende, 2016).

There are patents proposing the use of steam treatment to crude coffee beans to improve *C. canephora* brews quality (Becker, Schlabs, & Ag, 1991; Dar, Bruckmann, & Kelly, 1985; Manfred, 1997). In a preliminary research (Kalschne, Viegas, De Conti, Corso, & Benassi, 2017), it was described a positive impact on the volatile profile and sensory acceptance by using the steam treatment on *C. canephora* PVA beans. However, the use of steam process can also affect the composition profile, by solubilization or thermal degradation of some components. There is a lack of information on literature regarding bioactive composition and antioxidant activity of PVA *Coffea canephora* roasted beans, as well as on the effect of the steam treatment on PVA coffee composition.

The aim of this study was to verify the effect of steam treatment on crude defective *C. canephora* beans (black, immature and source) regarding the bioactive compounds profile and on the antioxidant activity of the roasted coffee.

3.2 Material and Methods

3.2.1 Reagents, standards and equipment

Reagents, solvents and analytical grade materials employed: HPLC grade acetonitrile (J.T. Baker, Phillipsburg, USA); acetic acid (purity ≥ 99.8%, Sigma Aldrich, St. Louis, USA); potassium hydroxide (purity ≥ 85%, Vetec, Duque de Caxias, Brazil); ethanol (purity ≥ 96%, Merck, Darmstadt, Germany); methyl tert-butyl ether (purity ≥ 99%, Acros Organics, Geel, Belgium); ABTS (2,2-azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) (purity ≥ 98%, Sigma Chemical, St. Louis, USA); Trolox (6-hydroxy-2,5,7,8,-tetramethylchromane-2-carboxylic acid) (Sigma Aldrich); potassium persulfate (purity ≥ 99%, Acros Organics); potassium dihydrogen phosphate (purity ≥ 99%, F. Maia, Cotia, Brazil); 0.22 μm nylon filter membrane and 0.22 μm syringe filter (Millipore, São Paulo, Brazil). The following chromatographic standards were used: caffeine, trigonelline, 5-caffeoylquinic acid (5-CQA), and 16-O-methylcafestol (Sigma Aldrich); cafestol; and kahweol (Axxora, San Diego, USA). A Spherisorb ODS-1 column (150 x 4.6 mm, 3 μm) (Waters, Darmstadt, Germany) were also used.

The water used to prepare standards and solutions was obtained through Purelab Option-Q (Elga, High Wycombe, United Kingdom).

Bioactive compounds analyses were carried out in an ultra-high performance liquid chromatography (Ultimate 3000, Thermo Scientific, Germering, Germany), equipped with

an automatic sample injector, quaternary pump, oven and diode array (DAD) detector, and, controlled by Chromeleon 7.0 software. The following equipment were also used: infrared moisture analyzer MB200 (Ohaus, Mumbai, India); colorimeter CR-410 (Konica Minolta, Tokyo, Japan) with standard illuminant C and 10-degree observer; vacuum oven Q819V2 (Quimis, São Paulo, Brazil), Burr bench grinder GVX2 (Krups, Solingen, Germany); Rod-Bel gas pilot roaster (São Paulo, Brazil), and UV-visible spectrophotometer Libra S22 (Biochrom, Cambridge, UK).

3.2.2 Sample preparation

C. canephora beans from the 2014 crop from Rondônia State (Brazil) was subjected to selection on an electronic sorter until approximately 20 kg of rejected coffee was obtained for the study. This material was characterized as 34.9% black, 8.9% immature, 37.9% sour, 15.7% non-defective beans, and 2.6% other (including shells and sticks, excluding stones).

Batches of 1.5 kg of the crude rejected beans were submitted to steam treatment following a full factorial design (FFD) 2^2 with 2 repetitions on the central point, with the content of bioactive compounds and the antioxidant activity of the roasted coffees as responses (Table 3.1). The range in the steam pressure (x_1) (2 to 8 bar) and process time (x_2) (3 to 29 min) applied were defined in a preliminary study (Kalschne et al., 2017).

The steam treatments were performed in a basket consisting of a cylindrical stainless steel container AISI standard (1200 mm height x 60 mm diameter). The container surface has holes of 3.0 mm in diameter, resulting in a 20% area opened for steam passage. The container was placed inside a pressure vessel where the steam was injected from a biomass boiler. The inlet pressure in the vessel was adjusted by a controlling valve installed in the steam pipe and a manometer recorded the applied pressure. The temperatures in the top of the vessel ranged from approximately 133 °C for 2 bar to around 175 °C for 8 bar. An untreated sample was maintained as a control.

The control had a moisture content of 8.6 ± 0.1 g 100 g^{-1}. After the steam treatment, all samples presented high moisture (from 32.0 to 49.8 g 100 g^{-1}), so the steamed coffee beans were previously dried in vacuum oven at 70 °C until reaching the moisture of the control (average moisture of 8.8 ± 0.8 g 100 g^{-1}). Subsequently, of each dry steamed coffee (SC) (100 g) was subjected to a roasting process at approximately 230 °C (Mendes, Menezes, & Silva, 2001). The roasting process time was standardized based on color

parameters (L* value) to obtain a medium-dark roast degree. The roasting time varied between 5 min (SC 4) and 14 min (SC 1); the control was roasted for 14 min 30 sec.

The roasted coffees were middle ground just before each analysis, whenever necessary it was and stored in plastic flasks at −18 °C until analysis. Roasted and ground steamed coffees (SC) had an average L* of 26.4 ± 1.1 and a moisture content of 1.5 ± 1.2 g 100 g^{-1}.

Additional information on the steam treatment equipment and characteristics of color, weight loss and moisture content of each SC is described in Kalschne et al. (2017).

3.2.3 Determination of bioactive compounds

The simultaneous analysis of trigonelline, caffeine and total chlorogenic acids was performed according to Alves, Dias, Benassi, & Scholz (2006). Samples (0.5000 g) were extracted with 30 mL of water at 80 °C for 10 min and filtered; the same aqueous extract was used for melanoidins and antioxidant activity estimation. An aliquot of 1 mL of the extract was diluted in 5.0 mL of acetic acid:water (5:95 v/v). The extract was filtered and injected (20 μL) into the chromatograph. Triplicate independent extractions were performed. The mobile phase was composed of acetic acid/ultrapure water (5:95 v/v) (A) and acetonitrile (B), and the following gradient elution was used: 1 min, 5% B; 5 min, 13% B; flow rate of 0.6 mL min^{-1}. Detection was set at 260 nm for trigonelline, 272 nm for caffeine and 320 nm for chlorogenic acids.

A direct hot saponification procedure was employed for diterpenes analyses, following Dias et al. (2010) and Mori et al. (2016). Samples (0.2000 g) were saponified with 2.0 mL of potassium hydroxide (2.5 mol L^{-1}) in ethanol (96% v/v) in a water bath at 80 °C for 1 h. For the extraction of the unsaponifiable matter, 2.0 mL of methyl tert-butyl ether were added. After stirring and centrifugation (2 min at 3000 rpm and room temperature), the organic phase was collected. The last step was repeated three times. Ultrapure water (2.0 mL) was added for cleaning and the organic extract was collected and evaporated to dryness in a water bath (at 80 °C for 2 h). After resuspension in 4 mL of the mobile phase, the extract was filtered and injected (20 μL) into the chromatograph. Triplicate independent extractions were performed. An isocratic elution with acetonitrile/ water (55:45 v/v) at a flow rate of 0.9 ml min^{-1} was applied. Detection was set at 230 nm, for cafestol and 16-O-methylcafestol, and at 290 nm for kahweol.

For UPLC analysis, the identification of the compounds was based on the retention times and UV spectra. Quantification was performed by external standardization using 6-point analytical curves with triplicate measurements ($R^2 \geq 0.999$ and $p < 0.001$). The concentration ranges for each compound were: 1 to 15 μg mL^{-1} for trigonelline; 20 to 60 μg mL^{-1} for caffeine; 0,5 to 30 μg mL^{-1} for 5-CQA; 40 to 200 ug mL^{-1} for cafestol; 2 to 80 ug mL^{-1} for kahweol and 8 to 120 ug mL^{-1} for 16-O-methylcafestol. The total chlorogenic acids (CGA) content was estimated by the sum of areas of detected compounds at 320 nm based on Corso, Vignoli, & Benassi (2016), using 5-CQA as a standard for quantification. All results were expressed on dry base as mg of a compound 100 g^{-1} of coffee.

The melanoidins were estimated by diluting 0.6 mL of the aqueous extract (used for hydrosoluble compounds analysis) in 1.4 mL of ultrapure water to achieve a concentration of 5 mg of coffee mL^{-1}. The analyses were carried out in genuine triplicate. Absorbance was measured at 420 nm (Almeida & Benassi, 2011) and results were expressed in AU.

3.2.4 Antioxidant activity

The ABTS radical scavenging activity was estimated according to Vignoli et al. (2014). ABTS radical cations (ABTS+•) were produced by reacting 7 mmol L^{-1} of ABTS stock solution with 2.45 mmol L^{-1} of potassium persulfate. The mixture stood in a dark flask at room temperature for 12-16 h before use. The ABTS+• solution was diluted with phosphate buffered saline (pH 7.4) to an absorbance of 0.700 ± 0.002 at 730 nm. SC aqueous extracts (detailed previously) or Trolox standard (10 μL) was added to 4 mL of the diluted ABTS+• solution. Readings were taken at 730 nm after a reaction time of 6 min. A 6-point calibration curve with triplicate measurements in the concentration range from 1 to 8 μmol L^{-1} was used. The results were expressed in g of Trolox equivalent antioxidant capacity (TEAC) 100 g^{-1} of coffee.

3.2.5 Statistical analysis

The contents of the bioactive compounds and the antioxidant activity (AA) on the SC obtained through the factorial design (Table 3.1) were analyzed by Experimental Design procedure of Statistica 8.0 software. The adequacy of the models generated by the factorial design was assessed, compounds with significant effects ($p \leq 0.05$) and $R^2 \geq 0.87$ were considered (Table 3.3). In order to optimize the responses of the factorial design, the global desirability function was determined using the Response Desirability Profiling

procedure. The goal was to maximize the content of the bioactive compounds and AA. The overall desirability value varied from 0, outside an acceptable region, to 1, where the response is at its goal or target. The higher value indicated the more desirable treatment, which is considered as the optimal solution.

3.3 Results and discussions

3.3.1 Composition profile and antioxidant activity of roasted coffees

Table 3.1 shows the content of bioactive compounds and the antioxidant activity for the SC coffees compared to the control (untreated).

The content of water-soluble bioactive compounds were lower in SC coffees compared to the control: caffeine varied from 863 to 1385 mg 100 g^{-1} (1694 mg 100 g^{-1} in the control); trigonelline, from 186 to 368 mg 100 g^{-1} (393 mg 100 g^{-1} in the control); and melanoidin, from 0.403 to 0.567 AU (0.608 UA in the control) (Table 3.1). An exception occurred with the total chlorogenic acids (CGA) content which varied on a wide range of values for SC, from 1221 to 2445 mg 100 g^{-1}, being the control content in this range (2024 mg 100 g^{-1}) (Table 3.1).

The compounds of coffee unsaponifiable fraction showed lower variations among treatments (Table 3.1). Kahweol and cafestol contents in the control (16.2 mg 100 g^{-1} and 491 mg 100 g^{-1}, respectively) were within the range observed for the different SC: between 10.3 and 33.1 mg of kahweol 100 g^{-1} and between 396 and 497 mg of cafestol 100 g^{-1}. 16-O-methylcafestol was present in SC in lower contents (from 76 to 103 mg 100 g^{-1}) compared to that observed for the control (120.2 mg 100 g^{-1}).

Antioxidant activity varied between 6991 and 10592 mg TEAC 100 g^{-1} in the SC, with lower values than that observed for the control (11150 mg TEAC 100 g^{-1}) (Table 3.1).

No data were reported by the literature regarding the composition of bioactive and antioxidant activity for roasted PVA *C. canephora*; in general, data availability for the composition of *C. canephora* is highly limited, notably for roasted products. Roasted *C. canephora* coffee from different origins and several degrees of roasting showed caffeine contents between 1567 and 2690 mg 100 g^{-1} (Casal, Oliveira, & Ferreira, 2000; Daglia, Cuzzoni, & Dacarrot, 1994; Hečimović, Belščak-Cvitanović, Horžić, & Komes, 2011; Perrone, Donangelo, & Farah, 2008a; Ramalakshmi, Rao, Takano-Ishikawa, & Goto, 2009; Silva et al., 2015; Souza & Benassi, 2012); trigonelline contents between 70 and 991

mg 100 g^{-1} (Casal et al., 2000; Daglia et al., 1994; Perrone et al., 2008a; Souza & Benassi, 2012); CGA contents between 543 and 7946 mg 100 g^{-1} (Hečimović et al., 2011; Perrone, Farah, Donangelo, Paulis, & Martin, 2008b; Ramalakshmi et al., 2009); kahweol contents between 0 and 20 mg 100 g^{-1} (Campanha, Dias, & Benassi, 2010; Finotello et al., 2017; Lercker et al., 1996; Mori et al., 2016; Souza & Benassi, 2012); cafestol contents between 76 and 360 mg 100 g^{-1} (Campanha et al., 2010; Dias, Faria-Machado, Mercadante, Bragagnolo, & Benassi, 2014; Finotello et al., 2017; Lercker et al., 1996; Mori et al., 2016; Schievano, Finotello, De Angelis, Mammi, & Navarini, 2014; Souza & Benassi, 2012); and 16-O-methylcafestol contents between 26 and 224 mg 100 g^{-1} (Finotello et al., 2017; Mori et al., 2016). For *C. canephora* antioxidant activity, values between 4255 and 11263 mg TEAC 100 g^{-1} (equivalent to 17 and 27 m mol TEAC/L) were reported (Hečimović et al., 2011; Ludwig et al., 2012; Ludwig, Sánchez, Peña, & Cid, 2014).

For commercial coffees (usually blends of *C. arabica* and *C. canephora*) the following compounds content were reported: 1050 to 2417 mg of caffeine 100 g^{-1} (Bartel, Mesias, & Morales, 2015; López-Galilea, De Peña, & Cid, 2008; Perrone et al., 2008a; Ribeiro, Leitão, Ramalho, & Lidon, 2014; Souza, Canuto, Dias, & Benassi, 2010), 220 to 740 mg of trigonelline 100 g^{-1} (López-Galilea et al., 2008; Perrone et al., 2008a; Ribeiro et al., 2014; Souza et al., 2010), 347 to 1594 mg of CGA 100 g^{-1} (Bartel et al., 2015; Monteiro & Trugo, 2005; Ribeiro et al., 2014), values between 0.349 and 0.690 AU for melanoidin (Almeida & Benassi, 2011), 100 to 780 mg of kahweol 100 g^{-1} (Campanha et al., 2010; Dias, Trindade, & Benassi, 2013; Souza et al., 2010), 247 to 490 mg of cafestol 100 g^{-1} (Campanha et al., 2010; Souza et al., 2010); and 5 to 131 mg of 16-O-methylcafestol 100 g^{-1} (Schievano et al., 2014). Antioxidant activity values between 2960 and 10938 mg TEAC 100 g^{-1} were described (Almeida & Benassi, 2011; Bartel et al., 2015; Cämmerer & Kroh, 2006; Contreras-Calderón et al., 2016).

Literature data comparison shows that, in general, bioactive compounds contents and antioxidant activity (Table 3.1) of the SC coffees (even considering the variations among treatments) are in the range described for *C. canephora* roasted coffees and commercial roasted coffees. This shows that although the raw material included defective crude coffees beans and the addition of extra stages to the process – steam treatment and drying – the composition profile of SC coffees showed no difference comparing to the regular roasted ones.

Table 3.1 – Full Factorial Design 2^2 Matrix with real and codified variables and content of bioactive compounds and antioxidant activity of roasted coffees.

Run/SC	Steam pressure (x_1) (bar)	Process time (x_2) (min)	Caffeine*	Trigonelline*	Chlorogenic Acids*	Melanoidins**	Kahweol*	Cafestol*	16-O-methylcafestol*	Antioxidant activity***
SC 1	-1 (2)	-1 (3)	1385 ± 25	365 ± 11	2042 ± 33	0.567 ± 0.002	9.7 ± 0.4	451 ± 11	93 ± 3	10592 ± 135
SC 2	+1 (8)	-1 (3)	1280 ± 37	342 ± 7	1773 ± 40	0.570 ± 0.003	10.3 ± 0.1	451 ± 1	98 ± 1	9618 ± 149
SC 3	-1 (2)	+1 (29)	863 ± 33	186 ± 3	1221 ± 44	0.403 ± 0.003	14.5 ± 0.2	396 ± 3	76 ± 0	6991 ± 83
SC 4	+1 (8)	+1 (29)	1075 ± 10	368 ± 12	2445 ± 22	0.530 ± 0.004	33.1 ± 0.5	497 ± 7	103 ± 1	9162 ± 137
SC 5	0 (5)	0 (16)	998 ± 15	286 ± 6	1628 ± 7	0.453 ± 0.003	11.6 ± 0.2	454 ± 10	93 ± 1	8037 ± 63
SC 6	0 (5)	0 (16)	1070 ± 9	299 ± 3	1705 ± 4	0.459 ± 0.002	14.9 ±0.3	464 ± 11	92 ± 1	8504 ± 32
Control (untreated)			1694 ± 34	393 ± 1	2024 ± 40	0.608 ± 0.004	16.2 ± 0.3	491 ± 8	120 ± 1	11150 ± 52

Mean ± standard deviation, expressed as * mg 100 g^{-1} (dry basis), **AU at 420 nm, *** mg TEAC 100 g^{-1} (dry basis).

3.3.2 Effect of steam treatment on the content of bioactive compounds and antioxidant activity of roasted coffees

The steam treatment significantly affected ($p \leq 0.05$) the content of the bioactive compounds studied (Table 3.2), except kahweol ($p > 0.05$), but had no significant impact on the AA of SC coffees ($p > 0.10$). The literature reports that, for different treatments in coffee products (roasting process and aqueous extraction with high temperature in the production of instant coffee), composition profile changes are more relevant than those observed in the antioxidant activity (Corso et al., 2016; Vignoli, Bassolli, & Benassi, 2011; Vignoli et al., 2014).

Valid linear models (with $R^2 \geq 0.87$ and adjusted $R^2 \geq 0.78$) were obtained for all the compounds significantly affected by steam treatment (Table 3.3). Non-significant effects were incorporated into the residue of the models, except in cases in which the effect influenced negatively the adjusted R^2. Corresponding surface responses are shown in Figure 3.1.

Table 3.2 – Effects of the factors studied on FFD for the response bioactive compounds.

Bioactive compounds	Factor	Effect	Standard error	t test	p-Value
Caffeine (mg 100 g^{-1})	Mean	1111.8	36.2	30.7	<0.000*
	x_2	-363.5	88.7	-4.1	0.026*
	x_1 by x_2	158.5	88.7	1.8	0.172
Trigonelline (mg 100 g^{-1})	Mean	307.7	8.0	38.3	0.001*
	x_1	79.5	19.7	4.0	0.056
	x_2	-76.5	19.7	-3.9	0.060
	x_1 by x_2	102.5	19.7	5.2	0.035*
CGA (mg 100 g^{-1})	Mean	1802.3	59.6	30.3	<0.000*
	x_1	477.5	145.9	3.3	0.047*
	x_1 by x_2	746.5	145.9	5.1	0.014*
Melanoidins (AU)	Mean	0.518	0.002	243.952	0.003*
	Curvature	-0.123	0.007	-16.738	0.038*
	x_1	0.065	0.004	15.321	0.041*
	x_2	-0.102	0.004	-24.042	0.026*
	x_1 by x_2	0.062	0.004	14.614	0.043*
Cafestol (mg 100 g^{-1})	Mean	452.2	3.4	132.3	<0.000*
	x_1	50.5	8.4	6.0	0.009*
	x_1 by x_2	50.5	8.4	6.0	0.009*
16-O-methylcafestol (mg 100 g^{-1})	Mean	92.4	0.3	322.3	<0.000*
	x_1	15.6	0.7	22.1	0.002*
	x_2	-6.4	0.7	-9.0	0.012*
	x_1 by x_2	11.3	0.7	16.0	0.004*

* $p \leq 0.05$; x_1 = steam pressure (bar), x_2 = process time (min).

Table 3.3 – Anova of linear models for the prediction of the bioactive compounds.

Bioactive compounds	Source of variation	Sum of squares	Degrees of freedom	Mean square	$F_{calculated}$	$F_{tabulated}$	p-Value	R^2	R^2(adj)
Caffeine (mg 100 g^{-1})	Regression	157254.5	2	78627.3	10.0	9.6	0.047*	0.87	0.78
	Residual	23628.3	3	7876.1					
	Total	180882.8	5						
Trigonelline (mg 100 g^{-1})	Regression	22678.8	3	7559.6	19.5	19.2	0.049*	0.97	0.92
	Residual	774.6	2	387.3					
	Total	23453.3	5						
Total of CQA (mg 100 g^{-1})	Regression	785268.5	2	392634.3	18.4	9.6	0.021*	0.92	0.87
	Residual	63866.8	3	21288.9					
	Total	849135.3	5						
Melanoidins (AU)	Regression	0.024	4	0.006	326.6	224.6	0.041*	0.99	0.99
	Residual	0.000	1	0.000					
	Total	0.024	5						
Cafestol (mg 100 g^{-1})	Regression	5100.5	2	2550.3	36.4	9.6	0.008*	0.96	0.93
	Residual	210.3	3	70.1					
	Total	5310.8	5						
16-O-methylcafestol (mg 100 g^{-1})	Regression	408.7	3	136.2	276.4	19.2	0.004*	0.99	0.99
	Residual	1.0	2	0.5					
	Total	409.7	5						

* $p \leq 0.05$; x_1 = steam pressure (bar), x_2 = time (min).

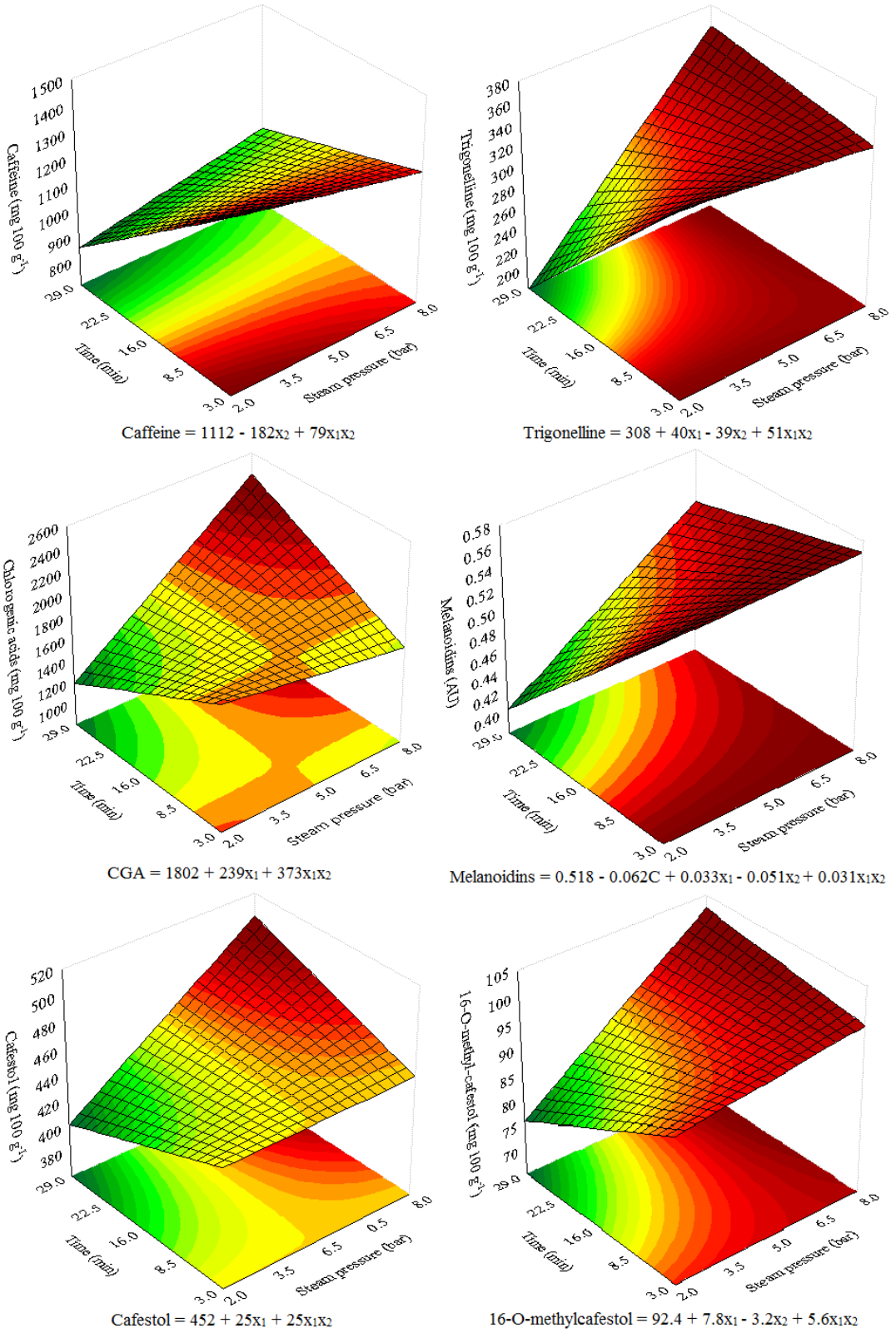

Figure 3.1 – Response surface plot of bioactive compounds.

x_1 = steam pressure (bar), x_2 = process time (min), C = 1 (central point, for x_1 and x_2 = 0) or C = 0 (other points).

It is important to emphasize that the effects discussed next are based on the models developed from composition data of the steamed roasted coffees, so the final content of each compound corresponds to the sum of the effects of the steam treatment and the subsequent roasting process. According to the described in sample preparation, as the roasting degree was standardized by the L* value, the most drastic process regarding the steam treatment (SC 4, 8 bar/29 min) took less time (5 min) to reach the desired roasting degree compared to the longer time (14 min) taken by the run SC 1 (2 bar/3 min). Intermediate roasting times were used for other design conditions.

Caffeine showed a different response to the treatment compared to the other compounds (Figure 3.1), being notably negatively affected by process time (x_2) (Table 3.2 and Figure 3.1). Similarly, De Conti (2013) reported the reduction in caffeine content in steamed coffees. Considering the thermal stability of caffeine (Dias & Benassi, 2015; Rosa et al., 2016; Vignoli et al., 2014), the reduction in the content of this bioactive was attributed to leaching. Caffeine shows water solubility of around 21 to 22 g L^{-1} (Merck, 2001; National Center for Biotechnology Information - NCBI, 2017); so probably due to the long contact with the steam associated to the high pressure (positive interaction between x_1 and x_2, Table 3.2), part of the caffeine was gradually solubilized and leached. The loss in steamed coffees is similar to what occurs in water decaffeination, in which caffeine leaching is the process that allows the production of decaffeinated coffee (Heilmann, 2001).

For other two bioactive compounds with different characteristics - trigonelline (hydrosoluble compound with low thermal stability) and melanoidins (developed in the thermal process) – it was observed similarity in the models and surfaces obtained: negative effect of the process time (x_2) and positive effects of the steam pressure (x_1) and x_1x_2 interaction (Table 3.2). The worst treatment condition for these bioactive would be working under low pressures at longer times. Both the maintenance of trigonelline content as well as the development of melanoidins were favored by the increase in pressure and reduction in process time in steam treatment.

Considering that the trigonelline is partially degraded with thermal treatments (Dias & Benassi, 2015; Rosa et al., 2016; Vignoli et al., 2014) and shows high solubility - around 1000 g/L (NCBI, 2017) – the effect observed could be explained by the two processes applied - steam treatment and roasting process. As the roasting degree was standardized by L*, more drastic steam processes (with temperatures reaching 175 °C) were followed by

roasting processes (in which higher temperatures around 230 °C, were applied) at shorter times, allowing for a good retention of this bioactive. The steam process at a short time and low pressure (followed by a longer roasting process) also had a positive effect on the maintenance of trigonelline (Table 3.2 and Figure 3.1).

The development of melanoidins was more expressive with a short steam process, increasing as pressure increased (Table 3.2). As short-time steamed coffees, regardless of the pressure, were roasted for long times, it seems that the roasting process had more impact on melanoidin content than the steam treatment. Compared to the steam treatment (2 bar ≅ 133 °C; 8 bar ≅ 175 °C), the low water activity and high temperature (230 °C) during roasting favored the development of the Maillard reaction, justifying the greater development of melanoidins.

CGA (hydrosoluble compounds with low thermal stability) and the liposoluble diterpenes cafestol and 16-O-methylcafestol showed similar models, with positive effects of steam pressure (x_1) and x_1x_2 interaction. With a lower impact, it was also observed for 16-O-methylcafestol an additional negative effect of the process time (x_2) (Table 3.2). Thus, the maintenance of these compounds was favored by the increase in pressure and steam processing time.

Compared to the other hydrosoluble bioactive studied (caffeine and trigonelline), the CGA show lower solubility, 40 mg L^{-1}. The CGA also stand out for their low thermal stability, notably in the *C. canephora* matrix (Dias & Benassi, 2015; Vignoli et al., 2014). During roasting, CGA participate in the Maillard reaction, degrading and becoming part of the melanoidins (Rosa et al., 2016). Greater CGA preservation occurred under the most drastic steam process conditions (8 bar/29 min) combined with a shorter roasting process, since the roasting process had greater impact in the CGA reduction than the steam treatment. It can be attested by observing that, contrary to other hydrosoluble bioactive, CGA contents in some SC were similar or superior to the control (Table 3.1), which was not steam treated and so it was subjected to a longer roasting process (14 min 30 sec).

Good retention of the diterpenes cafestol and 16-O-methylcafestol was observed in the SC coffees (Table 3.1), due to the low solubility and relative thermal stability of these compounds (Dias et al., 2014). Higher pressures and longer process time favored retention; higher contents of these diterpenes were observed under the most drastic processing conditions (8 bar/29 min) (Table 3.2). No significant effects of the treatment were observed for kahweol in the range studied; however higher concentration (33.1 mg 100 g^{-1})

was also got under the most drastic condition (8 bar/29 min), reinforcing the behavior observed for the other diterpenes in the present study (Table 3.1). According to the literature, coffee lipids content increases with roasting process proportionally to the reduction in the content of thermolabile components in the matrix (Dias et al., 2014; Rosa et al., 2016). Taking into account the greater thermal stability of the diterpenes compared to the other bioactive studied and the fact that they do not solubilize, the maintenance or increase in their contents with drastic steam treatment may be related to this proportional increase in lipid fraction.

Considering the set of bioactive studied, the desirability profiling was evaluated to maximize the content of bioactive compounds in roasted SC coffees**.**

By desirability results, steam treatments with longer times and low pressures should be avoided to prevent the loss of bioactive compounds and a consequent lower antioxidant activity (Figure 3.2, Table 3.1).

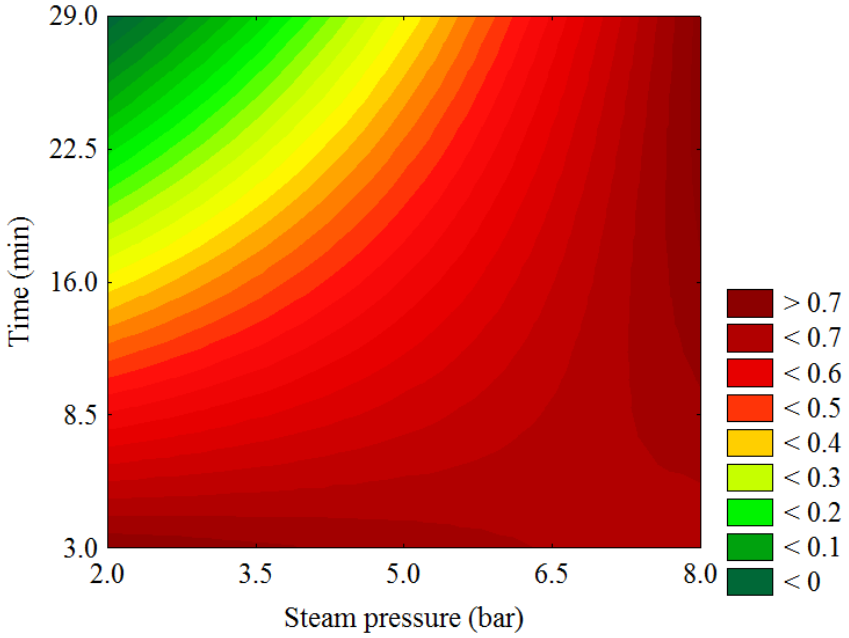

Figure 3.2 – Desirability plot of bioactive compounds.

On the other hand, both conditions a drastic (longer time at high pressure, 8 bar/29 min) or a mild (short time at low pressure, 2 bar/3 min) steam treatments allowed to maintain the bioactive compounds content in roasted PVA coffee (Figure 3.2). We emphasize again that the effects described are related to the composition of SC roasted coffees, so extreme steam treatment conditions (SC 4 e SC 1) was compensated by the

differentiated roasting process to which each SC was submitted. Samples treated under drastic steam conditions demanded shorter roasting process to get the same roasting degree (color-based), due to the previous occurrence of Maillard reactions in the SC. On the other hand, mild steam treatment conditions demanded longer roasting. For many bioactive, there was an indication that the roasting process impact was greater than the steam treatment.

If relevant, the choice of the steam treatment condition may also be based on favoring some specific compound inside the most desirable region (Figure 3.2). The mild steam treatment condition (short time at low pressure) favors caffeine retention while more aggressive processes (long time at high pressure), would be more efficient retaining CGA and diterpenes (Figure 3.1).

3.4 Conclusions

Caffeine, trigonelline, chlorogenic acid, melanoidin, cafestol and 16-O-methylcafestol bioactive contents in roasted SC coffee were affected by steam treatment conditions; however, no significant effect was observed for the antioxidant activity. SC roasted coffee compounds profile depended on both steam treatment and the subsequent roasting process. In the range of steam pressure and process time studied, the content of bioactive compounds in roasted steamed coffee was higher in conditions that combined the most drastic steam treatment (8 bar/29 min) followed by a short roasting process, or the milder steam treatment (2 bar/3 min), followed by a longer roasting process. Steam treatments with longer times and low pressures should be avoided to prevent the loss of bioactive compounds.

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CAPÍTULO 4

SENSORY CHARACTERIZATION AND ACCEPTANCE OF COFFEE BREWS OF *C. arabica* AND *C. canephora* BLENDED WITH STEAMED DEFECTIVE COFFEE.

(Artigo a ser submetido para avaliação na revista LWT- Food Science and Technology).

Sensory characterization and acceptance of coffee brews of *C. arabica* and *C. canephora* blended with steamed defective coffee.

Abstract

Steam treatment has been reported as an alternative to improve the cup quality of coffee; in this research, it was applied to *C. canephora* defective coffee. The aim of the study was to evaluate the sensory perception of steamed defective *C. canephora* coffee (SDC) in roasted coffee blends, by Flash Profile and acceptance tests. SDC was produced by steam treatment (5 bar, 16 min) and a standardized roasting process was applied to all coffees. Four samples were prepared as follows: AB (100% *C. arabica*), CB (100% *C. canephora*), ASDB and CSDB, both blends with 50% of SDC and *C. arabica* or *C. canephora*, respectively. Coffee brews were prepared through percolation (50 g coffee/ 500 mL water at 92 °C). Coffee species were more relevant in sensory discrimination of the brews than SDC addition. AB and ASDB were characterized as having brown color, fruity/herbal/green bean aroma and coffee/residual coffee flavor. CB and CSDB were described as viscous, with foam, black color, bitter taste, and aroma/taste related to the roasting process. With SDC addition, typical sensory characteristics of each species were maintained, but the intensity of the attributes was reduced. Coffee brews blends with 50% SDC *C. canephora* are well accepted.

Keywords: PVA beans, Flash Profile, acceptance, Preference Mapping, robusta.

4.1 Introduction

Brazil is the world's largest coffee producer and exporter, producing the two most important commercial species. In 2017, it will produce up to 35.4 million bags (60 kg) of *C. arabica* and up to 10.1 million bags of *C. canephora* (CONAB, 2017). Defective coffee beans represent about 15 to 20% of the total coffee produced in Brazil due mainly to strip-picking harvesting conditions applied. The presence of defective beans reduces the cup quality and the price of the product marketed in Brazil and limit the exportations (Farah & Santos, 2015; Franca & Oliveira, 2015; Toledo, Pezza, Pezza, & Toci, 2016; Wei & Tanokura, 2015).

The PVA coffee beans, from the Portuguese "preto" (black beans), "verde" (green or immature beans) and "ardido" (sour beans) are the main intrinsic defects related to the

coffee cup quality decrease. The presence of black beans is usually associated with bitter taste and unclean and pungent flavors. Sour beans contribute to sour taste and oniony flavor, and immature beans impart astringency to the coffee brew (Franca & Oliveira, 2015; Mendonça, Franca, & Oliveira, 2009).

Some patents described the use of steam treatment to raw coffee beans to improve *C. canephora* brews quality (Becker, Schlabs, & Ag, 1991; Dar, Bruckmann, & Kelly, 1985; Manfred, 1997). Similarly, Kalschne, Viegas, De Conti, Corso, and Benassi (2017a) described a positive impact on the volatile profile and sensory acceptance by using steam treatment (5 bar/16 min) on *C. canephora* PVA beans. The authors described that the steam treatment could affect (by solubilization and/or thermal degradation) the content of some bioactive compounds (caffeine, trigonelline, chlorogenic acids, melanoidins, and diterpenes) that are also important in coffee flavor (Kalschne, Viegas, Conti, Corso, & Benassi, 2017b). Thus, the changes in the volatile profile and composition of PVA with steam treatment could provide brews with singular sensory characteristics.

In Brazil, the roasted and ground coffee is used to make the drip/filtered coffee brew, the most popular type of coffee preparation. In 2015 the Brazilian consumption per capita of this coffee was about 81 L, corresponding to 819 mil ton of roasted coffee, and it is estimated for 2019 an increase up to 906 mil ton (ABIC, 2015, 2016).

Considering the expressive consumption of roasted and ground coffee and the unavoidable presence of defective coffee beans in Brazilian products, the steam treatment of PVA beans is a possibility to improve the sensory quality of the coffee. Regarding the sensory techniques, the Flash Profile is a descriptive analysis that combines the survey of attributes of the Free Profile with the use of a ranking procedure providing a rapid description and discrimination of a set of samples (Kobayashi & Benassi, 2012; Terhaag & Benassi, 2010). In a different approach, on acceptance tests, the consumers indicate which sample or set of samples were accepted by them (Lawless & Heymann, 2010). Analyzing the results jointly, the most/least accepted sample could be identified by the acceptance test while the Flash Profile could define the attributes that characterize each sample.

Although sensory characteristics of *C. arabica* brews are well described in the literature, there is a lack of information regarding coffee brews produced with *C. canephora* or PVA coffee (from *C. arabica* or *C. canephora*). In a preliminary study, it was observed that up to 45% of steamed PVA coffee could be added to *C. arabica* coffee without causing sensory rejection (Kalschne et al., 2017a).

The aim of this study was to evaluate the sensory perception of steamed defective *C. canephora* coffee analyzing coffee brews of *C. arabica* and *C. canephora* blended with 50% of SDC. In addition, pure *C. arabica* and *C. canephora* brews were evaluated for comparison; all the coffee brews were characterized by Flash Profile, and the acceptance was also evaluated.

4.2 Materials and Methods

4.2.1 Coffees

To obtain defective beans for the study, *C. canephora* raw coffee from the 2014 crop from Rondônia State (Brazil) was subjected to selection on an electronic sorter. The raw material was characterized as 34.9% black, 8.9% immature, 37.9% sour, 15.7% non-defective beans, and 2.6% other (including shells and sticks, excluding stones). Three batches of 1.5 kg of the rejected beans (81.7% PVA beans) were submitted to steam treatment at 5 bar for 16 min, previously defined as the best condition to improve the volatile profile of the SDC coffee, maximizing the contents of volatiles with positive impact and minimizing those with negative impact (Kalschne et al., 2017a). The equipment and the steam treatment employed were described in detail by (Kalschne et al., 2017a). The steamed defective coffee (SDC) was dried in vacuum oven (Q819V2, Quimis, São Paulo, Brazil) at 70 °C until moisture reached 8.8 ± 0.6 g 100 g^{-1}.

A high quality *C. arabica* coffee (NY 2) (AC) from 2015 crop from Paraná State (Brazil) and a high quality *C. canephora* (neutral brew, very light robusta flavor) (CC) from 2015 crop from Rondônia State (Brazil) were also used.

All coffees were subjected to a medium roasting process (Rod Bel, São Paulo, Brazil) at approximately 230 °C, suggested as optimal for *C. canephora* (Mendes, De Menezes & Silva, 2001). The roasted coffees were subsequently middle ground (0.7% retained in sieve size 1.18 mm; 73% retained in sieve size 0.60 mm, and 20% passing a sieve size 0.60 mm) in a Burr Grinder GVX2 (Krups, Solingen, Germany). The roasted coffees were ground just before each analysis and coffee brews preparation, whenever necessary it was stored in plastic flasks at −18 °C.

The length of the roasting process was standardized based on L* value of roasted and ground coffee, obtaining 26.5 ± 0.2 for AC, 26.1 ± 0.1 for CC, and 26.2 ± 0.0 for SDC. The roasting weight loss was 15.4 ± 0.5% for AC, 17.0 ± 1.1% for CC, and 13.4 ± 1.3 for

SDC. A final moisture of 1.2 ± 0.0 g 100 g^{-1} for AC, 0.9 ± 0.1 g 100 g^{-1} for CC, and 1.7 ± 0.1 g 100 g^{-1} for SDC, were observed.

4.2.2 Coffee brews preparation

Four coffee brews were studied as follows:

- AB: a pure arabica coffee brew using 100% of *C. arabica* (AC);

- ASDB; a blended coffee brew using 50% of AC and 50% of steamed defective coffee (SDC);

- CB: a pure robusta coffee brew using 100% of *C. canephora* (CC);

- CSDB: a blended coffee brew using 50% of CC and 50% of SDC.

The coffee brews were prepared through percolation with Melitta 102 paper filters (Guaíba, Brazil) following the proportion of 50 g of roasted and ground coffee for 500 mL of mineral water (Ouro Fino, Campo Largo, Brazil) at 92 °C. No sugar was added to the coffee brews for the Flash Profile analysis. For the acceptance evaluation, the coffee brews were sweetened with 6% of sucrose (Alto Alegre, Presidente Prudente, Brazil), the amount of sucrose was defined in preliminary tests (data not shown). The coffee brews were stored in thermal bottles and kept for up to 2 h, until being served at 70 ºC (Corso, Vignoli & Benassi, 2016).

4.2.3 Coffee brews characterization

Caffeine, trigonelline, and 5-caffeoylquinic acid (5-CQA) were determined by ultra-high performance liquid chromatographic as previously described by Kalschne et al. (2017b). The coffee brews were diluted (1 μL) in 10 mL of 5% acetic acid solution (v/v) and filtered (0.22 μm syringe filter, Millipore, São Paulo, Brazil). An aliquot of 20 μL was injected into the liquid chromatograph (Ultimate 3000, Thermo Scientific, Germering, Germany) equipped with an automatic sample injector, quaternary pump, column oven, and diode array detector, controlled by Chromeleon 7.0 software. A Spherisorb ODS-1 column (150 x 4.6 mm, 3 μm) (Waters, Darmstadt, Germany) was employed. A mobile phase of acetic acid/ultrapure water (5:95 v/v) (A) and acetonitrile (B) was used following the gradient elution: 1 min 5% B; 6 min 13% B, with a flow rate of 0.6 mL min^{-1}. Detection was set at 260 nm for trigonelline, 272 nm for caffeine and 320 nm for 5-CQA. Analyses were carried out at 25 °C in triplicate. The identification of the compounds was based on the retention times and UV spectra. Quantification was performed by external

standardization using 6-point analytical curves with triplicate measurements ($R^2 \geq 0.999$ and $p < 0.001$). The concentration ranges from 1 to 15 μg mL^{-1} for trigonelline; 12 to 38 μg.mL^{-1} for caffeine; and 0.5 to 30 μg.mL^{-1} for 5-CQA. The results were expressed in mg of compound mL^{-1} of coffee brew.

Melanoidins were analyzed by diluting 200 μL of the coffee brew in 3.8 mL of ultrapure water to achieve a concentration of 5 mg of coffee mL^{-1}. Samples were read in a spectrophotometer (Perkin Elmer, Lambda XLS, Beaconsfield, UK) at a wavelength of 420 nm (Almeida & Benassi, 2011). The melanoidins content was estimated based on the absorptivity value of 1.1289 L g^{-1} cm^{-1} proposed by Tagliazucchi, Verzelloni & Conte (2010). A coffee brew was used as a source of melanoidins to obtain an analytical curve (7 points in triplicate, $R^2 = 1$) in the concentration from 1 to 7 mg of coffee mL^{-1}, corresponding to an absorbance range of 0.092 to 1.001 AU. The analysis was performed in triplicate, and the results were expressed in mg of melanoidins mL^{-1} of coffee brew.

The total soluble solids content was determined in triplicate using a pocket refractometer (Atago PAL-BX/RI, Tokyo, Japan) with automatic temperature compensation. The results were expressed as ºBrix.

The pH was determined directly in coffee brews, in triplicate, using a pHmeter (21 pH/mv meter, Hanna, São Paulo, Brazil).

The titratable acidity was determined according to Kobayashi & Benassi (2012). The coffee brews (20 mL) were diluted in distilled water (20 mL), and titration was carried out with NaOH 0.1 M until pH 8.2. The analysis was performed in triplicate and results were expressed as mL de NaOH 0.1 M in 20 mL of coffee brew.

The data were analyzed by one-way ANOVA and Tukey test ($p \leq 0.05$) using the software Statistica 8.0 (Tulsa, USA).

4.2.4 Sensory analysis

All participants were coffee consumers, and before the trials, they answered a self-administered questionnaire on socio-demographic data and consumption habits. Tests were conducted in a sensory analysis laboratory, in individual booths, under white light. Coffee brews, approximately 50 mL, were served at 70 ºC, in styrofoam cups (100 mL) codified with three randomized digits. The presentation order of the samples was randomized for each assessor. Assessors were required to clean the palate with water before and between

samples. This study was authorized by the Ethics Committee of Universidade Estadual de Londrina (Certificate of Ethical Evaluation Presentation 36840214.0.0000.5231).

4.2.5 Flash Profile

Forty coffee consumers (18 females and 22 males) were recruited for the Flash Profile (FP) test. The participants presented diversity in age (60% aged between 18 and 25, 18% between 26 and 35, 10% between 36 and 45 and 12% above 46 years old) and in their educational level (55% completed high school, 12% undergraduate and 33% graduate). All participants were regular coffee consumers: 75% consumed daily, and 25% at least 3 times a week. They usually consumed filtered coffee brews (36%), espresso/gourmet/single-dose capsule (29%), instant coffee (19%) and cappuccino (16%). Most participants declare themselves as consumers of sweetened coffee (55% with sugar and 7% with sweetener).

The test was applied according to Kobayashi & Benassi (2012). The four coffee brews were simultaneously served, and the assessors were requested to record attributes considering the similarities and differences between samples regarding appearance, aroma, flavor, texture and mouthfeel perceptions. After attributes development, an individual score sheet and a corresponding list of attributes definition for each attribute were defined for each assessor. Once again coffee brews were simultaneously presented, and the assessors ordered the samples by the increased intensity of each attribute.

The data were analyzed by the Generalized Procrustes Analysis, using the software Senstools version 2.3.28 (OP & P Product Research, Utrecht, Netherlands). Data were provided as 40 individual matrices (one per assessor) with four lines (samples) and a different number of columns ranging from 5 to 14, according to the number of attributes used by each assessor.

4.2.6 Acceptance test

A panel of 142 coffee consumers (67 females and 75 males) was used. The participants were younger (89% aged between 18 and 25, 9% between 26 and 36 and 2% above 36 years old) and less educated (86% completed high school, 10% were undergraduate, and 4% were graduate) than the FP panel. All participants were coffee consumers: 71% consumed daily, 23% at least 3 times a week, and 6% occasionally. They consumed filtered coffee brews (31%), cappuccino (21%), espresso/gourmet/decaffeinated/single-dose capsule (29%) and instant coffee (19%). The

panel was mainly composed by sweetened coffee consumers (77% with sugar and 7% with sweetener).

A 10-cm hybrid hedonic scale anchored with verbal terms (0=disliked extremely, 5=neither liked, nor disliked, 10=like extremely) (Villanueva, Petenate & Silva, 2005) was used to the assess acceptance of the coffee brews regarding the attributes of color, aroma, texture, flavor, and overall acceptance. The purchase intent was evaluated using a similar 10-cm hybrid hedonic scale anchored with verbal terms (0=definitely not buy, 5=maybe buy/maybe not buy, 10=definitely buy). The assessors were asked to report the most/least appreciated sensory characteristics. The samples were presented monadically according to an experimental design of complete balanced blocks, in a randomized order.

Data were analyzed by two-way ANOVA (considering the samples and consumers as sources of variation) and Tukey test ($p \leq 0.05$) using the software Statistica 8.0. Internal preference mapping was carried out by Multidimensional Scaling and Cluster analysis using the software Senstools version 2.3.28. A Pearson's correlation was employed to correlate sensory acceptance with physico-chemical parameters using the software Statistica 8.0.

4.3 Results and Discussion

4.3.1 Coffee brew characteristics

The analyzed coffee brews showed different characteristics in regards to composition (Table 4.1), in function of the coffee species used and the presence of SDC in blends.

Brews with the presence of *C. arabica* (AB and ASDB) were characterized by higher contents of trigonelline and 5-CQA, and by lower pH and higher acidity (Table 4.1). A positive correlation was observed between 5-CQA and trigonelline contents and the acidity of the coffee brews ($r \geq 0.97$, $p \leq 0.05$) and a negative correlation of these parameters with the pH ($r \geq - 0.95$, $p \leq 0.05$) (Supporting Table 4.3S).

C. canephora brews, CB and CSDB (emphasizing that the SDC of the blend was also originated from *C. canephora*) showed higher caffeine and melanoidin contents (Table 4.1). A positive correlation between caffeine and melanoidin contents were observed ($r \geq 0.95$, $p = 0.05$) (Supporting Table 4.3S). This behavior is in agreement with the literature, in which lower caffeine content and higher contents of trigonelline and 5-CQA are

reported for roasted arabica coffees compared to robusta ones (Dias & Benassi, 2015; Gloess et al., 2014; Vignoli, Viegas, Bassoli, & Benassi, 2014).

Regardless of the coffee species present in the blend, with the addition of SDC (ASDB and CSDB), it was observed lower contents of trigonelline, 5-CQA, melanoidin and total solids as well as lower acidity and higher pH, compared to pure coffee brews (AC, CC) (Table 4.1). This behavior can be explained by the fact that a preliminary study showed that, in function of the steam treatment applied to SDC, there is a partial leaching and/or thermal degradation of these hydrosoluble components (Kalschne et al., 2017b).

As for caffeine, a compound that showed greater diversity in function of coffee species among the compounds studied, the SDC addition effect was dependent on the species: it increased caffeine content in the ASDB and reduced in the CSDB (Table 4.1). Even with the partial leaching of caffeine in the steam treated coffee (Kalschne et al., 2017b), as the SDC was originated from the CC species, it still showed more caffeine than AC.

Table 4.1 - Characterization of the coffee brews* (without sugar).

Parameters**	AB	ASDB	CB	CSDB
Caffeine (mg mL^{-1})	$1.43^{d} \pm 0.02$	$1.63^{c} \pm 0.08$	$2.62^{a} \pm 0.5$	$2.12^{b} \pm 0.03$
Trigonelline (mg mL^{-1})	$0.96^{a} \pm 0.01$	$0.72^{b} \pm 0.02$	$0.52^{c} \pm 0.20$	$0.45^{d} \pm 0.01$
5-CQA (mg mL^{-1})	$0.87^{a} \pm 0.01$	$0.80^{b} \pm 0.02$	$0.60^{c} \pm 0.02$	$0.55^{d} \pm 0.01$
Melanoidins (mg mL^{-1})	$11.21^{c} \pm 0.08$	$10.64^{d} \pm 0.09$	$14.59^{a} \pm 0.05$	$12.57^{b} \pm 0.13$
Total soluble solids (° Brix)	$2.83^{b} \pm 0.06$	$2.40^{d} \pm 0.02$	$2.97^{a} \pm 0.06$	$2.70^{c} \pm 0.02$
pH	$5.54^{d} \pm 0.01$	$5.57^{c} \pm 0.01$	$5.68^{b} \pm 0.01$	$5.76^{a} \pm 0.01$
Acidity (mL NaOH 0.1M 20 mL^{-1})	$3.03^{a} \pm 0.03$	$2.68^{b} \pm 0.03$	$2.52^{c} \pm 0.03$	$2.23^{d} \pm 0.03$

*AB: 100% *C. arabica* (AC); ASDB: 50% AC and 50% steamed defective coffee (SDC); CB: 100% *C. canephora* (CC); CSDB: 50% CC and 50% SDC.

**Mean ± standard deviation (n = 3); different letters in the same line indicate significant differences between samples by Tukey test ($p \leq 0.05$).

4.3.2 Flash Profile

A two-dimensional consensual solution accounted for 70.2% of the variance (Figure 4.1); similar values (from 67 to 88% of variance in two or three dimensional solutions) were described in Flash Profile studies with coffee brews and different matrices (Dairou & Sieffermann, 2002; Kobayashi & Benassi, 2012; Mamede & Benassi, 2016; Terhaag & Benassi, 2010; Veinand, Godefroy, Adam, & Delarue, 2011). Maximum residual variance by assessor was 0.59%, according to values of 0.6% to 0.75% described

in the literature (Mamede & Benassi, 2016; Terhaag & Benassi, 2010), showing the panel consensus in describing the coffee brews.

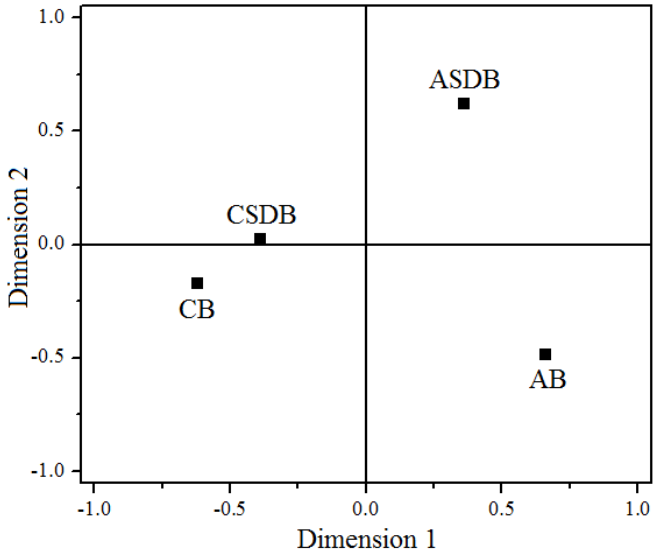

Figure 4.1 – Flash Profile two-dimensional consensus plot for coffee brews.

AB: 100% *C. arabica* (AC); ASDB: 50% AC and 50% steamed defective coffee (SDC); CB: 100% *C. canephora* (CC); CSDB: 50% CC and 50% SDC.

The descriptive terms used in the brews characterization that were better correlated with Dimensions 1 and 2 of the samples configuration ($r \geq |0.6|$) are described in Table 4.1S (in Supporting information) for each assessor. Despite the addition of SDC, most sensory attributes were common to those reported by other researches with descriptive sensory analysis of coffee brews (*C. arabica*, commercial instant coffee and decaffeinated coffees) (Kitzberger, Scholz, Silva, Benassi, & Pereira, 2016; Kobayashi & Benassi, 2012; Mamede, Perazzo, & Maciel, 2010). These attributes include brown color, black color, transparency, the presence of foam, brightness, coffee aroma, roasted/burned aroma, fruity aroma, green herb aroma, coffee flavor, roasted/burned flavor, bitter taste, acid taste, sweet taste, astringency, viscosity and full-bodied, among other less mentioned attributes (Supporting Table 4.1S).

Dimension 1 (accounted for 44% of the variance) was associated with coffee species and allowed the discrimination of the brews with AC (AB and ASDB) from those with only CC (CB and CSDB) in the composition (Figure 4.1). This dimension was positively correlated with brown color, fruity aroma/herb/green bean, coffee flavor/residual coffee flavor, and negatively correlated to black color, burned/roasted aroma,

chocolate/caramel/sweet aroma, bitter taste, roasted flavor, residual bitter taste/burned flavor, presence of foam and viscosity (in the cup and mouth) (Supporting Table 4.1S).

Dimension 2 (26% of variance) was associated with the presence of steam treated defects and allowed the discrimination of brews with SDC (ASDB and CSDB) from those without it (AB e CB) (Figure 4.1). This dimension was positively correlated to brown color and coffee flavor, and negatively correlated to black color, fruity/herb/green bean aroma, acid/sour/residual acid flavor, bitter taste and viscosity (in the cup and mouth) (Supporting Table 4.1S).

Coffee species had greater relevance in differentiating brews by sensory characteristics than the addition of SDC. Thus, there was a similarity between coffee robusta brews (CB and CSDB), regardless of the SDC addition (since the defect was of the same species). AB, without the addition of SDC, was the most different brew among the studied (Figure 4.1).

CB and CSDB brews were described as viscous, with black color and with a bitter taste, which differed from brews with AC by the presence of foam and intense aroma and flavors associated with the roasting process (burned/roasted aroma, chocolate/caramel/sweet aroma, roasted flavor and residual bitter taste/burned flavor) (Supporting Table 4.1S, Figure 4.1). As described before, the CC brews had high soluble solids contents, which contribute to the viscosity (Table 4.1). Compared to AC, they also showed higher caffeine content associated with a bitter taste (Ramalakshmi & Raghavan, 1999; Gloess et al., 2014) - and melanoidins, related to color and flavors and aromas associated with roasting (Table 4.1). These characteristics refer to robusta coffee, so the presence of SDC in a blend caused a little alteration in their sensory profile. Compared to CB, CSDB had less typical robusta coffee brew characteristics, probably due to the leaching of some components as described previously (Figure 4.1, Supporting Table 4.1S).

AB and ASDB brews were characterized by a brown color, fruity/herb/ green bean aroma and coffee/residual coffee flavor (Supporting Table 4.1S, Figure 4.1). These beverages showed higher 5-CQA content and acidity (Table 4.1), characteristics of the presence of arabica coffee (Gloess et al., 2014). In this case, it was observed a greater sensory effect of the addition of SDC, with sensory differentiation between AB and ASDC brews (Figure 4.1). ASDC blend showed clear color and lower viscosity, consistent with a lower content of total soluble solids and melanoidins (Supporting Table 4.1S, Figure 4.1, Table 4.1). AB brew (with the highest content of 5-CQA and titratable acidity) was

described as having the greatest intensity of fruity/herb/green bean aroma and acid/sour/residual acid taste (Supporting Table 4.1S, Figure 4.1), compared to ASDC. Similarly that observed for CC brews, the addition of SDC allowed to keep the typical sensory characteristics of the arabica coffee brews, being reduced only the intensity of these attributes.

4.3.3 Acceptance test and Internal preference mapping

The CB, ASDB and ACDB brews showed scores grades superior to 6 for overall acceptance and attributes acceptance (color, texture, aroma and flavor), showing good acceptance and purchase intent, with no significant difference (Table 4.2). The AB brew showed lower scores regarding texture and flavor, which probably justifies its low acceptance and, at the same time, less purchase intent (Table 4.2).

The color was cited as the most appreciated attribute in the brews, with percentages between 42.8% (CB) and 25.1% (AB) (Supporting Table 4.2S). Color acceptance was positively correlated with caffeine and melanoidins content ($r \geq 0.95$, $p = 0.05$) (Supporting Table 4.3S), favored by the presence of robusta coffee.

The flavor was the second attribute most appreciated (from 36.8% for ASDB to 22.0% for CB), followed by the aroma (from 21.1% for ASDB to 15.6% for CB) and the texture (from 18.1% for AB to 12.9% for ASDB) (Supporting Table 4.2S). The acceptance regarding flavor and texture was positively correlated to overall acceptance ($r \geq 0.97$, $p \leq 0.03$) and purchase intent ($r \geq 0.96$, $p \leq 0.04$), reinforcing the impact of these attributes on the brew's general assessment (Table 4.3S).

Table 4.2 - Sensory acceptance and purchase intent for the coffee brews.

Parameters*	AB	ASDB	CB	CSDB
Color	$7.4^{b} \pm 1.9$	$7.3^{b} \pm 1.7$	$8.0^{a} \pm 1.7$	$7.6^{ab} \pm 1.6$
Texture	$5.2^{b} \pm 3.0$	$6.7^{a} \pm 1.9$	$6.3^{a} \pm 2.2$	$6.4^{a} \pm 2.0$
Aroma	$6.9^{a} \pm 2.1$	$7.1^{a} \pm 2.0$	$7.1^{a} \pm 1.9$	$7.0^{a} \pm 1.9$
Flavor	$5.6^{b} \pm 2.9$	$6.6^{a} \pm 2.4$	$6.0^{ab} \pm 2.4$	$6.5^{a} \pm 2.3$
Overall acceptance	$5.9^{b} \pm 2.4$	$6.7^{a} \pm 2.0$	$6.4^{ab} \pm 1.9$	$6.6^{a} \pm 1.9$
Purchase intent	$5.1^{b} \pm 3.0$	$6.3^{a} \pm 2.5$	$5.9^{a} \pm 2.5$	$6.2^{a} \pm 2.5$

*Mean ± standard deviation (n = 142); AB: 100% *C. arabica* (AC); ASDB: 50% AC and 50% steamed defective coffee (SDC); CB: 100% *C. canephora* (CC); CSDB: 50% CC and 50% SDC.

Hedonic scale: 0 = disliked extremely, 5 = neither liked, nor disliked, 10 = like extremely; Purchase intent: 0 = definitely not buy, 5 = maybe buy/maybe nor buy, 10 = definitely buy; different letters in the lines indicate significant difference by Tukey test ($p \leq 0.05$).

Individual results assessment by the Internal preference mapping allows verifying the segmentation of the consumers (Figure 4.2). The two-dimensional solutions accounted for 76% and 78% of the variance for overall acceptance and purchase intent, respectively (Figure 4.2).

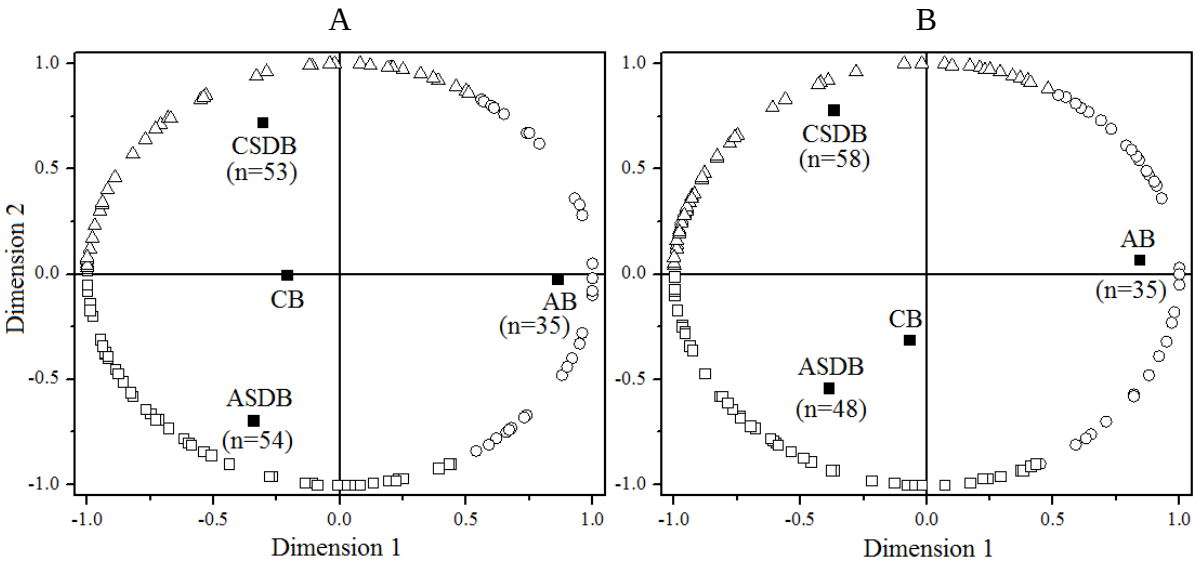

Figure 4.2 - Internal preference mapping for coffee brews: configuration of the samples and consumers considering overall acceptance (A) and purchase intent (B).

AB: 100% *C. arabica* (AC); ASDB: 50% AC and 50% steamed defective coffee (SDC); CB: 100% *C. canephora* (CC); CSDB: 50% CC and 50% SDC.

Group 1 (○): prefer AB; Group 2 (□): prefer ASDB; Group 3 (Δ): prefer CSDB.

n = number of consumers that prefer the sample indicated.

AB brew was well discriminated from the others, located separately on the right side of the Internal preference maps (Figures 4.2A and 4.2B). Despite its lowest global acceptance scores (Table 4.2), a group of consumers (group 1, 35 assessors, 25% of the total) preferred it and had high intention to purchase it (Figures 4.2A and 4.2B).

An opposite behavior was verified for the CB brew that showed similar acceptance to the SDC brews (Table 4.2), and was located next to these brews on the Maps, but it was not indicated as preferred by any consumer (Figure 4.2).

Consumers considered the brews with SDC as the preferred ones and having higher purchase intent (Figure 4.2). Group 2 consumers preferred the arabica coffee blend ASDB (54 and 48 assessors corresponding to 38% and 34% of the total, for overall acceptance and purchase intent, respectively). Group 3 consumers preferred the robusta coffee blend

CSDB (53 and 58 assessors corresponding to 37% and 41% of the total, for overall acceptance and purchase intent, respectively) (Figure 4.2).

The average grades of acceptance and purchase intent and the behavior of consumers' in the preference maps showed an excellent potential for the use of up to 50% of *C. canephora* SDC in blends with *C. arabica* and *C. canephora.*

4.4 Conclusion

In general, the addition of 50% of defective steam treated *C. canephora* coffee to *C. arabica* and *C. canephora* did not generate in the blends different sensory attributes from those used to describe pure coffee brews. *C. arabica* brews (pure or blend) were described as having a brown color, fruity/herb/green bean aroma and coffee/residual coffee flavor. *C. canephora* brews were characterized by a black color, greater viscosity with greater presence of foam, bitter taste, and with aromas and flavors associated with the roasting process. The coffee species used had more relevance towards differentiating sensory characteristics of the brews than the addition of SDC. Therefore, it was observed similarity among *C. canephora* brews, while the pure *C. arabica* brew (without SDC addition) was the one with the most differentiated sensory characteristics. With the addition of SDC, the most typical sensory characteristics of each species were kept, reducing only the intensity of the attributes. Coffee brews with SDC were well accepted by consumers (indicated as preferred and with greater purchase intent) thus showing a potential for the addition of up to 50% of SDC of *C. canephora* in blends with *C. arabica* and *C. canephora.* The coffee blend with SDC is an excellent proposal, since it presents sensory characteristics and acceptance with good potential for commercialization in Brazil and creates an appropriate, realizable and efficient solution for PVA beans.

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Supplementary Material

Table 4.1S - Attributes best correlated ($r \geq |0.6|$) with the dimension 1 and 2 of each assessor on Flash Profile.

A*	Dimension 1	Dimension 2
1	Pepper aroma (0.75); Coffee aroma (-0.75); Astringency (0.75); Coffee flavor (0.88); Bitter taste (0.88); Residual bitter taste (0.75); Burned flavor (0.88);	Brown color (-0.80);
2	Roasted coffee aroma (-0.85); Acid taste (0.85); Fruity aroma (0.85)	Black color (-0.80);
3	Black color (-0.85); Burned aroma (-0.98); Coffee aroma (0.98); Bitter taste (-0.98); Coffee flavor (0.98); Residual bitter taste (-0.75);	Foam (0.80);
4		Coffee aroma (0.80); Acid taste (-0.97); Coffee flavor (0.97); Burned/roasted flavor (-0.97); Residual bitter taste (-0.87); Residual acid taste (-0.87);
5	Bitter taste (-0.85); Black color (-0.98); Brown color (0.98); Astringency (0.75); Viscosity (-0.98); Coffee aroma (-0.88); Coffee flavor (-0.98);	Burned flavor (-0.64); Residual flavor (0.97);
6	Black color (-0.66); Residual astringent flavor (0.88); Coffee aroma (0.88)	
7	Black color (-0.85); Yellow color (edge) (0.85); Transparency (0.85); Roasted aroma (0.98); Sugarcane liquor aroma (-0.98); Roasted flavor (0.98); Residual bitter taste (0.88); Bitter taste (0.98); Coffee flavor (0.98); Coffee aroma)0.98); Sugarcane liquor flavor (-0.98);	
8	Black color (-0.85); Yellow color (edge) (-0.85); Watery (-0.98); Coffee aroma (0.88); Coffee flavor (0.98); Bitter taste (-0.85);	Transparency (0.80); Burned flavor (0.97); Residual bitter taste (0.64); Residual burned flavor (0.64);
9	Watery (0.98); Brown color (0.98); Black color (-0.98); Viscosity (mouth) (-0.98); Coffee aroma (-0.88);	Bitter taste (-0.80); Astringency (-0.80); Coffee flavor (-0.80);
10	Brown color (0.98); Black color (-0.98); Viscosity (cup) (-0.98); Herb aroma (tea) (0.88); Coffee aroma (-0.88); Bitter aroma (0.88); Bitter taste (0.88); Coffee flavor (-0.88); Herb flavor (tea) (0.88); Residual bitter taste (-0.85);	Viscosity (mouth) (-0.64);
11	Coffee aroma (-0.66); Brown color (0.98); Black color (-0.98); Bitter taste (-0.85); Watery (0.85); Viscosity (cup) (-0.85);	Coffee flavor (0.97); Burned aroma (-0.87); Roasted flavor (-0.64);
12	Black color (-0.88); Coffee aroma (0.66); Roasted flavor (-0.98); Residual bitter taste (-0.85);	Coffee flavor (-0.80); Foam (-0.80); Oiliness (-0.80); Viscosity (cup) (-0.80); Watery (-0.80); Fruity aroma (-0.97);
13	Green branch aroma (-0.85); Coffee flavor (-0.88); Sour flavor (0.98); Black color (-0.88); Roasted coffee flavor (0.88); Velvety texture (-0.75);	Coffee aroma (-0.64);
14	Brown color (-0.66); Caramel color (edge) (-0.66); Transparency (0.66); Viscosity (-0.98); Pepper flavor (0.88); Foam (-0.85); Residual pepper flavor (0.88); Caramel aroma (-0.88);	
15		Coffee aroma (-0.87); Coffee flavor (-0.97); Black color (-0.87); Green color (edge) (-0.87); Watery flavor (-0.97); Bitter taste (-0.80); Residual coffee flavor (-0.97); Viscosity (cup) (-0.80);
16	Yellow color (edge) (0.75); Bitter taste (-0.66); Residual coffee flavor (0.66); Residual bitter taste (-0.88); Burned favor (-0.75);	Black color (-0.87); Coffee aroma (-0.87); Coffee flavor (-0.87); Viscosity (cup) (-0.87); Viscosity (mouth) (-0.97); Watery (0.87);

17	Fruity aroma (0.98); Viscosity (cup) (0.66); Bitter taste (-0.85);	Brown color (0.87); Coffee aroma (0.97); Sweet taste (-0.87); Coffee flavor (0.87);
18	Black color (-0.85); Viscosity (cup) (-0.75); Characteristic coffee aroma (-0.75); Roasted aroma (-0.85); Burned aroma (-0.75); Green bean aroma (0.88); Coffee flavor (-0.75); Green bean flavor (-0.88); Sweet taste (0.75);	
19	Black color (-0.88); Viscosity (cup) (-0.66); Coffee aroma (-0.88); Sweet aroma (-0.88); Biological yeast aroma (0.88); Watery (0.66); Residual rancid flavor (0.98); Astringency (-0.75); Residual bitter taste (-0.98);	Coffee flavor (0.64); Residual metallic flavor (0.97); Sour flavor (-0.64);
20	Black color (-0.75); Brown color (0.75); Coffee aroma (-0.88); Roasted aroma (0.98); Bitter taste (-0.98); Sweet taste (0.98); Residual flavor (0.66);	Foam (-0.87); Coffee flavor (0.80);
21	Black color (-0.98); Bitter taste (0.75); Coffee flavor (0.85); Coffee aroma (-0.75); Roasted aroma (0.88); Brown color (edge) (0.75);	Residual bitter taste (-0.64); Residual coffee flavor (0.80);
22	Black color (-0.98); Brown color (0.98); Coffee aroma (-0.98); Green bush aroma (stevia) (0.88); Sweet aroma (-0.88); Bitter taste (0.88); Astringency (0.88); Residual coffee flavor (-0.98); Chocolate aroma (-0.98);	Residual bitter taste (-0.64); Viscosity (cup) (-0.87); Foam (-0.97);
23	Black color (-0.98); Brown color (0.85); Yellow color (edge) (-0.98); Foam (-0.75); Coffee aroma (-0.85); Woody aroma (0.98); Coffee flavor (-0.98); Woody flavor (0.98); Viscosity (0.75); Transparency (-0.66); Residual powder (0.85); Residual coffee flavor (-0.85); Residual woody flavor (0.85);	
24	Black color (-0.85); Burned flavor (-0.85);	Coffee aroma (0.87); Yellow color (edge) (-0.87); Coffee flavor (0.97); Residual bitter taste (0.87); Roasted aroma (0.87);
25	Green color (edge) (-0.98); Coffee aroma (0.66); Residual coffee flavor (0.66); Astringency (-0.66);	Black color (-0.87); Bitter taste (-0.80); Sweet taste (0.80); Viscosity (-0.87); Transparency (-0.97);
26	Black color (-0.85); Viscosity (-0.75); Coffee aroma (-0.75); Coffee flavor (-0.75); Bitter taste (-0.75); Residual coffee flavor (-0.88);	Brown color (0.87);
27	Roasted aroma (-0.88); Coffee aroma (-0.88); Roasted flavor (-0.88); Viscosity (mouth) (-0.98); Residual burned flavor (-0.98);	Black color (-0.87); Brown color (0.87);
28	Viscosity (cup) (0.66); Brown color (-0.75); Coffee aroma (0.88); Residual coffee flavor (0.66); Residual burned flavor (-0.66);	Coffee flavor (-0.87);
29	Coffee aroma (-0.98); Burned aroma (0.88); Burned flavor(0.88);	Yellow color (0.97); Residual coffee flavor (0.64); Coffee flavor (0.97);
30	Coffee aroma (-0.66); Green bean flavor (0.98); Residual coffee flavor (0.98); Foam (0.98);	Black color (0.97); Coffee flavor (0.97);
31	Green color (edge) (0.88); Brown color (-0.88); Coffee aroma (0.98); Foam (-0.88); Watery (0.88); Coffee flavor (-0.66); Earth aroma (-0.98); Viscosity (mouth) (0.88); Chocolate aroma (-0.88); Coffee powder aroma (0.98); Espresso flavor (0.88);	Bitter taste (-0.64);
32	Coffee aroma (0.98); Coffee flavor (0.98); Residual bitter taste (0.66); Foam (-0.98); Metallic aroma (-0.88); Metallic flavor (-0.88);	Caramel color (-0.80); Transparency (-0.80); Burned aroma (0.87); Burned flavor (0.87);
33	Sweet aroma (-0.66);	Residual bitter taste (-0.97); Coffee aroma (0.64); Herb aroma (spices) (-0.87); Coffee flavor (0.64); Sweet taste (0.87); Bitter taste (-0.87);

34	Brown color (-0.98); Brightness (-0.75); Earth/wood aroma (-0.75); Coffee aroma (-0.85); Foam (-0.85); Earth flavor (-0.75); Sour flavor (0.98);	Residual bitter taste (-0.97); Sour flavor (-0.64); Viscosity (mouth) (-0.87);
35	Yellow/green color (edge) (0.85); Viscosity (0.75); Coffee flavor (0.98); Residual coffee flavor (0.98);	Coffee aroma (-0.64);
36	Coffee aroma (0.98); Pungent aroma (earth) (-0.98); Coffee flavor (0.85); Pungent flavor (earth) (-0.85); Residual bitter taste (-0.85); Residual coffee flavor (0.98);	Brown color (-0.64); Viscosity (-0.80);
37	Black color (-0.66);	Fruity aroma (-0.97); Coffee aroma (0.64); Acid taste (-0.80); Coffee flavor (0.64); Astringency (-0.80); Residual acid taste (-0.80); Coffee flavor (0.80);
38	Black color (-0.85); Brown color (0.85); Viscosity (-0.85); Coffee aroma (-0.98); Roasted aroma (-0.98); Green bean aroma (0.98); Coffee flavor (-0.88); Roasted flavor (-0.85); Green bean flavor (0.98); Transparency (0.85); Residual coffee flavor (-0.85);	
39	Coffee flavor (0.85); Bitter taste (-0.85); Residual bitter taste (-0.85); Residual coffee flavor (0.85);	Transparency (0.97); Black color (-0.97); Brown color (0.97); Green bean aroma (-0.97); Roasted aroma (0.97); Green bean flavor (-0.97); Viscosity (-0.87);
40	Coffee flavor (0.85); Bitter taste (-0.85); Residual bitter taste (-0.85);	Transparency (0.97); Black color (-0.97); Brown color (0.97); Green bean aroma (-0.97); Roasted aroma (0.97); Green bean flavor (-0.97); Viscosity (-0.87);

***** Assessor.

Table 4.2S - Most appreciated sensory characteristics of each coffee brew and frequency of reporting (%).

AB	ASDB	CB	CSDB
Color (34.4%)	Color (25.1%)	Color (42.8%)	Color (33.3%)
Aroma (19.4%)	Aroma (21.1%)	Aroma (17.9%)	Aroma (17.9%)
Texture (18.1%)	Texture (12.9%)	Texture (15.6%)	Texture (13.0%)
Flavor (24.4%)	Flavor (36.8%)	Flavor (22.0%)	Flavor (32.1%)

AB: 100% *C. arabica* (AC); ASDB: 50% AC and 50% steamed defective coffee (SDC); CB: 100% *C. canephora* (CC); CSDB: 50% CC and 50% SDC.

Table 4.3S - Pearson correlation among physicochemical data, sensory acceptance and purchase intent ($p \leq 0.05$).

	Caffeine	Trigonelline	5-CQA	Melanoidin	Soluble solids	pH	Acidity	Acceptance					Purchase Intent
								Color	Texture	Aroma	Flavor	Overall acceptance	
Caffeine	1.00												
Trigonelline	-0.83	1.00											
5-CQA	-0.86	0.97*	1.00										
Melanoidins	0.95*	-0.67	-0.76	1.00									
TSS	0.54	-0.10	-0.31	0.76	1.00								
pH	0.75	-0.93	-0.98*	0.65	0.26	1.00							
Acidity	-0.69	0.97*	0.94	-0.50	0.04	-0.95*	1.00						
Acceptance Color	0.95*	-0.63	-0.72	>0.99*	0.77	0.60	-0.45	1.00					
Texture	0.44	-0.72	-0.52	0.15	-0.52	0.46	-0.71	0.14	1.00				
Aroma	0.57	-0.58	-0.40	0.35	-0.26	0.25	-0.46	0.37	0.88	1.00			
Flavor	0.14	-0.60	-0.40	-0.16	-0.74	0.42	-0.69	-0.19	0.91	0.62	1.00		
Overall	0.35	-0.71	-0.52	0.05	-0.60	0.49	-0.74	0.03	0.99*	0.78	0.97*	1.00	
Purchase Intent	0.39	-0.75	-0.56	0.10	-0.56	0.53	-0.77	0.07	0.99*	0.78	0.96*	>0.99*	1.00

CONCLUSÃO GERAL E CONSIDERAÇÕES

A combinação do tratamento com vapor e subsequente processo de torra modificou o perfil de composição de compostos voláteis e de bioativos no café torrado PVA.

Considerando o perfil de compostos voláteis, a melhor condição foi definida como o tratamento 5 bar/16 min (SC 5) onde foi observado um aumento na concentração dos voláteis de impacto positivo e decréscimo nos compostos voláteis de impacto negativo. Outros dois tratamentos demostraram potencial: o mais brando (2 bar/3 min), que também apresentou bom balanço de compostos voláteis positivos e negativos, e o mais drástico (8 bar/29 min), que se destacou pela redução nos voláteis de impacto negativo.

No geral, tratamentos com vapor com tempos longos e baixas pressões devem ser evitados para melhor preservação dos compostos bioativos e atividade antioxidante. Caso o objetivo fosse a manutenção específica de algum bioativo, seria interessante a opção por tratamentos diferenciados: tempos mais curtos para preservação de cafeína, trigonelina e melanoidinas (os dois últimos empregando condição de pressão mais alta), e tratamento mais drástico (tempo longo e alta pressão) para ACGs e diterpenos (cafestol e 16-O-metilcafestol). No tratamento mais promissor em termos do perfil volátil (5 bar/16 min) obteve-se adequada preservação da atividade antioxidante e dos bioativos de um modo geral .

Numa avaliação preliminar para definição de possíveis porcentagens de adição do tratamento com maior potencial (SC5) em *blends*, verificou-se que até 30% de SC 5 não gerou rejeição ou percepção da presença do defeito em bebida produzida com *blend* de café arábica, e que o *blend* com 30% de SC 5 teve boa aceitação, concluindo-se assim pela viabilidade da aplicação e a possibilidade de uso de porcentagens superiores a 30% de PVA tratado.

As bebidas produzidas a partir de *blends* de café arábica e café canéfora com 50% de SC 5 apresentaram um perfil adequado de compostos bioativos e foram bem aceitas e descritas como apresentando características sensoriais similares as bebidas puras, sendo indicadas como preferidas e com maior intenção de compra pelos consumidores.

Concluiu-se que o tratamento com vapor de grãos de café canéfora PVA preliminarmente ao processo de torra é uma proposta promissora, com boas perspectivas de emprego no Brasil. Destaca-se por ser uma solução apropriada e eficiente na melhora da qualidade e agregação de valor do café PVA e aplicável para as indústrias beneficiadoras de café.