UNIVERSIDADE FEDERAL DE SANTA MARIA
CENTRO DE CIÊNCIAS RURAIS
PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIA E TECNOLOGIA DOS ALIMENTOS

Rosane Teresinha Heck

REPLACEMENT OF ANIMAL FAT BY RESTRUCTURED VEGETABLE OILS IN BURGERS: NUTRITIONAL, SENSORY AND TECHNOLOGICAL IMPACTS

SUBSTITUIÇÃO DA GORDURA ANIMAL POR ÓLEOS VEGETAIS REESTRUTURADOS EM HAMBÚRGUERES: IMPACTOS NUTRICIONAIS, SENSORIAIS E TECNOLÓGICOS

Santa Maria, RS
2021

Rosane Teresinha Heck

REPLACEMENT OF ANIMAL FAT BY RESTRUCTURED VEGETABLE OILS IN BURGERS: NUTRITIONAL, SENSORY AND TECHNOLOGICAL IMPACTS

SUBSTITUIÇÃO DA GORDURA ANIMAL POR ÓLEOS VEGETAIS REESTRUTURADOS EM HAMBÚRGUERES: IMPACTOS NUTRICIONAIS, SENSORIAIS E TECNOLÓGICOS

Thesis presented to
the Graduate Program in Science and
Food Technology, Area
Concentration in Science and Technology of
Food, Federal University of Santa
Maria (UFSM, RS), as a partial requirement
for obtaining a **degree of Doctor in
Food Science and Technology.**

Tese apresentada ao
Programa de Pós-Graduação em Ciências e
Tecnologia dos Alimentos, Área de
Concentração em Ciência e Tecnologia de
Alimentos, Universidade Federal de Santa
Maria (UFSM, RS), como exigência parcial
para obter o grau de Doutor em
Ciência e Tecnologia de Alimentos.

Advisor/Orientador: Prof. Dr. Paulo Cezar Bastianello Campagnol
Co-Advisor/Coorientador: Prof. Dr. Cristiano Ragagnin de Menezes

Santa Maria, RS
2021

This study was financied in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) – Finance Code 001

Heck, Rosane Teresinha
REPLACEMENT OF ANIMAL FAT BY RESTRUCTURED VEGETABLE OILS IN BURGERS: NUTRITIONAL, SENSORY AND TECHNOLOGICAL IMPACTS / Rosane Teresinha Heck.- 2021.
110 p.; 30 cm

Orientador: Paulo Cezar Bastianello Campagnol
Coorientador: Cristiano Ragagnin de Menezes
Tese (doutorado) - Universidade Federal de Santa Maria, Centro de Ciências Rurais, Programa de Pós Graduação em Ciência e Tecnologia dos Alimentos, RS, 2021

1. Microencapsulation 2. Gelation 3. Bioactive compounds 4. Healthier meat products 5. Healthy oil I. Campagnol, Paulo Cezar Bastianello II. de Menezes, Cristiano Ragagnin III. Título.

Sistema de geração automática de ficha catalográfica da UFSM. Dados fornecidos pelo autor(a). Sob supervisão da Direção da Divisão de Processos Técnicos da Biblioteca Central. Bibliotecária responsável Paula Schoenfeldt Patta CRB 10/1728.

Declaro, ROSANE TERESINHA HECK, para os devidos fins e sob as penas da lei, que a pesquisa constante neste trabalho de conclusão de curso (Tese) foi por mim elaborada e que as informações necessárias objeto de consulta em literatura e outras fontes estão devidamente referenciadas. Declaro, ainda, que este trabalho ou parte dele não foi apresentado anteriormente para obtenção de qualquer outro grau acadêmico, estando ciente de que a inveracidade da presente declaração poderá resultar na anulação da titulação pela Universidade, entre outras consequências legais.

Rosane Teresinha Heck

REPLACEMENT OF ANIMAL FAT BY RESTRUCTURED VEGETABLE OILS IN BURGERS: NUTRITIONAL, SENSORY AND TECHNOLOGICAL IMPACTS

Thesis presented to
the Graduate Program in Science and
Food Technology, Area
Concentration in Science and Technology of
Food, Federal University of Santa
Maria (UFSM, RS), as a partial requirement
for obtaining a **degree of Doctor in**
Food Science and Technology.

Approved on August 18, 2021.

_____

Paulo Cezar Bastianello Campagnol, Dr. (UFSM)
(Chairperson / Advisor)

_____

Cristiano Ragagnin de Menezes, Dr. (UFSM)
(Co-Advisor/Coorientador)

_____

Jose Manuel Lorenzo Rodriguez Dr. (UVIGO)

_____

Marco Antonio Trindade Dr. (USP)

_____

Marise Aparecida Rodrigues Pollonio Dra. (Unicamp)

_____

Roger Wagner, Dr. (UFSM)

Santa Maria, RS
2021

NUP: 23081.066401/2021-16 **Prioridade:** Normal

Homologação de ata de banca de defesa de pós-graduação

134.332 - Bancas examinadoras: indicação e atuação

COMPONENTE

Ordem	Descrição	Nome do arquivo
4	Folha de aprovação	Folha de aprovação.pdf

Assinaturas

15/09/2021 09:27:20

PAULO CEZAR BASTIANELLO CAMPAGNOL (PROFESSOR DO MAGISTÉRIO SUPERIOR)

03.10.07.00.0.0 - CURSO-PROGRAMA PG e CIÊNCIA E TECNOLOGIA DE ALIMENTOS - CPPGCTA

15/09/2021 10:06:10

CRISTIANO RAGAGNIN DE MENEZES (PROFESSOR DO MAGISTÉRIO SUPERIOR)

03.39.00.00.0.0 - DEPARTAMENTO DE TECNOLOGIA E CIÊNCIA ALIMENTOS - DTCA

15/09/2021 14:18:56

Marco Antonio Trindade (Pessoa Física)

Usuário Externo (096.***.***-**)

15/09/2021 18:32:46

Jose Manuel Lorenzo (Pessoa Física)

Usuário Externo (PAI******)

21/09/2021 19:48:47

ROGER WAGNER (PROFESSOR DO MAGISTÉRIO SUPERIOR)

03.39.00.00.0.0 - DEPARTAMENTO DE TECNOLOGIA E CIÊNCIA ALIMENTOS - DTCA

30/09/2021 14:25:12

Marise Aparecida Rodrigues Pollonio (Pessoa Física)

Usuário Externo (024.***.***-**)

DEDICATÓRIA

Minha querida mãe Amélia (in memoriam) e meu pai Silfredo, cujos esforços para me educar sempre estiveram em primeiro lugar. Aqui estão os resultados de seus esforços. E também a meu marido Ilson por seu apoio incondicional. A minha filha Gabriela que ainda está em meu ventre, mas já amamos tanto! Gratidão infinita!

AGRADECIMENTOS

Durante os quatro anos de doutorado, só tenho a agradecer a todos que passaram pelo meu caminho e que com certeza deixaram um pouco de si. É muito difícil transformar sentimentos em palavras, mas serei eternamente grata a vocês, pessoas imprescindíveis para a realização e conclusão deste trabalho.

Primeiramente, agradeço ao meu orientador, o professor Paulo Cezar Bastianello Campagnol, um excelente exemplo de profissional e de ser humano. Só tenho a agradecer aos seus ensinamentos, orientações, palavras de incentivo, paciência e dedicação. Você é uma pessoa ímpar, onde busco inspirações para me tornar melhor em tudo faço e irei fazer daqui para frente. Meus mais sinceros agradecimentos.

Ao Professor Cristiano Ragagnin de Menezes, muito obrigada pela ajuda, ensinamentos, orientações e contribuições. Por me receber em seu laboratório de portas abertas e sempre estar à disposição, e me incentivando a acreditar que as microcápsulas dariam certo. Realmente, deram, e você é parte essencial desse trabalho. Gatidão!

Ao professor Juliano Barin por me encorajar a encarar o mundo do empreendedorismo e o desafio de transformar o "Paper" em "PIB" ao fundar a *startup* w3 Special Ingredients.

Agradeço à Universidade Federal de Santa Maria (UFSM) e ao Curso de Pós-Graduação em Ciência e Tecnologia dos Alimentos (PPGCTA) por viabilizarem a realização deste trabalho.

À minha família, pelo amor, apoio, confiança e motivação incondicional, que sempre me impulsionaram em direção às vitórias dos meus desafios. E, por compreenderem minhas ausências.

Aos professores Alexandre José Cichoski e Roger Wagner por gentilmente cederem espaço em seus laboratórios e colaboração inestimável.

As colegas de pós-graduação Raquel Vendruscolo, Mariane Bittencourt Fagundes, Daniele Ferreira e Jordana Lima por toda a ajuda, companheirismo, compreensão, carinho e amizade. Valeu mesmo!

A bolsista de iniciação cientifica (IC) Leticia Correa, que colaborou neste trabalho, muito obrigada!

Por fim, agradeço à Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Código de Financiamento 001 pelo auxílio financeiro.

"*Antes que você possa alcançar o topo de uma árvore e entender os brotos e as flores, você terá de ir fundo nas raízes, porque o segredo está lá. E, quanto mais fundo vão as raízes, mais alto vai a árvore"*.

(Nietzsche)

RESUMO

Devido ao alto teor de ácidos graxos saturados o consumo excessivo de produtos cárneos pode aumentar o risco do surgimento de doenças crônicas. Desta forma, para conferir características mais saudáveis aos produtos cárneos, este estudo teve por objetivo melhorar o seu perfil lipídico. Óleos com altos teores de ácidos graxos ômega-3 foram aplicados na forma microencapsulada e hidrogelificada como substitutos de gordura animal em hambúrgueres. Na primeira parte do estudo foi avaliada a estabilidade oxidativa de óleo de chia enriquecido com alecrim por extração assistida por ultrassom (EAU) e por maceração convencional (CME). Nesta etapa foi possível observar alta concentração de compostos bioativos presentes nos óleos macerados, tanto por EAU quanto por CME. Na sequência, o óleo de chia foi microencapsulado e usado para substituir 50 % de gordura em hambúrgueres. Os resultados obtidos através da incorporação direta do alecrim no óleo de chia por EAU antes da microencapsulação mostraram que esta é uma alternativa viável para reduzir a oxidação lipídica. A redução da oxidação lipídica das microcápsulas possibilitou substituir a gordura animal em hambúrgueres por óleos ricos em n-3 PUFAs, sem comprometer sua qualidade oxidativa e sensorial. Na segunda etapa foram elaborados hambúrgueres (20 % de toucinho) com a substituição de 0, 20, 40, 60, 80 e 100 % de toucinho por emulsão hidrogelificada (HE) de óleos de chia e de linhaça (50:50). A substituição do toucinho suíno por HE não afetou as propriedades tecnológicas dos hambúrgueres. O perfil sensorial demonstrou que é possível substituir até 60 % de toucinho por HE produzido neste estudo. No entanto, melhorias na estabilidade oxidativa da HE de óleos de chia e linhaça se fizeram necessárias. Desta maneira, na terceira etapa, a HE foi enriquecida nas concentrações de 6, 8 e 10 % com um extrato aquoso de casca de jabuticaba (ECJ) obtido por extração por micro-ondas por hidrodifusão e gravidade (MHG). O HE enriquecido com ECJ foi aplicado como substituto de 60 % do teor de gordura de hambúrgueres. A adição de 10 % de ECJ na HE foi eficaz em manter uma oxidação lipídica semelhante ao controle até o 60º dia de armazenamento. Além disso, a incorporação do JPE no HE reduziu os defeitos sensoriais causados pela reformulação lipídica. Buscando melhorias através da incorporação de antioxidantes naturais, na quarta etapa deste trabalho, resíduos de extratos de casca de erva-mate (BMBE) foram testados para aplicação no HE e posterior substituição lipídica nos hambúrgueres. A qualidade sensorial dos hambúrgueres não foi afetada pela adição de HE de óleo de chia enriquecido com BMBE. Além disso, a adição de BMBE ao HE reduziu parcialmente o aumento da oxidação lipídica causado pelo aumento na razão PUFA / SFA. Desta maneira, no presente estudo foram desenvolvidas com sucesso novas estratégias e

soluções tecnológicas consistentes no âmbito industrial para redução da gordura saturada em hambúrgueres, resultando em produtos mais saudáveis e de elevada qualidade sensorial.

Palavras-chave: ácidos graxos saturados (SFA); ácidos graxos monoinsaturados (MUFA); ácidos graxos poliinsaturados (PUFA); microencapsulação; gelatinização; compostos bioativos; produtos cárneos saudáveis; qualidade nutricional; óleos saudáveis.

ABSTRACT

Due to the high content of saturated fatty acids, the excessive consumption of meat products can increase the risk of chronic diseases. Thus, to confer healthier characteristics to meat products, this study aimed to improve their lipid profile. Oils with high levels of omega-3 fatty acids were applied in microencapsulated and hydrogelified form as animal fat substitutes in burgers. In the first part of the study the oxidative stability of rosemary-enriched chia oil was evaluated by ultrasound-assisted extraction (UAE) and conventional maceration (CME). In this step it was possible to observe a high concentration of bioactive compounds present in the macerated oils, both by UAE and CME. Then, the chia oil was microencapsulated and used to replace 50% of fat in burgers. The results obtained by directly incorporating rosemary into chia oil by UAE prior to microencapsulation showed that this is a viable alternative to reduce lipid oxidation. The reduction of lipid oxidation in microcapsules made it possible to replace animal fat in burgers with oils rich in n-3 PUFAs, without compromising their oxidative and sensory quality. In the second step, burgers (20 % pork back fat) were prepared with the replacement of 0, 20, 40, 60, 80 and 100 % pork back fat by hydrogelified emulsion (HE) of chia and flaxseed oils (50:50). The substitution of pork back fat with HE did not affect the technological properties of the burgers. The sensory profile showed that it is possible to replace up to 60 % of fat with HE produced in this study. However, improvements in the oxidative stability of HE from chia and flaxseed oils were necessary. Thus, in the third step, HE was enriched at concentrations of 6, 8 and 10 % with an aqueous jabuticaba peel extract (JPE) obtained by microwave extraction by hydrodiffusion and gravity (MHG). The HEenriched with JPE was applied to replace 60 % of animal fat content of the burgers. The addition of 10 % JPE in HE was effective in maintaining lipid oxidation similar to the control until the 60th day of storage. Furthermore, incorporation of JPE into HE reduced sensory defects caused by lipid reformulation. Seeking improvements through the incorporation of natural antioxidants, in the fourth step of this work, residues of yerba mate bark extracts (BMBE) were tested for application in HE and subsequent lipid replacement in burgers. The sensory quality of the burgers was not affected by HE addition of chia oil enriched with BMBE. Furthermore, the addition of BMBE to HE partially reduced the increase in lipid oxidation caused by the increase in PUFA / SFA ratio. Thus, in the present study, new strategies and consistent technological solutions were successfully developed at the industrial level for reducing saturated fat in burgers, resulting in healthier products with high sensory quality.

Keywords: saturated fatty acid (SFA); monounsaturated fatty acids (MUFA); polyunsaturated fatty acid (PUFA); microencapsulation; gelation; bioactive compounds; healthier meat products; nutritional quality; healthy oil.

.

SUMÁRIO

1. APRESENTAÇÃO

Esta tese de doutorado segue as normas estabelecidas na Estrutura e Apresentação de Monografias, Dissertações e Teses - MDT da UFSM (UFSM, 2015). Os resultados obtidos durante os anos de doutoramento são apresentados na forma de seis artigos científicos e um capítulo de livro.

O primeiro capítulo apresentará um artigo de revisão publicado na revista *Current Opinion in Food Science* no ano de 2021 intitulado "Microencapsulation of healthier oils: an efficient strategy to improve the lipid profile of meat products" que apresenta informações sobre estudos recentes sobre a microencapsulação de óleos mais saudáveis como estratégia para melhorar o perfil lipídico de produtos cárneos.

Da mesma forma, o segundo capítulo intitulado "Replacement of saturated fatty acids by healthy oils to improve the nutritional quality of meat products" é um capítulo que será publicado no livro intitulado "Food lipids: sources, health implications, and future trends" da Editora Elsevier. Este capítulo aborda as estratégias recentes utilizadas para incorporar óleos saudáveis em produtos cárneos e o efeito na saúde de diferentes tipos de ácidos graxos e a qualidade nutricional dos principais óleos saudáveis usados em formulações de produtos cárneos.

O terceiro capítulo intitulado "Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques" publicado em 2018 na revista *Meat Science* apresenta um estudo sobre a estabilidade oxidativa de óleos de chia enriquecidos com alecrim por extração assistida por ultrassom (UAE) e por maceração convencional (CME). O óleo de chia enriquecido com alecrim pelos UAE ou CME foi microencapsulado e usado para substituir 50 % de gordura em hambúrgueres.

Na mesma linha, o quarto capítulo intitulado "Volatile compounds and sensory profile of burgers with 50 % fat replacement by microparticles of chia oil enriched with Rosemary" trará um artigo publicado na revista *Meat Science* em 2019 que trata da incorporação direta de alecrim no óleo de chia (CO) realizada por extração assistida por ultrassom (UAE) e extração por maceração convencional (CME). O CO microencapsulado foi utilizado em hambúrgueres e os compostos voláteis e as propriedades sensoriais (Check-All That-Apply e aceitação global) foram avaliadas nos dias 1 e 120 de armazenamento sob congelamento.

No quinto capítulo da tese será encontrado o artigo intitulado “Hydrogelled emulsion from chia and linseed oils: A promising strategy to produce low-fat burgers with a healthier lipid profile” publicado na revista *Meat Science* em 2019 e que traz a substituição de 0, 20, 40, 60, 80 e 100 % de toucinho por emulsão hidrogelificada (HE) dos óleos de chia e linhaça.

No sexto capítulo intitulado “Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: A green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils” publicado em 2020 na revista *Meat Science* foi realizada a inserção do extrato de casca de jabuticaba (JPE) obtido por extração de microondas por hidrodifusão e gravidade (MHG) em emulsão hidrogelificada e posterior aplicação em hambúrgueres.

Finalizando os capítulos desta tese, o sétimo capítulo publicado na revista *Meat Science* em 2021 intitula-se “Lipid oxidation and sensory quality of Omega-3 rich buffalo burgers enriched with chlorogenic acids from the mate (*Ilex paraguariensis*) tree harvesting residues”. Neste capitulo foi realizada a adição do extrato liofilizado da casca de ramos de erva-mate (BMBE) ao óleo de chia sob forma de emulsão hidrogelificada e usado para substituir 50 % de toucinho em hambúrgueres.

Ao final desta tese, encontram-se os itens discussão geral, conclusão geral e considerações finais a respeito dos resultados apresentados nos artigos científicos.

2. INTRODUÇÃO

O hambúrguer é um produto cárneo amplamente consumido em diversos países. Uma prova disto é que um único *fast food* vende anualmente mais de 100 bilhões de unidades ao redor do mundo com uma taxa de 75 hambúrgueres por segundo (SPENCER; FRANK; MCINTOSH, 2005). Como principais razões para este alto consumo, se pode citar as suas características sensoriais agradáveis, praticidade de consumo e alto teor proteico (RAMADHAN; HUDA; AHMAD, 2011). Apesar da legislação brasileira permitir a adição de uma infinidade de aditivos e ingredientes, os mais comumente encontrados em hambúrgueres brasileiros são condimentos (alho e cebola), proteínas de origem vegetal (principalmente proteínas derivadas da soja), aromatizantes, estabilizantes (fosfato e seus derivados), antioxidantes (ascorbato e seus derivados) e corantes naturais.

No entanto, a qualidade nutricional deste produto é questionada por especialistas da área da saúde, já que contém uma alta quantidade de gordura animal (até 30%). Além de aumentar o valor calórico, esta gordura animal também aumenta a concentração de ácidos graxos saturados (SFA) do produto. Assim, o consumo frequente de hambúrgueres pode aumentar a incidência de obesidade, doenças cardiovasculares e alguns tipos de câncer (KAEFERSTEIN; CLUGSTON, 1995). Além disso, pelo fato de possuir uma maior proporção de n-6 PUFAs do que n-3 PUFAs (VALENCAK et al., 2015), a gordura animal também aumenta a relação n-6/n-3 PUFAs. Este desequilíbrio nos teores de PUFAs pode ocasionar o surgimento de várias doenças crônicas (BEECHER, 1999).

Desta forma, a substituição de uma parte da gordura animal por óleos com altos teores de ácidos graxos ômega-3 pode conferir aos produtos cárneos características mais saudáveis, pois o consumo deste tipo de ácido pode reduzir os fatores de risco relacionados com o surgimento de doenças cardiovasculares (CHAUDHARY et al., 2016). Os óleos de chia e de linhaça são interessantes para essa substituição, uma vez que contêm cerca de 60 % de ácido α-linolênico em seu perfil lipídico (BODOIRA; PENCI; RIBOTTA; MARTÍNEZ, 2017; RUBILAR et al.; 2012). Alguns autores investigaram esses óleos, incluindo Gómez-Estaca et al. (2019) e Heck et al. (2017), que usaram óleo de chia e linhaça como substituto de gordura animal em produtos cárneos cozidos. Os produtos cárneos apresentaram uma alta relação PUFA / SFA e baixa relação n-6/ n-3, demonstrando assim que os óleos de chia e de linhaça podem ser uma boa alternativa para melhorar o perfil lipídico. No entanto, apesar dos potenciais benefícios para a saúde, alguns problemas tecnológicos e sensoriais foram relatados.

Alguns estudos sobre estratégias para aumentar a estabilidade oxidativa e prevenir a degradação térmica de óleos ricos em ácidos graxos n-3 já foram realizados. Uma das alternativas é utilizar a microencapsulação, que consiste basicamente na produção de micropartículas onde o material do núcleo é revestido com um agente microencapsulador (CHAMPAGNE; FUSTIER, 2007). Dentre as diversas técnicas de microencapsulação, a gelificação iônica externa utilizando alginato como agente microencapsulante permitiu a produção de micropartículas resistentes a altas temperaturas e com liberação controlada dos compostos ativos no intestino humano (HECK et al, 2017). O uso de óleos microencapsulados pode ser uma alternativa viável para o enriquecimento de produtos cárneos com ácidos graxos n-3. Outro método que tem mostrado bons resultados no aprisionamento de óleos vegetais é o da emulsão hidrogelificada (HE). HEs são sistemas semi-sólidos, nos quais uma fase líquida hidrofóbica é imobilizada em um ambiente tridimensional de sólidos lipofílicos (agentes gelificantes) (CO; MARANGONI, 2012). Entre as diferentes maneiras de preparar HE, as emulsões óleo-em-água (O / W) são as mais estudadas devido à relativa facilidade de preparo e baixo custo (SANDOVAL-SALCEDO et al., 2015). Além disso, a carragena usada como agente de gelificação é descrita na literatura como um importante agente para melhorar a estabilidade e a textura de produtos cárneos com baixo teor de gordura (AYADI; KECHAOU; MAKNI; ATTIA, 2009; LURUENA-MARTÍNEZ; VIVAR-QUINTANA; REVILLA, 2004).

Além de utilizar estratégias para reter os óleos ricos em ácidos graxos n-3, uma constante preocupação dos pesquisadores é em relação a estabilidade oxidativa destes óleos no interior da matriz cárnea. Uma alternativa seria o uso de antioxidantes naturais e, assim, possibilitar a aplicação desses óleos em alimentos com um perfil de ácidos graxos nutricionalmente desfavorável (FERNANDES; TRINDADE; LORENZO; DE MELO, 2018; MUNEKATA et al.; 2017). Os métodos mais comuns de extração de antioxidantes naturais empregam grandes quantidades de solventes tóxicos, como o etanol e o metanol (YARA-VARÓN; LI; BALCELLS; CANELA- GARAYOA; CHEMAT, 2017). As técnicas de extração verde foram desenvolvidas e otimizadas nos últimos anos. Neste contexto, a extração assistida por ultrassom (EAU) tem sido utilizada com sucesso para extração de compostos bioativos. As vantagens da EAU, quando comparadas aos métodos de extração por solvente, incluem economia de tempo e aumento da atividade antioxidante (ZBANCIOC; MANGALAGIU; MOLDOVEANU, 2015; ZHOU et al., 2016). A cavitação na superfície do material de origem pode levar a descamação superficial, erosão e quebra de partículas, aumentando assim o processo de transferência de massa (CHEMAT et al., 2017). A extração em micro-ondas por hidrodifusão e gravidade (MHG) é outro método eficaz para extrair compostos bioativos de

matrizes vegetais. Neste método a força da gravidade é utilizada para acelerar o processo de extração. De uma maneira geral, a extração por MHG consiste em colocar o material vegetal em um forno de micro-ondas e o seu aquecimento interno irá provocar o rompimento das células onde se encontram os óleos essenciais. Da mesma maneira que o método da EAU, o método de extração por MHG é conhecido como ambientalmente amigável, pois reduz a geração de resíduos e o consumo de energia e elimina ou reduz o uso de solventes tóxicos (CHEMAT et al., 2017).

Diante do exposto anteriormente, a inserção de óleos ricos em ácidos graxos n-3 na forma de microcápsulas ou estruturados como emulsão hidrogelificada pode efetivamente mimetizar a funcionalidade da gordura animal em produtos cárneos, com o diferencial de ter um perfil lipídico mais saudável (KOUZOUNIS; LAZARIDOU; KATSANIDIS, 2017). No entanto, há poucos relatos sobre o efeito que o tratamento térmico provoca no perfil de ácidos graxos dos óleos microencapsulados e hidrogelificados. Além disso, os efeitos que os óleos microencapsulados e hidrogelificados provocam no perfil sensorial de hambúrgueres são pouco explorados na literatura, o que dificulta estratégias para melhorar a qualidade sensorial desse tipo de produto. Por outro lado, a incorporação de antioxidantes naturais em óleos ricos em ácidos graxos n-3 através das técnicas de extração sem solvente EAU e MHG tem sido pouco explorada. Deste modo, a realização deste estudo é um importante passo para preencher lacunas e nortear a reformulação lipídica de produtos cárneos cozidos.

REFERÊNCIAS

AYADI, M.; KECHAOU, A.; MAKNI, I.; ATTIA, H. Influence of carrageenan addition on turkey meat sausages properties. *Journal of Food Engineering*, v. 93, n. 3, p. 278–283, 2009.

BEECHER, G. R. Phytonutrients' role in metabolism: Effects on resistance to degenerative processes. *Nutrition Reviews*, n. 57, p. 53–56, 1999.

BODOIRA R. M.; PENCI, M. C.; RIBOTTA, P.D.; MARTÍNEZ, M.L. Chia (Salvia hispanica L.) oil stability: study of the effect of natural antioxidants. LWT - *Food Science and Technology*, v. 75, p. 107–113, 2017.

CHAMPAGNE, C. P., & FUSTIER, P. Microencapsulation for the improved delivery of bioactive compounds into foods. *Current Opinion in Biotechnology*, v. 18, p. 184–190, 2007.

CHAUDHARY, R., SAADIN, K., BLIDEN, K. P., HARRIS, W. S., DINH, B., SHARMA, T., GURBEL, P. A. Risk factors associated with plasma omega-3 fatty acid levels in patients with suspected coronary artery disease. *Prostaglandins, Leukotrienes and Essential Fatty Acids*, v. 113, p. 40–45, 2016.

CHEMAT, F.; ROMBAUT, N.; MEULLEMIESTRE, A.; TURK, M.; PERINO, S.; FABIANO-TIXIER, A. S.; ABERT-VIAN, M. Review of Green Food Processing techniques. Preservation, transformation, and extraction. *Innovative Food Science and Emerging Technologies*, v. 41, 357–377, 2017.

CO, E. D.; MARANGONI, A. G. Organogels: An alternative edible oil-structuring method. *Journal of the American Oil Chemists Society*, v. 89, n. 5, p. 749–780, 2012.

FERNANDES, R. P. P.; TRINDADE, M. A.; LORENZO, J. M.; DE MELO, M. P. Assessment of the stability of sheep sausages with the addition of different concentrations of Origanum vulgare extract during storage. *Meat Science*, v. 137, p. 244–257, 2018.

GÓMEZ-ESTACA, J.; HERRERO, A. M.; HERRANZ, B.; ÁLVAREZ, M. D.; JIMÉNEZ-COLMENERO, F.; COFRADES, S. Characterization of ethyl cellulose and beeswax oleogels

and their suitability as fat replacers in healthier lipid pâtés development. *Food Hydrocolloids*, v. 87, p. 960–969, 2019.

HECK, R. T.; VENDRUSCOLO, R. G.; DE ARAÚJO ETCHEPARE, M.; CICHOSKI, A. J.; DE MENEZES, C. R.; BARIN, J. S.; LORENZO, J. M.; WAGNER, R.; CAMPAGNOL, P. C. B. Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science*, v. 130, p. 16–25, 2017.

KAEFERSTEIN, F. K.; CLUGSTON, G. A. Human health problems related to meat production and consumption. Fleischwirtschaft, v. 75, p. 889–892, 1995.

KOUZOUNIS, D.; LAZARIDOU, A.; KATSANIDIS, E. Partial replacement of animal fat by oleogels structured with monoglycerides and phytosterols in frankfurter sausages. *Meat Science*, v. 130, p. 38–46, 2017.

LURUENA-MARTÍNEZ, M. A.; VIVAR-QUINTANA, A. M.; REVILLA, I. Effect of locust bean/xanthan gum addition and replacement of pork fat with olive oil on the quality characteristics of low-fat frankfurters. *Meat Science*, v. 68, n. 3, p. 383–389, 2004.

MUNEKATA, P. E. S.; DOMINGUEZ, R.; FRANCO, D.; BERMÚDEZ, R.; TRINDADE, M. A.; LORENZO, J. M. Effect of natural antioxidants in Spanish salchichón elaborated with encapsulated n-3 long chain fatty acids in konjac glucomannan matrix. *Meat Science*, v. 124, p. 54–60, 2017.

RAMADHAN, K.; HUDA, N.; AHMAD, R. Physicochemical characteristics and sensory properties of selected Malaysian commercial chicken burgers. International Food Research Journal v. 18, p. 1349–1357, 2011.

RUBILAR, M.; MORALES, E.; CONTRERAS, K.; CEBALLOS, C.; ACEVEDO, F.; VILLARROEL, M.; SHENE, C. Development of a soup powder enriched with microencapsulated linseed oil as a source of omega-3 fatty acids. *European Journal of Lipid Science and Technology*, v. 114, n. 4, p. 423–433, 2012.

SANDOVAL-SALCEDO, L.; COFRADES, S.; RUIZ-CAPILLAS, C.; MATALANIS, A.; MCCLEMENTS, D. J.; DECKER, E. A.; JIMENEZ-COLMENERO, F. Oxidative stability of n-3 fatty acids encapsulated in filled hydrogel particles and of pork meat systems containing them. *Food Chemistry*, v. 184, p. 207–213, 2015.

SPENCER, E.H.; FRANK, E.; MCINTOSH, N. F. Potential effects of the next 100 billion hamburgers sold by McDonalds. *American Journal of Preventive Medicine*, v. 28, p. 379–381, 2005.

YARA-VARÓN, E.; LI, Y.; BALCELLS, M.; CANELA-GARAYOA, R.; FABIANO-TIXIER, A.-S.; CHEMAT, F. Vegetable oils as Alternative Solvents for Green Oleo-Extraction, Purification and Formulation of Food and Natural Products. *Molecules*, v. 22, n. 9, p. 1474, 2017.

VALENCAK, T. G.; GAMSJÄGER, L.; OHRNBERGER, S.; CULBERT, N. J.; RUF, T. Healthy n−6/n−3 fatty acid composition from five European game meat species remains after cooking. BMC Research Notes, v. 8, p. 273, 2015.

ZBANCIOC, G.; MANGALAGIU, I. I.; MOLDOVEANU, C. Ultrasound assisted synthesis of imidazolium salts: An efficient way to ionic liquids. *Ultrasonics - Sonochemistry*, v. 23, p. 376–384, 2015.

ZHOU, M.; LIU, J.; ZHOU, Y.; HUANG, X.; LIU, F.; PAN, S.; HU, H. Effect of high intensity ultrasound on physicochemical and functional properties of soybean glycinin at different ionic strengths. *Innovative Food Science and Emerging Technologies*, v. 34, p. 205–213, 2016.

3. OBJETIVOS

3.1 OBJETIVOS GERAIS

Avaliar a aplicação de óleos ricos em ácidos graxos n-3 na forma microencapsulada e hidrogelificada nas propriedades nutricionais, oxidativas, sensoriais e tecnológicas de hambúrgueres com reduzido teor de gordura.

3.2. OBJETIVOS ESPECIFICOS

- Avaliar o efeito da substituição de toucinho por microcápsulas de óleo de chia enriquecidas com compostos bioativos do alecrim através da extração assistida por ultrassom (EAU) e maceração convencional (CME) na qualidade oxidativa de hambúrgueres durante o armazenamento.
- Investigar as mudanças nos compostos voláteis e no perfil sensorial de hambúrgueres causados pela substituição de 50 % de toucinho por microcápsulas de óleo de chia enriquecido com alecrim por EAU e CME.
- Avaliar as propriedades físico-químicas, oxidativas, tecnológicas, nutricionais e sensoriais de hambúrgueres com substituição parcial e total de toucinho por emulsão hidrogelificada (HE) de óleo de chia e de linhaça.
- Investigar os parâmetros de qualidade, oxidação lipídica e perfil sensorial de hambúrgueres com substituição de 60 % de gordura animal por HE enriquecida com extrato de casca de jabuticaba obtido por MHG.
- Avaliar os principais atributos de qualidade tecnológica, oxidativa e sensorial de hambúrgueres com substituição de 50 % de toucinho por HE adicionada de extrato elaborado com resíduos da colheita de erva-mate.

4. CAPÍTULO 1- MICROENCAPSULATION OF HEALTHIER OILS: AN EFFICIENT STRATEGY TO IMPROVE THE LIPID PROFILE OF MEAT PRODUCTS

Rosane Teresinha Heck, José Manuel Lorenzo, Bibiana Alves Dos Santos, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Paulo Cezar Bastianello Campagnol

Current Opinion in Food Science
Vol. 40, p. 6-12, 2021
https://doi.org/10.1016/j.cofs.2020.04.010

Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Food Science

Microencapsulation of healthier oils: an efficient strategy to improve the lipid profile of meat products

Rosane Teresinha Heck[1], José Manuel Lorenzo[2], Bibiana Alves Dos Santos[1], Alexandre José Cichoski[1], Cristiano Ragagnin de Menezes[1] and Paulo Cezar Bastianello Campagnol[1]

Meat products have high SFAs levels and a high n-6/n-3 ratio, with negative effects on human health. The replacement of animal fat by oils rich in n-3 PUFAs is an efficient strategy to improve the lipid profile of meat products. However, the technological properties, oxidative stability, and sensory quality of meat products can be impaired by this lipid reformulation. Recently, microencapsulation has stood out as a promising tool to improve the oxidative stability of oils, allowing their application in meat products. Considering the growing interest in this topic among the population, this short-review aims to highlight the recent studies on the microencapsulation of healthier oils as a strategy to improve the lipid profile of meat products.

Addresses
[1] Universidade Federal de Santa Maria, CEP 97105-900, Santa Maria, Rio Grande do Sul, Brazil
[2] Centro Tecnológico de la Carne de Galicia, Parque Tecnológico de Galicia, San Cibrán das Viñas, Rúa Galicia N 4, Ourense, Spain

Corresponding author:
Campagnol, Paulo Cezar Bastianello (paulo.campagnol@ufsm.br)

Current Opinion in Food Science 2020, **40**:6–12

This review comes from a themed issue on **Innovations in food science**

Edited by **Jose Manuel Lorenzo**

https://doi.org/10.1016/j.cofs.2020.04.010

Introduction

Pork back fat is the main source of fat used in the manufacture of meat products. The level of addition depends on the type of meat product, which can reach up to 50% in extreme cases. The improvement in the sensory and technological quality of products and the reduction of production costs are the main reasons for the wide use of pork back fat in the meat industry. However, pork back fat has a high content (~20–28%) of palmitic acid (16:0) and a high n-6 polyunsaturated fatty acids (PUFAs)/ n-3 PUFAs (n-6/n-3) ratio (~10–17) [1•,2]. Therefore, excessive consumption of meat products with a high content of pork back fat may increase risk factors related to the onset of cardiovascular diseases [3–5].

Several studies have been carried out in an attempt to improve the nutritional quality of meat products by modifying their fatty acid profile through the incorporation of healthier oils [6–11]. An effective approach is to partially replace pork back fat with oils rich in n-3 PUFAs [12–17]. The main advantage of this approach is the increase in the PUFA/saturated fatty acids ratio (PUFA/SFA) and the reduction of n-6/n-3 ratio [18–22]. However, the technological and sensory quality of the final product is dramatically impaired when liquid oils are used in the formulations. In addition, liquid oils are quickly oxidized during the processing of meat products, which brings double damage to the consumers' health since besides degrading the n-3 PUFAs, the oxidation can also generate toxic products [23].

Microencapsulation is a physical barrier to protect bioactive compounds from the degradation reactions [24]. This technology can be considered one of the most promising alternatives to increase oxidative stability and prevent the thermal degradation of oils rich in n-3 PUFAs. In this context, healthier oils can be applied to meat products that have a nutritionally unfavorable fatty acid profile. Oil microparticles have a functional quality similar to pork back fat, which is important for maintaining the technological properties of the product [25,26]. Although various microencapsulation techniques have been used worldwide, the selection of a method that best adapts to the specific conditions of each food is required. Concerning the cooked meat products, microparticles should be resistant to high temperatures, which is not necessary for raw cured meat products. The microparticles should also resist the pH and moisture conditions of the product, and must breakdown when passing through the human gut.

Based on the growing interest in the incorporation of n-3 PUFAs in meat products, the main objective of this short review is to highlight and summarize the recent studies on the microencapsulation of healthier oils as a strategy to improve the lipid profile of meat products.

Microencapsulation techniques used for oils rich in n-3 PUFAs

Microencapsulation has emerged as a new strategy for the protection of n-3 PUFAs against lipid oxidation. For the formation of microparticles, oil droplets embedded in a homogeneous or heterogeneous matrix are dispersed in a medium, leading to the formation of a physical barrier between the oil and the environment, reducing contact with oxidizing agents [27]. The main techniques used to produce healthier oil microparticles used in meat products include spay-drying, freeze-drying, complex coacervation, and external ionic gelation (extrusion). In this short review, some considerations about these microencapsulation techniques will be briefly presented and discussed to understand the mechanisms and the processes involved. More details about other techniques (emulsification, coaxial electrospray system, coacervation, *in situ* polymerization, fluidized-bed-coating, and supercritical fluid technology) also used for the microencapsulation of marine, vegetable, and essential oils can be consulted in the review of Bakry *et al.* [28].

The encapsulation by spray-drying has been the most used technique, probably due to the low cost of processing, as well as the fast and continuous process [29]. Another advantage of this encapsulation technique is the small size of the particles produced (<40 μm), with positive effects on the texture of food products, besides the production of dry particles that can readily be used in food [30•]. The spray-drying technique involves the dispersion of core material (oils or other materials) into a polymeric solution, forming an emulsion or dispersion, which is atomized into a medium and dehydration of particles to produce the microparticles [31]. Studies to enrich meat products with n-3 PUFAs have evaluated the fish oil microparticles produced by spray-drying [32]. However, the use of microparticles produced by spray-drying in cooked meat products is not recommended, as the wall materials commonly used, such as gum Arabic, maltodextrin and modified starch, disintegrate when subjected to temperatures above 50°C. In contrast, spray-dried microparticles can be successfully used in raw meat products since this technique is very effective in protecting bioactive compounds.

The freeze-drying process uses low temperatures and consists of freezing the samples followed by vacuum dehydration (removal of ice by sublimation), until the formation of a stable freeze-dried material [33]. This procedure can be used for oils containing high levels of n-3 PUFAs, thus preserving their nutritional quality since it can be performed at low temperatures. Freeze-drying is indicated for the production of microparticles for use in a variety of food products, such as bread, instant foods, yogurt, cheese, butter, and cream. In contrast, the complex coacervation method uses an oil-in-water emulsion containing a biopolymer solution. In complex coacervation, there is an interaction between two oppositely charged polymers (wall material), with the formation of a complex around droplets or particles of the active material (filling), leading to the formation of microparticles [34]. The microparticles produced by freeze-drying or complex coacervation techniques cannot be used in cooked meat products, as they can rupture at temperatures below the cooking point of meat products (72°C), especially when using gelatin as a coating material.

The microparticles produced by the external ionic gelation are formed by a suspension of core particles in the coating liquid, which passes through a two-fluid atomizing nozzle, resulting in a thin-coated film [35]. Unlike previous techniques, this technique allows heating without compromising the structure of the microcapsule and can be used in cooked meat products. Further details on the principle of the different microencapsulation techniques can be found in the reviews reported by Feizollahi *et al.* [36] and Pérez-Palacios *et al.* [37].

Application of healthier oil microparticles in meat products

Recently, studies have used different techniques to encapsulate oils rich in n-3 PUFAs for application in meat products (Table 1). An interesting approach was proposed by Domínguez *et al.* [38] who produced frankfurter sausages with reduced-fat and a healthier fatty acid profile by replacing 50% of pork back fat with microencapsulated fish oil by spray-drying and a non-encapsulated mixture of olive oil and fish oil (1:1). As expected, the lipid reformulation increased monounsaturated fatty acids (MUFAs) and n-3 PUFAs levels. However, surprisingly, the samples with microencapsulated fish oil showed higher lipid oxidation when compared to the samples with the non-encapsulated mixture of olive oil and fish oil. It is worth noting that the sausages were pasteurized at 90°C for 30 min, which may have led to the breakdown of microparticles and the consequent oxidation of fish oil. In a similar study, Lorenzo *et al.* [22] evaluated the effect of the partial replacement (25, 50 and 75%) of pork back fat by fish oil microencapsulated by spray-drying on physico-chemical properties, lipid oxidation and fatty acid profile of Spanish *salchichón*. The incorporation of microencapsulated fish oil increased the eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) content and decreased the n-6/n-3 PUFA ratio. However, the batches where pork back fat was replaced with microencapsulated fish oil showed higher TBARs values, reflecting an increase of the lipid oxidation. Keenan *et al.* [39] also reported higher lipid oxidation index in beef burgers with replacement of animal fat by microencapsulated fish oil. In addition, the authors reported a higher degree of oxidation for burgers containing microencapsulated fish oils compared to their unencapsulated counterparts, which was possibly related to oxygen exposure during the encapsulation process, temperature of drying process, susceptibility of fish PUFAs to heat, and the large surface to oxygen contact. These results demonstrate the difficulty of

Table 1

Recent studies on the microencapsulation of healthier oils as a strategy to improve the lipid profile of meat products

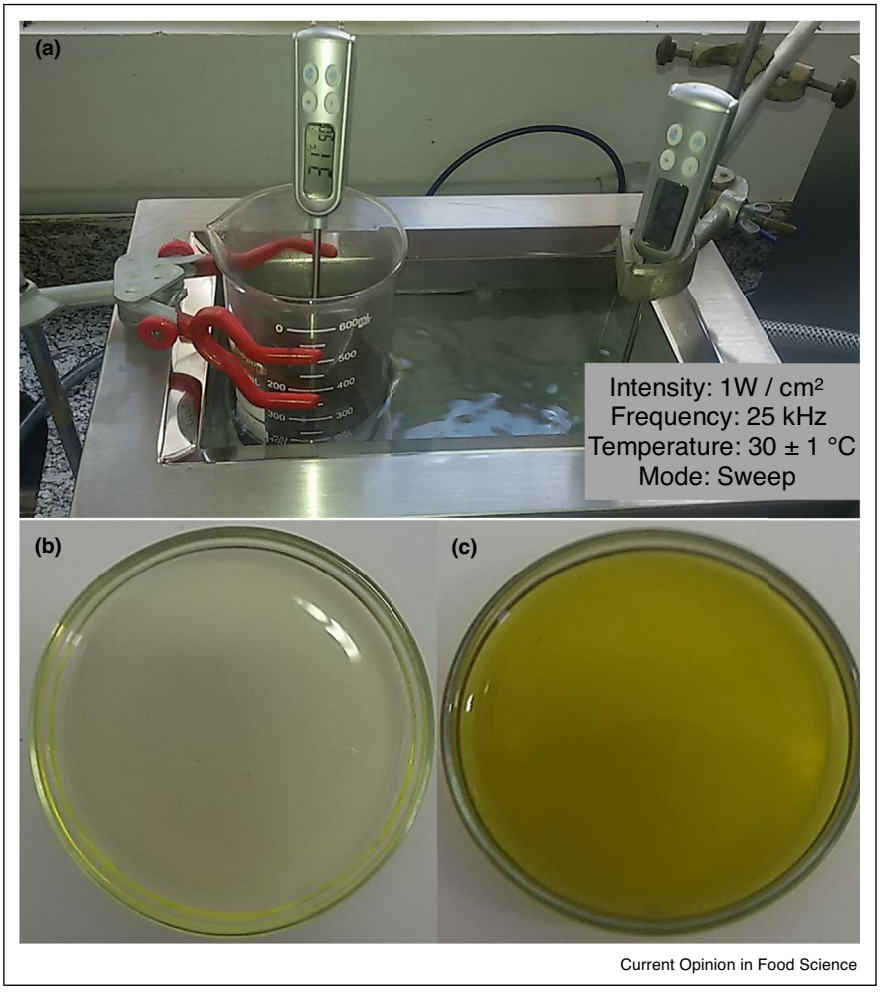
Oil	Technique of microencapsulation	Meat product	Level used	Main results	Ref.
Fish	Spray-drying	Frankfurter sausages	50% fat replacement	• The lipid reformulation increased MUFAs and n-3 PUFAs levels. • The microencapsulation process increased the lipid oxidation. • The microencapsulated oil batch presented the highest TBARS values and volatile compounds derivate from lipid oxidation.	[38]
Fish	Spray-drying	Spanish *salchichón*	25, 50 and 75% fat replacement	• Increased EPA and DHA content. • Decreased the n-6/n-3 PUFAs ratio. • Higher TBARS values, reflecting an increase of the lipid oxidation.	[22]
Fish	Spray-drying	Burger	$\leq$15% fat replacement	• TBARS values increased over storage. • Optimization predicted a burger formulation with 7.8% substitution in beef-fat with encapsulated fish oil. • Panelists scored the optimized burger formulation lower than controls for overall acceptability.	[39]
Fish	Spray-drying	Chicken nuggets	50% fat replacement	• Nuggets with microencapsulated fish oil did not differ from control ones with respect to any sensory trait. • Higher levels of lipid and protein oxidation indicators and of volatile compounds from fatty acid oxidation were found in microencapsulated fish oil samples.	[32]
Fish	Spray-drying	Burger	5% Enrichment	• After storage and cooking, EPA and DHA were better preserved in the burgers with microencapsulated fish oil. • The encapsulated fish oil group showed the best scores with respect to the control.	[40]
Fish	Spray-drying	Dry-cured sausages	2.75% and 5.26 % Enrichment	• Cooked and dry-cured products susceptible to be labeled as 'source of omega-3 fatty acids'. • Without influencing physico-chemical characteristics, oxidative stability, and acceptability.	[41•]
Chia and linseed	External ionic gelation	Burger	50% fat replacement	• Decrease fat content of the burgers by up to 50%. • Increase of up to 90% in the PUFA/SFA ratio. • Decrease of up to 18.5 and 14.7% in the atherogenicity and thrombogenicity indices, respectively. • Modified burgers presented a healthy n-6/n-3 PUFAs ratio. • Higher lipid oxidation in burgers containing microencapsulated chia oil. • Replacement of 50% pork back fat by linseed oil microparticles did not affect the sensory quality of the burgers.	[42•]
Tiger nut, chia and linseed	Spray-drying	Deer pâté	50% fat replacement	• Decrease in fat and cholesterol contents. • Decreasing the total amount of SFAs and increasing PUFAs (chia and linseed pâtés) or MUFAs contents (tiger nut pâtés). • Pâtés prepared with oils that contained high n-3 PUFAs levels displayed higher TBARS values.	[1•]
Chia enriched with rosemary	External ionic gelation	Burger	50% fat replacement	• Higher Eh and TBARS values were found in burgers containing chia oil microparticles without rosemary. • The burgers produced with chia oil microparticles enriched with rosemary showed greater oxidative stability than other treatments, mainly after cooking. • Groups enriched with rosemary presented an increase in terpenic volatiles and were characterized by the descriptors herbal and pleasant aroma and ideal texture.	[44•,45]

incorporating oils rich in n-3 PUFAs in meat products, especially those subjected to intense heat treatment.

Pérez-Palacios *et al.* [32] also used fish oil to improve the lipid profile of chicken nuggets. The authors compared the oxidative stability between non-encapsulated and spray-dried fish oil and reported that the treatments with the addition of microencapsulated fish oil showed less lipid and protein oxidation during storage when compared to treatments with non-encapsulated oil. Similarly, Aquilani *et al.* [40] studied non-encapsulated and spray-dried fish oil as a fat substitute in pork burgers. The authors compared the changes in EPA and DHA levels during the storage of the burgers under refrigeration and freezing. The results showed that microencapsulation was effective to prevent EPA and DHA oxidation in both refrigerated and frozen burgers. In addition, higher TBARs values were found for the samples with non-encapsulated oil, while the samples with microencapsulated oil showed values similar to the control (made with pork back fat). Another interesting approach to fish oil encapsulation has been proposed by González *et al.* [41•]. Those authors encapsulated fish oil by spray-drying using different coating materials (lecithin + maltodextrin and lecithin + chitosan + maltodextrin) and applied in cooked and raw cured meat products. As expected, the addition of the microparticles increased EPA and DHA levels, enabling the products to be labeled as a 'source of n-3 PUFAs'. However, the most curious result was that microencapsulation masked the flavor and aroma of fish oil, and also protected the oil from oxidative reactions. This approach solved two of the main problems that hinder the application of fish oil in meat products and contributed to further studies on this subject.

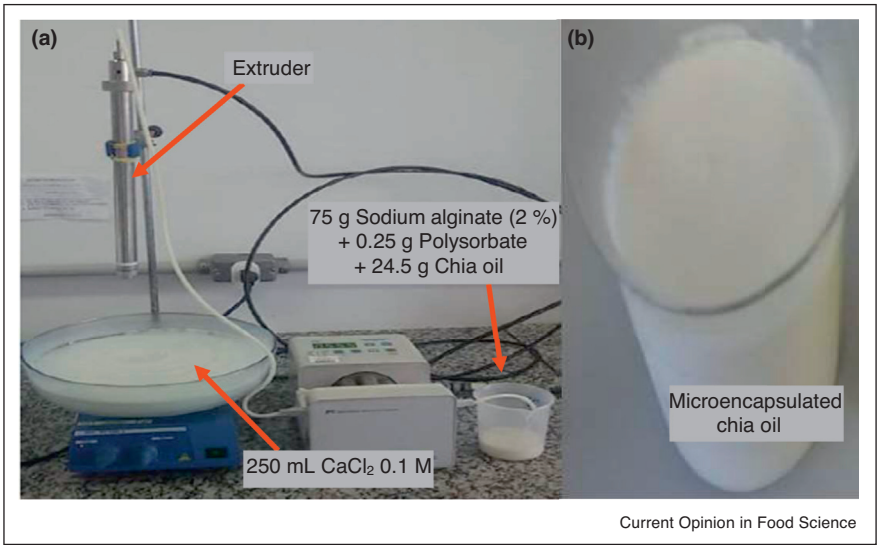
However, those authors did not report the thermal stability of the microparticles, which does not allow concluding whether n-3 PUFAs remained inside the microparticles

Figure 1



(a) Direct incorporation of rosemary into chia oil by ultrasound. (b) Chia oil. (c) Chia oil with rosemary.

Figure 2



(a) Production of microparticles by the external ionic gelation technique. (b) Rosemary enriched chia oil microparticles.

after cooking. In addition, for the health benefit of n-3 PUFAs, it is necessary for microparticles to rupture in the human intestine. In this context, Heck *et al.* [42•] were the pioneers in evaluating the effect of temperature and pH on the stability of chia and linseed oil microparticles produced by external ionic gelation. The authors proved that the microparticles remained intact after being subjected to 72°C, with no disruption at the pH values commonly found in most meat products (4.5 and 6.0). However, at pH 7.5, the microparticles ruptured after 180 min, which suggests that the oils would only be released during passage through the human intestine. The authors used microparticles as a substitute for 50% of pork back fat in burgers. Besides contributing to the fat reduction in burgers, this strategy increased the PUFA/SFA ratio and decreased the n-6/n-3 ratio and the atherogenicity and thrombogenicity indices. However, microencapsulation was not able to completely protect chia oil from lipid oxidation reactions, which impaired the sensory and nutritional quality. In agreement with these results, Ramella *et al.* [1•] studied the addition of chia and linseed oil microparticles in deer pâté, and also found greater lipid oxidation, which was observed in the treatments containing the chia oil microparticles, probably due to the higher content of n-3 PUFAs in chia oil when compared to linseed oil [43].

An innovative approach to improve the oxidative stability of microparticles containing oils rich in n-3 PUFAs was proposed by Heck *et al.* [44•,45]. The chia oil was enriched with bioactive compounds from rosemary through green extraction techniques before the microencapsulation process (Figures 1 and 2). Then, the microparticles with rosemary-enriched chia oil were used in burger formulations, and the oxidative stability and the sensory quality were evaluated. Higher TBARS values were found in burgers containing chia oil microparticles without the presence of rosemary. In contrast, burgers with the addition of microparticles containing rosemary-enriched chia oil showed greater oxidative stability, especially after cooking. In addition, the incorporation of rosemary antioxidants into chia oil reduced the sensory defects caused by the lipid reformulation. Thus, these results demonstrate that the combination of healthy oils and natural antioxidants is an effective way to improve the oxidative stability of microparticles, enabling the production of healthier and safe meat products [46,47].

Conclusions and future directions

Microencapsulation can be considered an effective tool to incorporate healthy oils in meat products. However, it cannot completely protect oils rich in n-3 PUFAs from the oxidative reactions. Thus, the direct incorporation of natural antioxidants in these oils before microencapsulation may be a promising alternative to be explored in future studies. In addition, future research is needed to investigate the integrity of microparticles when subjected to the same pH and temperature conditions of meat processing, which is essential to ensure the breakdown of microparticles only in the human intestine. Another challenge is the development of more economically viable microparticles, as a major obstacle to this technology is the high cost of equipment.

Conflict of interest statement

Nothing declared.

Acknowledgements

National Council for Scientific and Technological Development (CNPq) (grant number 425577/2018-0), and Coordination for the Improvement of Higher Education Personnel (CAPES finance code 001) have supported this work. P.C.B Campagnol and J.M. Lorenzo are members of the HealthyMeat network, funded by CYTED (ref. 119RT0568).

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest

1. • Ramella MV, Pateiro M, Barba FJ, Franco D, Campagnol PCB, Munekata PES, Tomasevic I, Domínguez R, Lorenzo JM: **Microencapsulation of healthier oils to enhance the physicochemical and nutritional properties of deer pâté**. *LWT - Food Sci Technol* 2020, **125**:109223

Recent article showing that microencapsulation cannot completely protect oils rich in n-3 PUFAs from the oxidative reactions.

2. Jiang J, Xiong YL: **Natural antioxidants as food and feed additives to promote health benefits and quality of meat products: a review**. *Meat Sci* 2017, **120**:107-117.

3. Boada LD, Henríquez-Hernández LA, Luzardo OP: **The impact of red and processed meat consumption on cancer and other health outcomes: epidemiological evidences**. *Food Chem Toxicol* 2016, **92**:236-244.

4. Klurfeld DM: **Research gaps in evaluating the relationship of meat and health**. *Meat Sci* 2017, **109**:86-95.

5. Hartmann GL, Teixeira F, Soares JM, Silva KA: **Effect of fat replacement by fructooligosaccharide in hamburger: physicochemical, technological and sensorial analysis**. *Int J Innov Educ Res* 2020, **8**:15-25.

6. Afshari R, Hosseini H, Khaneghah AM, Khaksara R: **Physico–chemical properties of functional low–fat beef burgers: fatty acid profile modification**. *LWT - Food Sci Technol* 2017, **78**:325-331.

7. Torquato AS, Silva–Buzanello RA, Canan C, Bittencourt PRS, Murakami AE, Gonçalves TR, Matsushita MK: **Fatty acid profile of meat from broiler chickens fed with different oil sources**. *Arc Zool* 2018, **67**:532-540.

8. Da Silva SL, Amaral JT, Ribeiro M, Sebastião EE, Vargas C, De Lima FF, Schneider G, Lorenzo JM, Fries LLM, Cichoski AJ, Campagnol PCB: **Fat replacement by oleogel rich in oleic acid and its impact on the technological, nutritional, oxidative, and sensory properties of Bologna–type sausages**. *Meat Sci* 2019, **149**:141-148.

9. Renna M, Brugiapaglia A, Zanardi E, Destefanis G, Prandini A, Moschini M, Sigolo S, Lussiana C: **Fatty acid profile, meat quality and flavour acceptability of beef from double–muscled Piemontese young bulls fed ground flaxseed**. *Ital J Anim Sci* 2019, **18**:355-365.

10. Franco D, Martins AJ, López-Pedrouso M, Cerqueira MA, Purriños L, Pastrana LM, Lorenzo JM: **Evaluation of linseed oil oleogels to partially replace pork backfat in fermented sausages**. *J Sci Food Agric* 2020, **100**:218-224.

11. Vargas FC, Gómez B, Mousavi AK, Strozzi I, Gavahian M, Barba FJ, Lorenzo JM: **Assessment of the suitability of pitanga leaf extract as a natural antioxidant for enhancing canola oil stability: monitoring lipid oxidation parameters**. *European J Lipid Sci Technol* 2019, **121**:1800447.

12. Domínguez R, Pateiro M, Agregán R, Lorenzo JM: **Effect of the partial replacement of pork backfat by microencapsulated fish oil or mixed fish and olive oil on the quality of frankfurter type sausage**. *J Food Sci Technol* 2017, **54**:26-37.

13. De Carvalho FAL, Munekata PE, Pateiro M, Campagnol PC, Domínguez R, Trindade MA, Lorenzo JM: **Effect of replacing backfat with vegetable oils during the shelf-life of cooked lamb sausages**. *LWT - Food Sci Technol* 2020, **122**:109052.

14. Carvalho JB, Munekata PE, De Carvalho FAL, Pateiro M, Barba FJ, Domínguez R, Lorenzo JM: **Use of tiger nut (*Cyperus esculentus* L.) oil emulsion as animal fat replacement in beef burgers**. *Foods* 2020, **9**:44.

15. De Carvalho FAL, Lorenzo JM, Pateiro M, Bermúdez Purriños L, Trindade MA: **Effect of guarana (*Paullinia cupana*) seed and pitanga (*Eugenia uniflora* L.) leaf extracts on lamb burgers with fat replacement by chia oil emulsion during shelf life storage at 2 °C**. *Food Res Int* 2019, **125**:108554.

16. Munekata PES, Domínguez R, Franco D, Bermúdez R, Trindade MA, Lorenzo JM: **Effect of natural antioxidants in Spanish salchichón elaborated with encapsulated n-3 long chain fatty acids in konjac glucomannan matrix**. *Meat Sci* 2017, **124**:54-60.

17. Domínguez R, Pateiro M, Munekata PES, Campagnol PCB, Lorenzo JM: **Influence of partial pork backfat replacement by fish oil on nutritional and technological properties of liver pate**. *Eur J Lipid Sci Technol* 2016, **119** FALTA PG.

18. Fagundes DTO, Lorenzo JM, Dos Santos BA, Fagundes MB, Heck RT, Cichoski AJ, Campagnol PCB: **Pork skin and canola oil as strategy to confer technological and nutritional advantages to burgers**. *Czech J Food Sci* 2017, **35**:352-359.

19. Gómez–Estaca J, Herrero AM, Herranz B, Álvarez MD, Jiménez–Colmenero F, Cofrades S: **Characterization of ethyl cellulose and beeswax oleogels and their suitability as fat replacers in healthier lipid pâtés development**. *Food Hydrocoll* 2019, **87**:960-969.

20. Heck RT, Saldaña E, Lorenzo JM, Correa LP, Fagundes MB, Cichoski AJ, Menezes CR, Wagner R, Campagnol PCB: **Hydrogelled emulsion from chia and linseed oils: a promising strategy to produce low–fat burgers with a healthier lipid profile**. *Meat Sci* 2019, **156**:174-182.

21. Martins AJ, Vicente AA, Pastrana LM, Cerqueira MA: **Oleogels for development of health–promoting food products**. *Food Sci Hum Wellness* 2020, **9**:31-39.

22. Lorenzo JM, Munekata PES, Pateiro M, Campagnol PCB, Domínguez R: **Healthy Spanish salchichón enriched with encapsulated n− 3 long chain fatty acids in konjac glucomannan matrix**. *Food Res Int* 2016, **89**:289-295.

23. Domínguez R, Pateiro M, Gagaoua M, Barba FJ, Zhang W, Lorenzo JM: **A comprehensive review on lipid oxidation in meat and meat products**. *Antioxidants* 2019, **8**:429.

24. Gómez B, Barba FJ, Domínguez R, Putnik P, Kovačević DB, Pateiro M, Lorenzo JM: **Microencapsulation of antioxidant compounds through innovative technologies and its specific application in meat processing**. *Trends Food Sci Technol* 2018, **82**:135-147.

25. Fernandes RPP, Trindade MA, Lorenzo JM, De Melo MP: **Assessment of the stability of sheep sausages with the addition of diff ;erent concentrations of *Origanum vulgare* extract during storage**. *Meat Sci* 2018, **137**:244-257.

26. Kouzounis D, Lazaridou A, Katsanidis E: **Partial replacement of animal fat by oleogels structured with monoglycerides and phytosterols in frankfurter sausages**. *Meat Sci* 2017, **130**:38-46.

27. Rios–Mera JD, Saldaña E, Ramírez Y, Auquiñivín EA, Alvim ID, Contreras–Castillo CJ: **Encapsulation optimization and pH– and temperature–stability of the complex coacervation between soy protein isolate and inulin entrapping fish oil**. *LWT - Food Sci Technol* 2019, **116**:108555.

28. Bakry AM, Abbas S, Ali B, Majeed H, Abouelwafa MY, Mousa A, Liang L: **Microencapsulation of oils: a comprehensive review of benefits, techniques, and applications**. *Compr Rev Food Sci Food Saf* 2016, **15**:143-182.

29. Franco D, Antequera T, Pinho SCD, Jiménez E, Pérez-Palacios T, Fávaro-Trindade CS, Lorenzo JM: **The use of microencapsulation by spray-drying and its aplication in meat products**. *Strat Obtain Health Foods* 2017:334-362.

30. • Comunian TA, Favaro–Trindade CS: **Microencapsulation using biopolymers as an alternative to produce food enhanced with phytosterols and omega–3 fatty acids: a review**. *Food Hydrocoll* 2016, **61**:442-457

This study highlights the different microencapsulation methods, with emphasis on the microencapsulation of n-3 PUFAs and unstable bioactive compounds.

31. Martínez JA, González MJ, Miranda MA, Carrillo S, Castillo R: **Sustitución De Aceite De Soya Por Aceite De Atún En La Dieta De Pollos Como Alternativa Para Enriquecer La Carne Con Ácidos Grasos Omega–3**. *Interciencia* 2016, **41**:851-856.

32. Pérez-Palacios T, Ruiz–Carrascal J, Jiménez–Martín E, Solomando JC, Antequera T: **Improving the lipid profile of ready–to–cook meat products by addition of omega–3 microcapsules: effect on oxidation and sensory analysis**. *J Sci Food Agric* 2018, **98**:5302-5312.

33. Hoz L, D'Arrigo M, Cambero I, Ordóñez JA: **Development of an n–3 fatty acid and αtocopherol enriched dry fermented sausage**. *Meat Sci* 2004, **67**:485-495.

34. Zuanon LAC, Malacrida CR, Telis VRN: **Production of turmeric oleoresin microcapsules by complex coacervation with gelatin–gum arabic**. *J Food Proc Eng* 2013, **36**:364-373.

35. Jiménez–Martín E, Antequera–Rojas T, Gharsallaoui A, Ruiz Carrascal J, Pérez–Palacios T: **Fatty acid composition in double and multilayered microcapsules of ω–3 as aff ;ected by storage conditions and type of emulsions**. *Food Chem* 2016, **194**:476-486.

36. Feizollahi E, Hadian Z, Honarvar Z: **Food fortification with omega–3 fatty acids, microencapsulation as an addition method**. *Curr Nutr Food Sci* 2018, **14**:90-103.

37. Pérez–Palacios T, Ruiz–Carrascal J, Solomando JC, Antequera T: **Strategies for enrichment in ω–3 fatty acids aiming for healthier meat products**. *Food Rev Int* 2019, **35**:485-503.

38. Domínguez R, Pateiro M, Agregán R, Lorenzo JM: **Effect of the partial replacement of pork backfat by microencapsulated fish oil or mixed fish and olive oil on the quality of frankfurter type sausage**. *J Food Sci Technol* 2017, **54**:26-37.

39. Keenan DF, Resconi VC, Smyth TJ, Botinestean C, Lefranc C, Kerry JP, Hamill RM: **The effect of partial-fat substitutions with encapsulated and unencapsulated fish oils on the technological and eating quality of beef burgers over storage**. *Meat Sci* 2015, **107**:75-85.

40. Aquilani C, Pérez–Palacios T, Sirtori F, Jiménez–Martín E, Antequera T, Franci O, Acciaioli A, Bozzi R, Pugliese C: **Enrichment of Cinta Senese burgers with omega–3 fatty acids. Effect of type of addition and storage conditions on quality characteristics**. *Grasas Y Aceites* 2018, **69**:235.

41. González JCS, Antequera T, Pérez–Palacios T: **Evaluating the use of fish oil microcapsules as omega–3 vehicle in cooked and dry–cured sausages as affected by their processing, storage and cooking**. *Meat Sci* 2020, **162**:108031
The microencapsulation process proposed in this study was effective to reduce the sensory and oxidative defects caused by fish oil. This approach can be used in other meat products.

42. Heck RT, Vendruscolo RG, De Araújo Etchepare M, Cichoski AJ, De Menezes CR, Barin JS, Lorenzo JM, Wagner R, Campagnol PCB: **Is it possible to produce a low–fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties?** *Meat Sci* 2017, **130**:16-25
The authors used the external ionic gelation technique to microencapsulate oils rich in n-3 PUFAs. This approach can be applied to other cooked meat products.

43. Ghena MM, Amany MB: **Chia (*Salvia Hispanica L.*) seed oil a new source of omega–3**. *Plant Arch* 2020, **20**:2678-2683.

44. Heck RT, Lucas BN, Dos Santos DJP, Pinton MB, Fagundes MB, Etchepare MA, Cichoski AJ, Menezes CR, Barin JS, Wagner R, Campagnol PCB: **Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green–extraction techniques**. *Meat Sci* 2018, **146**:147-153
The article presents an interesting approach to improving the oxidative stability of healthier oil microparticles by the addition of natural antioxidants.

45. Heck RT, Fagundes MF, Cichoski AJ, De Menezes CR, Barin JS, Lorenzo JM, Wagner R, Campagnol PCB: **Volatile compounds and sensory profile of burgers with 50 % fat replacement by microparticles of chia oil enriched with rosemary**. *Meat Sci* 2018, **148**:164-170.

46. Munekata PES, Rocchetti G, Pateiro M, Lucini L, Domínguez R, Lorenzo JM: **Addition of plant extracts to meat and meat products to extend shelf-life and health-promoting attributes: an overview**. *Curr Opin Food Sci* 2020, **31**:81-87.

47. Granato D, Barba FJ, Kovačević DB, Lorenzo JM, Cruz AG, Putnik P: **Functional foods: product development, technological trends, efficacy testing, and safety**. *Ann Rev Food Sci Technol* 2020, **11**:93-118.

5. CAPÍTULO 2: REPLACEMENT OF SATURATED FATTY ACIDS BY HEALTHY OILS TO IMPROVE THE NUTRITIONAL QUALITY OF MEAT PRODUCTS

Rosane Teresinha Heck, Bibiana Alves Dos Santos, José Manuel Lorenzo, Claudia Ruiz-Capillas, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Paulo Cezar Bastianello Campagnol

Food lipids: sources, health implications, and future trends
Chapter 18, Ed. Elsevier, 2022, In press

Abstract

In general, meat products usually have an unfavorable lipid profile associated with health implications. The replacement of animal fats (especially saturated fatty acids) by healthy oils (rich in polyunsaturated fatty acid) may be an efficient strategy to improve the nutritional quality of meat products. However, the addition of healthy oils to the formulations can negatively affect the technological, oxidative, and sensory quality and reduce the shelf life of meat products. Thus, strategies for incorporating healthy oils should be studied with caution. Considering the growing interest in this issue, this chapter will assess the recent strategies used to incorporate healthy oils in meat products and the health effect of different types of fatty acids and the nutritional quality of the main healthy oils used in meat product formulations.

Keywords: saturated fatty acid (SFA); monounsaturated fatty acids (MUFA); polyunsaturated fatty acid (PUFA); microencapsulation; gelation; bioactive compounds; healthier meat products, nutritional quality, healthy oil.

1 Introduction

The fat content of most processed meat products is greater than 30 %, which represents a high amount of saturated fatty acids (SFA) in these products. Several studies have shown an association between a diet rich in SFA and the incidence of obesity, cardiovascular disease, and some types of cancer (Boada, Henríquez-Hernández, & Luzardo, 2016). The consumption of monounsaturated fatty acids (MUFA), among which oleic acid stands out due to its high prevalence, has also been related to a reduction in LDL cholesterol and triglycerides in the blood. However, recently there are a controversy in the real effect of the SFA and MUFA in human health (Liu et al., 2017; Ruiz-Capillas and Herrero, 2021).

Meat products also have a high ratio of dietary n-6/n-3 polyunsaturated fatty acids (PUFA) (15-20:1), which is nutritionally unfavorable for human health (Wood et al., 2004). Thus, frequent consumption of meat products can lead to inflammation and the appearance of several chronic diseases (Klurfeld, 2017).

Therefore, improving the lipid profile of meat products is necessary to meet the demands of health-conscious consumers. The replacement of animal fat, mainly SFA, by healthy oils is one of the most effective approaches to increase MUFA and the PUFA/SFA ratio and to reduce the n-6/n-3 PUFA ratio. Traditionally, the incorporation of this healthy oil for the development of a healthier lipid meat product has been done by adding it directly in liquid form. However, the use of oils liquid can negatively affect important technological and sensory attributes of meat products, as well as reducing their shelf life due to increased lipid oxidation (Triki, Herrero, Rodríguez-Salas, Jimenez-Colmenero, & Ruiz-Capillas, 2013).

In order to avoid this important problem, different techniques have been tried (pre-emulsion, emulsion gelled, microencapsulation, etc.) (Jimenez-Colmenero *et al.,* 2015; Herrero and Ruiz-Capillas, 2021). The microencapsulation technique is one of the available alternatives to protect oils from lipid oxidation, which basically consists of the production of microparticles, in which the core material is coated with an encapsulating agent (Champagne & Fustier, 2007). Gelation is another strategy that improves the oxidative stability of oils. This technique retains and locks oil and lead to a reduction of fat migration and control phase separation (water and oil), providing properties similar to solid fats. The kinds of gels formed depends on the polarity of the liquid phase, and can be classified as hydrogels, emulgels, and oleogels/organogels (Jimenez-Colmenero *et al.,* 2015). Hydrogels are formed when water is used as the continuous phase, while emulgel corresponds to the formation of a biphasic emulsion (Dickinson, 2012). In turn, oleogels/organogels are formed from the dispersion and structuring of the oil by an organogelator (Balasubramanian, Damodar, & Sughir, 2012).

Heck et al. (2017, 2019) and Gómez-Estaca et al. (2019) used microencapsulation and gelation techniques to incorporate oils rich in n-3 PUFA (polyunsaturated fatty acids) in meat products, aiming at the use of natural products and healthy ingredients in the food industry. The results showed that the

application of these oils was effective to nutritionally improve meat products. However, the use of healthy oils should be carried out with caution even for microencapsulated or gelled oils, once these techniques do not completely protect PUFA from lipid oxidation. Recently, new strategies have been studied to improve the oxidative stability of healthy oils applied to meat products. Promising results have been reported by some authors on the incorporation of bioactive compounds in healthy oils through emerging green extraction techniques, which have been developed and optimized in recent years (Heck et al. 2019, 2020; Pintado et al., 2021).

In this context, this chapter will present the health benefits of reducing SFA and increasing PUFA intake. The fatty acid composition of the main oils used in meat products will also be presented. In addition, the main approaches to improve the nutritional quality of meat products through the replacement of saturated fatty acid by healthy oils will be discussed. Studies about the use of liquid, pre-emulsified, gelled, and microencapsulated oils as substitutes for animal fat in meat products, as well as the strategies to enrich healthy oils with bioactive compounds will be presented.

2 Health benefits of reducing SFA and increasing PUFA intakes

The World Health Organization (WHO) recommends that total fat should not exceed 30 % of the total energy intake, and the total consumption of SFA should not exceed 10 %. Therefore, SFA should be replaced by MUFA and PUFA in foods (Prado, Matsushita, Visentainer, & Souza, 2004). Each SFA affects differently the cholesterol concentrations in the different plasma lipoprotein fractions. For example, lauric (C12:0), myristic (C14:0) and palmitic (C16:0) acids increase LDL (Low Density Lipoprotein) cholesterol, which is not observed for stearic acid (C18:0). Some studies have shown that replacing SFA (C12:0-C16:0) with PUFA can lead to a decrease in total and LDL cholesterol concentrations and an increase in HDL (High Density Lipoprotein) cholesterol, which reduces the risk of cardiovascular disease (CVD) (IOM, 2005). To achieve these goals, the recommended minimum intake for PUFA is around 6 % of the total energy intake. According to the statements by both the WHO and FAO, the recommended proportion of SFA and PUFA in diets should be between 0.4 and 1.0 (WHO, 2003). The substitution of SFA (C12:0 - C16:0) by MUFA can also confer a similar effect than with PUFA, although on a smaller scale (WCRF/AICR, 2007).

The PUFA Linoleic (LA, C18: 2) (n-6/omega 6) and α-linolenic (ALA, C18: 3) (n-3/omega 3) acids are considered essential acids, as they cannot be synthesized by humans. It is recommended that 2.5 % and 0.5 % of the dietary energy come from LA and ALA, respectively (WHO, 2003, WCRF/AICR, 2007). In addition, the intake of n-3 long-chain PUFA, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), can also decrease the risk of CVD and possibly other aging-related degenerative diseases. To obtain these benefits, the consumption of 0.25 g of EPA + DHA per day is recommended. However, some studies have shown that high n-3 PUFA concentrations can

increase lipid peroxidation and reduce cytokine production (NHMRC, 2006). Therefore, it is recommended to consume up to 2 g of EPA + DHA as an acceptable daily limit.

Currently, in most Western countries, there is a higher consumption of n-6 PUFA when compared with n-3 PUFA, with an n-6/n-3 ratio around 20:1. The high n-6 PUFA and low n-3 PUFA intake is related to the increased incidence of inflammatory and autoimmune diseases, various types of cancer, and cardiovascular diseases (Lee et al., 2006). This increased risk is due to a high intake of LA leads to an increase in arachidonic acid concentration (AA), thus increasing the production of 2- and 4-series eicosanoids (prostaglandin E2 and leukotriene B4) through the cyclooxygenase and lipoxygenase pathways, respectively. A high production of eicosanoids is related to the occurrence of immunological disorders, cardiovascular, and inflammatory diseases. On the other hand, the intake of fatty acids from the n-3 PUFA family, such as ALA, EPA, or DHA, which compete with AA for the same enzymatic routes, competitively inhibits the oxidation of arachidonic acid by cyclooxygenase (COX) to prostaglandins and its conversion into leukotrienes (LTs) via 5-lipoxygenase (LOX) (Bhangle, & Kolasinski, 2011).

Thus, an n-6/n-3 ratio below 4 is recommended for a healthy diet (Simopoulos, 2002). Likewise, the atherogenicity (IA) and thrombogenicity (TI) indexes indicate the relationship between the main classes of SFA, MUFA, and PUFA and according to Turan, Sonmez and Kaya (2007), indicate potential for stimulating platelet aggregation. Thus, the lower these indexes, the greater the potential of the lipid fraction to protect the organism against the occurrence of CVD (Senso, Suárez, Ruiz-Cara, Garcia-Gallego, 2007).

3 Fatty acids profile of the main oils used in meat products

The lipid reformulation in meat products can be performed by replacing animal fat for other lipids containing higher MUFA and PUFA contents (Weiss, Gibis, Schuh, & Salminen, 2010). Various vegetable and marine oils can be used for this purpose, leading to an improvement of the nutritional profile of meat products. The vegetable oils used in meat products include soybean, cotton, canola, linseed, and chia oils, which are sources of MUFA and PUFA fatty acids, in addition to being cholesterol-free (Pelser, Linssen, Legger, & Houben, 2007). Long-chain fatty acids (EPA, docosapentaenoic acid (DPA), and DHA) are found mainly in fish and algae oils (Weiss et al., 2010). In this context, several studies have reported the improvement of the lipid profile of meat products by replacing animal fat for oils of vegetable and marine origin. The fatty acid profile of the main oils used in meat products will be presented below.

3.1 Oils rich in monounsaturated fatty acids (MUFA)

Vegetable oils rich in MUFA are used in meat products to increase the oleic acid content, once regular consumption of this fatty acid (FA) is associated with the prevention of diseases and clinical conditions such as type 2 diabetes, infections, neurological, and cardiovascular disorders (Bagdatli, 2018; Câmara et al., 2020).

Olive oil is one of the most used oils in meat products due to its high oleic acid content, which corresponds to about 70 to 80 % of the total FA and also contain 8-9 % PUFA (linoleic, and linolenic acids) (Fernandes et al., 2020). It also contains lower concentrations in SFA (13%) (mainly palmitic and stearic). It is worth mentioning that the FA composition of olive oil is strongly related to environmental conditions, including soil, altitude, and temperature. The unsaponifiable fraction of olive oil has more than 200 constituents at lower concentrations, such as α- and γ-tocopherols, tocotrienols, β-carotene, phytosterols, flavonoids, and hydrophilic phenolic compounds, which are responsible for the antioxidant and inflammatory effects of this oil (Fernandes et al., 2020). In addition, the European Food Safety Authority (Turck et al., 2018) authorized health claim for olive oil due to its nutritional benefits to the health of consumers.

Palm oil is another example of vegetable oil rich in MUFA used successfully in the reformulation of meat products. It presents a 50/50 balance of SFA and MUFA in the composition, and can be used in technological applications as a semi-solid lipid base at room temperature (Chaves, Barrera-Arellano, & Ribeiro, 2018). In addition, palm oil contains a high palmitic acid content, which distinguishes it from other types of vegetable oils and provides differentiated crystallization characteristics.

Tiger nut oil can also be used in meat product formulations. It contains high oleic acid (67–69 %) content, followed by palmitic acid (14 %), linoleic acid (10 %) and stearic acid (3.5-5 %) (Roselló-Solo et al., 2018). Tiger nut oil is also rich in α-tocopherol (5–87 μg/g) and phytosterols, such as β-sitosterol (43-61 mg/100 g), campesterol (11-17 mg/100 g), and stigmasterol (17-21 mg/100 g) (Lopéz-Cortés, Salazar-García, Malheiro, Guardiola, & Pereira, 2013).

Recently, high oleic sunflower oil has been developed through conventional plant breeding through the use of chemical mutagenesis. It contains about 75-88 % oleic acid and low SFA levels (3-5 % palmitic acid, 2-6 % stearic acid). It presents oxidative stability about 10 times greater than soybean, canola, and sunflower oils due to its lower content of linolenic acid (< 1 %) (Chaves, Barrera-Arellano, & Ribeiro, 2018). Currently, studies have been directed towards the production of ultra-high oleic sunflower oil, which contains more than 90 % oleic acid and presents greater thermo-oxidative stability when compared to high oleic sunflower oil (Alberio et al., 2016).

Hazelnut oil is also rich in MUFA, with oleic acid (73.48-81.57 %) as the major FA, followed by linoleic acid (10.76-14.95 %). It also has a lower concentration of linolenic, palmitic, and stearic acids (Balta et al., 2006).

3.2 Oils rich in n-6 PUFA

Vegetable oils are a rich source of n-6 PUFA, that in an adequate relationship with the n-3 PUFA presents health benefits well as was documented in the literature. Soybean oil is one of the main oils used to increase PUFA and reduce SFA in meat products due to its high availability and lower cost when compared to other oils (Chaves, Barrera-Arellano, & Ribeiro, 2018). Soybean oil is mainly composed of n-6 PUFA (> 50 % linoleic acid), and also has a considerable oleic acid level (~ 20 %) and a small amount of n-3 PUFA (<8 %).

Sunflower oil is another vegetable oil used in meat products. It contains high concentrations of linoleic acid (63.3 %) and oleic acid (28.3 %) (Flagella, Rotunno, Tarantito, Caterina, & Caro, 2002). Cotton oil has also been used to enrich meat products with n-6 PUFA, as it has linoleic acid as the major FA (~ 55 % of the total FA). Sesame oil is also considered an interesting alternative to be used in meat products, as it is rich in n-6 PUFA (~ 40 % linoleic acid), has high concentrations of MUFA (~40 % oleic acid) and also contains sesamoline and sesamol that increase its oxidative stability (Sowmya, Jeyarani, Jyotsna, & Indrani, 2009). Grape seed oil, which stands out for being produced from the residue of the wine and/or grape production industry, has also been used successfully in meat products. Its lipid composition rich in linoleic acid has attracted attention as a source of n-6 PUFA (Beveridge, Girard, Kopp, & Drover, 2005).

3.3 Oils rich in n-3 PUFA

Oils rich in n-3 PUFA include oils of chia, linseed, canola, Echium, seaweed, and fish. Regular consumption of these oils can bring health benefits, such as reduced LDL levels (low density lipoproteins), inhibition of type I and I diabetes, prevention of cardiovascular diseases, and colon and breast cancer (Kajla, Sharma, & Sood, 2015).

Linseed contains 37.1 % oil, 28.9 % carbohydrates, 20.3 % protein, 4.8 % dietary fiber, 6.5 % moisture, and 2.4 % minerals (Singh, Mridula, Rehal, & Barnwal, 2011). The lipid portion is rich in α-linolenic acid (50–60 %), tocopherols (20-70 mg/100 g) and carotenoids (~7 ppm) (Goyal, Sharma, Upadhyay, Gill, & Sihag, 2014). Similarly, chia seeds (*Salvia hispanica* L.) contain 25-40 % oil, 17-24 % protein, and 18-30 % dietary fiber (Timilsena, Vongsvivut, Adhikari, & Adhikari, 2017). The lipid fraction of chia seeds is rich in n-3 PUFA (~75 %). They also have antioxidant compounds such as tocopherols, phytosterols, and carotenoids (Fernández-López, Viuda-Martos, & Pérez- Alvarez, 2020). Both linseed and chia oils have n-3 PUFA levels three times higher than n-6 PUFA, with an n-6/n-3 PUFA ratio between 1 to 2 (Ramcharitar, Badrie, Mattfeldt-Beman, Matsuo, & Ridley, 2005).

Echium plantagineum L. is an herbaceous plant that produces numerous seeds containing about 30 % oil (Gray, Payne, McClements, Decker, & Lad, 2010). *Echium* oil is rich in n-3 PUFA, such as alpha-linolenic acid (30 % of total FA) and stearidonic acid (13 % of total FA) (Miquel, 2008). Canola

oil can be another alternative among the vegetable oils rich in n-3 PUFA, and is considered one of the main terrestrial sources of this type of fatty acid (Nguyen, Malau-Auduli, Cavalieri, & Nichols, 2018). Canola oil has about 6 to 14 % linolenic acid and an n-6/n-3 PUFA ratio, which ranges from 2 to 4. In addition, it has a low SFA content (<7 %) (Giese, 1996).

Marine vegetable oils have also been used to produce meat products with a healthier lipid profile. Marine plants, especially single-celled organisms, have the capacity to elongate the PUFA chain and desaturate α-linolenic acid to produce EPA and DHA. Depending on the type of seaweed oil, the fatty acid profile varies a lot and they also have other beneficial compounds. For example, algal oil DHA ™ -S present docosahexaenoic acid-DHA content ⩾ 350 mg / g oil and rosemary extract, tocopherols and ascorbyl palmitate as antioxidants (López-López et al., 2009).

The long-chain PUFA from fish oil come from a diet rich in microalgae, which explains the abundance of EPA and DHA fatty acids in some fish oils of marine origin (Weiss et al., 2010). Fish oils contain approximately 22 % EPA, 3 % DPA, and 22 % DHA, and are one of the richest sources of long-chain n-3 PUFA (Pelser et al., 2007).

4 Approaches for the addition of healthy oils in meat products

The improvement of the lipid profile of meat products can bring numerous health benefits to consumers. However, many studies have shown that the replacement of animal fat by vegetable and/or marine oils can affect the technological characteristics and the sensory quality, and decrease the oxidative stability of meat products. In this context, different ways of use of vegetable and/or marine oils have been studied to improve the final quality of reformulated products. The most studied applications will be presented below.

4.1 Addition of liquid healthy oils to meat products

Numerous studies have evaluated the replacement of animal fat by vegetable and marine oils in liquid form in meat products (Table 1). In many cases, the technological, oxidative, and sensory qualities are impaired by this reformulation.

López-López, Cofrades, Ruiz-Capillas, and Jiménez-Colmenero (2009) observed that a healthier lipid formulation (seaweed and olive oils and the reducing animal fat) in frankfurters produced a good balance of MUFA/SFA, PUFA/SFA and n−6/n−3 ratios and constitute a good means to produce low-sodium products with important dietary fibre content, with better Na/K ratios and rich in Ca. However, had little effect on amino acid profiles of frankfurters. In a similar study, Domínguez, Agregán, Gonçalves, and Lorenzo (2016) also improved the nutritional quality of pâté by replacing animal fat with olive oil.

Domínguez, Pateiro, Munekata, Campagnol, and Lorenzo (2017) observed an increase in volatile compounds related to lipid oxidation in liver pate with the addition of fish oil. In Bologna sausages, the use of *Echium* oil was efficient to improve the technological quality and the lipid profile of the products. However, the sensory quality of the products was negatively affected by the replacement of 100 % animal fat by *Echium* oil (Pires, Santos, Barros, & Trindade, 2019).

Barbut, Wood, and Marangoni (2016) replaced animal fat by liquid canola oil in sausages and reported that the strategy impaired the sensory quality of the products. Similar results were reported by Alejandre, Astiasarán, Ansorena, and Barbut (2019), who used liquid canola oil as a fat substitute in emulsion type meat batters. In another study, Barbut and Marangoni (2019) studied the effects of replacing animal fat by canola, soybean, and linseed oils in liquid form, and also reported a reduction in the oxidative stability, with negative effects on the sensory quality of products.

4.2 Addition of pre-emulsified healthy oils to meat products

The use of pre-emulsified vegetable and/or marine oils is well documented in the literature (Table 2). Bloukas, Paneras, and Fournitzis (1997) were one of the pioneers in this approach, by replacing pork fat for olive oil pre-emulsified with soy protein isolate (SPI) in fermented sausages. The authors concluded that the substitution of up to 20 % did not affect the technological quality of the products. Similarly, Muguerza, Gimeno, Ansorena, Bloukas, and Astiasaran (2001) reported that it is possible to replace up to 25 % animal fat by olive oil emulsified with SPI without changing the sensory characteristics of the fermented sausages.

Delgado-Pando, Cofrades, Ruiz-Capillas, Triki, and Jiménez-Colmenero (2012) observed that the low-fat pork liver pâtés enriched with n − 3 PUFA/konjac gel were formulated by replacing (50% and 100%) pork backfat by a combination of healthier oils (olive, linseed and fish oils) and konjac gel improved the nutritional characteristics (mainly PUFA) in this pate but noted a lower oxidative stability. Likewise, Jiménez-Colmenero, Herrero, Pintado, Sola, and Ruiz-Capillas (2010) observed a lower hardness, cohesiveness and chewiness and poorer ($P < 0.05$) adhesiveness than control (all pork fat) frankfurters formulated with oil-in-water emulsions stabilized with various protein systems (sodium caseinate, soy protein isolate, meat protein and microbial transglutaminase). On the other hand, Tudose, Iordachescu, Stan, Cercel, and Alexe (2014) reported an increase in lipid oxidation of cooked meat emulsion produced by replacing animal fat for pre-emulsified olive oil and palm oil. These results demonstrates the difficulty of incorporating healthy oils in cooked meat products due to the acceleration of lipid oxidation reactions through heating.

In another interesting approach, Ansorena and Astiasarán (2004) used emulsified linseed oil as a substitute for animal fat in fermented sausages. The authors reported that the strategy improved the lipid profile and did not affect the oxidative stability of the products. Similar results have been reported by Pelser et al. (2007) who studied canola and linseed oils pre-emulsified with soy protein isolate as a

substitute for 10, 15, and 20 % animal fat in fermented sausages. In another approach, Choi et al. (2013) also obtained promising results in cooked sausages with a replacement of 20 % fat for sunflower oil pre-emulsified with dietary fibers. More recently, de Carvalho et al. (2020a) produced cooked lamb sausages with replacement of animal fat by pre-emulsified healthy oils (chia, linseed and olive). In addition to improving nutritional quality, the proposed lipid reformulation did not affect the color and aroma of the products during storage. In addition, among the oils tested, linseed oil showed the best technological and sensory results. In another study, Lima et al. (2021) concluded that the inclusion of pre-emulsified linseed oil as animal fat replacement improved the technological quality and texture of sheep sausages.

Another promising strategy to improve the lipid profile of meat products has been carried out by Barros et al. (2021), who produced low-fat burgers enriched with emulsions of algae oils and/or wheat germ. This strategy was effective in improving important nutritional indices such as the n-6/n-3 PUFA and PUFA/SFA ratios. In addition, the use of algae oil emulsion did not affect the technological and sensory quality of the burgers. Barros et al. (2020) also evaluated the effect of replacing animal fat by tiger nut oil emulsion and reported an increase in oleic acid contents, with no changes in the texture parameters and sensory acceptance of the burgers.

The use of pre-emulsified grape seed oil in meat products has been reported by Kim, Yong, Jung, Kim, and Choi, (2020). The authors reported that in addition to improving the lipid profile, the use of pre-emulsified grape seed oil as a substitute for animal fat also improved the technological quality of meat emulsions. In another similar study, Kim et al. (2021) used emulsions made from corn, grape seed, soybean, and olive oils as animal fat replacer in duck meat emulsions. The authors reported no effects on the oxidative stability, and an improvement in the nutritional quality of the products. Urgu-Ozturk, Ozturk-Kerimoglu, and Serdaroglu (2020) used pre-emulsified hazelnut oil as animal fat replacer to produce healthier sausages. The authors reported that sausages produced with up to 100 % replacement of animal fat by pre-emulsified hazelnut oil showed excellent technological quality and high sensory acceptance.

4.3 Addition of healthy oils microencapsulated to meat products

Several studies have proven the effectiveness of microencapsulation as a way to increase the oxidative stability of healthy oils in meat products (Table 3). An interesting study was carried out by Pérez-Palacios, Ruiz-Carrascal, Jiménez-Martín, Solomando, and Antequera (2018) who compared the effect of the addition of unencapsulated and encapsulated fish oil to chicken nuggets. The authors reported that the microencapsulation was effective in reducing lipid and protein oxidation during storage. In a similar study, Aquilani, et al (2018) produced low-fat burgers enriched with non-encapsulated and encapsulated fish oil and evaluated changes in EPA and DHA contents during storage. The results showed that microencapsulation protected these important FA from the oxidative reactions during refrigerated and frozen storage.

Domínguez, Pateiro, Agregán, and Lorenzo (2017) produced low-fat sausages enriched with microencapsulated fish oil and a blend (1:1) of olive and non-encapsulated fish oils and reported promising results. As expected, the reformulated products presented an increase in MUFA and n-3 PUFA levels. However, the products containing fish oil microparticles presented higher lipid oxidation. It is worth noting that the sausages were pasteurized at 90 °C for 30 min, which may have led to the rupture of microparticles and the consequent oxidation of fish oil. This result highlights the difficulty of incorporating oils rich in n-3 PUFA in meat products, especially those subjected to severe heat treatment. In addition, the depreciation of sensory quality is another challenge to incorporate fish oil in meat products. This trend was also reported by Lorenzo, Munekata, Pateiro, Campagnol, and Domínguez (2016) in fermented sausages made with microencapsulated fish oil. To minimize the sensory problems, González, Antequera, and Pérez-Palacios (2020) produced fish oil microparticles using different coating materials (lecithin, maltodextrin, and chitosan). The strategy was effective in reducing the sensory defects caused by fish oil in meat products, and also protected fish oil from degradation during storage. In addition, the use of these microparticles in cooked and raw cured meat products increased the EPA and DHA levels, enabling the products to be labeled as a "source of n-3 PUFA".

Heck et al. (2017) proposed another effective strategy for encapsulating healthier oils. The authors produced chia and linseed oil microparticles to be used as an animal fat replacer in burgers. The great innovation of this study was the evaluation of the thermal stability of the microparticles. The results showed no rupture of the microparticles during cooking of the burgers, which is very important to prevent oxidation of MUFA and PUFA. In addition, the evaluation of the microparticles stability at different pH values evidenced the rupture of the particles only at the pH value of the human intestine, thus guaranteeing the absorption of these FA with health benefits. Although the addition of microparticles has significantly improved the lipid profile of the burgers, an increase in lipid oxidation was observed for the products made with the addition of chia oil microparticles.

4.4 Addition of gelled healthy oils to meat products

The use of healthy oils in gel form is another promising strategy to improve the lipid profile of meat products (Table 4). Jiménez-Colmenero, Triki, Herrero, Rodríguez-Salas, and Ruiz-Capillas (2013) study the healthy oil combination (olive, linseed and fish oil) stabilized in a Hydrogel (konjac matrix) as pork fat replacement in dry fermented sausages. The replacement incorporation of this oil-in-konjac matrix reduced SFA and increased PUFA contents, improving the n–6/n–3 ratio in dry sausage, however, was observed a decreased of hardness, increased cohesiveness and also decreased sensorial parameters. Triki et al. (2013) for the same combination studied observed an increased in the lipid oxidation with storage time, more noticeably in samples with higher PUFA content. Recently, Franco et al. (2020) used ɣ-oryzanol and b-sitosterol to structure flaxseed oil. Despite the nutritional improvement, the use of gelled oil as animal fat replacer in fermented sausages impaired the sensory quality of the

products. However, in another study, Alejandre, Poyato, Ansorena, and Astiasarán (2016) obtained good results when using gelled linseed oil to replace animal fat in fermented sausages. In addition to the improvement in the lipid profile (2.32g ω-3 FA/100 g product), the reformulated products exhibited high oxidative stability and good sensory acceptance.

Kouzouni, Lazaridou, and Katsanidis (2017) used monoglycerides and phytosterols to structure sunflower oil. The replacement of 50 % of animal fat by the gelled oil did not affect the lipid oxidation and the sensory quality of sausages. In another study, Moghtadaei, Amir, and Goli (2018) used sesame oil gelled with beeswax to replace 50 % of animal fat in burgers. Although the sensory quality of the burgers was not affected, the reformulation led to an increase in hardness of the products.

Paglarini et al. (2019) evaluated the effect of replacing animal fat by gelled soybean oil on the quality of Bologna sausages. Although the reformulation has nutritionally improved the fatty acid profile of the products, the color and texture were negatively affected. More recently, Paglarini et al. (2021) tested the used of inulin-based emulsion gel as fat substitute to improve the fatty acid profile and reduce the salt in bologna saudage.

Wolfer, Acevedo, Prusa, Sebranek, and Tarté (2018) also used gelled soybean oil as a fat replacer in sausages. The results indicated that although the lipid profile may have health benefits, further studies are required to improve the technological quality and oxidative stability of the products. Tarté, Paulus, Acevedo, Prusa, and Lee (2020) evaluated the use of gelled conventional soybean oil and high oleic soybean oil in bologna sausages. The addition of gelled high oleic soybean oil nutritionally improved the fatty acid profile of the products, which presented acceptable technological and sensory qualities. More recently, Vargas-Ramela et al. (2020) produced dry-fermented deer sausage with replacement of animal fat by gelled oils (olive, canola and soy). The authors concluded that this approach was very efficient for the production of products with high nutritional, oxidative and sensory quality.

Another interesting approach has been proposed by Da Silva et al. (2019), who used high oleic sunflower oil gelled by pork skin as a fat replacer in Bologna sausages. The reformulation improved the nutritional quality of the products by increasing the oleic acid contents. In addition, no changes in oxidative stability were observed and the products had good sensory acceptance up to the level of 50 % replacement. In a similar study, Fagundes et al. (2017) used pork skin to structure canola oil, using the gelled oil to replace 50 % animal fat in burgers. The authors reported that the reformulation was effective to confer technological and nutritional advantages to burgers.

4.5 Addition of healthy oils enriched with bioactive compounds to meat products

Apart the natural bioactive compounds presents in the healthy oils, the addition of other bioactive compounds has been used to improve the oxidative stability and sensory quality of meat products enriched with healthy oils (Table 5). Munekata et al. (2017) used agro-industry residues (beer residue, chestnut leaves and peanut skin) to improve the lipid stability of pâté produced with healthier

oils (fish and olive). Natural antioxidants decreased the level of hexanal at the beginning of storage, however, no additional protection against lipid oxidation was observed. Another interesting approach to improve the oxidative stability of healthy oils has been proposed by Heck et al. (2018a, 2018b). In those studies, chia oil was enriched with bioactive compounds from rosemary using green extraction techniques. This enriched chia oil was microencapsulated and used to replace 50 % of animal fat in burger formulations. The results showed that the combination of healthy oils and natural antioxidants was effective to improve the oxidative stability of the microparticles, thus enabling the production of healthier and safe meat products. In another similar study, Heck et al. (2020) used jabuticaba peel extract (JPE) to increase the oxidative stability of healthier oils (chia and linseed), which were gelled and used as animal fat replacer in burgers. This strategy was efficient to maintain the lipid oxidation level of the reformulated burgers below the sensorially detectable limit during 90 days of storage at -18 °C. In addition, the use of JPE improved the sensory quality of the burgers. In another study, de Carvalho et al. (2019) added natural antioxidants to lamb patties enriched with chia oil. Guarana seed and pitanga leaf extracts decreased the lipid and protein oxidation without harmful the sensory quality. Likewise, de Carvalho et al. (2020b) used turmeric extract (Curcuma longa L.) to improve the lipid stability of fresh lamb sausages enriched with tiger nut oil. Turmeric extract decreased the formation of volatile compounds from lipid oxidation and improved the aroma of sausages.

Another efficient strategy to increase the oxidative stability of oils in meat products has been reported by Alejandre, Ansorena, Calvo, Cavero, and Astiasarían (2019). The authors used blackthorn branch extract (*Prunus spinosa* L.) to enrich microalgal oil, which was then gelled and used as a fat substitute in burgers. The results showed that the high amount of polyphenolic compounds in the extract led to an increase in the oxidative stability of microalgal oil, making viable its application in burgers. Moreover, recently Pintado et al (2021), check the used of two different solid polyphenol extracts from grape seed (R-EPG) or grape seed and olive (R-EPGO)] in emulsion gel with olive oil as animal fat replacers in the development of frankfurters. The incorporation of EGs improved their lipid content, particularly, and the levels of phenolic compounds (hydroxytyrosol and flavanols) and the frankfurters were well accepted by the panellists.

In a recent study, Zhu, Guo, Tang, and Yang (2020) enriched olive oil with Jerusalem artichoke powder (JAP), which is a natural ingredient rich in phenolic compounds. Subsequently, the JAP-enriched olive oil was pre-emulsified and used as fat replacer in Harbin dry sausages. The results proved that the strategy was effective to increase the oxidative stability and the sensory quality of the products. In another approach, Berasategi et al. (2014) produced Bologna sausages with substitution of animal fat for an oil-in-water emulsion made with a mixture of algae and linseed oils stabilized with Melissa extract. In addition to not affecting technological quality, the proposed reformulation did not affect the oxidative stability and the sensory quality of the products.

5. Final remarks

The replacement of animal fat by healthy oils may be an effective strategy to improve the nutritional quality of meat products. However, this approach can negatively affect the technological, oxidative, and sensory quality and decrease the shelf life of meat products. Emulsification, gelation, and microencapsulation technologies have proven to be very effective in solving most problems caused by the addition of liquid healthy oils to meat products. However, these technologies fail to completely protect healthy oils from oxidative reactions. Thus, the use of additional bioactive compounds may be an alternative to control lipid oxidation of meat products, allowing the use of healthy oils to improve the nutritional quality of the products.

References

Alberio, C. et al. (2016) "A new sunflower high oleic mutation confers stable oil grain fatty acid composition across environments," European Journal of Agronomy. Argentina: Elsevier B.V., 73, pp. 25–33. doi: 10.1016/j.eja.2015.10.003.

Alejandre, M. et al. (2016) "Linseed oil gelled emulsion: A successful fat replacer in dry fermented sausages," Meat Science. Spain: Elsevier Ltd, 121, pp. 107–113. doi: 10.1016/j.meatsci.2016.05.010.

Alejandre, M., Ansorena, D., et al. (2019) "Influence of a gel emulsion containing microalgal oil and a blackthorn (Prunus spinosa L.) branch extract on the antioxidant capacity and acceptability of reduced-fat beef patties," Meat Science. Spain: Elsevier Ltd, 148, pp. 219–222. doi: 10.1016/j.meatsci.2018.05.022.

Alejandre, M., Astiasarán, I., et al. (2019) "Using canola oil hydrogels and organogels to reduce saturated animal fat in meat batters," Food Research International. Spain: Elsevier Ltd, 122, pp. 129–136. doi: 10.1016/j.foodres.2019.03.056.

Ansorena, D. and Astiasarán, I. (2004) "The use of linseed oil improves nutritional quality of the lipid fraction of dry-fermented sausages," Food Chemistry. Spain, 87(1), pp. 69–74. doi: 10.1016/j.foodchem.2003.10.019.

Aquilani, C. et al. (2018) "Enrichment of Cinta Senese burgers with omega-3 fatty acids. Effect of type of addition and storage conditions on quality characteristics," Grasas y Aceites. Departmento de Publicaciones del CSIC, p. 235. doi: 10.3989/gya.0671171.

Bağdatli, A. (2018) "The influence of Quinoa (Chenopodium Quinoa Willd.) flour on the physicochemical, textural and sensorial properties of beef meatball," Italian Journal of Food

Science. Turkey: Chiriotti Editori, 30(2), pp. 280–288. Available at: http://www.chiriottieditori.it/ojs/index.php/ijfs/article/view/945/380.

Balasubramanian, R., Sughir, A. A. and Damodar, G. (2012) "Oleogel: A promising base for transdermal formulations," Asian Journal of Pharmaceutics. Libyan Arab Jamahiriya, 6(1), pp. 1–9. doi: 10.4103/0973-8398.100118.

Balta, M. F. et al. (2006) "Determination of fatty acid compositions, oil contents and some quality traits of hazelnut genetic resources grown in eastern Anatolia of Turkey," Journal of Food Composition and Analysis. Turkey, 19(6–7), pp. 681–686. doi: 10.1016/j.jfca.2005.10.007.

Barbut, S. and Marangoni, A. (2019) "Organogels use in meat processing – Effects of fat/oil type and heating rate," Meat Science. Canada: Elsevier Ltd, 149, pp. 9–13. doi: 10.1016/j.meatsci.2018.11.003.

Barbut, S., Wood, J. and Marangoni, A. (2016) "Potential use of organogels to replace animal fat in comminuted meat products," Meat Science. Canada: Elsevier Ltd, 122, pp. 155–162. doi: 10.1016/j.meatsci.2016.08.003.

Barros, J. C. et al. (2020) "Use of tiger nut (Cyperus esculentus L.) oil emulsion as animal fat replacement in beef burgers," Foods. Brazil: MDPI Multidisciplinary Digital Publishing Institute, 9(1). doi: 10.3390/foods9010044.

Barros, J. C. et al. (2021) "Healthy beef burgers: Effect of animal fat replacement by algal and wheat germ oil emulsions," Meat Science. Brazil: Elsevier Ltd, 173. doi: 10.1016/j.meatsci.2020.108396.

Berasategi, I. et al. (2014) "Healthy reduced-fat Bologna sausages enriched in ALA and DHA and stabilized with Melissa officinalis extract," Meat Science. Spain, 96(3), pp. 1185–1190. doi: 10.1016/j.meatsci.2013.10.023.

Beveridge, T. H. J. et al. (2005) "Yield and composition of grape seed oils extracted by supercritical carbon dioxide and petroleum ether: Varietal effects," Journal of Agricultural and Food Chemistry. Canada, 53(5), pp. 1799–1804. doi: 10.1021/jf040295q.

Bhangle, S. and Kolasinski, S. L. (2011) "Fish Oil in Rheumatic Diseases," Rheumatic Disease Clinics of North America. United States, 37(1), pp. 77–84. doi: 10.1016/j.rdc.2010.11.003.

Bloukas, J. G., Paneras, E. D. and Fournitzis, G. C. (1997) "Effect of replacing pork backfat with olive oil on processing and quality characteristics of fermented sausages," Meat Science. Greece: Elsevier Ltd, 45(2), pp. 133–144. doi: 10.1016/S0309-1740(96)00113-1.

Boada, L. D., Henríquez-Hernández, L. A. and Luzardo, O. P. (2016) "The impact of red and processed meat consumption on cancer and other health outcomes: Epidemiological evidences," Food and Chemical Toxicology. Spain: Elsevier Ltd, 92, pp. 236–244. doi: 10.1016/j.fct.2016.04.008.

Câmara, A. K. F. I. et al. (2020) "Reducing phosphate in emulsified meat products by adding chia (Salvia hispanica L.) mucilage in powder or gel format: A clean label technological strategy," Meat Science. Brazil: Elsevier Ltd, 163. doi: 10.1016/j.meatsci.2020.108085.

de Carvalho, F. A. L. et al. (2019) "Effect of guarana (Paullinia cupana) seed and pitanga (Eugenia uniflora L.) leaf extracts on lamb burgers with fat replacement by chia oil emulsion during shelf life storage at 2 °C," Food Research International. Elsevier BV, p. 108554. doi: 10.1016/j.foodres.2019.108554.

de Carvalho, F. A. L., Munekata, P. E. S., Pateiro, M., et al. (2020) "Effect of replacing backfat with vegetable oils during the shelf-life of cooked lamb sausages," LWT, 122, p. 109052. doi: 10.1016/j.lwt.2020.109052.

de Carvalho, F. A. L., Munekata, P. E. S., Lopes de Oliveira, A., et al. (2020) "Turmeric (Curcuma longa L.) extract on oxidative stability, physicochemical and sensory properties of fresh lamb sausage with fat replacement by tiger nut (Cyperus esculentus L.) oil," Food Research International. Elsevier BV, p. 109487. doi: 10.1016/j.foodres.2020.109487.

Champagne, C. P. and Fustier, P. (2007) "Microencapsulation for the improved delivery of bioactive compounds into foods," Current Opinion in Biotechnology. Canada, 18(2), pp. 184–190. doi: 10.1016/j.copbio.2007.03.001.

Chaves, K. F., Barrera-Arellano, D. and Ribeiro, A. P. B. (2018) "Potential application of lipid organogels for food industry," Food Research International. Brazil: Elsevier Ltd, 105, pp. 863–872. doi: 10.1016/j.foodres.2017.12.020.

Choi, Y. S. et al. (2013) "Quality characteristics of reduced-fat frankfurters with pork fat replaced by sunflower seed oils and dietary fiber extracted from makgeolli lees," Meat Science. South Korea, 93(3), pp. 652–658. doi: 10.1016/j.meatsci.2012.11.025.

De Oliveira Fagundes, D. T. et al. (2017) "Pork skin and canola oil as strategy to confer technological and nutritional advantages to burgers," Czech Journal of Food Sciences. Brazil: Czech Academy of Agricultural Sciences, 35(4), pp. 352–359. doi: 10.17221/67/2017-CJFS.

Delgado-Pando, G. et al. (2012) "Enriched n-3 PUFA/konjac gel low-fat pork liver pâté: Lipid oxidation, microbiological properties and biogenic amine formation during chilling storage," Meat Science. Spain, 92(4), pp. 762–767. doi: 10.1016/j.meatsci.2012.06.035.

Dickinson, E. (2012) “Emulsion gels: The structuring of soft solids with protein-stabilized oil droplets,” Food Hydrocolloids. United Kingdom, 28(1), pp. 224–241. doi: 10.1016/j.foodhyd.2011.12.017.

Domínguez, R. et al. (2016) “Effect of fat replacement by olive oil on the physico-chemical properties, fatty acids, cholesterol and tocopherol content of pâté,” Grasas y Aceites, 67(2), p. e133. doi: 10.3989/gya.0629152.

Domínguez, R. et al. (2017) “Effect of the partial replacement of pork backfat by microencapsulated fish oil or mixed fish and olive oil on the quality of frankfurter type sausage,” Journal of Food Science and Technology. Spain: Springer India, 54(1), pp. 26–37. doi: 10.1007/s13197-016-2405-7.

Fernandes, J. et al. (2020) “Is olive oil good for you? A systematic review and meta-analysis on anti-inflammatory benefits from regular dietary intake,” Nutrition. Portugal: Elsevier Inc., 69. doi: 10.1016/j.nut.2019.110559.

Fernández-López, J., Viuda-Martos, M. and Pérez-Alvarez, J. A. (2021) “Quinoa and chia products as ingredients for healthier processed meat products: technological strategies for their application and effects on the final product,” Current Opinion in Food Science. Elsevier BV, pp. 26–32. doi: 10.1016/j.cofs.2020.05.004.

Flagella, Z. et al. (2002) “Changes in seed yield and oil fatty acid composition of high oleic sunflower (Helianthus annuus L.) hybrids in relation to the sowing date and the water regime,” European Journal of Agronomy. Elsevier BV, pp. 221–230. doi: 10.1016/s1161-0301(02)00012-6.

Franco, D. et al. (2020) “Evaluation of linseed oil oleogels to partially replace pork backfat in fermented sausages,” Journal of the Science of Food and Agriculture. Spain: John Wiley and Sons Ltd, 100(1), pp. 218–224. doi: 10.1002/jsfa.10025.

Gómez-Estaca, J. et al. (2019) “Characterization of ethyl cellulose and beeswax oleogels and their suitability as fat replacers in healthier lipid pâtés development,” Food Hydrocolloids. Spain: Elsevier B.V., 87, pp. 960–969. doi: 10.1016/j.foodhyd.2018.09.029.

Goyal, A. et al. (2014) “Flax and flaxseed oil: an ancient medicine & modern functional food,” Journal of Food Science and Technology. India: Springer, 51(9), pp. 1633–1653. doi: 10.1007/s13197-013-1247-9.

Gray, D. A. et al. (2010) “b). Volatile compounds and sensory profile of burgers with 50 % fat replacement by microparticles of chia oil enriched with rosemary,” European Journal of Lipid Science and Technology, 112, pp. 164–170.

Heck, R. T. et al. (2017) “Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties?,” Meat Science. Brazil: Elsevier Ltd, 130, pp. 16–25. doi: 10.1016/j.meatsci.2017.03.010.

Heck, R. T. et al. (2018) “Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques,” Meat Science. Brazil: Elsevier Ltd, 146, pp. 147–153. doi: 10.1016/j.meatsci.2018.08.009.

Heck, R. T., Saldaña, E., et al. (2019) “Hydrogelled emulsion from chia and linseed oils: A promising strategy to produce low-fat burgers with a healthier lipid profile,” Meat Science. Brazil: Elsevier Ltd, 156, pp. 174–182. doi: 10.1016/j.meatsci.2019.05.034.

Heck, R. T., Fagundes, M. B., et al. (2019) “Volatile compounds and sensory profile of burgers with 50% fat replacement by microparticles of chia oil enriched with rosemary,” Meat Science. Brazil: Elsevier Ltd, 148, pp. 164–170. doi: 10.1016/j.meatsci.2018.10.017.

Heck, R. T. et al. (2020) “Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: A green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils,” Meat Science. Brazil: Elsevier Ltd, 170. doi: 10.1016/j.meatsci.2020.108230.

Herrero, A. M. and Ruiz-Capillas, C. (2021) “Novel lipid materials based on gelling procedures as fat analogues in the development of healthier meat products,” Current Opinion in Food Science. Spain: Elsevier Ltd, 39, pp. 1–6. doi: 10.1016/j.cofs.2020.12.010.

Hulya, T. (2007) “Fatty acid profile and proximate composition of the thornback ray (Raja clavata, L. 1758) from the Sinop coast in the Black Sea,” Journal of Fisheries Sciences.com. Scitechnol Biosoft Pvt. Ltd., pp. 97–103. doi: 10.3153/jfscom.2007012.

Jiménez-Colmenero, F. et al. (2010) “Influence of emulsified olive oil stabilizing system used for pork backfat replacement in frankfurters,” Food Research International. Elsevier BV, pp. 2068–2076. doi: 10.1016/j.foodres.2010.06.010.

Jiménez-Colmenero, F. et al. (2013) “Healthy oil combination stabilized in a konjac matrix as pork fat replacement in low-fat, PUFA-enriched, dry fermented sausages,” LWT - Food Science and Technology. Spain, 51(1), pp. 158–163. doi: 10.1016/j.lwt.2012.10.016.

Jimenez-Colmenero, F. et al. (2015) “Novel applications of oil-structuring methods as a strategy to improve the fat content of meat products,” Trends in Food Science and Technology. Spain: Elsevier Ltd, 44(2), pp. 177–188. doi: 10.1016/j.tifs.2015.04.011.

Kajla, P., Sharma, A. and Sood, D. R. (2015) “Flaxseed—a potential functional food source,” Journal of Food Science and Technology. India: Springer India, 52(4), pp. 1857–1871. doi: 10.1007/s13197-014-1293-y.

Kim, T. K. et al. (2020) “Effects of replacing pork fat with grape seed oil and gelatine/alginate for meat emulsions,” Meat Science. South Korea: Elsevier Ltd, 163. doi: 10.1016/j.meatsci.2020.108079.

Kim, T. K. et al. (2021) “Physiochemical properties of reduced-fat duck meat emulsion systems: effects of preemulsification with vegetable oils and duck skin,” Poultry Science. South Korea: Elsevier Inc., 100(2), pp. 1291–1298. doi: 10.1016/j.psj.2020.10.044.

Klurfeld, D. M. (2015) “Research gaps in evaluating the relationship of meat and health,” Meat Science. United States: Elsevier Ltd, 109, pp. 86–95. doi: 10.1016/j.meatsci.2015.05.022.

Kouzounis, D., Lazaridou, A. and Katsanidis, E. (2017) “Partial replacement of animal fat by oleogels structured with monoglycerides and phytosterols in frankfurter sausages,” Meat Science. Greece: Elsevier Ltd, 130, pp. 38–46. doi: 10.1016/j.meatsci.2017.04.004.

Lee, S. et al. (2006) “Effect of antioxidants and cooking on stability of n-3 fatty acids in fortified meat products,” Journal of Food Science. United States: Blackwell Publishing Inc., 71(3), pp. C233–C238. doi: 10.1111/j.1365-2621.2006.tb15623.x.

Lima, T. L. S. et al. (2021) “Pre-emulsioned linseed oil as animal fat replacement in sheep meat sausages: Microstructure and physicochemical properties,” Journal of Food Processing and Preservation. Brazil: Blackwell Publishing Ltd, 45(1). doi: 10.1111/jfpp.15051.

Liu, A. G. et al. (2017) “A healthy approach to dietary fats: Understanding the science and taking action to reduce consumer confusion,” Nutrition Journal. United States: BioMed Central Ltd., 16(1). doi: 10.1186/s12937-017-0271-4.

Lopéz-Cortés, I. et al. (2013) “Chemometrics as a tool to discriminate geographical origin of Cyperus esculentus L. based on chemical composition,” Industrial Crops and Products. Spain, 51, pp. 19–25. doi: 10.1016/j.indcrop.2013.08.061.

López-López, I. et al. (2009) “Design and nutritional properties of potential functional frankfurters based on lipid formulation, added seaweed and low salt content,” Meat Science. Spain, 83(2), pp. 255–262. doi: 10.1016/j.meatsci.2009.05.014.

Lorenzo, J. M. et al. (2016) “Healthy Spanish salchichón enriched with encapsulated n − 3 long chain fatty acids in konjac glucomannan matrix,” Food Research International. Spain: Elsevier Ltd, 89, pp. 289–295. doi: 10.1016/j.foodres.2016.08.012.

Mir, M. (2008) “Echium oil: A valuable source of n-3 and n-6 fatty acids,” OCL - Oleagineux Corps Gras Lipides. Spain, 15(4), pp. 252–256. doi: 10.1684/ocl.2008.0203.

Moghtadaei, M., Soltanizadeh, N. and Goli, S. A. H. (2018) “Production of sesame oil oleogels based on beeswax and application as partial substitutes of animal fat in beef burger,” Food Research International. Iran: Elsevier Ltd, 108, pp. 368–377. doi: 10.1016/j.foodres.2018.03.051.

Muguerza, E. et al. (2001) “Effect of replacing pork backfat with pre-emulsified olive oil on lipid fraction and sensory quality of Chorizo de Pamplona - a traditional Spanish fermented sausage,” Meat Science. Spain, 59(3), pp. 251–258. doi: 10.1016/S0309-1740(01)00075-4.

Munekata, P. E. S. et al. (2017) “Effect of natural antioxidants on physicochemical properties and lipid stability of pork liver pâté manufactured with healthy oils during refrigerated storage,” Journal of Food Science and Technology. Brazil: Springer India, 54(13), pp. 4324–4334. doi: 10.1007/s13197-017-2903-2.

Nguyen, D. V. et al. (2018) “Supplementation with plant-derived oils rich in omega-3 polyunsaturated fatty acids for lamb production,” Veterinary and Animal Science. Australia: Elsevier B.V., 6, pp. 29–40. doi: 10.1016/j.vas.2018.08.001.

Paglarini, C. S. et al. (2019) “Functional emulsion gels as pork back fat replacers in Bologna sausage,” Food Structure, 20.

Pelser, W. M. et al. (2007) “Lipid oxidation in n - 3 fatty acid enriched Dutch style fermented sausages,” Meat Science. Netherlands, 75(1), pp. 1–11. doi: 10.1016/j.meatsci.2006.06.007.

Pérez-Palacios, T. et al. (2018) “Improving the lipid profile of ready-to-cook meat products by addition of omega-3 microcapsules: effect on oxidation and sensory analysis,” Journal of the Science of Food and Agriculture. Spain: John Wiley and Sons Ltd, 98(14), pp. 5302–5312. doi: 10.1002/jsfa.9069.

Pintado, T. et al. (2021) “Phenolic compounds in emulsion gel-based delivery systems applied as animal fat replacers in frankfurters: Physico-chemical, structural and microbiological approach,” Food Chemistry. Spain: Elsevier Ltd, 340. doi: 10.1016/j.foodchem.2020.128095.

Pires, M. A. et al. (2019) “Effect of replacing pork backfat with Echium oil on technological and sensory characteristics of bologna sausages with reduced sodium content,” LWT. Brazil: Academic Press, 109, pp. 47–54. doi: 10.1016/j.lwt.2019.04.009.

Prado, I. N. et al. (2004) Carne bovina brasileira: realidade e perspectivas. Revista Nacional da Carne.

Ramcharitar, A. et al. (2005) “Consumer acceptability with Muffind flaxseed (linum usitatissimum),” Journal of food Science, 70.

Roselló-Soto, E. et al. (2018) “Tiger nut and its by-products valorization: From extraction of oil and valuable compounds to development of new healthy products,” Innovative Food Science and Emerging Technologies. Spain: Elsevier Ltd, 45, pp. 306–312. doi: 10.1016/j.ifset.2017.11.016.

Ruiz-Capillas, C. and Herrero, A. M. (2021) “Development of Meat Products with Healthier Lipid Content: Vibrational Spectroscopy,” Foods. MDPI AG, 10(2), p. 341. doi: 10.3390/foods10020341.

Senso, L. et al. (2007) “On the possible effects of harvesting season and chilled storage on the fatty acid profile of the fillet of farmed gilthead sea bream (Sparus aurata),” Food Chemistry. Elsevier BV, pp. 298–307. doi: 10.1016/j.foodchem.2006.01.036.

da Silva, S. L. et al. (2019) “Fat replacement by oleogel rich in oleic acid and its impact on the technological, nutritional, oxidative, and sensory properties of Bologna-type sausages,” Meat Science. Brazil: Elsevier Ltd, 149, pp. 141–148. doi: 10.1016/j.meatsci.2018.11.020.

Simopoulos, A. P. (2002) “The importance of the ratio of omega-6/omega-3 essential fatty acids,” Biomedicine and Pharmacotherapy. United States: Elsevier Masson SAS, 56(8), pp. 365–379. doi: 10.1016/S0753-3322(02)00253-6.

Singh, K. K. et al. (2011) “Flaxseed: A potential source of food, feed and fiber,” Critical Reviews in Food Science and Nutrition. India, 51(3), pp. 210–222. doi: 10.1080/10408390903537241.

Solomando, J. C., Antequera, T. and Perez-Palacios, T. (2020) “Evaluating the use of fish oil microcapsules as omega-3 vehicle in cooked and dry-cured sausages as affected by their processing, storage and cooking,” Meat Science. Spain: Elsevier Ltd, 162. doi: 10.1016/j.meatsci.2019.108031.

de Souza Paglarini, C. et al. (2021) “Using inulin-based emulsion gels as fat substitute in salt reduced Bologna sausage,” Journal of the Science of Food and Agriculture. Brazil: John Wiley and Sons Ltd, 101(2), pp. 505–517. doi: 10.1002/jsfa.10659.

Sowmya, M. et al. (2009) “Effect of replacement of fat with sesame oil and additives on rheological, microstructural, quality characteristics and fatty acid profile of cakes,” Food Hydrocolloids. India, 23(7), pp. 1827–1836. doi: 10.1016/j.foodhyd.2009.02.008.

Tarté, R. et al. (2020) “High-oleic and conventional soybean oil oleogels structured with rice bran wax as alternatives to pork fat in mechanically separated chicken-based bologna sausage,” LWT. United States: Academic Press, 131. doi: 10.1016/j.lwt.2020.109659.

Timilsena, Y. P. et al. (2017) “Physicochemical and thermal characteristics of Australian chia seed oil,” Food Chemistry. Australia: Elsevier Ltd, 228, pp. 394–402. doi: 10.1016/j.foodchem.2017.02.021.

Triki, M. et al. (2013) “Chilled storage characteristics of low-fat, n-3 PUFA-enriched dry fermented sausage reformulated with a healthy oil combination stabilized in a konjac matrix,” Food Control. Elsevier BV, pp. 158–165. doi: 10.1016/j.foodcont.2012.10.008.

Tudose, C. et al. (2014) “Influence of animal fat replacement with vegetable oils on the sensorial perception of meat emulsified products,” Annals of the University Dunarea de Jos of Galati, Fascicle VI: Food Technology. Romania: Galati University Press, 38(2), pp. 94–103. Available at: http://www.ann.ugal.ro/tpa/Anale%202014/8_Tudose.pdf.

Turck, D. et al. (2018) “Guidance for the scientific requirements for health claims related to antioxidants, oxidative damage and cardiovascular health: (Revision 1),” EFSA Journal. undefined: Wiley-Blackwell Publishing Ltd, 16(1). doi: 10.2903/j.efsa.2018.5136.

Urgu-Ozturk, M., Ozturk-Kerimoglu, B. and Serdaroglu, M. (2020) “Design of healthier beef sausage formulations by hazelnut-based preemulsion systems as fat substitutes,” Meat Science, 167.

Vargas-Ramella, M. et al. (2020) “Inclusion of healthy oils for improving the nutritional characteristics of dry-fermented deer sausage,” Foods. Brazil: MDPI AG, 9(10). doi: 10.3390/foods9101487.

WCRF/AICR (2007) Food, Nutrition, Physical Activity, and the Prevention of Cancer: a Global Perspective.

Weiss, J. et al. (2010) “Advances in ingredient and processing systems for meat and meat products,” Meat Science. Germany, 86(1), pp. 196–213. doi: 10.1016/j.meatsci.2010.05.008.

WHO (2003) “Diet, Nutrition and the Prevention of Chronic Diseases,” WHO Technical Report Series, 916, pp. 1–149.

Wolfer, T. L. et al. (2018) “Replacement of pork fat in frankfurter-type sausages by soybean oil oleogels structured with rice bran wax,” Meat Science. United States: Elsevier Ltd, 145, pp. 352–362. doi: 10.1016/j.meatsci.2018.07.012.

Wood, J. G. et al. (2004) “Sirtuin activators mimic caloric restriction and delay ageing in metazoans,” Nature. United States, 430(7000), pp. 686–689. doi: 10.1038/nature02789.

Zhu, Y. et al. (2020) “Beneficial effects of Jerusalem artichoke powder and olive oil as animal fat replacers and natural healthy compound sources in Harbin dry sausages,” Poultry Science. China: Elsevier Inc., 99(12), pp. 7147–7158. doi: 10.1016/j.psj.2020.08.058

Figure 1: Lipid Reformulation in Meat Products

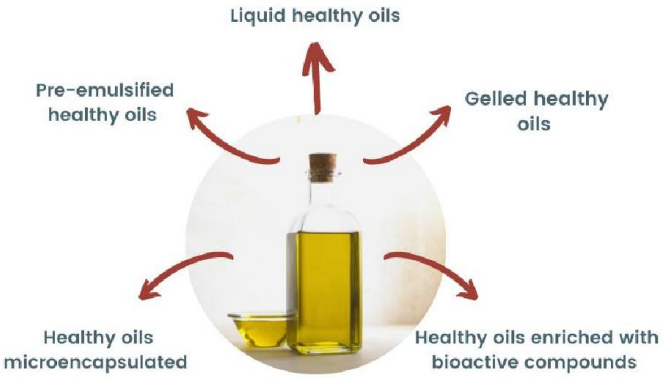
The fat content of most processed meat products is greater than 30 %, which represents a high amount of saturated fatty acids (SFA) in these products.

Several studies have shown an association between a diet rich in SFA and the incidence of obesity, cardiovascular disease, and some types of cancer

Meat products also have a high ratio of dietary n-6/n-3 polyunsaturated fatty acids (PUFAs) (15-20:1), which is nutritionally unfavorable for human health.

Improving the lipid profile of meat products is necessary to meet the demands of health-conscious consumers. The replacement of animal fat by healthy oils is one of the most effective approaches to increase the PUFA/SFA ratio and to reduce the n-6/n-3 PUFAs ratio.

Alternatives to animal fat replacement



Approaches to improve the nutritional quality of meat products by replacing animal fat with healthy oils demonstrate health benefits of reducing SFA and increasing PUFAs intake.

Figure 2:

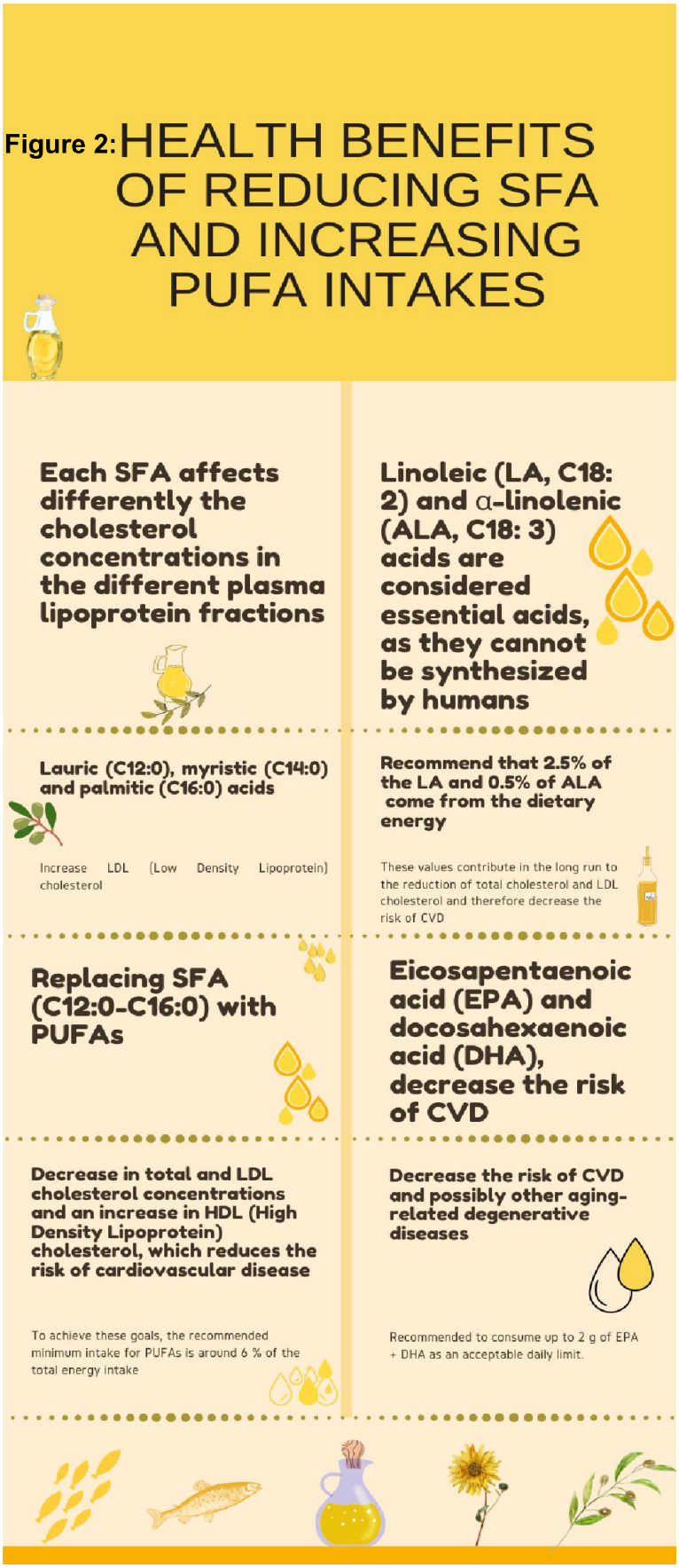

HEALTH BENEFITS OF REDUCING SFA AND INCREASING PUFA INTAKES
Each SFA affects differently the cholesterol concentrations in the different plasma lipoprotein fractions
Linoleic (LA, C18: 2) and α-linolenic (ALA, C18: 3) acids are considered essential acids, as they cannot be synthesized by humans
Lauric (C12:0), myristic (C14:0) and palmitic (C16:0) acids
Increase LDL (Low Density Lipoprotein) cholesterol
Recommend that 2.5% of the LA and 0.5% of ALA come from the dietary energy
These values contribute in the long run to the reduction of total cholesterol and LDL cholesterol and therefore decrease the risk of CVD
Replacing SFA (C12:0-C16:0) with PUFAs
Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), decrease the risk of CVD
Decrease in total and LDL cholesterol concentrations and an increase in HDL (High Density Lipoprotein) cholesterol, which reduces the risk of cardiovascular disease
To achieve these goals, the recommended minimum intake for PUFAs is around 6 % of the total energy intake
Decrease the risk of CVD and possibly other aging-related degenerative diseases
Recommended to consume up to 2 g of EPA + DHA as an acceptable daily limit.

Table 1: Addition of liquid healthy oils in meat products.

Meat product	Used oil	% of substitution	Advantages	Disadvantages	References
Pork liver pâté	Olive	50 and 100	• Nutritional improvement; • Increase in the number of PUFAs.	• Less oxidative stability.	Delgado-Pando, Cofrades, Ruiz-Capillas, Triki, & Jiménez-Colmenero, 2012.
Liver pâté	Fish	50 and 75	• Decrease in SFA and increase in PUFAs.	• Increase in volatiles derived from lipid oxidation.	Domínguez, Pateiro, Munekata, Campagnol, &Lorenzo, 2017.
Bologna sausages	Echium	50 and 100	• Technologically feasible; • Higher content of n3-PUFAs and a decreased n6 / n3 ratio.	• Less sensory acceptance in the T100 treatment.	Pires, Santos, Barros, & Trindade, 2019.
Frankfurters	Canola	50	• Lower content of saturated fat.	• Greater hardness; • Very small fat globules.	Barbut, Wood, & Marangoni, 2016.
Emulsion type meat batters	Canola	21	• Reduction of saturated fat content.	• Increased hardness and lightness.	Alejandre, Astiasarán, Ansorena, & Barbut, 2019.
Comminuted meat system	Canola, soya and linseed	25	• Increase in n3-PUFAs.	• Increase in cooking losses; • Lipid oxidation	Barbut & Marangoni, 2019.

Table 2: Addition of pre-emulsified healthy oils in meat products.

Meat product	Used oil	% of substitution	Advantages	Disadvantages	References
Fermented sausages	Olive	10 and 20	• Do not affected the processing and quality of sausages.	• Higher lightness and yellowness values; • Higher TBA values.	Bloukas, Paneras, & Fournitzis, 1997.
Fermented sausages (Chorizo de Pamplona)	Olive	0, 10, 15, 20, 25 and 30	• No changes in sensory characteristics up to 25%; •Increased fraction of MUFAs and PUFAs and reduced cholesterol content.	• 30 % substitution was unacceptable due to fat loss during ripening.	Muguerza, Gimeno, Ansorena, Bloukas, & Astiasaran, 2001.
Emulsified meat product	Olive and palm	25	• Texture and sensory acceptability were not impaired.	• Lipid oxidation increased.	Tudose, Iordachescu, Stan, Cercel, & Alexe, 2014.
Dry-fermented sausages	Linseed	25	• Similar characteristics to the control and lipid oxidation problems were not detected.	• Hexanal and nonanal presented the highest values in products containing linseed oil.	Ansorena & Astiasarán, 2004.
Fermented sausages	Linseed and canola	10, 15 and 20	• Increase in the PUFAs / SFA ratio and decrease in the n-6 / n-3 ratio.	• Lipid oxidation increased during storage.	Pelser, Linssen, Legger, & Houben, 2007.
Frankfurters	Sunflower	0, 5, 10, 15, and 20	• Reduced fat content and improved technological and sensory characteristics.	• Lower redness and yellowness values.	Choi et al., 2013.
Burgers	Algae	100	• Effective in improving important nutritional indices such as the n-6/n-3 PUFAs and PUFA/SFA ratios.	• Sensory differences were observed in the flavour and overall quality parameters.	Barros et al., 2021.
Burgers	Tiger nut (*Cyperus esculentus* L.)	50 and 100	• Increase in oleic acid contents, with no changes in the texture parameters and sensory acceptance of the burgers.	• The TN100 samples were considered as acceptable by consumers.	Barros et al., 2020.
Sausages	Grape seed	50	• Improved the technological quality of meat emulsions.	• Sausages pre-emulsified with olive oil and alginate exhibite da harder texture than the control sample.	Kim, Yong, Jung, Kim, & Choi, 2020.
Duck meat emulsions	Corn, grape seed, soybean and olive	20	• No effects on the oxidative stability, and an improvement in the nutritional quality of the products.	• Lower thermal stability than control.	Kim et al., 2021.
Sausages	Hazelnut	0, 50 and 100	• Showed excellent technological quality and high sensory acceptance.	• Higher amount of lipids.	Urgu-Ozturk, Ozturk-Kerimoglu, & Serdaroglu, 2020.

Table 3: Addition of healthy microencapsulated oils.

Meat product	Used oil	% of substitution	Technique used	Advantages	Disadvantages	References
Dutch-style fermented sausages	Fish	15 and 30	Spray drying	• Accepted sensorially.	• Lipid oxidation parameters (propanal and hexanal) showed higher values.	Josquin, Linssen, & Houben, 2012.
Chicken nuggets	Fish	25	Spray drying	• Oxidative protection without changes in sensory quality.	• Liquid oil showed greater juiciness and salty.	Pérez-Palacios, Ruiz-Carrascal, Jiménez-Martín, Solomando, & Antequera, 2018.
Pork burgers-drying	Fish	4.6	Spray-drying	• Microencapsulation was efficient to prevent oxidation of EPA and DHA in both refrigerated and frozen hamburgers.	• Higher values of TBARs were found in the samples with non-encapsulated oil.	Aquilani, et al., 2018.
Frankfurter sausages	Fish	50	Spray-drying	• Increased levels of MUFAs and n-3 PUFAs.	• Higher lipid oxidation.	Domínguez, Pateiro, Agregán, & Lorenzo, 2017.
Fermented sausages ("Chorizo")	Olive, linseed and fish	50	Matrix konjac	• Reduced the SFA content and increased the PUFAs content, making the n-6 / n-3 ratio healthier.	• The sensorial quality of the products was impaired by the reformulation.	Jiménez-Colmenero, Triki, Herrero, Rodríguez-Salas, & Ruiz-Capillas, 2013.
Fermented sausages (Spanish salchichón)	Fish	25, 50 and 75	Matrix konjac	• The total amount of PUFAs increased by 2.3 %.	• Increased lipid oxidation.	Lorenzo, Munekata, Pateiro, Campagnol, & Domínguez, 2016.
Cooked and dry-cured sausages	Fish	5	Spray-drying and freeze-drying	• The quality of enriched meat products does not appear to be impaired after storage.	• Refrigerated storage influenced the usual changes in lipid oxidation and hedonic scores.	González, Antequera, & Pérez-Palacios, 2020.
Burger	Chia and linseed	50	External ionic gelation	• It did not affect hardness and improved important technological properties; • Healthier n6 / n3 ratio and atherogenicity and thrombogenicity indexes.	• Hamburgers with chia oil microparticles showed higher lipid oxidation and lower sensory quality.	Heck et al., 2017.

Table 4: Addition of gelled healthy oils.

Meat product	Used oil	% of substitution	Technique used	Advantages	Disadvantages	References
Frankfurter sausages	Sunflower	50	Oleogels	• No changes were observed in the oxidation and sensory analysis parameters.	• Lower values of hardness, brittleness, gumminess and chewiness.	Kouzouni, Lazaridou, & Katsanidis, 2017.
Burgers	Sesame	25 and 50	Oleogels	• 11 % reduction in cooking loss and 1.6 % reduction in fat absorption.	• Shrinkage in cooking and lipid oxidation increased.	Moghtadaei, Amir, & Goli, 2018.
Fermented sausages (Spanish salchichón)	Linseed	20-40	Oleogels	• The fatty acid profile has improved nutritionally.	• Changes in sensory quality.	Franco et al., 2020.
Dry fermented sausages	Linseed	26.3, 32.8 and 39.5	Gelled emulsion	• Increase in the supply of polyunsaturated fatty acids (PUFA) (up to 10.3 %) and reductions in the n-6 / n-3 ratio.	• Formation of volatile aldehydes derived from the oxidation of lipids.	Alejandre, Poyato, Ansorena, & Astiasarán, 2016.
Bologna sausage	Soya	50 and 100	Gelled emulsion	• Nutritionally improved fatty acid profile.	• Affected the color by increasing L* and reducing a*; • Greater hardness, chewabilitymand shear strength.	Paglarini et al., 2019.
Frankfurters	Soya	10	Oleogel	• No change in the attributes of firmness, chewability and elasticity.	• In the sensory evaluation, the frankfurters aroma and flavor was significantly reduced in relation to the Control.	Wolfer, Acevedo, Prusa, Sebranek, & Tarté, 2018.
Bologna sausage	Oleic soya	41.9	Oleogel	• More favorable fatty acid profile	• Color parameters (a*, b*, c*) and sensory color intensity were higher, and L* lower in reformulated treatments.	Tarté, Paulus, Acevedo, Prusa, & Lee, 2020.
Bologna-type sausages	Oleic sunflower	25, 50, 75 and 100	Oleogel	• Emulsion stability increased and cooking loss decreased; • Increased the proportion of oleic acid within the lipid fraction by up to 20 % and decreased the proportion of linoleic acid by up to 10 %, without changes in oxidative stability.	• The sensory quality of bologna-type sausages was impaired by over 50 % substitution.	Da Silva et al., 2019.
Burgers	Canola	50	Oleogel	• Effective strategy to confer technological and nutritional advantages to low-fat burgers.	• Increase in TPA parameters (hardness, gumminess, and chewiness) and lightness (L*)	Fagundes et al., 2017.

Table 5: Addition of healthy oils enriched with bioactive compounds.

Meat product	Used oil	Antioxidant	Extraction technique	Main effects	References
Burger	Chia	Rosemary extract (*Rosmarinus officinalis L.*)	Ultrasound-assisted extraction (UAE)	• Greater oxidative stability, especially after cooking; • Reduced sensory defects caused by lipid reformulation.	Heck et al., 2018a, 2018b.
Burger	Chia and linseed	Peel jabuticaba (*Myrciaria cauliflora*)	Extraction of microwave assisted hydrodiffusion by gravitational action (MHG)	• The addition of 10 % JPE to HE in burgers controlled lipid oxidation until the 60th day of storage; • Reduced sensory defects caused by lipid reformulation.	Heck et al., 2020.
Burgers	Microalgal	Blackthorn branch extract (*Prunus spinosa L.*).	Hydroalcoholic	• Showed that the high amount of polyphenolic compounds in the extract led to an increase in the oxidative stability of microalgal oil, making viable its application in burgers.	Alejandre, Ansorena, Calvo, Cavero, & Astiasarían, 2019.
Harbin dry sausages	Olive	Jerusálem artichoke	Powder	• The results proved that the strategy was effective to increase the oxidative stability and the sensory quality of the products.	Zhu, Guo, Tang, & Yang, 2020.
Bologna sausages	Seaweed and linseed	Melissa	Lyophilized	• Healthier lipid profile; • No technological, sensory and lipid oxidation problems were detected during 32 days of refrigerated storage. Lightness, yellowness and chroma increased.	Berasategi et al., 2014.

6. CAPÍTULO 3 - OXIDATIVE STABILITY OF BURGERS CONTAINING CHIA OIL MICROPARTICLES ENRICHED WITH ROSEMARY BY GREEN-EXTRACTION TECHNIQUES

Rosane Teresinha Heck, Bruna Nichelle Lucas, Daniele Jorge Paiva Dos Santos, Mariana Basso Pinton, Mariane Bittencourt Fagundes, Mariana de Araújo Etchepare, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Juliano Smanioto Barin, Roger Wagner, Paulo Cezar Bastianello Campagnol

Meat Science

Vol. 146, p. 147-153, 2018

https://doi.org/10.1016/j.meatsci.2018.08.009

Meat Science 146 (2018) 147–153

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques

Rosane Teresinha Heck, Bruna Nichelle Lucas, Daniele Jorge Paiva Dos Santos, Mariana Basso Pinton, Mariane Bittencourt Fagundes, Mariana de Araújo Etchepare, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Juliano Smanioto Barin, Roger Wagner, Paulo Cezar Bastianello Campagnol*

Universidade Federal de Santa Maria, CEP 97105-900 Santa Maria, Rio Grande do Sul, Brazil

ARTICLE INFO

Keywords:
Natural antioxidant
Ultrasound-assisted extraction
Omega 3
Microencapsulation
Healthier meat products

ABSTRACT

In the first part of this study, the oxidative stability of chia oils enriched with rosemary by ultrasound-assisted extraction (UAE) and by a conventional maceration extraction (CME) was evaluated. In the second part, chia oil enriched with rosemary by UAE or CME was microencapsulated and used to replace 50% fat in burgers. The oxidative and sensory quality of burgers were evaluated during 120 days of storage at −18 °C. Chia oil enriched with rosemary by UAE presented a higher oxidative stability compared to CME. Higher Eh and TBARS values were found in burgers containing chia oil microparticles without rosemary. The burgers produced with chia oil microparticles enriched with rosemary by UAE showed greater oxidative stability than other treatments, mainly after cooking. Furthermore, the incorporation of rosemary antioxidants to chia oil reduced the sensory defects caused by the lipid reformulation.

1. Introduction

The partial replacement of animal fat by vegetable oils is one of the most effective strategies to reduce the saturated fatty acid content of meat products, which has gained importance due to the consumer's concern about the harmful effects of saturated fat. The addition of oils rich in n-3 PUFAs to the formulations may be an effective tool to produce meat products with a healthier lipid profile since studies have shown that the consumption of these fatty acids may reduce the risk factors for cardiovascular disease (Chaudhary et al., 2016). In this context, chia oil can be considered as a good alternative since it contains about 70% α-linolenic acid (C18:3^{9,12,15}) (Ayerza & Coates, 2005). However, the direct incorporation of oils rich in n-3 PUFAs into meat products can adversely affect important technological and sensory parameters (Monteiro, Souza, Costa, Faria, & Vicente, 2017; Xiong et al., 2016).

Heck et al. (2017) reported that the microencapsulation by the external ionic gelation technique may be effective to incorporate oils rich in n-3 PUFAs into cooked meat products. These researchers used chia and linseed oil microparticles as a substitute for animal fat in hamburgers. In addition to improving the PUFA/SFA and n-6/n-3 ratios and reducing the atherogenicity and thrombogenicity rates, this alternative lipid reformulation did not cause major changes in the sensory attributes of the processed products. However, the microencapsulation could not completely protect plant oils from the oxidative processes. This fact was observed mainly for the chia oil microparticles, since chia oil contains a higher amount of n-3 PUFAs compared to the linseed oil. Thus, alternatives to improve the oxidative stability of these microparticles should be investigated.

Although the addition of synthetic or natural antioxidants is considered the best alternative to reduce the lipid oxidation in meat products, the use of synthetic antioxidants has aroused criticism from the scientific community due to their suspected long-term toxicity. There is a vast number of natural compounds with antioxidant activity, and rosemary stands out especially for use in cooked meat products (Shah, Bosco, & Mir, 2014).

Green extraction techniques have been developed and optimized in recent years. These techniques used for the extraction of bioactive compounds are known as environmentally friendly because they reduce waste generation and energy consumption and eliminate or reduce the use of toxic solvents (Chemat et al., 2017). In this context, ultrasound (US) has been successfully used in the extraction of bioactive compounds. The advantages of ultrasound-assisted extraction (UAE) when compared to solvent extraction methods include time-saving and

* Corresponding author.
E-mail address: paulo.campagnol@ufsm.br (P.C.B. Campagnol).

https://doi.org/10.1016/j.meatsci.2018.08.009
Received 2 February 2018; Received in revised form 31 July 2018; Accepted 15 August 2018
Available online 18 August 2018
0309-1740/

increased antioxidant activity (Zbancioc, Mangalagiu, & Moldoveanu, 2015; Zhou et al., 2016). The US acts directly on the plant cell to release metabolites through the cavitation phenomenon, which increases the shear force in the environment. The cavitation on the surface of the source material can lead to surface peeling, erosion and particle breakdown, thus increasing the mass transfer process (Chemat et al., 2017).

To date, the incorporation of natural antioxidants into oils rich in n-3 PUFAs through the UAE, without the use of solvents has been little explored. Thus, in the first part of this study, the response surface methodology (RSM) was used to optimize the UAE of rosemary bioactive compounds directly in chia oil. The oxidative stability of chia oil enriched with rosemary by UAE optimized by RSM and by a conventional maceration extraction (CME) was compared. In a second step, chia oils incorporated with rosemary bioactive compounds obtained by both the UAE and CME were microencapsulated and used as a substitute for animal fat in burgers. The effect of this reformulation on the oxidative and sensory quality of the burgers was evaluated during 120 days of storage at −18 °C.

2. Materials and methods

2.1. Experiment 1. Direct incorporation of rosemary into chia oil by green-extraction techniques and production of the rosemary-enriched chia oil microparticles

2.1.1. Material

Fresh rosemary leaves (*Rosmarinus officinalis* L.) were harvested in February 2017 in the city of Santa Maria (Southern Brazil). The leaves were washed in running water and sanitized by immersion in a sodium hypochlorite solution (200 mg × kg^{-1}) for 10 min. The leaves were cut into small pieces and dried at 50 °C for 24 h in an air circulation oven (MDH, DeLeo, Brazil). The dried leaves were kept during 24 h at room temperature protected from light prior to use. The leaves were milled at the time of use in an analytical mill (MA630/1, Marconi, Brazil) until a fine powder was obtained. Chia oil was purchased from Giroil S.A. (Santo Ângelo, Brazil). The fatty acid composition of chia oil was as follows: 13.8% SFA; 7.1% MUFA (6.89% oleic acid); 79% PUFA; 20.25% n-6 PUFA (linoleic acid: 20.16%); and 58.7% n-3 PUFA (linolenic acid: 58.65%) (Heck et al., 2017).

2.1.2. Ultrasound-assisted extraction

The direct incorporation of the rosemary antioxidants into the chia oil was carried out in an ultrasonic bath (TI-H-10, Elma, Germany) operating at 1 W/cm^2, a frequency of 25 kHz, temperature of 30 ± 1 °C, sweep operating mode, and 100% amplitude. Chia oil (100 g) was mixed with different proportions of rosemary leaves (5 to 20%). After homogenization, the mixture was subjected to the ultrasonic bath for a time ranging from 300 to 900 S. *chia* oil containing the natural antioxidants extracted from the rosemary leaves was filtered on a filter paper and stored in amber flasks at 4 ± 1 °C until use (approximately 24 h).

2.1.2.1. Surface response methodology. The ultrasound-assisted extraction parameters were optimized through the Surface Response Methodology (RSM) using a Central Composite Design (CCD). The extraction time (x1) and the rosemary concentration (x2) were considered as the independent variables (Table 1). The experimental design consisted of 9 factorial experiments with 3 replicates at the central point. The antioxidant capacity of chia oil determined by the ORAC (Oxygen Radical Absorbance Capacity) assay was considered as a dependent variable. The ORAC assay was performed according to the methodology described by Ou, Hampsch-Woodill, and Prior (2001), in triplicate, and the results were expressed as μg of Trolox equivalent g^{-1} chia oil. The results of the CCD and the predicted responses were analyzed by the software Statistica 10 (Stasoft Inc., Tulsa, USA).

Table 1
Levels of the independent variables of the CCD for the direct incorporation of rosemary antioxidant compounds into chia oil using ultrasound-assisted extraction.

	−1.41	−1	0	1	+1.41
x_1: Extraction time (seg)	300	447	600	753	900
x_2: Rosemary concentration (%)	5	9.19	12.5	16.81	20

Analysis of variance (ANOVA) was used to determine the significance of the models, at a level of significance of 95%.

2.1.2.2. Validation of the study. To validate the model, three experiments were performed. The ORAC assay was performed in chia oil samples containing 5, 12.5, and 20% rosemary and subjected to UAE for 600 s. The percentage error between the actual value obtained in the ORAC assay and the value predicted by the model was calculated as follows: Error (%) = [Actual values - Predicted values / Predicted values] x 100. The model can be considered acceptable when present an error lower than 10% (Bimakr et al., 2013).

2.1.3. Conventional maceration extraction

The conventional maceration extraction consisted of the addition of 12.5 g of dehydrated rosemary leaves to 100 g chia oil. The mixture was homogenized in an Ultra Turrax homogeneizer (Model T18, IKA, Brazil) at 10000 rpm for 2 min and allowed to stand in the absence of light for four hours. Thereafter, the mixture was filtered and the flavored oil was stored in an amber bottle under refrigeration (4 ± 1 °C) during 24 h.

2.1.4. Determination of the antioxidant capacity and induction period of chia oils enriched or not with rosemary

The antioxidant capacity of the three oils (chia oil without rosemary, chia oil enriched with rosemary by UAE and chia oil enriched with rosemary by CME) was evaluated by the ORAC assay (Ou et al., 2001). Also, an accelerated oxidation test was performed by measuring the Rancimat induction period (892, Metrohm, Herisau, Switzerland) at 110 °C and an air flow rate of 20 L/h. All analyses were performed in triplicate.

2.1.5. Production of the microparticles

The microparticles were produced by the external ionic gelation technique (Etchepare et al., 2016). For that, chia oil was mixed with a 2% sodium alginate solution. The mixture was homogenized in an Ultra Turrax homogeneizer (Model T18, IKA, Brazil) at 10000 rpm for 2 min. After complete homogenization, the mixture was atomized in 0.1 M $CaCl_2$ solution using a dual fluid atomizer nozzle (0.1 mm) at a distance of 12 cm from the solution, under air pressure of 0.125 kg/cm. After atomization, the microparticles were kept under constant stirring for 30 min, and then sieved in a wire mesh sieve (150 μm) and washed with sterile distilled water. Three types of chia oil were used for the production of the microparticles, as follows: microparticles M1 produced with chia oil without rosemary; microparticles M2 produced with rosemary-enriched chia oil using the UAE, and microparticles M3 produced with rosemary-enriched chia oil using a CME. The microparticles were stored under refrigeration (4 ± 1 °C) during 12 h. The three types of microparticles had a microencapsulation efficiency of approximately 87%, which was similar to other studies (Chang, Yarankovich, & Nickerson, 2016; Heck et al., 2017).

2.2. Experiment 2. Evaluation of the oxidative stability and sensory acceptance of the burgers with replacement of 50% fat by the rosemary-enriched chia oil microparticles

2.2.1. Production of burgers

Three independent replicates of each batch were prepared. Beef

(*rectus femoris*), pork back fat, and spices (salt and garlic) were purchased in a local market. The pork back fat and beef were ground separately using a 3 mm disc (Model PJ22, Jamar Ltda, São Paulo, Brazil). Then, beef was mixed with salt for extraction of the myofibrillar proteins. After this step, the remaining ingredients were added and mixed. Burgers (100 g), 11 cm in diameter and 2.5 cm thick were produced, and immediately frozen and stored at −18 °C during 120 days.

The Control sample was made with 78.4% beef, 20% pork back fat, 1.5% salt, and 0.1% garlic. For the treatment HCO, 50% fat of the Control sample was replaced by the unencapsulated chia oil ($2.5\,g \times 100\,g^{-1}$) and water ($7.5\,g \times 100\,g^{-1}$). For the treatments HM1, HM2, and HM3, 50% back fat was replaced by the microparticles M1, M2, and M3, respectively.

After 1, 30, 60, 90 and 120 days of storage, the frozen samples were thawed at 4 °C in a refrigerator for 12 h. After, the samples were cooked in an electric grill (Multi Grill, Britânia, Brazil) preheated to 150 °C, until reaching an internal temperature of 72 °C in the geometric center of each burger, which was measured by a spit thermometer (HM-600, Highmed, Brazil).

2.2.2. Determination of pH and redox potential (Eh)

The pH and the redox potential (Eh) were determined in triplicate in the raw and cooked hamburgers after 1, 30, 60, 90, and 120 days of storage. For that, 5 g sample was homogenized with 50 mL of distilled water and the pH and Eh were measured using a digital pH meter (DM-23-DC, Digimed, Brazil) equipped with pH penetration electrode and redox platinum electrode, respectively.

2.2.3. Color measurements

The color of raw and cooked hamburgers was measured after 1, 30, 60, 90, and 120 days of storage. A Minolta CR-400 colorimeter (Minolta Sensing Inc. Konica, Japan) was used with spectral reflectance included as calibration mode, illuminant D65, and an observation angle of 10°, operating in the CIELAB system. Color variables were measured at six points on each side of three samples per treatment. The L* (lightness), a* (intensity of the red color), b* (intensity of the yellow color) values were determined.

2.2.4. Determination of proximate composition and thiobarbituric acid reactive substances (TBARS)

The proximate composition (moisture, protein and fat) was determined in triplicate in raw and cooked burgers after 1 day of storage (AOAC, 2005). The determination of TBARS was performed in triplicate in raw and cooked burgers according to Bruna, Ordonez, Fernández, Herranz, and Hoz (2001), using trichloroacetic acid instead of perchloric acid as solvent. The results were expressed in milligrams of malonaldehyde per kg of the sample. The analyses were performed after 1, 30, 60, 90, and 120 days of storage.

2.2.5. Consumer study

This study was approved by the Research Ethics Committee of the Federal University of Santa Maria (RS, Brazil) (CAAE: 57433316.8.0000.5346). The samples were coded with three digits, and served to the assessors in a monadic order, following a balanced design (Ares, Barreiro, Deliza, Giménez, & Gámbaro, 2010). The sensory evaluation was performed in individual booths with fluorescent lighting. The cooked burgers were cut into 4 cm × 4 cm × 2.5 cm, individually wrapped in foil, and kept at 60 °C in an oven. Water at room temperature and cracker biscuits were provided to consumers for palate cleansing. The analysis was performed on the first day of storage and after 120 days of storage. One hundred burger consumers (64% female, 36% male at day 1; and 56% female, 44% male at day 120), aged 18–55 participated in the tests. A sensory acceptance test was performed using a structured hedonic scale of nine points, ranging from “disliked very much” to “liked very much” (Stone, Bleibaum, & Thomas, 2012). The attributes color, aroma, flavor, texture, and overall acceptance were evaluated (Meilgaard, Carr, & Civille, 2006).

Table 2
Coded and real levels of the extraction time (x1) and rosemary concentration (x2) and ORAC values for different levels of experimental design.

Run	Extraction time (s) (x1)	Rosemary concentration (x2)	ORAC (μg Trolox/g)
1	(−1) 447	(−1) 9.19	106.78
2	(1) 753	(−1) 9.19	223.51
3	(−1) 447	(1) 16.81	97.97
4	(1) 753	(1) 16.81	171.65
5	(−1.41) 300	(0) 12.5	53.31
6	(+1.41) 900	(0) 12.5	114.21
7	(0) 600	(−1.41) 5	113.84
8	(0) 600	(+1.41) 20	123.57
9	(0) 600	(0) 12.5	231.52
10	(0) 600	(0) 12.5	261.96
11	(0) 600	(0) 12.5	205.12
12	(0) 600	(0) 12.5	234.28

2.2.6. Statistical analysis

A randomized complete block design was used and the experiment was repeated three times. Data were analyzed using a general linear model considering the treatments as a fixed effect and the replicates as a random effect. Significant differences were analyzed by the Tukey's test at the 5% level of significance. Data were analyzed using the SPSS statistical program (SPSS, Chicago, IL, USA).

3. Results and discussion

3.1. Experiment 1. Direct incorporation of rosemary into chia oil by green-extraction techniques

3.1.1. Optimization of the experimental conditions for the direct incorporation of rosemary in chia oil using UAE and validation of the study

The antioxidant capacity of chia oil enriched with different rosemary concentrations using the UAE ranged from 53.31 to 261.96 μg Trolox/g (Table 2). These data were used to obtain the response surface, correlating the parameters concentration and the extraction time (Fig. 1). The analysis of variance ($R^2 = 0.9147$) showed that the linear and the quadratic term of the extraction time, and the quadratic term of the rosemary concentration were significant ($P < .001$) for the extraction of the antioxidant compounds from rosemary leaves. The application of RSM (Fig. 1) suggested that the combination of approximately 600 to 700 s of extraction time and 10 to 14% concentration of rosemary leaves provided greater antioxidant capacity to chia oil. Thus, the conditions of the central point (12.5% of rosemary leaves and 600 s of extraction time) were chosen to enrich the chia oil used in the production of microparticles.

The validation of the study was performed with the purpose of verifying the accuracy of the models, and the percentage error allowed comparing the experimental and predicted values. The percentage error of the present study ranged from 0.34 to 1.14% (data not shown). Thus, it can be stated that the model was adequate to identify the optimal US time and rosemary concentration, since according to Bimakr et al. (2013) < 10% error is considered acceptable.

3.1.2. Determination of the antioxidant activity and induction period of chia oils enriched or not with rosemary

The results of the antioxidant activity determined by the ORAC assay and the oxidative stability measured through the induction period of chia oils enriched or not with rosemary are presented in Table 3. As expected, chia oils enriched with rosemary by UAE or CME presented a higher oxidative stability than chia oil without rosemary. The UAE process was more effective in incorporating the rosemary antioxidant compounds in chia oil when compared to the CME. In agreement with

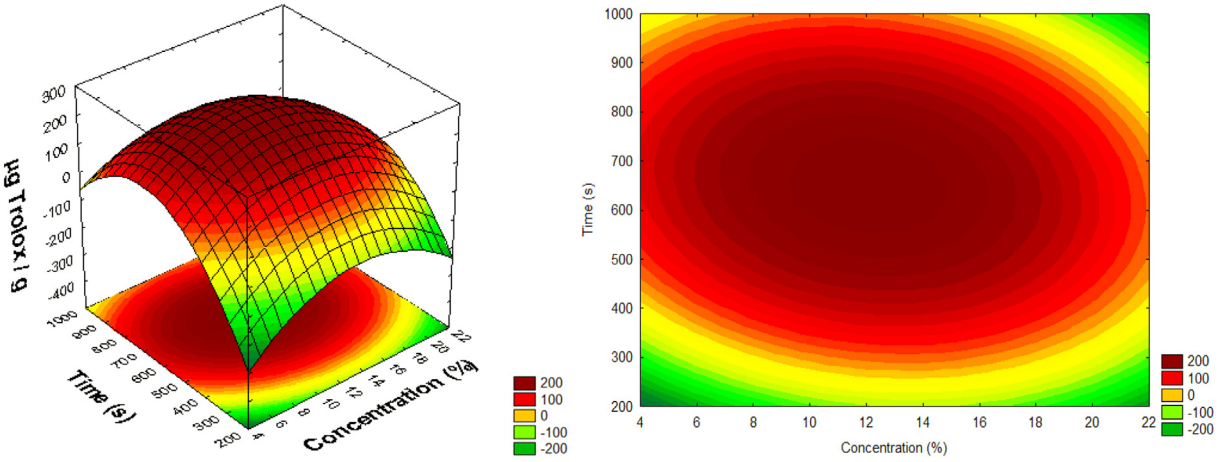


Fig. 1. 3D and 2D graphics for the effects of rosemary concentration and extraction time on the antioxidant capacity of chia oil enriched with different rosemary concentrations using the ultrasound-assisted extraction.

Table 3
ORAC and Rancimat values of chia oils enriched or not with rosemary by ultrasound-assisted extraction (UAE) and conventional maceration extraction (CME).

Treatments	Chia	Chia UAE	Chia CME	SEM	Sig
ORAC (µg Trolox/g)	131.90^{c}	228.86^{a}	204.13^{b}	23.96	*
Induction time (hours)	0.08^{c}	6.29^{a}	4.81^{b}	0.33	**

SEM- Standard error of the mean.
Treatments: Chia: chia oil without rosemary; Chia UAE: chia oil enriched with rosemary by UAE; Chia CME: chia oil enriched with rosemary by CME.
** $P < .001$.
* $P < .01$.

the results of the antioxidant activity, UAE increased the induction period ($P < .001$), conferring a greater oxidative stability to chia oil, probably due to the cavitation bubbles from the changes in temperature and pressure. The implosion of these bubbles can lead to a rupture of the cell walls of the plant, favoring the release of bioactive compounds (Dolatowski & Stasiak, 2012; Tiwari, 2015).

3.2. Experiment 2. Evaluation of the oxidative stability and sensory acceptance of burgers with replacement of 50% fat by the rosemary-enriched chia oil microparticles

3.2.1. Determination of pH and Eh

The lipid reformulation significantly affected the pH values (Table 4). The addition of the microparticles increased ($P < .001$) the pH of the raw and cooked burgers when compared to the Control at the beginning of storage (day 1), probably due to the alkaline pH of the sodium alginate solution used in the production of the microparticles. However, the addition of non-encapsulated chia oil (HCO) caused a significant ($P < .001$) decrease in pH of the raw and cooked burgers at day 1, probably due to the use of unrefined chia oil. An increase in pH after 30 days of storage can be observed in the raw and cooked burgers, except in HM1 and HM2 cooked burgers, due to the release of alkaline compounds from the protein degradation reactions (Lu, Zhang, Liu, Wang, & Ding, 2011). After 60 days of storage, a decrease in pH values was observed for all treatments, except for HM1 cooked burgers, which remained practically stable until the end of storage (day 120). However, it is noteworthy that despite the changes caused by the lipid reformulation and storage, the pH values were considered adequate for this type of meat product (Comi, Tirloni, Andyanto, Manzano, & Iacumin, 2015; Kryževičūtė, Jaime, Diez, Rovira, & Venskutonis, 2017).

The redox potential (Eh) is an effective parameter to evaluate the oxidative stability of meat products (Latoch & Stasiak, 2015). It can be defined as the substrate's ability to gain or lose electrons, that is, its oxidation or reduction capacity, respectively. Thus, the higher the Eh, the more oxidized the substrate. The lipid reformulation proposed in this study led to important changes in Eh (Table 4). In the raw burgers, the treatment HCO presented lower Eh values ($P < .001$) when compared to the control at days 1, 30, and 120 of storage, which suggests a lower oxidation. Similar behavior was observed for the treatment containing chia oil microparticles without the antioxidants (HM1). However, an opposite behavior was observed after cooking, since the treatments HCO and HM1 presented higher Eh values ($P < .001$) than the Control during the whole period of storage. This result demonstrates that the addition of oils rich in n-3 high PUFAs in cooked meat products may be a barrier since heating accelerates the oxidation of these fatty acids (Malheiro et al., 2009). On the other hand, the raw burgers of the treatments HM2 and HM3 had lower Eh values ($P < .001$) during storage when compared to the Control. In the cooked burgers, the treatments HM2 and HM3 presented lower Eh values ($P < .001$) when compared to the control at days 1, 30, and 120 of storage. These results suggest that the direct incorporation of rosemary extract into chia oil using UAE or CME was effective to reduce the oxidative reactions.

3.2.2. Instrumental color

The results of the L*, a* and b* values of the raw and cooked burgers are presented in Table 5. The addition of chia oil microparticles (HM1, HM2 and HM3) did not affect the L* values during the storage of the raw burgers when comparing to the Control. In addition, the raw burgers of the treatments HM2 and HM3 did not present significant differences ($P > .05$) in relation to the Control in the a* values during the storage. However, lower a* values were found in the raw burgers of the treatment HM1 in relation to the control after 60, 90 and 120 days of storage. In addition, the raw burgers of the treatment HCO presented in relation to the control a lower a* values in the end of storage (day 120). This result suggests that chia oil in both the encapsulated and non-encapsulated form accelerated the oxidative processes. Regarding the b* values of the raw burgers, higher values were observed after 1 and 30 days of storage for the treatments HM1, HM2, and HM3 when compared to the Control, probably due to the yellowish color of the microparticles. HM2 and HM3 also presented a higher b* values than Control after 120 and 60 days of storage, respectively. A significant decrease of the b* values of the raw burgers was observed in all treatments after 120 days compared to beginning of storage (day 1). This behavior was also reported by Martínez et al. (2012) and Valencia, Ansorena, and Astiasarán (2006) in meat products with the addition of fish oil.

The cooked burgers HM1 and HM3 presented higher L* values

Table 4
pH e Eh (mV) values during storage of burgers.

	Days	Raw					SEM	Sig	Cooked					SEM	Sig
		Control	HCO	HM1	HM2	HM3			Control	HCO	HM1	HM2	HM3		
pH	1	5.77^{cB}	5.75^{dB}	5.80^{bBC}	5.79^{bB}	5.83^{aB}	0.003	***	5.96^{cB}	5.93^{dD}	6.03^{aA}	5.99^{bBC}	6.04^{aC}	0.003	***
	30	6.01^{dA}	6.04^{cA}	6.14^{bA}	5.95^{eA}	6.27^{aA}	0.003	***	6.06^{bA}	6.05^{bA}	5.98^{dAB}	6.02^{cB}	6.15^{aA}	0.004	***
	60	5.60^{dD}	5.70^{cC}	5.74^{bC}	5.73^{bC}	5.78^{aC}	0.004	***	5.94^{eC}	5.99^{bB}	5.97^{cAB}	6.07^{aA}	5.95^{dD}	0.002	***
	90	5.66^{cC}	5.67^{cD}	5.76^{bC}	5.73^{cC}	5.84^{aB}	0.005	***	5.95^{aC}	5.96^{aC}	5.96^{aB}	5.97^{aC}	5.95^{aD}	0.012	n.s.
	120	5.67^{eC}	5.69^{dCD}	5.83^{aB}	5.74^{bC}	5.72^{cD}	0.002	***	5.97^{dB}	5.99^{cB}	5.93^{eB}	6.01^{bBC}	6.07^{aB}	0.002	***
	SEM	0.007	0.008	0.022	0.008	0.017	–	–	0.004	0.005	0.022	0.013	0.010	–	–
	Sig	***	***	***	***	***	–	–	***	***	***	***	***	–	–
Eh (mV)	1	45.22^{aE}	40.77^{bE}	34.44^{cD}	34.44^{cE}	33.22^{cD}	0.75	***	49.55^{bD}	56.33^{aCD}	58.44^{aBC}	34.33^{cD}	27.22^{dD}	1.54	***
	30	75.44^{aD}	57.11^{cD}	71.77^{bC}	57.77^{cD}	57.22^{cB}	1.21	***	56.77^{cC}	66.88^{aB}	61.11^{bB}	46.22^{dC}	39.88^{eC}	1.01	***
	60	79.00^{aC}	77.77^{aC}	76.77^{aC}	73.77^{bC}	71.55^{bC}	1.00	***	48.22^{bD}	52.22^{aCD}	53.22^{aBC}	49.33^{bBC}	47.77^{bB}	0.97	***
	90	94.22^{aB}	91.77^{aB}	92.66^{aB}	82.33^{bB}	74.11^{cC}	1.75	***	49.22^{cC}	56.66^{bC}	66.55^{aB}	48.00^{cB}	47.77^{cB}	0.97	***
	120	152.33^{aA}	138.11^{bA}	137.77^{bA}	137.55^{bA}	134.44^{bA}	2.10	***	89.66^{bA}	96.66^{aA}	93.00^{aA}	82.88^{cA}	85.22^{cA}	1.64	***
	SEM	2.91	2.69	2.37	2.37	2.15	–	–	2.14	2.24	2.35	2.36	2.17	–	–
	Sig	***	***	***	***	***	–	–	***	***	***	***	***	–	–

SEM- Standard error of the mean. Mean values within the same line followed by the same lowercase letters did not show any significant difference ($P > .05$) by Tukey test. Mean values within the same column followed by the same upper case did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; HCO: 50% substitution of pork back fat by chia oil; HM1: 50% substitution of pork back fat by microparticles produced with chia oil without rosemary; HM2: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using the ultrasound-assisted extraction; HM3: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using a conventional maceration extraction.
Sig.: significance; n.s. (not significant).
*** $P < .001$.

($P < .001$) than Control at the beginning of storage (day 1). However, after 60, 90 and 120 days of storage, there was an increase ($P < .001$) in L* values in HM1, HM2, and HM3, suggesting a lower moisture retention in these formulations (Fernandes et al., 2014). In general, no significant differences were observed in the a* values for the treatments containing the microparticles (HM1, HM2, and HM3) when compared to the control. A lower a* value ($P < .001$) was observed in the HM2 compared to the control just in the 30th day of storage. In addition, a slight increase in b* values was found in the treatment HM1 in relation to control after 30, 90 and 120 days of storage. The treatments HCO and HM2 also showed a slight increase in b* values when compared to the control in the day 90 and 1 of storage, respectively. The cooking masked the changes in color caused by the lipid reformulation of the raw burgers. So, it can be concluded that the lipid reformulation did not

Table 5
Color values during storage of burgers.

		Raw							Cooked						
	Days	Control	HCO	HM1	HM2	HM3	SEM	Sig	Control	HCO	HM1	HM2	HM3	SEM	Sig
L_*	1	46.92^{aA}	47.12^{aA}	48.05^{aA}	49.01^{aA}	47.50^{aA}	1.31	n.s.	45.50^{bcA}	45.08^{cAB}	47.94^{aA}	47.43^{abA}	48.11^{aA}	0.71	***
	30	48.99^{aA}	44.41^{bA}	47.71^{abA}	48.44^{aA}	46.97^{abA}	1.33	***	44.49^{bA}	42.46^{cC}	43.99^{bcB}	47.20^{aA}	45.38^{bB}	0.62	***
	60	45.74^{abA}	45.04^{bA}	46.21^{abA}	48.80^{aA}	45.86^{abA}	1.14	***	44.20^{bA}	47.10^{aA}	46.11^{aAB}	46.52^{aA}	47.37^{aAB}	0.54	***
	90	47.33^{aA}	44.14^{aA}	47.13^{aA}	47.53^{aA}	46.41^{aA}	1.47	n.s.	42.63^{bA}	42.92^{bBC}	47.79^{aA}	47.72^{aA}	46.68^{aAB}	0.99	***
	120	43.61^{aA}	45.35^{aA}	45.94^{aA}	47.04^{aA}	47.56^{aA}	1.65	n.s.	43.53^{cA}	45.00^{bcAB}	46.14^{abAB}	48.14^{aA}	47.05^{abAB}	0.82	***
	SEM	2.19	1.10	1.06	1.25	1.43	–	–	1.18	0.87	0.88	0.58	0.77	–	–
	Sig	n.s.	n.s.	n.s.	n.s.	n.s.	–	–	n.s.	**	***	n.s.	***	–	–
a*	1	22.58^{aA}	23.62^{aA}	23.76^{aA}	23.91^{aA}	23.43^{aA}	0.67	n.s.	10.29^{abA}	10.27^{abC}	10.56^{aB}	10.41^{aA}	9.76^{bC}	0.20	***
	30	19.52^{abD}	18.26^{bB}	16.59^{bB}	20.48^{aB}	18.07^{bBC}	0.68	***	11.32^{abA}	10.85^{abcBC}	11.41^{aAB}	10.50^{cA}	10.79^{bcB}	0.20	***
	60	18.68^{abB}	17.37^{bcB}	16.38^{cB}	18.06^{bC}	19.64^{aB}	0.55	***	10.93^{abA}	11.26^{abAB}	11.30^{aA}	10.74^{bA}	10.87^{abAB}	0.18	**
	90	16.40^{aBC}	17.04^{aB}	13.93^{bC}	17.34^{aC}	17.21^{aC}	0.70	***	10.29^{aA}	10.82^{aBC}	10.72^{aAB}	10.75^{aA}	10.79^{aB}	0.40	n.s.
	120	15.41^{aC}	13.12^{bcC}	11.13^{cD}	16.35^{aC}	14.39^{abD}	0.75	***	11.18^{bcA}	11.85^{aA}	11.24^{abcAB}	10.65^{cA}	11.53^{abA}	0.23	***
	SEM	0.96	0.68	0.74	0.72	0.69	–	–	0.38	0.26	0.27	0.23	0.24	–	–
	Sig	***	***	***	***	***	–	–	n.s.	***	**	n.s.	***	–	–
b*	1	16.77^{bA}	17.96^{abA}	18.72^{aA}	19.22^{aA}	18.50^{aA}	0.48	***	15.82^{bA}	16.21^{bA}	16.67^{bA}	18.20^{aA}	16.41^{bA}	0.38	***
	30	13.41^{cBC}	15.51^{bB}	16.13^{bB}	17.62^{aB}	16.23^{bBC}	0.47	***	15.72^{bA}	15.68^{bA}	17.11^{aA}	16.84^{abB}	16.08^{abA}	0.41	***
	60	15.18^{bAB}	15.05^{bB}	14.97^{bBC}	15.94^{abC}	17.18^{aAB}	0.52	**	15.14^{aAB}	15.63^{aA}	16.07^{aA}	16.02^{aB}	15.49^{aA}	0.34	n.s.
	90	14.77^{aBC}	15.12^{aB}	15.59^{aB}	15.68^{aC}	16.20^{aBC}	0.60	n.s.	14.13^{bB}	16.60^{aA}	16.07^{aA}	15.84^{abB}	15.60^{abA}	0.63	***
	120	13.11^{bC}	13.07^{bC}	13.63^{bcC}	15.79^{aC}	14.88^{abC}	0.60	***	15.45^{bAB}	16.21^{abA}	16.95^{aA}	16.02^{abB}	16.34^{abA}	0.36	***
	SEM	0.66	0.60	0.59	0.56	0.57	–	–	0.54	0.43	0.49	0.44	0.44	–	–
	Sig	***	***	***	***	***	–	–	*	n.s.	n.s.	***	n.s.	–	–

SEM- Standard error of the mean. Mean values within the same line followed by the same lowercase letters did not show any significant difference ($P > .05$) by Tukey test. Mean values within the same column followed by the same upper case did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; HCO: 50% substitution of pork back fat by chia oil; HM1: 50% substitution of pork back fat by microparticles produced with chia oil without rosemary; HM2: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using the ultrasound-assisted extraction; HM3: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using a conventional maceration extraction.
Sig.: significance; n.s. (not significant).
*** $P < .001$.
** $P < .01$.
* $P < .05$.

Table 6
Proximate composition (moisture, protein and fat) and TBARS values during storage of burgers.

	Days	Raw					SEM	Sig	Cooked					SEM	Sig
		Control	HCO	HM1	HM2	HM3			Control	HCO	HM1	HM2	HM3		
Moisture (%)	1	62.6^{b}	66.9^{a}	67.5^{a}	67.53^{a}	69.35^{a}	0.95	***	55.21^{b}	52.81^{b}	59.14^{a}	58.16^{a}	58.55^{a}	0.96	***
Protein (%)	1	18.53^{a}	18.69^{a}	19.33^{a}	19.6^{a}	18.83^{a}	0.3	n.s.	24.87^{a}	24.45^{a}	25.26^{a}	25.38^{a}	24.99^{a}	0.66	n.s.
Fat (%)	1	18.22^{a}	8.43^{b}	8.52^{b}	8.55^{b}	8.46^{b}	0.16	***	15.06^{a}	6.71^{c}	9.08^{b}	9.18^{b}	9.19^{b}	0.16	***
TBARs	1	0.151^{bD}	0.20^{bC}	0.302^{aC}	0.174^{bC}	0.188^{bD}	0.019	***	0.404^{cdB}	0.452^{cD}	1.128^{aB}	0.395^{dC}	0.585^{bD}	0.016	***
(mg MDA/kg sample)	30	0.451^{bB}	0.438^{bB}	0.659^{aB}	0.231^{dC}	0.347^{cC}	0.015	***	0.438^{dB}	0.599^{bC}	0.870^{aC}	0.260^{eD}	0.530^{cE}	0.022	***
	60	0.344^{bcC}	0.636^{aA}	0.714^{aB}	0.244^{cC}	0.352^{bC}	0.034	***	0.445^{dB}	0.641^{cC}	1.141^{aB}	0.378^{eC}	0.720^{bC}	0.016	***
	90	0.406^{cB}	0.594^{aA}	0.561^{abBC}	0.435^{cB}	0.488^{bcB}	0.033	***	0.630^{cA}	1.609^{aA}	1.242^{bB}	0.856^{cA}	0.844^{cB}	0.073	***
	120	0.525^{bA}	0.653^{bA}	1.211^{aA}	0.579^{bA}	0.711^{bA}	0.089	***	0.569^{dA}	0.999^{cB}	1.596^{aA}	0.639^{dB}	1.293^{bA}	0.04338	***
	SEM	0.019	0.028	0.091	0.030	0.031	–	–	0.021	0.025	0.080	0.018	0.016	–	***
	Sig	***	***	***	***	***	–	–	***	***	***	***	***	–	

SEM- Standard error of the mean. Mean values within the same line followed by the same lowercase letters did not show any significant difference ($P > .05$) by Tukey test. Mean values within the same column followed by the same upper case did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; HCO: 50% substitution of pork back fat by chia oil; HM1: 50% substitution of pork back fat by microparticles produced with chia oil without rosemary; HM2: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using the ultrasound-assisted extraction; HM3: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using a conventional maceration extraction.
Sig.: significance; n.s. (not significant).
*** $P < .001$.

cause great modifications in the color of cooked burgers. 3.2.3 Proximate composition and determination of TBARS.

The proximate composition (moisture, protein and fat) and TBARS values of the raw and cooked burgers are shown in Table 6. The protein content was not affected ($P > .05$) by lipid reformulation in the raw and cooked burgers. As expected, the lipid reformulation increased moisture ($P < .001$) and reduced the lipid content ($P < .001$) of raw burgers. The reformulated burgers presented a reduction of approximately 53% in the fat content in relation to the Control before cooking. After cooking, a reduction in the moisture content was observed in all treatments, leading to an increase in the protein content. The Control and HCO treatment presented after cooking a reduction of about 17.34% and 20.4%, respectively, of the fat content. This result is expected, as the increase of temperature promote the coalescence of the fat globules and subsequent release of the product (Tabarestani & Tehrani, 2014). Probably, a reduction of fat content after cooking did not occur in the treatments HM1, HM2 and HM3 due to the high thermal stability of the microparticles as reported by Heck et al. (2017).

Heck et al. (2017) reported that chia oil microparticles produced by the external ionic gelation technique increased the TBARS values in burgers. This fact was also confirmed in this study since the treatment HM1 had higher TBARS values than the Control before and after cooking. These changes during storage can be related to the production of the microparticles, because the oxygen incorporated during the atomization stage can accelerate the lipid oxidation reactions (Pignitter et al., 2014).

The addition of non-encapsulated chia oil (HCO) increased the TBARS values of the raw burgers in relation to the control after 60 and 90 days of storage. This result was evidenced mainly after cooking, since an increase of 37, 44, and 155% TBARS was observed for the treatment HCO when compared to the control after 30, 60, and 90 days of storage, respectively. In agreement with these results, some researchers found higher TBARS values after the incorporation of n-3 PUFAs rich oils in meat products (Delgado-Pando, Cofrades, Ruiz-Capillas, Triki, & Jiménez-Colmenero, 2012; Domínguez, Pateiro, Munekata, Campagnol, & Lorenzo, 2016; Josquin, Linssen, & Houben, 2012; Lorenzo, Munekata, Pateiro, Campagnol, & Domínguez, 2016).

The direct incorporation of the rosemary antioxidants into chia oil by both UAE and CME was effective to reduce the lipid oxidation during the microencapsulation process. The treatments HM2 and HM3 presented similar TBARS values ($P > .05$) to the Control before cooking at days 1, 60, 90 and 120 of storage. However, the cooked burgers of the treatment HM3 presented higher ($P < .001$) TBARS values than control after 1, 30, 60 and 120 days of storage. On the other hand, the cooked burgers of the treatment HM2 showed lower ($P < .001$) TBARS values than control after 30 and 60 days of storage. In addition, no significant differences in TBARS values were observed in the cooked burgers of the treatment HM2 and the control at 1, 90 and 120 days of storage.

Table 7
Consumer sensory evaluation during storage of burgers.

	Day 1					SEM	Sig	Day 120					SEM	Sig
	Control	HCO	HM1	HM2	HM3			Control	HCO	HM1	HM2	HM3		
Color	6.74^{a}	6.83^{a}	6.31^{a}	6.67^{a}	6.72^{a}	0.27	n.s.	6.48^{a}	7.06^{a}	6.80^{a}	6.59^{a}	6.94^{a}	0.26	n.s.
Aroma	6.57^{a}	6.62^{a}	6.05^{b}	6.66^{a}	6.61^{a}	0.27	**	6.06^{c}	6.28^{c}	6.57^{bc}	6.91^{ab}	7.20^{a}	0.27	***
Flavor	6.78^{a}	6.97^{a}	6.02^{b}	6.59^{a}	6.42^{a}	0.30	*	6.21^{b}	6.81^{ab}	6.65^{ab}	6.60^{ab}	6.94^{a}	0.27	n.s.
Texture	6.55^{a}	6.71^{a}	6.06^{a}	6.46^{a}	6.47^{a}	0.28	n.s.	6.43^{a}	6.70^{a}	6.72^{a}	6.43^{a}	7.16^{a}	0.28	n.s.
Overall accaptance	6.67^{ab}	6.89^{a}	6.17^{b}	6.7^{ab}	6.49^{ab}	0.28	**	6.26^{b}	6.55^{ab}	6.47^{ab}	7.01^{a}	7.03^{a}	0.26	*

SEM- Standard error of the mean. Mean values within the same line followed by the same lowercase letters did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; HCO: 50% substitution of pork back fat by chia oil; HM1: 50% substitution of pork back fat by microparticles produced with chia oil without rosemary; HM2: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using the ultrasound-assisted extraction; HM3: 50% substitution of pork back fat by microparticles produced with rosemary-enriched chia oil using a conventional maceration extraction.
Sig.: significance; n.s. (not significant).
*** $P < .001$.
** $P < .01$.
* $P < .05$.

3.2.3. Consumer study

The results of the consumer study are presented in Table 7. As reported by Heck et al. (2017) the substitution of pork back fat by the chia oil microparticles reduced the sensory quality of the burgers. Similar findings were obtained in this study, since the treatment HM1 presented significantly lower scores for the attributes aroma and flavor after 1 day of storage, when compared to the Control. The lower sensory acceptance can be correlated with the high TBARS (1128 mg MDA/kg sample) values in HM1 at day 1. In addition, the presence of $CaCl_2$ in the microparticles may also have affected these results, since studies have shown that this salt impairs the sensory quality of meat products (Dos Santos et al., 2015; Horita, Morgano, Celeghini, & Pollonio, 2011). The direct incorporation of rosemary in chia oil was effective to solve the sensory problems caused by the microencapsulation process, since all sensory scores for the treatments HM2 and HM3 were similar to the Control at day 1. In addition, HM2 and HM3 presented higher scores than Control for the attributes aroma and overall acceptance at the end of storage (day 120). The incorporation of non-encapsulated chia oil (HCO) did not impair the sensory quality of the burgers after 1 and 120 days of storage in relation to Control. However, after 120 days of storage, a lower acceptance of the attribute aroma was found in HCO when compared to the treatments HM2 and HM3.

4. Conclusion

Chia oil enriched with rosemary by UAE presented higher oxidative stability than chia oil enriched with rosemary by CME. The replacement of 50% pork back fat by chia oil microparticles produced by the external ion gelation technique decreased the oxidative stability during the storage of the burgers. The direct incorporation of rosemary in chia oil by UAE before the microencapsulation proved to be a viable alternative to solve this problem. Thus, the present results showed that it is possible to replace the animal fat in burgers by oils rich in n-3 PUFAs, without compromising their oxidative and sensory quality.

References

AOAC (Association of Official Analytical Chemistry) (2005). *Official methods of analysis of AOAC International* (17th ed.). (Maryland: USA).

Ares, G., Barreiro, C., Deliza, R., Giménez, A., & Gámbaro, A. (2010). Application of a check-all-that-apply question to the development of chocolate milk desserts. *Journal of Sensory Studies, 96*, 47–55.

Ayerza, R., & Coates, W. (2005). Ground chia seed and chia oil effects on plasma lipids and fatty acids in the rat. *Nutrition Research, 25*, 995–1003.

Bimakr, M., Rahman, R. A., Taip, F. S., Adzahan, N. M., Sarker, M. Z., & Ganjloo, A. (2013). Supercritical carbon dioxide extraction of seed oil from winter melon (Benincasa hispida) and its antioxidant activity and fatty acid composition. *Molecules, 18*(1), 997–1014.

Bruna, J. M., Ordonez, J. A., Fernández, M., Herranz, B., & Hoz, L. (2001). Microbial and physico-chemical changes during the ripening of dry fermented sausages superficially inoculated with or having added an intracellular cell-free extract of Penicillium aurantiogriseum. *Meat Science, 59*, 87–96.

Chang, C., Yarankovich, N., & Nickerson, M. T. (2016). Microencapsulation of canola oil by lentil protein isolate-based wall materials. *Food Chemistry, 212*, 264–273.

Chaudhary, R., Saadin, K., Bliden, K. P., Harris, W. S., Dinh, B., Sharma, T., ... Gurbel, P. A. (2016). Risk factors associated with plasma omega-3 fatty acid levels in patients with suspected coronary artery disease. *Prostaglandins, Leukotrienes and Essential Fatty Acids, 113*, 40–45.

Chemat, F., Rombaut, N., Meullemiestre, A., Turk, M., Perino, S., Fabiano-Tixier, A. S., & Abert-Vian, M. (2017). Review of Green Food Processing techniques. Preservation, transformation, and extraction. *Innovative Food Science and Emerging Technologies, 41*, 357–377.

Chemat, F., Rombaut, N., Sicaire, A. G., Meullemiestre, A., Fabiano-Tixier, A. S., & Abert-Vian, M. (2017). Ultrasound assisted extraction of food and natural products. Mechanisms, techniques, combinations, protocols and applications. A review. *Ultrasonics - Sonochemistry, 34*, 540–560.

Comi, G., Tirloni, E., Andyanto, D., Manzano, M., & Iacumin, L. (2015). Use of bio-protective cultures to improve the shelf-life and the sensorial characteristics of commercial hamburgers. *LWT - Food Science and Technology, 62*(2), 1198–1202.

Delgado-Pando, G., Cofrades, S., Ruiz-Capillas, C., Triki, M., & Jiménez-Colmenero, F. (2012). Enriched n−3 PUFA/konjac gel low-fat pork liver pâté: Lipid oxidation, microbiological properties and biogenic amine formation during chilling storage. *Meat Science, 92*, 762–767.

Dolatowski, Z. J., & Stasiak, D. M. (2012). Ultrasonically assisted diffusion processes. In N. Lebovka, E. Vorobiev, & F. Chemat (Eds.). *Enhancing extraction processes in the food industry* (pp. 123–141). Boca Raton: CRC Press.

Domínguez, R., Pateiro, M., Munekata, P. E. S., Campagnol, P. C. B., & Lorenzo, J. M. (2016). Influence of partial pork backfat replacement by fish oil on nutritional and technological properties of liver pâté. *European Journal of Lipid Science and Technology, 118*, 1–7.

Dos Santos, B. A., Campagnol, P. C. B., da Cruz, A. G., Morgano, M. A., Wagner, R., & Pollonio, M. A. R. (2015). Is there a potential consumer market for low-sodium fermented sausages? *Journal of Food Science, 80*, 1093–1099.

Etchepare, M., Raddatz, G. C., Flores, E. M. M., Zepka, L. Q., Jacob-Lopes, E., Barin, J. S., & Menezes, C. R. (2016). Effect of resistant starch and chitosan on survival of Lactobacillus acidophilus microencapsulated with sodium alginate. *LWT - Food Science and Technology, 65*, 511–517.

Fernandes, R. P. P., Freire, M. T. A., De Paula, E. S. M., Kanashiro, A. L. S., Catunda, F. A. P., & Rosa, A. F. (2014). Stability of lamb loin stored under refrigeration and packed in different modified atmosphere packaging systems. *Meat Science, 96*, 554–561.

Heck, R. T., Vendruscolo, R. G., Etchepare, M. A., Cichoski, A. J., Menezes, C. R., Barin, J. S., ... Campagnol, P. C. B. (2017). Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science, 130*, 16–25.

Horita, C. N., Morgano, A. M., Celeghini, R. M. S., & Pollonio, M. A. R. (2011). Physico-chemical and sensory properties of reduced-fat mortadella prepared with blends of calcium, magnesium and potassium chloride as partial substitutes for sodium chloride. *Meat Science, 89*(4), 426–433.

Josquin, N. M., Linssen, J. P. H., & Houben, J. H. (2012). Quality characteristics of Dutchstyle fermented sausages manufactured with partial replacement of pork backfat with pure, pre-emulsified or encapsulated fish oil. *Meat Science, 90*(1), 81–86.

Kryževičūtė, N., Jaime, I., Diez, A. M., Rovira, J., & Venskutonis, P. R. (2017). Effect of raspberry pomace extracts isolated by high pressure extraction on the quality and shelf-life of beef burgers. *International Journal of Food Science and Technology, 52*(8), 1852–1861.

Latoch, A., & Stasiak, D. M. (2015). Effect of Mentha piperita on oxidative stability and sensory characteristics of cooked pork sausage. *Journal of Food Processing and Preservation, 39*, 1566–1573.

Lorenzo, J. M., Munekata, P. E. S., Pateiro, M., Campagnol, P. C. B., & Domínguez, R. (2016). Healthy Spanish salchichón enriched with encapsulated n−3 long chain fatty acids in konjac glucomannan matrix. *Food Research International, 89*, 289–295.

Lu, F., Zhang, J. Y., Liu, S. L., Wang, Y., & Ding, Y. T. (2011). Chemical, microbiological and sensory changes of dried Acetes chinensis during accelerated storage. *Food Chemistry, 127*(1), 159–168.

Malheiro, R., Oliveira, I., Vilas-Boas, M., Falcão, S., Bento, A., & Pereira, J. A. (2009). Effect of microwave heating with different exposure times on physical and chemical parameters of olive oil. *Food and Chemical Toxicology, 47*, 92–97.

Martínez, B., Miranda, J., Vázquez, B., Fente, C., Franco, C., Rodríguez, J., & Cepeda, A. (2012). Development of a hamburger patty with healthier lipid formulation and study of its nutritional, sensory, and stability properties. *Food and Bioprocess Technology, 5*(1), 200–208.

Meilgaard, M. C., Carr, T., & Civille, G. V. (2006). *Sensory evaluation techniques* (4th ed.). Boca Raton: CRC Press.

Monteiro, G. M., Souza, X. R., Costa, D. P. B., Faria, P. B., & Vicente, J. (2017). Partial substitution of pork fat with canola oil in Toscana sausage. *Innovative Food Science and Emerging Technologies, 44*, 2–8.

Ou, B., Hampsch-Woodill, M., & Prior, R. L. (2001). Development and validation of an improved oxygen radical absorbance capacity assay using fluorescein as the fluorescent probe. *Journal Agriculture Food Chemistry, 49*, 4619–4926.

Pignitter, M., Stolze, K., Gartner, S., Dumhart, B., Stoll, C., Steiger, G., ... Somoza, V. (2014). Cold fluorescent light as major inducer of lipid oxidation in soybean oil stored at household conditions for eight weeks. *Journal of Agricultural and Food Chemistry, 62*(10), 297–305.

Shah, M. A., Bosco, S. J. D., & Mir, S. A. (2014). Plant extracts as natural antioxidants in meat and meat products. *Meat Science, 98*(1), 21–33.

Stone, H., Bleibaum, R. N., & Thomas, H. A. (2012). *Sensory evaluation practices* (4th ed.). New York: Academic Press.

Tabarestani, H. S., & Tehrani, M. M. (2014). Optimization of physicochemical properties of low-fat hamburger formulation using blend of soy flour, split-pea flour and wheat starch as part of fat replacer system. *Journal of Food Processing and Preservation, 38*, 278–288.

Tiwari, B. K. (2015). Ultrasound: A clean, green extraction technology. *Trends in Analytical Chemistry, 71*, 100–109.

Valencia, I., Ansorena, D., & Astiasarán, I. (2006). Nutritional and sensory properties of dry fermented sausages enriched with n−3 PUFAs. *Meat Science, 72*(4), 727–733.

Xiong, G., Wang, P., Zheng, H., Xu, X., Zhu, Y., Zhou, G., & Xiong, G. (2016). Effects of plant oil combinations substituting pork back-fat combined with pre-emulsification on physicochemical, textural, microstructural and sensory properties of spreadable chicken liver PATE. *Journal of Food Quality, 39*(4), 331–341.

Zbancioc, G., Mangalagiu, I. I., & Moldoveanu, C. (2015). Ultrasound assisted synthesis of imidazolium salts: An efficient way to ionic liquids. *Ultrasonics - Sonochemistry, 23*, 376–384.

Zhou, M., Liu, J., Zhou, Y., Huang, X., Liu, F., Pan, S., & Hu, H. (2016). Effect of high intensity ultrasound on physicochemical and functional properties of soybean glycinin at different ionic strengths. *Innovative Food Science and Emerging Technologies, 34*, 205–213.

7. CAPÍTULO 4 - VOLATILE COMPOUNDS AND SENSORY PROFILE OF BURGERS WITH 50 % FAT REPLACEMENT BY MICROPARTICLES OF CHIA OIL ENRICHED WITH ROSEMARY

Rosane Teresinha Heck, Mariane Bittencourt Fagundes, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Juliano Smanioto Barin, José Manuel Lorenzo, Roger Wagner, Paulo Cezar Bastianello Campagnol

Meat Science
Vol. 148, p. 164-170, 2019
https://doi.org/10.1016/j.meatsci.2018.10.017

Meat Science 148 (2019) 164–170

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Volatile compounds and sensory profile of burgers with 50% fat replacement by microparticles of chia oil enriched with rosemary

Rosane Teresinha Heck[a], Mariane Bittencourt Fagundes[a], Alexandre José Cichoski[a], Cristiano Ragagnin de Menezes[a], Juliano Smanioto Barin[a], José Manuel Lorenzo[b], Roger Wagner[a], Paulo Cezar Bastianello Campagnol[a,*]

[a] Universidade Federal de Santa Maria, CEP 97105-900, Santa Maria, Rio Grande do Sul, Brazil
[b] Centro Tecnológico de la Carne de Galicia, Parque Tecnológico de Galicia, San Cibrán das Viñas, Rúa Galicia N 4, Ourense, Spain

ARTICLE INFO

Keywords:
Ultrasound-assisted extraction
Conventional maceration extraction
Green-extraction
Natural antioxidant
Microencapsulation
n-3 PUFAs
Healthier meat products

ABSTRACT

Direct incorporation of rosemary leaves into chia oil (CO) was performed by ultrasound-assisted extraction (UAE) and conventional maceration extraction (CME). CO was microencapsulated and used in burgers, as follows: control (20% pork back fat (PBF)); HCO (10% PBF + 7.5% water + 2.5% unencapsulated CO); HM1 (10% PBF + 10% CO microparticles); HM2 (10% PBF + 10% CO microparticles enriched by UAE) and HM3 (10% PBF + 10% CO microparticles enriched by CME). The volatile compounds and the sensory properties (Check-All-That-Apply and overall acceptability) of burgers were evaluated at days 1 and 120 of frozen storage. The control, HCO, and HM1 groups were characterized for volatile compounds produced by lipid and protein oxidation, and sensory descriptors related to lipid oxidation. HM2 and HM3 groups presented an increase in terpenic volatiles and were characterized by the descriptors herbal and pleasant aroma and ideal texture. In addition, liking scores were positively correlated to the descriptors that characterized the HM2 and HM3 groups.

1. Introduction

Burgers are meat products widely consumed in developed countries. However, these products generally contain a high animal fat content (20 to 30%) (Colmenero, 2000), which can positively affect the sensory characteristics, such as appearance, texture, aroma and flavor of the product, and contribute to the improvement of important technological attributes, including yield and reduction of cooking shrinkage (Tokusoglu & Ünal, 2003). In addition, the use of fat is also important in reducing production costs, enabling the production of economically viable meat products (Jayathilakan, Sultana, Radhakrishna, & Bawa, 2012).

However, it is widely known that intake of animal fat, which is high in saturated fatty acids, increases the risk factors related to the onset of cardiovascular diseases (Blekkenhorst et al., 2015). Thus, there is a growing concern of the meat industry to nutritionally improve the lipid profile of meat products, and in this context, the replacement of animal fat by oils rich in *n*-3 PUFAs may be a very effective alternative (Dominguez, Pateiro, Agregán, & Lorenzo, 2017; Lorenzo, Munekata, Pateiro, Campagnol, & Dominguez, 2016). Some studies have already evaluated the substitution of animal fat by oils rich in *n*-3 PUFAs in burgers such as linseed (Singh, Chatli, Biswas, & Sahoo, 2014) and canola (Afshari, Hosseini, Khaneghah, & Khaksar, 2017; Selani et al., 2016). Despite the potential health benefits, negative effects were observed with respect to the reduction of oxidative stability, and the technological and sensory quality of the products.

In an attempt to solve such problems, we studied the substitution of 50% pork back fat by microencapsulated chia oil in burgers (Heck et al., 2017). The lipid reformulation improved important technological attributes, such as cooking loss and fat retention, as well as improving the PUFA /SFA and *n*-6/*n*-3 ratio and the atherogenicity and thrombogenicity indices. However, the oxidative stability and the sensory quality was not improved, probably due to the formation of volatile compounds from the degradation of *n*-3 PUFAs, as these fatty acids are highly sensitive to lipid oxidation reactions (Elmore, Campo, Enser, & Mottram, 2002; Kosowska, Majcher, & Fortuna, 2017).

The use of natural antioxidants is an alternative to increase the oxidative stability of oils rich in *n*-3 PUFAs and thus, enable their application in foods with a nutritionally unfavorable fatty acid profile (Fernandes, Trindade, Lorenzo, & de Melo, 2018; Munekata et al., 2017). The most common extraction methods of natural antioxidants currently employ large amounts of toxic solvents, as ethanol and

* Corresponding author.
E-mail address: paulo.campagnol@ufsm.br (P.C.B. Campagnol).

https://doi.org/10.1016/j.meatsci.2018.10.017
Received 14 June 2018; Received in revised form 22 October 2018; Accepted 23 October 2018
Available online 25 October 2018
0309-1740/

methanol (Yara-Varón, Li, Balcells, Canela-Garayoa, & Chemat, 2017). Ultrasonic assisted extraction (UAE) has been used as an alternative to directly incorporate the antioxidant compounds from plant materials into vegetable oils without using toxic solvents (Chen et al., 2018; Goula, Ververi, Adamopoulou, & Kaderides, 2017). This extraction method causes a rupture of the cell wall due to the acoustic cavitation effect, achieving a high yield of bioactive compounds in a short time (Roselló-Soto et al., 2015). Conventional maceration extraction (CME) is another effective method for incorporating bioactive compounds directly into vegetable oils. However, it is more time consuming when compared to the ultrasonic-assisted extraction (Veillet, Tomao, & Chemat, 2010; Assami, Chemat, Meklati, and Chemat (2016).

In our previous study, we optimized the direct incorporation of rosemary leaves into chia oil by ultrasound-assisted extraction (UAE) and compared with a conventional maceration extraction (CME). In addition, we produced microparticles of chia oil enriched with rosemary by these green-extraction techniques and use as pork back fat substitute in burgers. This approach was useful to improve the oxidative stability during the frozen storage of burgers, however, important quality attributes were not addressed (Heck et al., 2018). Regardless of nutritional and oxidative quality, the sensory quality will always be the main determinant for the consumer acceptance for food products. Considering that there are well-known challenges regarding the addition of rosemary on the herbal flavor of the finished product, it is necessary to determine the impact of this lipid reformulation on the volatile compounds and the sensory profile. For this reason, the goal of this study was to evaluate the volatile compounds and the sensory profile of burgers with 50% pork back fat replacement by microparticles of chia oil enriched with rosemary by UAE and CME.

2. Material and methods

2.1. Direct incorporation of rosemary leaves into chia oil by ultrasound-assisted extraction and conventional maceration extraction

Fresh rosemary leaves (*Rosmarinus officinalis* L.) were purchased in the local market and dried at 50 °C for 24 h in an air circulation oven (MDH, DeLeo, Brazil). The dried leaves were milled in an analytical mill (MA630/1, Marconi, Brazil) until a fine powder was obtained. The finely ground rosemary leaves were incorporated directly into chia oil (Giroil S.A., Santo Ângelo, Brazil) by ultrasound-assisted extraction (UAE) and conventional maceration extraction (CME). The fatty acid composition of chia oil was as follows: 13.8% SFA; 7.1% MUFA (6.89% oleic acid); 79% PUFA; 20.25% *n*-6 PUFA (linoleic acid: 0.09%); and 58.7% *n*-3 PUFA (linolenic acid: 58.65%).

The UAE was carried out in an ultrasonic bath (TI-H-10, Elma, Germany) operating at a nominal power of 1000 W, at frequency of 25 kHz, sweep operating mode, and 100% amplitude. The water temperature of the ultrasonic bath was maintained at 30 °C during application of the ultrasound. The ultrasonic bath had an acoustic power of 230 W (Koda, Kimura, Kondo, & Mitome, 2003) and a volumetric power of 28 W/L (Feng, Barbosa-Cánovas, & Weiss, 2011). Chia oil (100 g) and the ground rosemary leaves (12.5 g) were sonicated for 10 min. The parameters of UAE were optimized in a previous study (Heck et al., 2018).

The CME was performed with the same amount of rosemary leaves (12.5 g) and chia oil (100 g) used in the UAE. The rosemary leaves and the chia oil were placed in an amber flask and homogenized. The mixture remained at rest for four hours without the incidence of light. For both extraction methods (UAE and CME), the mixture was filtered with a filter paper (Whatman # 40) and the rosemary-enriched chia oil was stored in amber flasks under refrigeration (4 ± 1 °C) for 24 h.

2.2. Production of the microparticles of chia oil enriched or not with rosemary

The microparticles were produced by the external ionic gelation process, as described by Etchepare et al. (2016). For this process, chia oil was mixed with a 2% sodium alginate solution. The mixture was atomized in 0.1 M $CaCl_2$ solution using a dual fluid atomizer nozzle (0.1 mm) at a distance of 12 cm from the solution, under air pressure of 0.125 kg/cm. After atomization, the microparticles were kept under constant stirring for 30 min, and then strained through a wire mesh sieve (150 μm) and washed with sterile distilled water. Three types of microparticles were produced, as follows: M1 (chia oil microparticles without the addition of rosemary); M2 (microparticles produced with rosemary-enriched chia oil by UAE); and M3 (microparticles produced with rosemary-enriched chia oil by CME). The microparticles had a lipid content of 25 ± 1.2% and an encapsulation efficiency of approximately 87%.

2.3. Burger manufacture

Three independent replicates of each batch were prepared. The control sample was made with 78.4% beef (*rectus femoris*), 20% pork back fat, 1.5% salt, and 0.1% garlic powder. The modified treatments were made with the same amount of beef, salt and garlic added in the the control. For the treatment HCO, 50% pork back fat of the control sample was replaced by the unencapsulated chia oil (2.5 g/100 g) and water (7.5 g/100 g). For the treatments HM1, HM2, and HM3, 50% pork back fat was replaced by the microparticles M1, M2, and M3, respectively. The pork back fat and beef were ground (Model PJ22, Jamar Ltda., São Paulo, Brazil) separately using a 3 mm disc. Then, beef was mixed (model MJ 35, Jamar Ltda., São Paulo, Brazil) with salt for extraction of the myofibrillar proteins, and the remaining ingredients were added and mixed. Burgers (100 g), 11 cm in diameter and 2.5 cm thick, were produced using a conventional burger-maker (HP 112, Picelli, São Paulo, Brazil). The burgers were individually placed in bags (20 × 14 cm) of high-density polyethylene films (50 μm thickness, with oxygen transmission rate 1434 cm^3/ m^2.day and water vapor transmission rate 0.6 g/ m^2.day). Bags were sealed using a Selovac 200B (Selovac, São Paulo, Brazil) packaging machine and the burgers were stored at −18 °C for 120 days. The control and modified raw burgers had a fat content of approximately 18.2% and 8.5%, respectively (Heck et al., 2018).

Prior to analyses (day 1 and 120), the burgers were thawed at 4 °C for 12 h in a domestic refrigerator. The samples were cooked on an electric grill (Multi Grill, Britânia, Brazil) preheated to 150 °C, until reaching an internal temperature of 72 °C in the geometric center of each burger, which was measured by a spit thermometer (HM-600, Highmed, Brazil).

2.4. Determination of volatile compounds

The volatile compounds were determined in triplicate in the raw and cooked burgers at the beginning (day 1) and at the end of storage (day 120). The sample (5 g) was placed in a 20 mL vial sealed with a PTFE facing silicone septum. Solid-phase microextraction (HS-SPME) was used for the extraction of volatile compounds. Isolation of the volatile compounds was performed with a fiber CarPDMS fiber (Carboxen-polydimethylsiloxane) (75 μm, 10 mm, Supelco, Bellefonte, PA, USA), and the separation and identification was performed in a gas chromatograph coupled to a mass spectrometer (GC/MS; SHIMADZU, QP-2010 Plus, Tokyo, Japan). For that, 1 μL sample was injected in the splitless mode using a ZP-WAX Plus (Chrompack, USA) column of 60 m × 0.25 mm internal diameter and 0.25 μm stationary phase thickness. Helium was used as carrier gas, under a constant flow rate of 1.0 mL/min. The initial column temperature was 35 °C, which was maintained for 3 min and then increased to 80 °C at a rate of 2 °C/min.

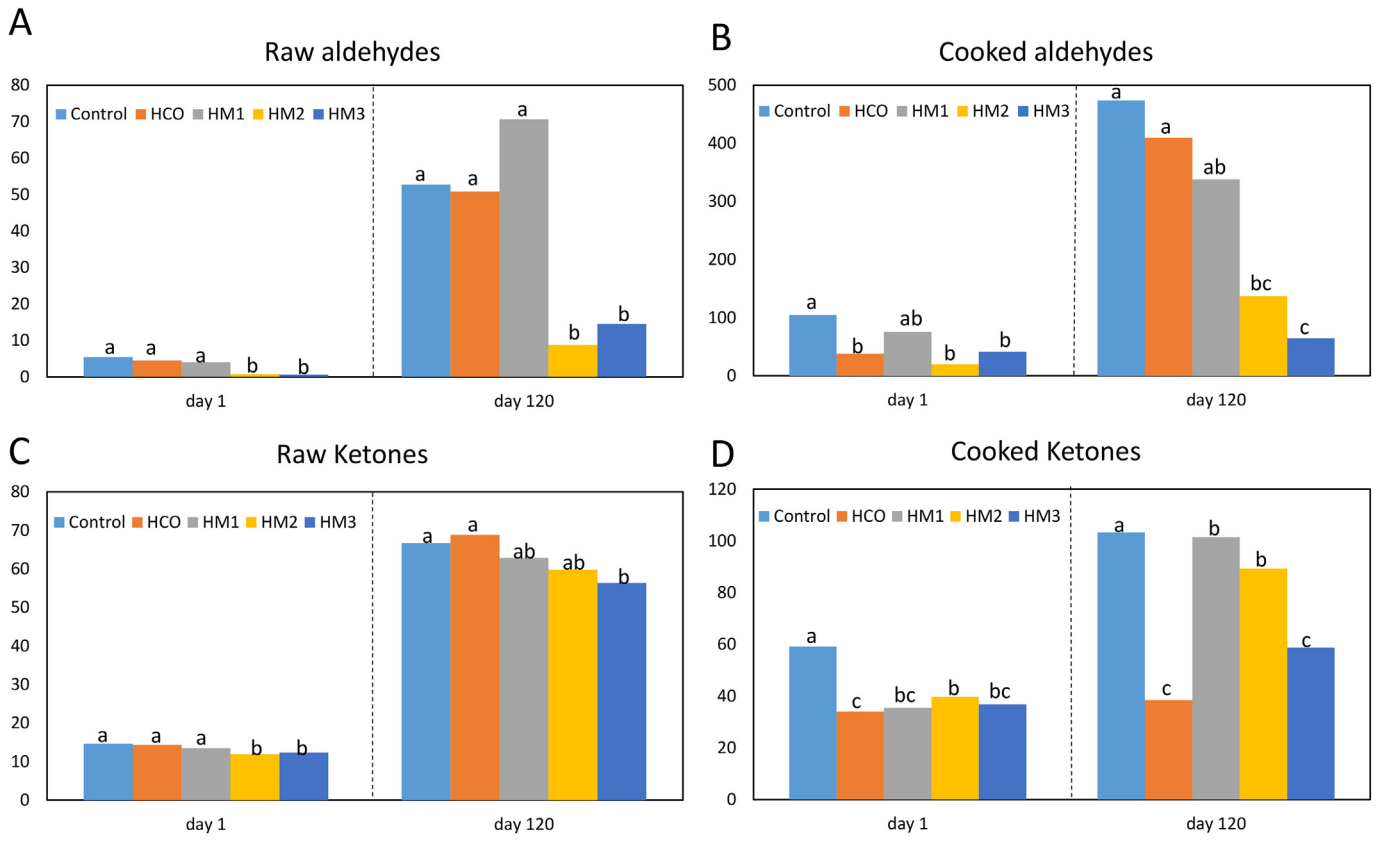


Fig. 1. Total aldehydes and ketones (average area $\times 10^6$) of raw and cooked burgers during frozen storage (days 1 and 120). Lots: Control: 20% pork back fat; HCO: 10% pork back fat, 7.5% water and 2.5% chia oil (CO); HM1: 10% pork back fat and 10% microparticles of CO; HM2: 10% pork back fat and 10% microparticles of CO enriched by ultrasound-assisted extraction: HM3: 10% pork back fat and 10% microparticles of CO enriched by conventional maceration extraction.

Then, the temperature increased from 5 °C/min to 230 °C, maintaining for 5 min. The identification of the volatile compounds was performed in a mass spectrometer operating in electron ionization mode (+70 eV) using a scan of 35–350 *m/z* and detector voltage of 0,96 kV. The identification was done according to Wagner and Franco (2012), by comparing the mass spectra with those obtained in the National Institute of Standards & Technology (NIST 05). The alkanes (C_6-C_{24}) were analyzed under the same chromatographic conditions, and the homologous series were used to calculate the linear retention index values, which was compared with that obtained in the literature (Acree & Heinrich, 2016; El-Sayed, 2016).

2.5. Sensory analysis

The CATA sensory test was applied to characterize the sensory profile of the reformulated burgers. This study was approved by the Research Ethics Committee of the Federal University of Santa Maria (RS, Brazil) (CAAE: 57433316.8.0000.5346). The sensory evaluation was performed in individual booths under fluorescent lights, with an intensity of approximately 350 lx. The samples were coded with randon three digits numbers, and evaluated in a monadic order, following a balanced design (Ares, Barreiro, Deliza, Giménez, & Gámbaro, 2010). The burgers were cooked as described in section 2.3. After, the cooked burgers were cut in pieces of approximately 4 × 4 × 2.5 cm and were individually wrapped in foil and served to consumers at 60 °C. The CATA sensory test was performed at the beginning (day 1) and at the end (day 120) of the storage. One hundred regular burger consumers, aged 18–55, participated in the study (64% female and 36% male at day 1, and 56% female and 44% male at day 120). The samples were characterized by the descriptors ideal color and oily appearance; aroma was characterized for greasy, rancid, acid, herbal, and pleasant. The flavor attribute was characterized by the descriptors rancid, pleasant, and unpleasant, and finally, texture was characterized using the descriptors ideal, rubbery, and juicy. These sensory descriptors were based on studies about low-fat meat products and its meaning was explained to each consumer before the test (Alves et al., 2016; Heck et al., 2017). At the end of the CATA test, the consumers were asked to give a score for overall acceptability of the products from 1 to 9.

2.6. Statistical analysis

The whole experiment was repeated three times. Data from the volatile compounds were evaluated by analysis of variance (ANOVA) using a general linear model considering the treatments as a fixed effect and the replicates as a random effect ($n = 3$). The Tukey's test was used at the 5% level of significance to compare the means between the treatments. Data were analyzed using the SPSS statistical program (SPSS Inc., Chicago, IL, USA). Principal Component Analysis (PCA) was used to evaluate the contribution of the volatile compounds during storage, using the statistical program Pirouette 3:11 (Infometrix, Washington, USA).

Correspondence Analysis based on chi-square distances was used to analyze the CATA question (Vidal, Tárrega, Antúnez, Ares, & Jaeger, 2015), calculated on the matrix with the frequency of each word for each sample. Principal Coordinates Analysis was calculated on the correlation matrix including CATA descriptors (tetrachoric correlation) and liking scores (biserial correlation). Correspondence Analysis and Principal Coordinates Analysis were calculated using the statistical program XLSTAT 2018.1 (Addinsoft, Paris, France).

3. Results and discussion

3.1. Volatile compounds of raw and cooked burgers at the beginning (day 1) and at the end of storage (day 120)

The volatile compounds detected in the raw and cooked burgers are shown in Table S1 and S2, respectively (supplementary material). The volatile compounds from lipid oxidation, such as aldehydes and ketones, are mainly responsible for the reduction of the sensory quality of cooked meat products (Domínguez, Gómez, Fonseca, & Lorenzo, 2014; Lorenzo et al., 2018). It is commonly reported in the literature that the addition of oils rich in *n*-3 PUFAs increases the volatile compounds from lipid oxidation in meat products (Ansorena & Astiasarán, 2004; Vieira et al., 2012). In this study, the control burgers were made with 20 g pork back fat/100 g. The replacement of 50% pork back fat by unencapsulated chia oil (2.5 g/100 g) and water (7.5 g/100 g) (HCO) did not cause significant ($P > .05$) changes in the total amount of aldehydes (Fig. 1a) and ketones (Fig. 1c) in the raw hamburgers at the beginning (day 1) and at the end of the storage period (day 120). Surprisingly, the HCO treatment had a lower amount of aldehydes ($P < .01$; Fig. 1b) and ketones ($P < .001$; Fig. 1d) when compared to the control after cooking. This result may be due to the oil release during cooking, as reported by Heck et al. (2017). The raw burgers of the treatment HM1 and of the control presented a similar ($P > .05$) content of aldehydes and ketones at the beginning (day 1) and at the end of the storage period (day 120) (Fig. 1a and c). However, after cooking, the treatment HM1 had a lower ($P < .001$) amount of ketones when compared to the control at day 1 of storage (Fig. 1d). This fact corroborates the results of Comunian and Favaro-Trindade (2016), who reported that the microencapsulation by the external ionic gelation technique is used to protect the oil from the oxidative reactions during heating. The direct incorporation of rosemary into chia oil by UAE and CME reduced ($P < .01$) the content of aldehydes in the HM2 and HM3 treatments in relation to the control, for both raw and cooked burgers (Fig. 1a and b), which reduced from 72% to 88% in raw burgers, and 71% to 86% in cooked burgers. HM2 and HM3 treatments presented a similar ($P > .05$) content of aldehydes in the raw and cooked burgers (days 1 and 120). However, HM3 presented a lower ($P < .001$) content of ketones than HM2 at day 120 of storage. This result suggests that UAE was more effective than CME at incorporating antioxidant compounds from rosemary leaves into chia oil.

3.2. Principal component analysis of volatile compounds in raw and cooked burgers at the beginning (day 1) and at the end of storage (day 120)

Principal Component Analysis (PCA) was applied to visually explore the impact of the substitution of pork back fat by the rosemary-enriched chia oil microparticles in the volatile profile of burgers. As shown in Table S1 and S2, the compounds that differed significantly among treatments by the Tukey's test were included in the PCA ($P < .05$).

The PCA of the volatile compounds of the raw burgers at the beginning (day 1) and at the end of the storage (day 120) is shown in Fig. 2. The first two principal components accounted for 64.4% of the total variance (PCI: 47.9% and *Pci*I: 16.5%). The PCI allowed discriminating the samples according to the processing time. Samples at the beginning of storage (day 1) were located in the left quadrant, while samples stored for 120 days were located in the right quadrant of PCI. The HM2 and HM3 treatments were discriminated through the *Pci*I, as they were grouped in the upper quadrant, whereas the control and the HCO and HM1 treatments were located in the lower quadrant of *Pci*I. At day 1 of storage, the raw burgers of the HM2 and HM3 treatments were characterized mainly by the compounds camphor, 4-terpinenol, verbenone, alpha-terpineol, pinocarvone, gamma-pinene, isobutyric acid, alpha terpinolene, and 2,3-methyl butanone. These compounds are characteristic of rosemary (Castro-Vázquez, Pérez-Coello, & Cabezudo, 2003) and were also found by Estévez, Ventanas, Ramírez, and Cava (2004) in a study using rosemary as an antioxidant in liver pate. In contrast, the raw burgers from the control and the HCO and HM1 treatments were characterized by the compounds: sulfide allyl methyl, phenol, propanoic acid, nonanoic acid, octanoic acid, butanoic acid ethyl ester, 2,3-butanedione, 3-methyl- 3-butenol, 1-butanol, benzaldehyde, and nonanal. The presence of lipid oxidation compounds, such as propanoic acid, nonanoic acid, and nonanal (Omonov, Kharraz, & Curtis, 2011) indicates that the treatments without the addition of rosemary presented higher lipid oxidation when compared to HM2 and HM3 groups shortly after manufacturing. The control, HCO, and HM1 treatments also showed a group of compounds from protein oxidation, such as 3-methyl-1-butanol, phenol, and benzaldehyde (Santos, Campagnol, Fagundes, Wagner, & Pollonio, 2015).

After 120 days of storage, raw burgers of the HM2 and HM3 treatments continued to be characterized by volatile terpenes, including eucalyptol, camphene, myrcene, limonene, sabinene, beta-caryophyllene, and alpha-pipene. In contrast, raw burgers of the other treatments (control, HCO, and HM1) were characterized by a large number of volatile compounds typical from lipid oxidation, such as hexanal, heptanal, 2-pentenal, and butanal (Im, Hayakawa, & Kurata, 2004). It is worth noting that hexanal is one of the main markers of lipid oxidation in meat products, giving the product a rancid taste when found in high concentrations (Brunton, Cronin, Monahan, & Durcan, 2000). These treatments were also characterized by compounds from protein oxidation, including dimethyldisulfide, 2 methyl-1butanol and 3-methyl butanal (Kosowska et al., 2017).

The PCA of the volatile compounds of the cooked burgers at the beginning (day 1) and at the end (day 120) of the storage is shown in Fig. 3. The first two principal components accounted for 64.82% of the total variance (PCI: 48.35% and *Pci*I: 16.47%), and the treatments were separated into three groups (Fig. 3A). At the beginning of storage (day 1), the cooked burgers from the control, HCO, HM1, HM2, and HM3 treatments were grouped in the lower left quadrant of PCI. After 120 days of storage, there was a separation between treatments with and without the addition of rosemary. The control and the HCO and HM1 treatments were grouped in the right quadrant of PC1, while HM2 and HM3 treatments were grouped in the upper left quadrant of PC1. As observed for raw burgers (Fig. 2), after 120 days of storage, the cooked burgers from the treatments without the addition of rosemary were characterized mainly by lipid oxidation compounds, such as aldehydes and ketones. On the other hand, the cooked burgers of the HM2 and HM3 treatments were again characterized mainly by terpene compounds after this period.

As can be seen in Fig. 3B, PCIII explained the discrimination between the treatments with and without addition of rosemary at the beginning of storage (day 1). The principal components accounted for 57.18% of the total variance (PCI: 48.35% and PCIII: 8.83%). At day 1 of storage, the HM2 and HM3 treatments were characterized by terpene compounds such as verbenone and caryophyllene. In contrast, the treatments without addition of rosemary (control, HCO, and HM1) were characterized by some terpene compounds, but were also characterized by compounds derived from lipid and protein oxidation (octanal and phenol, respectively).

3.3. Check-all-that-apply (CATA)

Correspondence analysis (CA) was used to evaluate the descriptors generated by the CATA questionnaire. The results of the beginning (day 1) and at the end (day 120) of the storage are presented in Fig. 4A. The CA explained 84.54% of the total variance, with 70.22% and 14.32% for the first and second dimensions, respectively. According to the first dimension, the treatments were separated into two distinct groups. The first group was formed by the HM2 and HM3 treatments, which were located in the right quadrant. The second group (Control, HCO and HM1) was located in the left quadrant. At days 1 and 120, the HM2 and HM3 treatments were characterized by the descriptor herbal aroma,

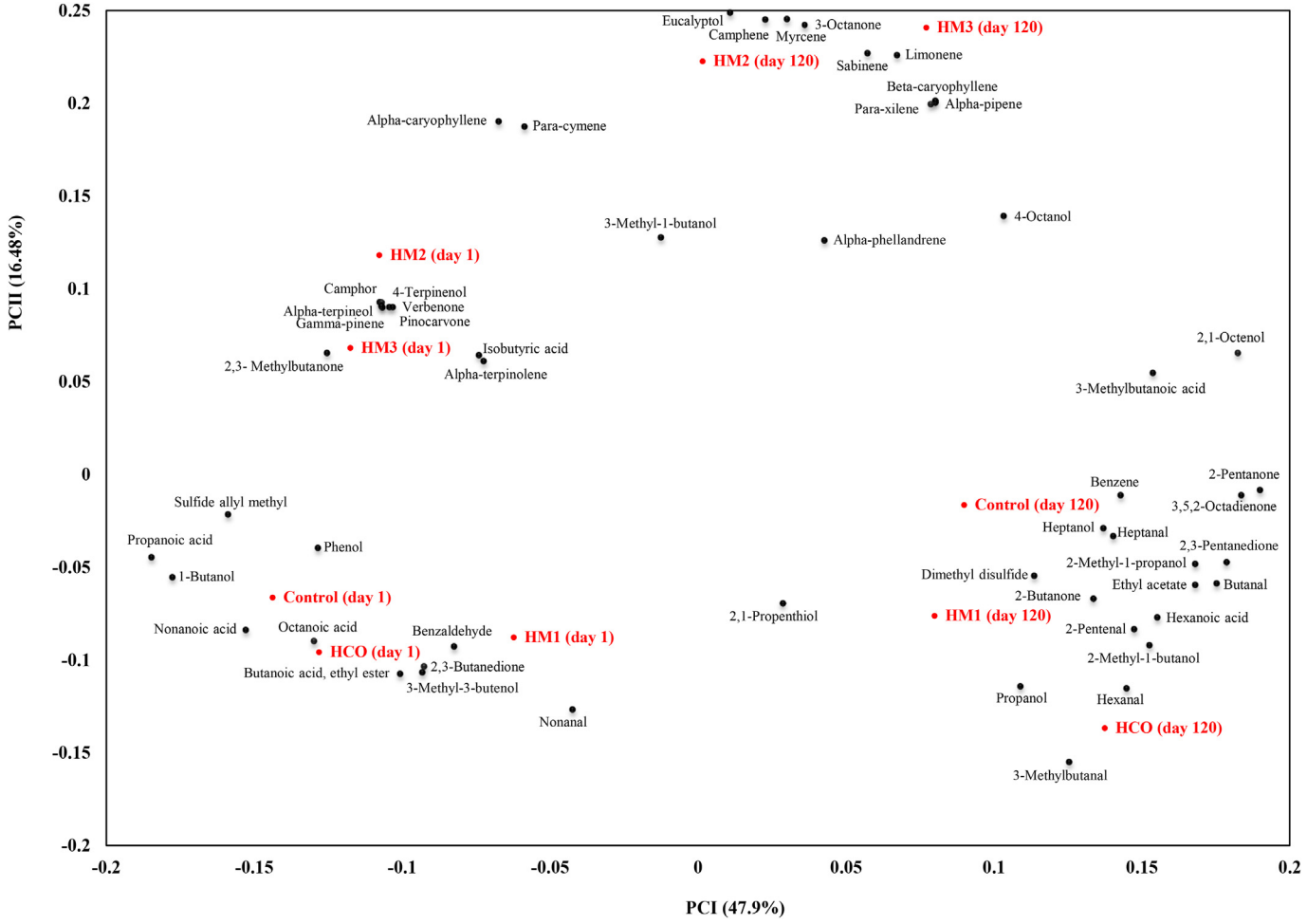


Fig. 2. Loadings of the first two main components (PC1–PC2) of the volatile compounds for the raw burgers during frozen storage (days 1 and 120). Lots: Control: 20% pork back fat; HCO: 10% pork back fat, 7.5% water and 2.5% chia oil (CO); HM1: 10% pork back fat and 10% microparticles of CO; HM2: 10% pork back fat and 10% microparticles of CO enriched by ultrasound-assisted extraction: HM3: 10% pork back fat and 10% microparticles of CO enriched by conventional maceration extraction.

which is well correlated with the high content of terpene compounds identified in these treatments (Figs. 2 and 3). In addition, they were also characterized by the descriptors pleasant aroma and ideal texture. It should be noted that the treatments containing rosemary-enriched chia oil microparticles (HM2 and HM3 groups) did not present any descriptors related to lipid oxidation. This result agrees with the low amount of aldehydes and ketones found in these treatments at beginning (day 1) and at the end (day 120) of the storage (Fig. 1). The second group (Control, HCO and HM1) was characterized by the descriptors rancid flavor, rancid aroma, unpleasant flavor, greasy aroma, and acid aroma. These descriptors are well correlated with the large content of volatile compounds from lipid oxidation identified in these treatments (Table S2, Fig. 3). In addition, these treatments were characterized by the descriptor oily, which is related to the oil release from the product during cooking.

Principal Coordinates Analysis was applied to the correlation matrix including descriptors and liking scores and the results are showed in a two dimensional map (Fig. 4B). The first two dimensions explains 46.4% of the variation and the scree plot indicated that the two first dimensions are sufficient to interpret relationships between descriptors. Liking scores seem to be positively correlated to the descriptors that characterized the treatments containing rosemary-enriched chia oil microparticles (HM2 and HM3 groups) in the CA (herbal aroma, pleasant aroma, and ideal texture) (Fig. 4A). In addition, liking scores seem to be inversely proportional to the descriptors rancid aroma, rancid flavor and oily that characterized the treatments without rosemary (Fig. 4a).

4. Conclusion

The chia oil did not have a major impact on the volatiles profile of raw and cooked burgers for both the unencapsulated and microencapsulated forms. On the other hand, a significant modification in the volatile profile of burgers was observed with the addition of microparticles of chia oil enriched with rosemary by UAE or CME techniques. This modification resulted in a decrease in volatiles from lipid and protein oxidation and an increase in terpenes at the beginning (day 1) and at the end of storage (day 120), before and after cooking. UAE may be considered a method of direct incorporation of bioactive compounds in chia oil more advantageous than CME, since in addition to the shorter extraction time, the burgers produced with microparticles of chia oil enriched with rosemary by UAE presented a lower content of ketones.

Sensory descriptors related to lipid oxidation were reported for burgers with the addition of chia oil microparticles. The direct incorporation of bioactive compounds from rosemary into chia oil by UAE or CME techniques before the microencapsulation process was effective to solve these sensory impacts. The sensory descriptors of the burgers from these treatments were positively correlated with liking scores. Therefore, it can be concluded that the lipid reformulation proposed in

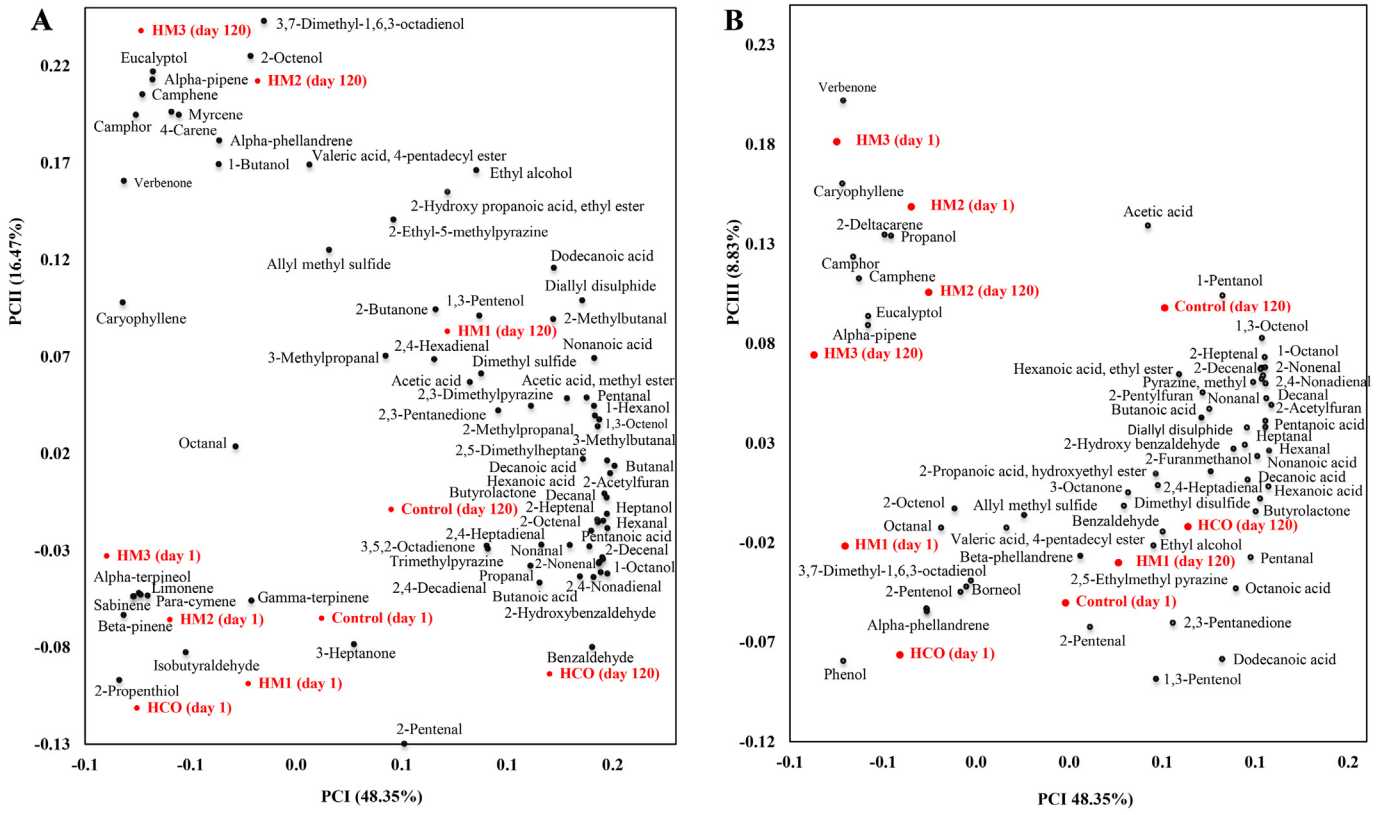


Fig. 3. A - Loadings of the first two main components (PC1–PC2) of the volatile compounds for the cooked burgers during frozen storage (days 1 and 120). **B-** Loadings of the first and third main components (PC1–PC3) of the volatile compounds for the cooked burgers during frozen storage (days 1 and 120). Lots: Control: 20% pork back fat; HCO: 10% pork back fat, 7.5% water and 2.5% chia oil (CO); HM1: 10% pork back fat and 10% microparticles of CO; HM2: 10% pork back fat and 10% microparticles of CO enriched by ultrasound-assisted extraction: HM3: 10% pork back fat and 10% microparticles of CO enriched by conventional maceration extraction.

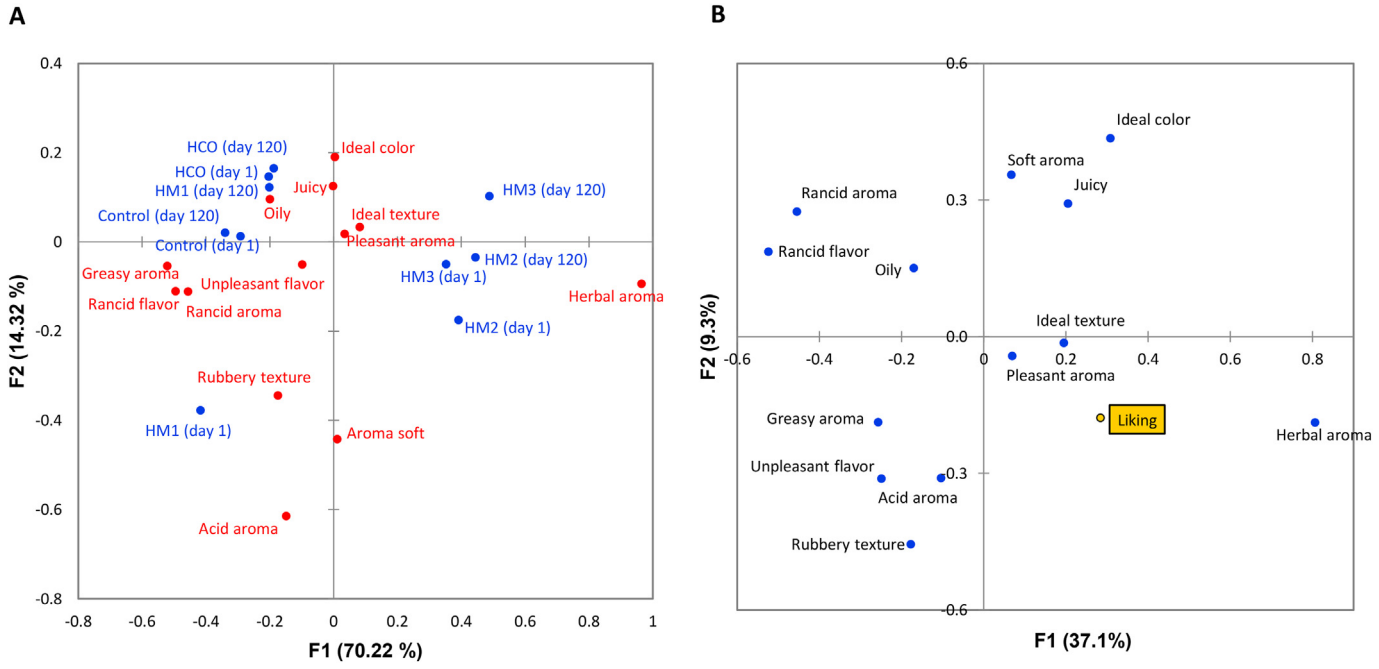


Fig. 4. A - Representation of the samples and the terms in the first and second dimensions of correspondence analysis performed on data questions check-all-that-apply (CATA) at the beginning (day 1) and at the end of storage (day 120). B - Principal Coordinates Analysis applied to the correlation matrix including CATA descriptors and liking scores. Lots: Control: 20% pork back fat; HCO: 10% pork back fat, 7.5% water and 2.5% chia oil (CO); HM1: 10% pork back fat and 10% microparticles of CO; HM2: 10% pork back fat and 10% microparticles of CO enriched by ultrasound-assisted extraction: HM3: 10% pork back fat and 10% microparticles of CO enriched by conventional maceration extraction.

this study is a promising alternative to incorporate healthy oils into cooked meat products.

Acknowledgements

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001. Jose M. Lorenzo is member of the MARCARNE network, funded by CYTED (ref. 116RT0503).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.meatsci.2018.10.017.

References

Acree, T., & Heinrich, A. (2016). Flavornet and human odor space. URL http://www.flavornet.org.

Afshari, R., Hosseini, H., Khaneghah, A. M., & Khaksar, R. (2017). Physico-chemical properties of functional low-fat beef burgers: Fatty acid profile modification. *LWT - Food Science and Technology, 78*, 325–331.

Alves, L. A. A. S., Lorenzo, J. M., Gonçalves, C. A. A., Santos, B. A., Heck, R. T., Cichoski, A. J., & Campagnol, P. C. B. (2016). Production of healthier bologna type sausages using pork skin and green banana flour as a fat replacers. *Meat Science, 121*, 73–78.

Ansorena, D., & Astiasarán, I. (2004). The use of linseed oil improves nutritional quality of the lipid fraction of dry-fermented sausages. *Food Chemistry, 87*(1), 69–74.

Ares, G., Barreiro, C., Deliza, R., Giménez, A., & Gámbaro, A. (2010). Application of a check-all-that-apply question to the development of chocolate milk desserts. *Journal of Sensory Studies, 96*, 47–55.

Assami, K., Chemat, S., Meklati, B., & Chemat, F. (2016). Ultrasound-Assisted Aromatisation with Condiments as an Enabling Technique for Olive Oil Flavouring and Shelf Life Enhancement. *Food Analytical Methods, 9*(4), 982–990.

Blekkenhorst, L. C., Prince, R. L., Hodgson, J. M., Lim, W. H., Zhu, K., Devine, A., ... Lewis, J. R. (2015). Dietary saturated fat intake and atherosclerotic vascular disease mortality in elderly women: A prospective cohort study. *The American Journal of Clinical Nutrition, 101*(6), 1263–1268.

Brunton, N. P., Cronin, D. A., Monahan, F. J., & Durcan, R. (2000). A comparison of solid-phase microextraction (SPME) fibres for measurement of hexanal and pentanal in cooked Turkey. *Food Chemistry, 68*(3), 339–345.

Castro-Vázquez, L., Pérez-Coello, M., & Cabezudo, M. (2003). Analysis of volatile compounds of rosemary honey. Comparison of different extraction techniques. *Chromatographia, 57*(3), 227–233.

Chen, C., Wang, L., Wang, R., Luo, X., Li, Y., Li, J., ... Chen, Z. (2018). Ultrasound-assisted extraction from defatted oat (Avena sativa L.) bran to simultaneously enhance phenolic compounds and β-glucan contents: Compositional and kinetic studies. *Journal of Food Engineering, 222*, 1–10.

Colmenero, J. F. (2000). Relevants factors in strategies for fat reduction meat products. *Food Science & Technology, 11*(2), 56–66.

Comunian, T. A., & Favaro-Trindade, C. S. (2016). Microencapsulation using biopolymers as an alternative to produce food enhanced with phytosterols and omega-3 fatty acids: A review. *Food Hydrocolloids, 61*, 442–457.

Domínguez, R., Gómez, M., Fonseca, S., & Lorenzo, J. M. (2014). Effect of different cooking methods on lipid oxidation and formation of volatile compounds in foal meat. *Meat Science, 97*(2), 223–230.

Dominguez, R., Pateiro, M., Agregán, R., & Lorenzo, J. M. (2017). Effect of the partial replacement of pork backfat by microencapsulated fish oil or mixed fish and olive oil on the quality of frankfurter type sausage. *Journal of Food Science and Technology, 54*, 26–37.

Elmore, J. S., Campo, M. M., Enser, M., & Mottram, D. S. (2002). Effect of lipid composition on meat-like model systems containing cysteine, ribose and polyunsaturated fatty acids. *Journal of Food Chemistry, 50*(5), 1126–1132.

El-Sayed, A. M. (2016). The Pherobase. Database of pheromones and semiochemicals. URL http://www.pherobase.com.

Estévez, M., Ventanas, S., Ramírez, R., & Cava, R. (2004). Analysis of volatiles in porcine liver pâtés with added sage and rosemary essential oils by using SPME-GC-MS. *Journal of Agricultural and Food Chemistry, 52*(16), 5168–5174.

Etchepare, M., Raddatz, G. C., Flores, E. M. M., Zepka, L. Q., Jacob-Lopes, E., Barin, J. S., ... Menezes, C. R. (2016). Effect of resistant starch and chitosan on survival of *Lactobacillus acidophilus* microencapsulated with sodium alginate. *LWT- Food Science and Technology, 65*, 511–517.

Feng, H., Barbosa-Cánovas, G. V., & Weiss, J. (2011). *Ultrasound technologies for food and bioprocessing.* New York: Springer Science + Business Media.

Fernandes, R. P. P., Trindade, M. A., Lorenzo, J. M., & de Melo, M. P. (2018). Assessment of the stability of sheep sausages with the addition of different concentrations of Origanum vulgare extract during storage. *Meat Science, 137*, 244–257.

Goula, A. M., Ververi, M., Adamopoulou, A., & Kaderides, K. (2017). Green ultrasound-assisted extraction of carotenoids from pomegranate wastes using vegetable oils. *Ultrasonics - Sonochemistry, 34*, 821–830.

Heck, R. T., Lucas, B. N., Dos Santos, D. J. P., Pinton, M. B., Fagundes, M. B., De Araújo Etchepare, M., ... Campagnol, P. C. B. (2018). Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques. *Meat Science, 146*, 147–153.

Heck, R. T., Vendruscolo, R. G., De Araújo Etchepare, M., Cichoski, A. J., De Menezes, C. R., Barin, J. S., ... Campagnol, P. C. B. (2017). Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science, 130*, 16–25.

Im, S., Hayakawa, F., & Kurata, T. (2004). Identification and sensory evaluation of volatile compounds in oxidized porcine liver. *Journal of Agricultural and Food Chemistry, 52*(2), 300–305.

Jayathilakan, K., Sultana, K., Radhakrishna, K., & Bawa, A. S. (2012). Use of by-products and waste from meat, poultry and fish processing industries: A review. *Journal of Food Science and Technology, 49*(3), 278–293.

Koda, S., Kimura, T., Kondo, T., & Mitome, H. (2003). A standard method to calibrate sonochemical efficiency of an individual reaction system. *Ultrasonics Sonochemistry, 10*, 149–156.

Kosowska, M., Majcher, A., & Fortuna, T. (2017). Volatile compounds in meat and meat products. *Food Science and Technology, 37*(1), 1–7.

Lorenzo, J. M., Munekata, P. E. S., Pateiro, M., Campagnol, P. C. B., & Dominguez, D. (2016). Healthy Spanish salchichón enriched with encapsulated n − 3 long chain fatty acids in konjac glucomannan matrix. *Food Research International, 86*, 289–295.

Lorenzo, J. M., Pateiro, M., Domínguez, R., Barba, F. J., Putnik, P., Kovačević, D. B., ... Franco, D. (2018). Berries extracts as natural antioxidants in meat products: A review. *Food Research International, 106*, 1095–1104.

Munekata, P. E. S., Dominguez, R., Franco, D., Bermúdez, R., Trindade, M. A., & Lorenzo, J. M. (2017). Effect of natural antioxidants in Spanish salchichón elaborated with encapsulated n-3 long chain fatty acids in konjac glucomannan matrix. *Meat Science, 124*, 54–60.

Omonov, T., Kharraz, E., & Curtis, J. (2011). Ozonolysis of Canola Oil: A Study of product yields and Ozonolysis Kinetics in Different Solvent Systems. *Journal of the American Oil Chemists' Society, 88*(5), 689–705.

Roselló-Soto, E., Galanakis, C. M., Brnčić, M., Orlien, V., Trujillo, F. J., Mawson, R., ... Barba, F. J. (2015). Clean recovery of antioxidant compounds from plant foods, by-products and algae assisted by ultrasounds processing. Modeling approaches to optimize processing conditions. *Trends in Food Science & Technology, 42*(2), 134–149.

Santos, B. A., Campagnol, P. C. B., Fagundes, M. B., Wagner, R., & Pollonio, M. A. R. (2015). Generation of volatile compounds in Brazilian low-sodium dry fermented sausages containing blends of NaCl, KCl, and $CaCl_2$ during processing and storage. *Food Research International, 74*, 306–314.

Selani, M. M., Shirado, G. A. N., Margiotta, G. B., Rasera, M. L., Marabesi, A. C., Piedade, S. M. S., ... Canniatti-Brazaca, S. G. (2016). Pineapple by-product and canola oil as partial fat replacers in low-fat beef burger: Effects on oxidative stability, cholesterol content and fatty acid profile. *Meat Science, 115*, 9–15.

Singh, R., Chatli, M., Biswas, A., & Sahoo, J. (2014). Quality of ω-3 fatty acid enriched low-fat chicken meat patties incorporated with selected levels of linseed flour/oil and canola flour/oil. *Journal of Food Science and Technology, 51*(2), 353–358.

Tokusoglu, Ö., & Ünal, M. K. (2003). Fat replacers in meat products. *Pakistan Journal of Nutrition, 2*(3), 196–203.

Veillet, S., Tomao, V., & Chemat, F. (2010). Ultrasound assisted maceration: An original procedure for direct aromatisation of olive oil with basil. *Food Chemistry, 123*(3), 905–911.

Vidal, L., Tárrega, A., Antúnez, L., Ares, G., & Jaeger, S. R. (2015). Comparison of correspondence analysis based on Hellinger and chi-square distances to obtain sensory spaces from check-all-that-apply (CATA) questions. *Food Quality and Preference, 43*, 106–112.

Vieira, C., Fernández-Diez, A., Mateo, J., Bodas, R., Soto, S., & Manso, T. (2012). Effects of addition of different vegetable oils to lactating dairy ewes' diet on meat quality characteristics of suckling lambs reared on the ewes' milk. *Meat Science, 91*(3), 277–283.

Wagner, R., & Franco, M. R. B. (2012). Effect of the variables time and temperature on volatile compounds extraction of salami by solid phase microextraction. *Food Analytical Methods, 5*(5), 1186–1195.

Yara-Varón, E., Li, Y., Balcells, M., Canela-Garayoa, R., Fabiano-Tixier, A.-S., & Chemat, F. (2017). Vegetable oils as Alternative Solvents for Green Oleo-Extraction, Purification and Formulation of Food and Natural Products. *Molecules, 22*(9), 1474.

8. CAPÍTULO 5 - HYDROGELLED EMULSION FROM CHIA AND LINSEED OILS: A PROMISING STRATEGY TO PRODUCE LOW-FAT BURGERS WITH A HEALTHIER LIPID PROFILE

Rosane Teresinha Heck, Erick Saldaña, José Manuel Lorenzo, Leticia Pereira Correa, Mariane Bittencourt Fagundes, Alexandre José Cichoski, Cristiano Ragagnin de Menezes, Roger Wagner, Paulo Cezar Bastianello Campagnol

Meat Science
Vol. 156, p. 174-182, 2019
https://doi.org/10.1016/j.meatsci.2019.05.034

Meat Science 156 (2019) 174–182

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Hydrogelled emulsion from chia and linseed oils: A promising strategy to produce low-fat burgers with a healthier lipid profile

Rosane Teresinha Heck[a], Erick Saldaña[b], José Manuel Lorenzo[c], Leticia Pereira Correa[a], Mariane Bittencourt Fagundes[a], Alexandre José Cichoski[a], Cristiano Ragagnin de Menezes[a], Roger Wagner[a], Paulo Cezar Bastianello Campagnol[a,*]

[a] Universidade Federal de Santa Maria, Santa Maria, CEP 97105-900 Rio Grande do Sul, Brazil
[b] Departamento de Agroindústria, Escola Superior de Agricultura "Luiz de Queiroz", Universidade de São Paulo, Alimentos e Nutrição, Av. Pádua Dias 11, 13418-900 Piracicaba, São Paulo, Brazil
[c] Centro Tecnológico de la Carne de Galicia, Parque Tecnológico de Galicia, San Cibrán das Viñas, Rúa Galicia N 4, Ourense, Spain

ARTICLE INFO

Keywords:
Healthier meat products
Hydrogelled emulsion
Chia oil
Linseed oil
Fatty acid profile
Consumer sensory characterization

ABSTRACT

Burgers (20% pork back fat) were produced with the replacement of 0, 20, 40, 60, 80, and 100% of pork back fat by hydrogelled emulsion (HE) from chia and linseed oils. No changes ($P > .05$) were observed for the moisture retention, diameter reduction, and cooking loss of the treatments, with a significant increase in the lipid retention ($P < .05$). Hardness increased ($P < .05$) with increasing the lipid replacement level, and a significant color difference (ΔE) was detected between the treatments and the control. In addition to reducing animal fat, a healthier fatty acid profile was reached after the lipid reformulation of the burgers, thus allowing the burgers to be labeled with health claims. The sensory tests (acceptance and Check-All-That-Apply) indicated that it is possible to replace up to 60% of pork back fat by HE.

1. Introduction

Burgers are meat products widely consumed worldwide, mainly due to their pleasant sensory characteristics and rapid preparation (Rios-Mera et al., 2019). However, the excessive consumption of these processed products can be detrimental to human health due to the presence of 20–30% of animal fat (Colmenero, 2000). Animal fat contains high contents of saturated fatty acids (SFA), and the intake of SFA may increase the risk factors related to the onset of cardiovascular diseases (Blekkenhorst et al., 2015). In contrast, polyunsaturated fatty acids (PUFAs) may contribute to the reduction of these risk factors, especially if the dietary n-6 to n-3 PUFA ratio is < 4 (Chaudhary et al., 2016). In this context, the meat industry has searched for alternatives to improve the lipid profile of the products, in an attempt to adapt the nutritional characteristics demanded by healthy consumers (Saldaña et al., 2018; Selani et al., 2016).

The replacement of animal fat by oils rich in n-3 PUFAs can be an alternative to obtain a healthier meat product. Chia and linseed oils are interesting for this substitution, since they contain around 60% α-linolenic acid in their lipid profile (Bodoira, Penci, Ribotta, & Martínez, 2017; Rubilar et al., 2012). Several authors have investigated these oils, including Gómez-Estaca et al. (2019) and Heck et al. (2018), who used linseed oil as an animal fat substitute in cooked meat products. The products presented a high PUFA/SFA and low n-6/n-3 ratio, thus demonstrating that linseed oil can be a good alternative for animal fat substitution. However, despite the potential health benefits, some technological and sensory aspects have been reported.

The hydrogelled emulsion (HE) method has shown good results in the entrapment of plant oils and may be an interesting alternative for the use of oils rich in n-3 PUFAs in meat products. HEs are semi-solid systems, in which a hydrophobic liquid phase is immobilized in a three-dimensional environment of lipophilic solids (gelling agents) (Co & Marangoni, 2012). Among the different ways of preparing HE, oil-in-water emulsions (O/W) are the most studied due to the relative ease of preparation and low cost (Salcedo-Sandoval et al., 2015). In addition, the carrageenan used as a gelling agent is described in the literature as an important agent to improve the stability and texture of low-fat meat products (Ayadi, Kechaou, Makni, & Attia, 2009; Luruena-Martínez, Vivar-Quintana, & Revilla, 2004).

The insertion of n-3PUFA rich oils in the structured form of HE can effectively mimic the functionality of pork back fat in meat products, with the differential of having a healthier lipid profile (Kouzounis,

* Corresponding author.
E-mail address: paulo.campagnol@ufsm.br (P.C.B. Campagnol).

https://doi.org/10.1016/j.meatsci.2019.05.034
Received 15 April 2019; Received in revised form 29 May 2019
Available online 05 June 2019

Lazaridou, & Katsanidis, 2017), thus allowing the production of healthier meat products. However, there are few reports on the effect of heat treatment on the fatty acid profile with the addition of HE rich in n-3 PUFAs. In addition, the effects of n-3 PUFAs on the sensory profile of burgers are little explored in the literature, which hinders strategies to improve the sensory quality of this type of product. Therefore, this study investigated the lipid reformulation of burgers by replacing 20, 40, 60, 80, and 100% pork back fat by HE rich in n-3 PUFAs. The physicochemical, oxidative, technological, and nutritional properties of the raw and cooked burgers were evaluated. The acceptance and the sensory profile of the burgers were also assessed by multivariate procedures through the application of the acceptance test and the Check-All-That-Apply (CATA) questionnaire.

2. Materials and methods

2.1. Production of HE

The HE was produced in triplicate using the following formulation: 12.5% chia oil (Giroil SA, Santo Angelo, Brazil), 12.5% linseed oil (Giroil SA), 70% water, 1% polysorbate 80 (Merck, São Paulo, Brazil) and 4% kappa carrageenan (NovaProm, Brazil). The carrageenan was homogenized with water for 1 min. The plant oils were homogenized with polysorbate for 1 min. The oily phase was slowly added to the aqueous phase and homogenization was carried out for a further 2 min. The homogenization was performed using a food mixer (BMX200P, Britânia, São Paulo, Brazil). After homogenization, the mixture was transferred to a water bath at 80 °C and incubated for 20 min with constant stirring to gelatinize. Then, the HE was cooled at room temperature for 2 h and stored at 4 °C for 12 h until the manufacture of the burgers.

2.2. Characterization of HE

2.2.1. Proximate composition, Aw and pH

The proximate composition, Aw and pH of HE were determined in triplicate. Moisture (AOAC Method 950.46), ash (AOAC Method 940.26), and proteins (AOAC Method 920.152) contents were determined according to AOAC International (2005). The lipid content was determined by the Bligh and Dyer (1959) method. Aw was determined using an Aqualab apparatus (Decagon Devices Inc., Pullman, EUA). The pH was determined using 5 g of sample, which was homogenized with 50 mL of distilled water, and the pH was measured using a digital pH meter (DM-23-DC, Digimed, Brazil).

2.2.2. Fatty acid profile

The fatty acid profile of the HE was determined in triplicate. Lipids were extracted according to the method described by Bligh and Dyer (1959). Subsequently, transesterification and methylation of the fatty acids were performed as reported by Hartman and Lago (1973). The fatty acid methyl esters (FAME) were analyzed by the injection of 1 μL into a gas chromatograph equipped with a Varian brand gas ionization detector (CG-DIC), Star model 3400CX (CA, USA) and 8200 Varian automatic sampler (CA, USA). The injector remained in the split mode with a 50:1 ratio and temperature of 250 °C. Hydrogen was used as a drag gas at a constant pressure of 15 psi. The FAMEs were separated on CP-Wax 52 CB capillary column (Middelburg, The Netherlands) (50 m × 0.32 mm × 0.20 μm). The initial column temperature was 50 °C, which remained for 1 min, increasing to 180 °C at 10 °C/min, after 200 °C with an increased rate of 2 °C/min, and then 10 °C/min until reaching 230 °C, maintaining in isotherm for 5 min. The detector was maintained at 240 °C. The FAME identification was performed by comparing the retention times of the analytes with FAME Mix-37 standards (P/N 47885-U; Sigma-Aldrich, St. Louis, USA). For the quantification of fatty acids, 200 μL of internal standard (23:0me) was added, reaching a concentration of 1 mg/mL of the sample. The chromatographic areas were corrected using equivalent chain length references and fatty acid methyl esters correction factor. The results were expressed as mg of fatty acids/100 g of product. The Atherogenicity (AI) and Thrombogenicity (TI) indexes were calculated as reported by Ulbricht and Southgate (1991), according to Eqs. (1) and (2), respectively.

$$AI = \frac{C12{:}\,0 + (4 * C14{:}\,0) + C16{:}\,0}{(\Sigma PUFA) + (\Sigma MUFA)} \quad (1)$$

$$TI = \frac{C14{:}\,0 + C16{:}\,0 + C18{:}\,0}{(0,5 * \Sigma MUFA) + (0,5 * n - 6) + (3 * n - 3) + \left(\frac{n-3}{n-6}\right)} \quad (2)$$

2.2.3. Penetration test of HE

The penetration analysis of HE was performed in five replicates. The tests were performed in a TA-TX2 texture analyzer (Stable Micro Systems Ltd., Surrey, England) equipped with a 25 kg load cell. The HE was cut into cylinders (2 cm height × 2 cm in diameter) and then a probe of 6 mm in diameter was moved at a constant velocity of 0.5 mm/s in 1 cycle of 50% compression, measuring the maximum strength at the point of springiness of HE (Hardness, N) (Bourne, 2002).

2.3. Manufacture of the burgers

The following ingredients were used for the production of the burgers: beef (*Rectus femoris*, 74.31% moisture; 22.75% protein; 4.41% lipid; and 1.13% ash), pork back fat (63.69% lipid; 25.48% moisture; 11.69% protein; and 0.34% ash), salt, and garlic purchased in the local market. The control was made with 78.4% beef, 20% pork back fat, 1.5% salt, and 0.1% garlic. For the treatments, a substitution of 20 (C20), 40 (C40), 60 (C60), 80 (C80), and 100% (C100) of pork back fat (20 g/100 g) by HE was performed.

Beef and pork back fat were ground separately in a grinder (PJ22, Jamar Ltda., São Paulo, Brazil) with a 3 mm disc. The beef was then homogenized with the salt for 3 min to extract myofibrillar proteins. After this step, the remaining ingredients were added and mixed for 3 min. The burgers (100 g, 11 cm in diameter and 2.5 cm thick) were formatted using a conventional burger-maker (HP 112, Picelli, São Paulo, Brazil). The burgers were individually placed in bags (20 × 14 cm) of high-density polyethylene film (50 μm thick, with the oxygen transmission rate of 1434 cm^3/m^2/day, and water vapor transmission rate 0.6 g/m^2/day). Bags were sealed under air using a Selovac 200B packaging machine (Selovac, São Paulo, Brazil) and stored at −18 °C until analysis (24 h). Three independent replicates of each batch were performed.

Prior to analyses, the burgers were thawed at 4 °C for 12 h. The samples were cooked in an electric grill (Multi Grill, Britânia, São Paulo, Brazil) until reaching the internal temperature of 72 °C, which was monitored with the aid of a spit-type thermometer (HM-600, High Paulo, Brazil).

2.4. Characterization of the burgers

2.4.1. Proximate composition, pH, Aw

The proximate composition (moisture, protein, lipid, and ash), Aw, and pH of the beef, pork back fat and the raw and cooked burgers were determined in triplicate according to procedures described in Section 2.2.1.

2.4.2. Lipid oxidation (TBARS)

TBARS were determined in triplicate in the raw and cooked burgers, according to the procedures described by Bruna, Ordonez, Fernández, Herranz, and Hoz (2001). The results were expressed in milligrams of malonaldehyde per kg of sample.

2.4.3. Instrumental color

The color of raw and cooked burgers was measured using a Minolta CR-400 colorimeter (Minolta Sensing Inc. Konica, Japan) with spectral reflectance included as a calibration mode, D65 illuminant, and 10° viewing angle, operating in the CIE system (L* a* b*). The color variables were measured at six points on each side of three samples by treatment. The values of L* (lightness), a* (redness) and b* (yellowness) were determined. The color difference (ΔE) was also determined according to the procedures described by Faria et al. (2015).

2.4.4. Fatty acid profile

The fatty acid profile of the beef, pork back fat and the raw and cooked burgers was determined in triplicate according to procedures described in Section 2.2.2.

2.4.5. Technological properties

The technological properties were determined in triplicate using three samples for each treatment. The raw samples were weighed and the diameter was measured. These procedures were repeated after cooking and cooling the samples to 25 °C. The cooking loss and diameter reduction was calculated according to Eqs. (3) and (4), respectively.

$$Cooking\ loss\,(\%) = \frac{weight_{raw} - weight_{cooked}}{weight_{raw}} \quad (3)$$

$$Diameter\ reduction\,(\%) = \frac{diameter_{raw} - diameter_{cooked}}{diameter_{raw}} \quad (4)$$

The moisture retention and the lipid retention were determined according to Eqs. (5) and (6), respectively.

$$Moisture\ retention\,(\%) = \frac{(100 - cooking\ loss\ (\%)) \times (moisture_{cooked})}{100} \quad (5)$$

$$Lipid\ retention\,(\%) = \frac{(weight_{cooked} \times lipid_{cooked})}{(weight_{raw} \times lipid_{raw})} \times 100 \quad (6)$$

2.4.6. Texture profile

For texture profile analysis (TPA), three burgers from each treatment were cooked and then cooled to 25 °C. Four cylinders (2 cm × 2 cm) were cut from each sample, totaling 12 cylinders per treatment. TPA analysis was performed in a TA-TX2 texture analyzer (Stable Micro Systems Ltd., Surrey, England) equipped with a 25 kg loading cell. A 36 mm probe was used, moving at a constant velocity of 1 mm/s and the cylinders were subjected to two consecutive cycles of 50% compression, as described by Bourne (1978). The parameters hardness (N), springiness (mm), cohesiveness, and chewiness (N) were determined.

2.4.7. Sensory analysis

This study was approved by the Research Ethics Committee of the Federal University of Santa Maria (RS, Brazil) (CAAE: 57433316.8.0000.5346). The burgers were cooked as described in Section 2.3, cut into pieces of 4 × 4 × 2.5 cm, individually wrapped, and kept in an oven at 60 °C. One hundred untrained and regular (at least once a month) burger consumers (62% female, 38% male, aged 18–55) received the samples coded with three random numbers in a sequential monadic way, following a Latin square design (Macfie, Bratchell, Greenhoff, & Vallis, 1989). The tests were performed in individual booths with fluorescent lighting (350 lx). Water and biscuits were provided to consumers to rinse their palate between the samples.

Firstly, consumers rated the overall acceptance of the burgers using a hedonic scale of nine points (1 = extremely disliked and 9 = extremely liked) (Stone, Bleibaum, & Thomas, 2012). Then, they answered a CATA questionnaire composed of 20 sensory descriptors. The descriptors were based on a preliminary study (Alves et al., 2016) and were related to the appearance (ideal color, pale color, strange appearance, and oily), aroma (soft, rancid, pleasant, and fat), flavor (rancid, pleasant, bitter, acid, unpleasant, seasoning in the right amount, and fish flavor) and texture (succulent, ideal, rubbery, dry, and hard).

2.5. Statistical analysis

2.5.1. Experimental design and instrumental data

The entire experiment was replicated three times ($n = 3$). Data (except the sensory data) were analyzed through analysis of variance (ANOVA) using a general linear model, considering the treatments as a fixed effect and the replicates as a random effect. Pairwise comparison was performed by the Tukey's test at 5% of significance. Data were analyzed using the SPSS statistical program (SPSS, Chicago, IL, USA).

2.5.2. Sensory analysis

Data set was represented by boxplot and ANOVA, considering treatments and consumers' as factors. The multi-dimensional representation of consumers acceptance of the burgers was performed by Internal Preference Mapping. This technique consisted of a principal component analysis (PCA) on the individual acceptance data (treatments in the rows and consumers in the columns), based on the Pearson correlation matrix (Saldaña et al., 2019a). To identify consumer segments, a hierarchical clustering on principal components (HCPC) was performed using the first five coordinates of the PCA, Euclidean distances, and Ward's agglomeration method (Saldaña et al., 2019a). A contingency table was obtained of the CATA results. Then, a correspondence analysis (CA) was performed using the X^2 distance, considering the average acceptance by clusters as supplementary variables. Confidence ellipses were built around the treatments by parametric bootstrapping, which indicates the multivariate similarity, that is, if two ellipses overlap, they are similar. All data processing was done in the R environment.

3. Results and discussion

3.1. Characterization of HE

The proximate composition of HE was: 69.9 ± 0.58% moisture, 23.1 ± 0.81% lipids, 0.96 ± 0.21% ash, and 0.84 ± 0.32% proteins (Table S1). These results indicated that the hydrogelled emulsion was able to retain practically all the oil (25%) used in the preparation of the emulsion. In addition, the HE showed Aw, pH, and hardness of 0.968; 6.00; and 138.35 N, respectively. HE contained 11.6 g/100 g PUFA, 3.2 g/100 g SFA, and 0.07 g/100 g MUFA. The levels of n-6 and n-3 fatty acids were 2.8 g/100 g and 8.7 g/100 g of product, respectively, with an n-6/n-3 ratio of 0.33 (Table S1).

3.2. Characterization of the burgers

3.2.1. Proximate composition, TBARS, pH, aw, and instrumental color

Significant differences were observed in the proximate composition of the reformulated burgers (Table 1). The replacement from 40% of pork back fat by HE caused an increase ($P < .001$) in moisture when compared to the control. It was also found a significant decrease in lipid content from 60% replacement of pork back fat. The raw burgers of the treatment C60 showed a ~ 25% reduction of lipids when compared to the control, while the raw burgers of the treatments C80 and C100 achieved ~ 50% reduction. The control presented higher ($P < .001$) protein values when compared to all raw burgers. The differences in the proximate composition of the raw burgers were expected, due to the different moisture contents, lipids, and proteins of pork back fat and HE (Table S1).

Concerning the cooked burgers, higher ($P < .001$) moisture contents were observed for the treatments C80 and C100. The control

Table 1
Effect of the partial and total replacement of pork back fat by hydrogelled emulsion from chia and linseed oils on the chemical composition, TBARS, pH, Aw, and color parameters of the beef burgers.

	Raw								Cooked							
	Control	C20	C40	C60	C80	C100	SEM	Sig.	Control	C20	C40	C60	C80	C100	SEM	Sig.
Chemical composition (%)																
Moisture	63.36^{d}	65.50^{cd}	67.60^{bc}	71.08^{ab}	73.35^{a}	71.33^{ab}	1.23	***	55.07^{b}	55.71^{b}	55.58^{b}	56.87^{b}	63.12^{a}	63.63^{a}	0.50	***
Ash	2.35^{a}	2.83^{a}	2.69^{a}	2.70^{a}	2.94^{a}	2.98^{a}	0.36	n.s.	3.72^{a}	3.07^{a}	3.72^{a}	3.03^{a}	3.13^{a}	3.35^{a}	0.28	n.s.
Protein	21.86^{a}	20.78^{b}	20.09^{b}	20.62^{b}	20.96^{b}	20.82^{b}	0.26	***	23.38^{a}	21.33^{b}	21.30^{b}	21.27^{b}	21.68^{b}	21.98^{b}	1.56	*
Fat	15.00^{a}	13.86^{a}	14.45^{a}	11.34^{b}	8.44^{c}	7.17^{c}	0.44	***	13.63^{a}	11.54^{ab}	10.93^{ab}	11.30^{ab}	7.82^{b}	7.08^{b}	1.36	*
Lipid oxidation																
TBARS	0.27^{d}	0.30^{d}	0.53^{c}	0.64^{b}	1.26^{a}	1.25^{a}	0.02	***	0.40^{e}	0.60^{d}	1.08^{c}	1.33^{b}	1.39^{b}	1.63^{a}	0.03	***
Physico-chemical parameters																
pH	5.78^{b}	5.74^{d}	5.75^{c}	5.76^{c}	5.78^{b}	5.81^{a}	0.003	***	5.78^{e}	5.88^{d}	5.94^{bc}	5.96^{b}	5.99^{a}	5.92^{c}	0.008	***
Aw	0.977^{a}	0.976^{ab}	0.968^{cd}	0.963^{d}	0.973^{ab}	0.971^{bc}	0.001	***	0.962^{a}	0.955^{ab}	0.957^{ab}	0.956^{ab}	0.951^{b}	0.953^{ab}	0.003	*
Color parameters																
L*	49.25^{a}	47.98^{a}	47.29^{a}	46.96^{a}	46.18^{a}	46.25^{a}	1.10	n.s.	45.93^{c}	46.81^{bc}	49.09^{ab}	50.86^{a}	50.76^{a}	47.84^{b}	0.84	***
a*	19.42^{ab}	18.45^{b}	18.42^{b}	19.14^{ab}	20.23^{a}	19.32^{ab}	0.58	**	11.33^{a}	10.83^{ab}	10.24^{bc}	10.42^{bc}	9.87^{c}	9.94^{c}	0.23	***
b*	15.59^{bc}	15.38^{c}	15.66^{bc}	16.75^{ab}	17.51^{a}	17.59^{a}	0.43	***	15.93^{b}	16.67^{ab}	16.06^{b}	17.25^{a}	17.50^{a}	16.93^{a}	0.41	**
ΔE	–	4.49^{b}	5.29^{ab}	5.33^{ab}	5.89^{a}	5.97^{a}	0.71	**	–	3.64^{b}	3.75^{b}	5.41^{ab}	5.35^{ab}	5.88^{a}	0.68	**

$^{a–d}$Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < .05$).
Batches: Control: 20% pork back fat; C20, C40, C60, C80 and C100: 20, 40, 60, 80 and 100% substitution of pork back fat by hydrogelled emulsion from chia and linseed oils, respectively.
Sig.: significance: ***($P < .001$), **($P < .01$), *($P < .05$), n.s. (not significant).

presented significantly higher protein levels ($P < .05$) when compared to all treatments. On the other hand, the treatments C80 and C100 presented lower ($P < .05$) lipid contents in relation to the control. These results are due to the lower moisture content and higher lipid and protein contents of the pork back fat when compared to HE (Table S1).

TBARS values increased significantly with the increase in the substitution level of pork back fat by HE (Table 1). However, only the treatments C80 and C100 showed TBARS values close to the threshold limit perceived by consumers (Wood et al., 2004). This increase in lipid oxidation of the reformulated burgers was expected due to the high concentration of PUFAs in HE (Table S1).

The pH and Aw values of the raw samples ranged from 5.74 to 5.81 and 0.963 to 0.977, respectively (Table 1). As expected, there was an increase in pH and a decrease in Aw after the cooking step, with significant differences between the treatments and the control. However, the lipid reformulation did not cause a great impact on these parameters, since the pH and Aw values of the reformulated burgers were within the range normally found for this type of meat product (Comi, Tirloni, Andyanto, Manzano, & Iacumin, 2015; Kryževičūtė, Jaime, Diez, Rovira, & Venskutonis, 2017; Selani et al., 2016).

The replacement of up to 60% pork back fat by HE did not affect the color parameters L*, a*, b* of the raw burgers (Table 1). However, the raw burgers with 80 and 100% substitution showed a more yellowness (higher b* values) color than the control, probably due to the differences in b* values of pork back fat and HE (Table S1). After cooking, the treatments with a substitution level of 40% pork back fat presented higher L* values and lower a* values when compared to the control. In addition, the substitution from 60% also increased b* values after cooking when compared to the control (Table 1). It should be noted that the differences in instrumental color between the control and the treatments are likely to be perceived by consumers in both raw and cooked burgers, once ΔE values were > 2 (Francis & Clydesdale, 1975).

3.2.2. Technological properties and texture profile

The cooking loss is a major concern when replacing animal fat with oils in meat products (Keenan et al., 2015). No significant differences were found in moisture retention, diameter reduction, and cooking loss between the control and the treatments (Table 2). Thus, the lipid reformulation was effective to maintain these important technological parameters. The treatments C60, C80, and C100 presented higher ($P < .001$) lipid retention than the control. The control burgers and the treatments with 20 (C20) and 40% (C40) substitution level showed a reduction of lipid between 1.3 and 3.5% after cooking, while the treatments with 60, 80, and 100% substitution presented lipid contents similar to the raw burgers. This result suggests that HE was effective in retaining plant oils after cooking. Similar results were found by Da Silva et al. (2019) and Wolfer, Acevedo, Prusa, Sebranek, and Tarté (2018) when using oleogels as lipid substitutes.

In this study, the lipid reformulation did not affect the springiness and cohesiveness of the burgers ($P > .05$). However, all reformulated treatments had a greater ($P < .001$) hardness and chewiness when compared to the control (Table 2). Hardness increased with an increase in the substitution level. The protein: lipid ratio of the control was 1.7 after cooking, while the modified samples exhibited a ratio ranging from 1.9 (C20) to 3.1 (C100) (Table 1). The higher protein: lipid ratio can explain the differences in hardness, once the proteins may have formed a dense protein network making the product harder (Youssef & Barbut, 2010), which increased the number of chewing cycles until swallowing, expressed as chewiness. Similarly, Fagundes et al. (2017) also reported a higher hardness of burgers made with a higher protein: lipid ratio.

3.2.3. Fatty acid profile

The fatty acid profile of raw and cooked burgers is shown in Tables 3 and 4, respectively. In quantitative terms, the main SFA found in raw and cooked burgers were palmitic acid (16:0) and stearic acid (18:0). The amount of SFA decreased ($P < .001$) gradually with the increase in the substitution level of pork back fat. The burgers of the treatments C60 (raw), C80 (raw and cooked), and C100 (raw and cooked) showed a reduction of > 30% in the SFA content when compared to the control. Therefore, these treatments can be claimed as “reduced in saturated fat” (European Parliament, 2006). The most abundant MUFA and PUFA of the raw and cooked burgers were oleic acid (18:1n-9c) and linoleic acid (C18:2n6c), respectively.

The modified treatments had a significantly higher linolenic acid content (C18:3n3) when compared to the control for both raw and cooked burgers. Raw and cooked burgers with 20% substitution of pork back fat by HE can be claimed as a “source of omega-3 fatty acids”, as they contain > 0.3 g linolenic acid per 100 g product. On the other hand, the burgers with a lipid substitution from 40% can be claimed as “high omega-3 fatty acids”, as they have at least 0.6 g linolenic acid per 100 g product (European Parliament, 2006).

Table 2
Effect of the partial and total replacement of pork back fat by hydrogelled emulsion from chia and linseed oils on technological and textural parameters of the beef burgers.

	Batches						SEM	Sig.
	Control	C20	C40	C60	C80	C100		
Technological parameters (%)								
Moisture retention	54.98^{a}	60.29^{a}	54.73^{a}	56.15^{a}	55.82^{a}	58.58^{a}	1.94	n.s.
Diameter reduction	20.61^{a}	18.05^{a}	16.93^{a}	16.80^{a}	17.16^{a}	17.94^{a}	2.95	n.s.
Cooking loss	36.78^{a}	31.79^{a}	33.46^{a}	31.60^{a}	35.18^{a}	34.39^{a}	1.96	n.s.
Lipid retention	57.42^{c}	60.73^{bc}	59.77^{c}	68.17^{a}	67.92^{a}	66.70^{ab}	1.89	***
Textural parameters (cooked burgers)								
Hardness (N)	68.31^{d}	92.85^{c}	117.32^{b}	104.60^{bc}	110.41^{bc}	140.31^{a}	6.02	***
Springiness	0.74^{a}	0.81^{a}	0.82^{a}	0.79^{a}	0.78^{a}	0.74^{a}	0.04	n.s.
Cohesiveness	0.46^{a}	0.52^{a}	0.47^{a}	0.49^{a}	0.51^{a}	0.45^{a}	0.02	n.s.
Chewiness (N)	31.61^{c}	48.49^{b}	55.76^{b}	51.03^{b}	57.00^{b}	68.97^{a}	3.15	***

$^{a–c}$Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < .05$).
Batches: Control: 20% pork back fat; C20, C40, C60, C80 and C100: 20, 40, 60, 80 and 100% substitution of pork back fat by hydrogelled emulsion from chia and linseed oils, respectively.
SEM: standard error of the mean
Sig.: significance: $^{***}(P < .001)$, $^{**}(P < .01)$, $^{*}(P < .05)$, n.s. (not significant).

The control exhibited a PUFA/SFA ratio of 0.39 and 0.37 for the raw and cooked burgers, respectively. This result is of concern for human health, once a frequent consumption of foods with a PUFA/SFA ratio lower than 0.45 may increase the incidence of cardiovascular diseases (Wood et al., 2004). The PUFA/SFA ratio increased significantly with the increase in the substitution level of pork back fat, which was above the minimum recommended to decrease the incidence of cardiovascular diseases for all treatments.

One of the main challenges for the development of healthier meat products is the reduction of n-6/n-3 ratio, once meat products should have a maximum ratio of 4:1 to be considered healthy (Simopoulos, 2011). The control presented a high n-6/n-3 ratio, with values of 13.34 and 10.37 for the raw and cooked burger, respectively, which was similar to that found in other studies (Baggio & Bragagnolo, 2006; Heck et al., 2017). However, all reformulated treatments presented a healthier n-6/n-3 ratio due to the high content of n-3 PUFAs from HE (Table

Table 3
Effect of the partial and total replacement of pork back fat by hydrogelled emulsion from chia and linseed oils on the fatty acid profile of the raw beef burgers.

	Raw (mg/100 g of sample)						SEM	Sig.
	Control	C20	C40	C60	C80	C100		
C12:0	6.35^{b}	5.79^{c}	7.06^{a}	0.00^{d}	0.00^{d}	0.00^{d}	0.09	***
C14:0	158.68^{a}	148.07^{a}	147.94^{a}	128.08^{b}	81.26^{c}	54.96^{d}	4.99	***
C14:1	8.62^{b}	9.17^{b}	12.82^{a}	11.64^{ab}	9.57^{ab}	8.52^{b}	1.06	**
C15:0	18.07^{ab}	17.07^{ab}	24.42^{a}	19.16^{ab}	16.02^{b}	12.96^{b}	2.20	***
C16:0	3272.72^{a}	2928.52^{b}	2893.64^{b}	2209.14^{c}	1386.87^{d}	939.01^{e}	48.82	***
C16:1	211.79^{a}	196.16^{ab}	190.68^{b}	152.45^{c}	98.30^{d}	56.25^{e}	4.73	***
C17:0	82.44^{a}	67.87^{b}	76.17^{ab}	69.94^{b}	52.67^{c}	32.39^{d}	3.18	***
C17:1	54.07^{a}	48.17^{b}	49.92^{ab}	40.35^{c}	31.35^{d}	22.39^{e}	1.45	***
C18:0	1872.02^{a}	1698.56^{b}	1661.99^{b}	1266.48^{c}	949.41^{d}	698.43^{e}	41.51	***
C18:1n9c	5729.45^{a}	5011.81^{b}	4988.57^{b}	3502.86^{c}	2579.09^{d}	1655.71^{e}	78.78	***
C18:2n6c	2179.50^{a}	2040.83^{a}	2167.27^{a}	1666.97^{b}	1070.73^{c}	871.49^{d}	49.02	***
C18:3n3	151.65^{e}	536.12^{d}	1033.82^{c}	1363.92^{bc}	1503.84^{b}	2264.97^{a}	104.11	***
C20:0	42.85^{a}	37.41^{b}	37.63^{b}	32.04^{c}	22.19^{d}	18.82^{d}	1.42	***
C20:1	107.22^{a}	88.26^{b}	96.62^{ab}	64.56^{c}	30.95^{d}	0.00^{e}	3.34	***
C20:2	105.95^{a}	97.99^{b}	92.88^{b}	59.29^{c}	29.94^{d}	0.00^{e}	2.25	***
C20:3n6	19.52^{a}	18.07^{b}	17.46^{b}	16.53^{b}	15.31^{bc}	13.1^{c}	1.72	***
C20:4n6	39.80^{ab}	48.61^{a}	49.36^{a}	41.63^{a}	22.34^{bc}	8.85^{c}	5.69	***
C20:3n3	19.68^{a}	19.37^{a}	19.18^{a}	17.30^{a}	11.21^{b}	10.02^{c}	1.51	***
C20:5n3	0.00^{c}	11.82^{b}	12.80^{b}	14.67^{ab}	15.10^{a}	17.21^{a}	1.89	***
C23:0	921.73^{a}	824.64^{a}	861.93^{a}	668.08^{b}	528.50^{c}	513.69^{c}	38.96	***
ΣSFA	6374.85^{a}	5727.91^{b}	5710.79^{b}	4392.92^{c}	3036.91^{d}	2270.25^{e}	78.34	***
ΣMUFA	6111.15^{a}	5353.56^{b}	5338.60^{b}	3771.87^{c}	2749.25^{d}	1742.87^{e}	83.91	***
ΣPUFA	2514.10^{c}	2772.80^{bc}	3392.76^{a}	3180.32^{ab}	2653.89^{c}	3158.80^{ab}	140.96	***
PUFA/SFA	0.39^{e}	0.49^{de}	0.60^{cd}	0.73^{bc}	0.88^{b}	1.40^{a}	0.05	***
n-6/n-3	13.34^{a}	3.76^{b}	2.16^{bc}	1.24^{bc}	0.74^{c}	0.39^{c}	0.79	***
AI	0.45^{a}	0.43^{ab}	0.40^{b}	0.39^{b}	0.32^{c}	0.24^{d}	0.01	***
TI	0.48^{a}	0.38^{b}	0.29^{c}	0.23^{d}	0.18^{d}	0.10^{e}	0.02	***

$^{a–e}$Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < .05$).
Batches: Control: 20% pork back fat; C20, C40, C60, C80 and C100: 20, 40, 60, 80 and 100% substitution of pork back fat by hydrogelled emulsion from chia and linseed oils, respectively.
SEM: standard error of the mean.
Sig.: significance: $^{***}(P < .001)$, $^{**}(P < .01)$.
SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; n-6 = omega-6; n-3 = omega-3. AI: atherogenic index; TI: thrombogenic index.

Table 4
Effect of the partial and total replacement of pork back fat by hydrogelled emulsion from chia and linseed oils on the fatty acid profile of the cooked beef burgers.

	Cooked (mg/100 g of sample)						SEM	Sig.
	Control	C20	C40	C60	C80	C100		
C12:0	7.91^{a}	6.80^{b}	4.16^{c}	4.96^{bc}	3.34^{c}	0.00^{d}	0.65	***
C14:0	157.20^{a}	117.00^{b}	115.22^{b}	110.79^{b}	78.20^{c}	52.50^{d}	3.26	***
C14:1	11.07^{a}	6.62^{b}	10.54^{a}	8.76^{ab}	8.35^{ab}	0.00^{c}	0.93	***
C15:0	19.92^{a}	13.26^{bc}	0.00^{d}	14.38^{b}	13.43^{bc}	10.83^{c}	1.00	***
C16:0	2936.27^{a}	2445.53^{b}	2205.83^{c}	2252.97^{c}	1553.68^{d}	1043.17^{e}	30.08	***
C16:1	205.33^{a}	152.62^{b}	145.74^{b}	142.14^{b}	99.71^{c}	63.27^{d}	3.78	***
C17:0	78.82^{a}	52.63^{b}	53.46^{b}	47.63^{c}	41.65^{d}	29.45^{e}	1.33	***
C17:1	56.01^{a}	38.41^{b}	40.03^{b}	39.58^{b}	31.65^{c}	0.00^{d}	1.63	***
C18:0	1648.66^{a}	1395.61^{b}	1346.08^{b}	1123.28^{c}	952.27^{d}	653.60^{e}	38.03	***
C18:1n9c	4972.75^{a}	4107.28^{b}	3679.04^{d}	3844.11^{c}	2628.85^{e}	1744.40^{f}	48.57	***
C18:2n6c	1771.50^{a}	1609.34^{ab}	1463.64^{b}	1597.20^{b}	1145.56^{c}	897.41^{d}	49.62	***
C18:3n3	145.50^{f}	372.42^{e}	680.50^{d}	879.89^{c}	1360.27^{b}	2013.96^{a}	52.46	***
C20:0	33.10^{a}	30.96^{b}	28.75^{c}	30.15^{bc}	22.98^{d}	19.80^{e}	0.44	***
C20:1	99.60^{a}	87.92^{b}	72.30^{d}	80.19^{c}	49.52^{e}	30.90^{f}	1.57	***
C20:2	97.50^{a}	75.79^{b}	59.06^{c}	66.34^{c}	34.76^{d}	14.90^{e}	2.29	***
C20:3n6	19.43^{a}	14.35^{b}	11.27^{c}	13.54^{b}	9.97^{c}	7.37^{d}	0.51	***
C20:4n6	56.32^{a}	38.66^{b}	34.48^{bc}	38.17^{b}	30.03^{c}	22.37^{d}	1.83	***
C20:3n3	17.18^{a}	13.79^{ab}	0.000^{d}	12.58^{b}	7.91^{c}	4.78^{c}	1.22	***
C20:5n3	15.70^{a}	0.00^{c}	0.00^{c}	0.00^{c}	14.96^{a}	8.76^{b}	0.52	***
C23:0	927.05^{a}	706.97^{b}	723.14^{b}	731.88^{b}	568.10^{c}	551.53^{c}	17.81	***
ΣSFA	5807.81^{a}	4761.96^{b}	4476.62^{c}	4316.04^{d}	3233.64^{e}	2368.79^{f}	43.30	***
ΣMUFA	5344.76^{a}	4392.84^{b}	3947.64^{c}	4114.79^{c}	2818.09^{d}	1838.56^{e}	51.28	***
ΣPUFA	2123.12^{c}	2124.35^{c}	2248.95^{c}	2607.72^{b}	2603.45^{b}	2969.55^{a}	70.16	***
PUFA/SFA	0.37^{e}	0.45^{de}	0.50^{d}	0.61^{c}	0.81^{b}	1.26^{a}	0.03	***
n-6/n-3	10.37^{a}	4.33^{b}	2.29^{c}	1.86^{c}	0.86^{d}	0.46^{d}	0.22	***
AI	0.48^{a}	0.45^{b}	0.43^{b}	0.40^{c}	0.34^{d}	0.24^{e}	0.01	***
TI	0.49^{a}	0.40^{b}	0.34^{c}	0.25^{d}	0.18^{e}	0.10^{f}	0.01	***

$^{a–f}$Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < .05$).
Batches: Control: 20% pork back fat; C20, C40, C60, C80 and C100: 20, 40, 60, 80 and 100% substitution of pork back fat by hydrogelled emulsion from chia and linseed oils, respectively.
SEM: standard error of the mean.
Sig.: significance: ***($P < .001$).
SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; n-6 = omega-6; n-3 = omega-3. AI: atherogenic index; TI: thrombogenic index.

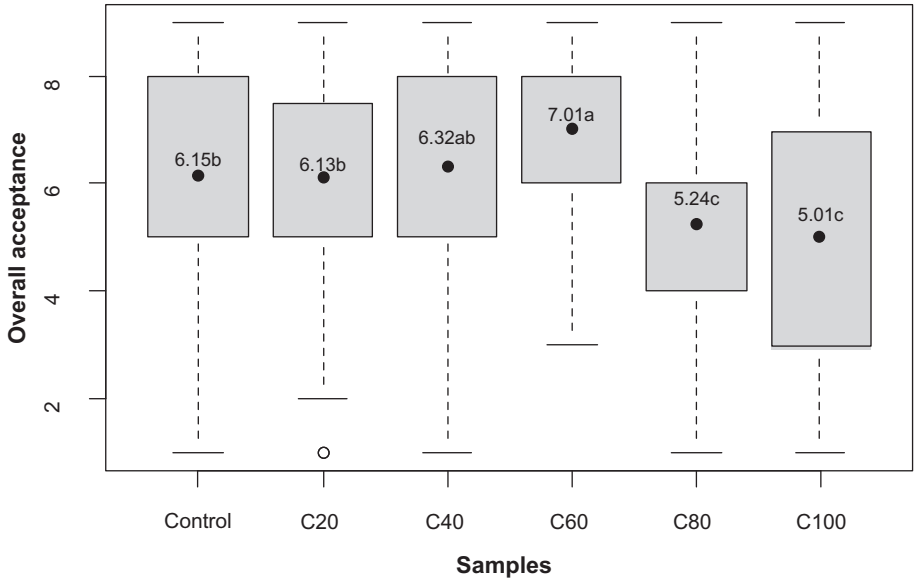


Fig. 1. Averages of the overall acceptance scores of the beef burgers using a nine-point hedonic scale. Different letters indicate significant differences according to the Tukey test.

S1). The lipid reformulation also resulted in a decrease in atherogenicity (AI) and thrombogenicity (TI) indices of both raw (Table 3) and cooked (Table 4) burgers. This fact also confers healthier characteristics to the product, once it suggests that the lipids present in the reformulated burgers can prevent the emergence of cardiovascular diseases (Ulbricht & Southgate, 1991).

3.2.4. Sensory evaluation

The treatment C60 presented the highest ($P < .001$) overall acceptance score (Fig. 1) among all treatments. The treatments C20 and

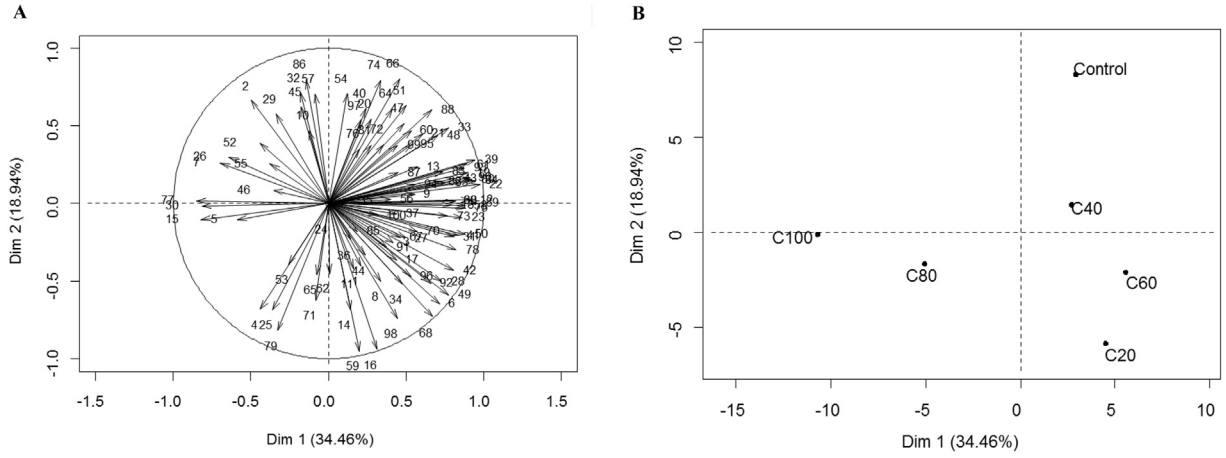


Fig. 2. Internal preference mapping: (A) Consumers; (B) Samples.

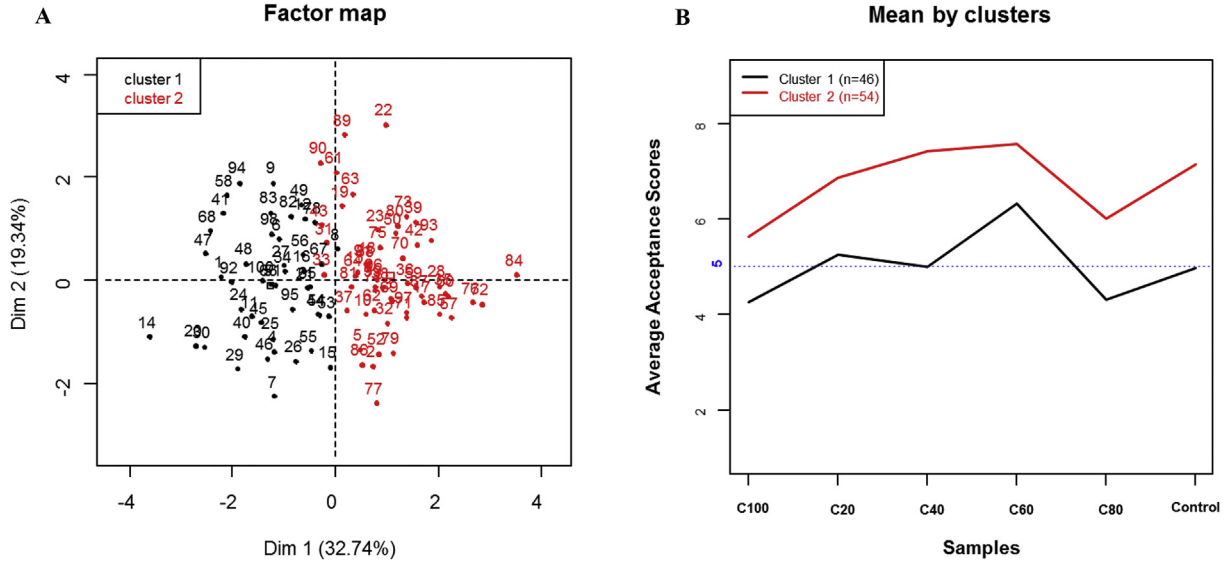


Fig. 3. Segmentation of consumers' panel. A: Factor map of consumers based on the two segments found; B: Representation of the average acceptance scores of each treatment by cluster.

C40 were similar to the control. Only the treatments C80 and C100 obtained lower overall acceptance scores ($P < .001$) than the control. However, the boxplot clearly indicated a high variability of the acceptance scores. Therefore, the results of the mean overall acceptance scores may have masked the preference of some consumers for the other treatments. Consumers (Fig. 2A) and treatments (Fig. 2B) were represented in the first two dimensions of the Internal Preference Mapping to show the heterogeneity of the consumers' acceptance (Saldaña et al., 2018). To identify the consumers of each segment, the HCPC technique was used (Fig. 3). Consumers were grouped into 2 different clusters (Fig. 3A), and the averages were calculated for the acceptance of each treatment by cluster (Fig. 3B). This analysis clearly indicated that the cluster 2 ($n = 54$), located at the top of the scale, was more tolerant towards the replacement of pork back fat by HE. The overall acceptance scores were > 5 for all treatments, indicating that consumers from cluster 2 were burger-lovers, thus evidencing a potential consumer market even for 100% replacement level. On the other hand, the cluster 1 ($n = 46$), located at the bottom of the scale, indicated that the treatments C80 and C100 were less accepted by consumers, as they presented scores below 5, while the treatments with up to 60% replacement of pork back fat showed a high overall acceptance.

The sensory profile of the burgers was represented in the first two dimensions of the CA, which explained 89.19% of the total variance (Dim 1: 79.98%, Dim 2: 9.21%) (Fig. 4). The confidence ellipses around the treatments were small, which shows a good stability of data (Saldaña et al., 2019b). The confidence ellipses were used to estimate the similarity between treatments over a multivariate perspective. It can be clearly seen that the treatments C20 and C40 were statistically similar to the control, while the treatment C60 was relatively close. In contrast, the treatments C80 and C100 were statistically similar to each other but completely different from the control. Clusters 1 and 2 presented a similar preference among them, once they were very close in the sensory map. Thus, the only difference between the clusters was the range used in the hedonic scale (Fig. 3B). The acceptance scores of the clusters 1 and 2 were positively related to the “succulent”, “ideal texture”, “pleasant aroma”, “pleasant flavor”, “seasoning in the right amount”, “ideal color”, and “soft aroma” attributes. These attributes were located in the negative quadrant of Dim 1 and characterized the control and the treatments C20, C40, and C60. In addition, the acceptance scores of the clusters 1 and 2 were inversely proportional to the descriptors that characterized the treatments C80 and C100, including “fatty flavor”, “rancid aroma”, “pale color”, “unpleasant flavor”, “fish flavor”, “oily”, “rancid flavor”, “acid flavor”, “bitter”, “rubbery texture”, and “hard”.

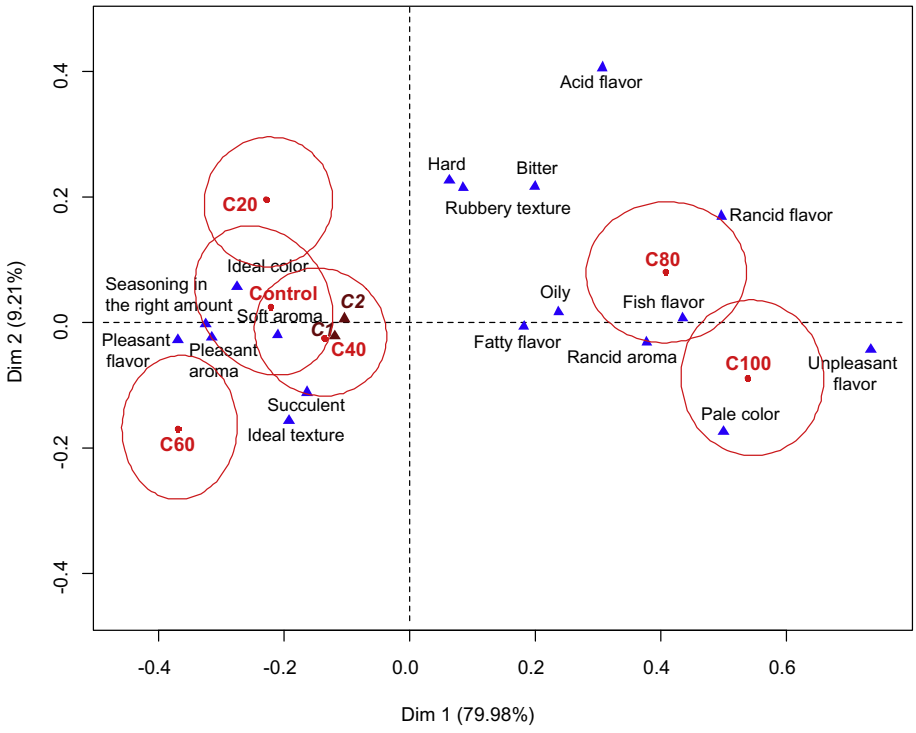


Fig. 4. Correspondence analysis on CATA questions of the beef burgers.

4. Conclusions

The replacement of pork back fat by HE did not adversely affect the technological properties of the burgers. However, the reformulation process led to an increase in hardness and chewiness of the samples due to increased protein: lipid ratio. The lipid reformulation reduced the lipid content and improved the fatty acid profile of the burgers, with no major changes in the lipid profile of the burgers after cooking. The substitution of only 20% pork back fat by HE has led to significant nutritional improvements. The acceptance scores and the sensory profile demonstrated that it is possible to replace up to 60% pork back fat by the HE produced in this study. Thus, the proposed lipid reformulation can be a promising strategy to produce low-fat burgers with a healthier lipid profile. However, to be used successfully in the meat industry, improvements in the oxidative stability of HE from chia and linseed oils are required.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.meatsci.2019.05.034.

Acknowledgements

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001 and by the Conselho Nacional de Desenvolvimento Científico e Tecnológico – CNPq [grant number 425577/2018-0]. E. Saldaña received the support from the "Consejo Nacional de Ciencia, Tecnología e Innovación Tecnológica – CONCYTEC" from Peru (CIENCIACTIVA programme, PhD scholarship contract agreement No. 104-2016-FONDECYT). P.C.·B Campagnol and J.M. Lorenzo are members of the HealthyMeat network, funded by CYTED (ref. 119RT0568).

Declarations of interest

None.

References

Alves, L. A. A. S., Lorenzo, J. M., Gonçalves, C. A. A., Santos, B. A., Heck, R. T., Cichoski, A. J., & Campagnol, P. C. B. (2016). Production of healthier bologna type sausages using pork skin and green banana flour as a fat replacers. *Meat Science, 121*, 73–78.

AOAC (Association of Official Analytical Chemistry) (2005). *Official methods of analysis of AOAC international* (17th ed.). Maryland, USA.

Ayadi, M., Kechaou, A., Makni, I., & Attia, H. (2009). Influence of carrageenan addition on Turkey meat sausages properties. *Journal of Food Engineering, 93*(3), 278–283.

Baggio, S. R., & Bragagnolo, N. (2006). The effect of heat treatment on the cholesterol oxides, cholesterol, total lipid and fatty acid contents of processed meat products. *Food Chemistry, 95*(4), 611–619.

Blekkenhorst, L. C., Prince, R. L., Hodgson, J. M., Lim, W. H., Zhu, K., Devine, A., ... Lewis, J. R. (2015). Dietary saturated fat intake and atherosclerotic vascular disease mortality in elderly women: A prospective cohort study. *The American Journal of Clinical Nutrition, 101*(6), 1263–1268.

Bligh, E. G., & Dyer, W. J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology, 37*(8), 911–917.

Bodoira, R. M., Penci, M. C., Ribotta, P. D., & Martínez, M. L. (2017). Chia (Salvia hispanica L.) oil stability: Study of the effect of natural antioxidants. *LWT - Food Science and Technology, 75*, 107–113.

Bourne, M. (2002). *Food texture and viscosity—Concept and measurement* (2nd ed.). San Diego: Academic Press0-12-119062-5 (Book Review).

Bourne, M. C. (1978). Texture profile analysis. *Food Technology, 32*(7), 62–66.

Bruna, J. M., Ordonez, J. A., Fernández, M., Herranz, B., & Hoz, L. (2001). Microbial and physico-chemical changes during the ripening of dry fermented sausages superficially inoculated with or having added an intracellular cell-free extract of *Penicillium aurantiogriseum*. *Meat Science, 59*, 87–96.

Chaudhary, R., Saadin, K., Bliden, K. P., Harris, W. S., Dinh, B., Sharma, T., & Gurbel, P. A. (2016). Risk factors associated with plasma omega-3 fatty acid levels in patients with suspected coronary artery disease. *Prostaglandins, Leukotrienes and Essential Fatty Acids, 113*, 40–45.

Co, E. D., & Marangoni, A. G. (2012). Organogels: An alternative edible oil-structuring method. *Journal of the American Oil Chemists Society, 89*(5), 749–780.

Colmenero, J. F. (2000). Relevants factors in strategies for fat reduction meat products. *Food Science and Technology, 11*(2), 56–66.

Comi, G., Tirloni, E., Andyanto, D., Manzano, M., & Iacumin, L. (2015). Use of bio-protective cultures to improve the shelf-life and the sensorial characteristics of commercial hamburgers. *LWT - Food Science and Technology, 62*(2), 1198–1202.

Da Silva, S. L., Amaral, J. T., Ribeiro, M., Sebastião, E. E., Vargas, C., De Lima, F. F., ... Campagnol, P. C. B. (2019). Fat replacement by oleogel rich in oleic acid and its impact on the technological, nutritional, oxidative, and sensory properties of Bologna-type sausages. *Meat Science, 149*, 141–148.

European Parliament (2006). Regulation (EC) no 1924/2006 of the European Parliament and of the council of 20 December 2006 on nutrition and health claims made on foods. *Official Journal of the European Union, L404*, 9–25.

Fagundes, D. T. O., Lorenzo, J. M., Dos Santos, B. A., Fagundes, M. B., Heck, R. T.,

Cichoski, A. J., ... Campagnol, P. C. B. (2017). Pork skin and canola oil as strategy to confer technological and nutritional advantages to burgers. *Czech Journal of Food Sciences, 35*(4), 352–359.

Faria, M. O., Cipriano, T. M., Cruz, A. G., Santos, B. A., Pollonio, M. A. P., & Campagnol, P. C. B. (2015). Properties of bologna-type sausages with pork back-fat replaced with pork skin and amorphous cellulose. *Meat Science, 104*, 44–51.

Francis, F. J., & Clydesdale, F. M. (1975). *Food colorimetry: Theory and applications.* Westport, CT: AVI Publishing Co.

Gómez-Estaca, J., Herrero, A. M., Herranz, B., Álvarez, M. D., Jiménez-Colmenero, F., & Cofrades, S. (2019). Characterization of ethyl cellulose and beeswax oleogels and their suitability as fat replacers in healthier lipid pâtés development. *Food Hydrocolloids, 87*, 960–969.

Hartman, L., & Lago, R. C. A. (1973). Rapid preparation of fatty acids methyl esters. *Laboratory Practice, 22*, 475–476.

Heck, R. T., Lucas, B. N., Dos Santos, D. J. P., Pinton, M. B., Fagundes, M. B., Etchepare, M. A., ... Campagnol, P. C. B. (2018). Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques. *Meat Science, 146*, 147–153.

Heck, R. T., Vendruscolo, R. G., Etchepare, M. A., Cichoski, A. J., Menezes, C. R., Barin, J. S., ... Campagnol, P. C. B. (2017). Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science, 130*, 16–25.

Keenan, D. F., Resconi, V. C., Smyth, T. J., Botinestean, C., Lefranc, C., Kerry, J. P., & Hamill, R. M. (2015). The effect of partial-fat substitutions with encapsulated and unencapsulated fish oils on the technological and eating quality of beef burgers over storage. *Meat Science, 107*, 75–85.

Kouzounis, D., Lazaridou, A., & Katsanidis, E. (2017). Partial replacement of animal fat by oleogels structured with monoglycerides and phytosterols in frankfurter sausages. *Meat Science, 130*, 38–46.

Kryževičūtė, N., Jaime, I., Diez, A. M., Rovira, J., & Venskutonis, P. R. (2017). Effect of raspberry pomace extracts isolated by high pressure extraction on the quality and shelf-life of beef burgers. *International Journal of Food Science and Technology, 52*(8), 1852–1861.

Luruena-Martínez, M. A., Vivar-Quintana, A. M., & Revilla, I. (2004). Effect of locust bean/xanthan gum addition and replacement of pork fat with olive oil on the quality characteristics of low-fat frankfurters. *Meat Science, 68*(3), 383–389.

Macfie, H. J., Bratchell, N., Greenhoff, K., & Vallis, L. (1989). Designs to balance the effect of order of presentation and first-order carry-over effects in hall tests. *Journal of Sensory Studies, 4*(2), 129–148.

Rios-Mera, J. D., Saldaña, E., Cruzado-Bravo, M. L. M., Patinho, I., Selani, M. M., Valentin, D., & Contreras-Castillo, C. J. (2019). Reducing the sodium content without modifying the quality of beef burgers by adding micronized salt. *Food Research Internacional, 121*, 288–295.

Rubilar, M., Morales, E., Contreras, K., Ceballos, C., Acevedo, F., Villarroel, M., & Shene, C. (2012). Development of a soup powder enriched with microencapsulated linseed oil as a source of omega-3 fatty acids. *European Journal of Lipid Science and Technology, 114*(4), 423–433.

Salcedo-Sandoval, L., Cofrades, S., Ruiz-Capillas, C., Matalanis, A., McClements, D. J., Decker, E. A., & Jimenez-Colmenero, F. (2015). Oxidative stability of n-3 fatty acids encapsulated in filled hydrogel particles and of pork meat systems containing them. *Food Chemistry, 184*, 207–213.

Saldaña, E., Garcia, A. O., Selani, M. M., Haguiwara, M. M. H., Almeida, M. A., Siche, R., & Contreras-Castillo, C. J. (2018). A sensometric approach to the development of mortadella with healthier fats. *Meat Science, 137*, 176–190.

Saldaña, E., Rios-Mera, J., Arteaga, H., Saldaña, J., Samán, C. M., Selani, M. M., & Villanueva, N. D. M. (2018). How does starch affect the sensory characteristics of mazamorra morada? A study with a dessert widely consumed by Peruvians. *International Journal of Gastronomy and Food Science, 12*, 20–22.

Saldaña, E., Saldarriaga, L., Cabrera, J., Behrens, J. H., Selani, M. M., Rios-Mera, J., & Contreras-Castillo, C. J. (2019a). Descriptive and hedonic sensory perception of Brazilian consumers for smoked bacon. *Meat Science, 147*, 60–69.

Saldaña, E., Saldarriaga, L., Cabrera, J., Behrens, J. H., Selani, M. M., Rios-Mera, J., & Contreras-Castillo, C. J. (2019b). Relationship between volatile compounds and consumer-based sensory characteristics of bacon smoked with different Brazilian woods. *Food Research International, 119*, 839–849.

Selani, M. M., Shirado, G. A. N., Margiotta, G. B., Saldanã, E., Spada, F. P., Piedade, S. M. S., & Canniatti-Brazaca, S. G. (2016). Effects of pineapple byproduct and canola oil as fat replacers on physicochemical and sensory qualities of low-fat beef burger. *Meat Science, 112*, 69–76.

Simopoulos, A. (2011). Evolutionary aspects of diet: The omega-6/omega-3 ratio and the brain. *Molecular Neurobiology, 44*(2), 203–215.

Stone, H., Bleibaum, R. N., & Thomas, H. A. (2012). *Sensory evaluation practices* (4th ed.). New York: Academic Press.

Ulbricht, T. L. V., & Southgate, D. A. T. (1991). Coronary heart disease: Seven dietary factors. *The Lancet, 338*, 985–992.

Wolfer, T. L., Acevedo, N. C., Prusa, K. J., Sebranek, J. G., & Tarté, R. (2018). Replacement of pork fat in frankfurter-type sausages by soybean oil oleogels structured with rice bran wax. *Meat Science, 145*, 352–362.

Wood, J. G., Rogina, B., Lavu, S., Ilowitz, K., Helfand, S. L., Tatar, M., & Sinclair, D. (2004). Sirtuin activators mimic caloric restriction and delay ageing in metazoans. *Nature, 430*, 686–689.

Youssef, M. K., & Barbut, S. (2010). Physicochemical effects of the lipid phase and protein level on meat emulsion stability, texture, and microstructure. *Journal of Food Science, 75*, 108–114.

9. CAPÍTULO 6 - JABUTICABA PEEL EXTRACT OBTAINED BY MICROWAVE HYDRODIFFUSION AND GRAVITY EXTRACTION: A GREEN STRATEGY TO IMPROVE THE OXIDATIVE AND SENSORY STABILITY OF BEEF BURGERS PRODUCED WITH HEALTHIER OILS

Rosane Teresinha Heck, Daniele Freitas Ferreira, Mariane Bittencourt Fagundes, Bibiana Alves Dos Santos, Alexandre José Cichoski, Erick Saldaña, José Manuel Lorenzo, Cristiano Ragagnin de Menezes, Roger Wagner, Juliano Smanioto Barin, Paulo Cezar Bastianello Campagnol

Meat Science
Vol. 170, p. 108230, 2020
https://doi.org/10.1016/j.meatsci.2020.108230

Meat Science 170 (2020) 108230

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: A green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils

Rosane Teresinha Heck[a], Daniele Freitas Ferreira[a], Mariane Bittencourt Fagundes[a], Bibiana Alves Dos Santos[a], Alexandre José Cichoski[a], Erick Saldaña[b], José Manuel Lorenzo[c], Cristiano Ragagnin de Menezes[a], Roger Wagner[a], Juliano Smanioto Barin[a], Paulo Cezar Bastianello Campagnol[a,*]

[a] Universidade Federal de Santa Maria, CEP 97105-900, Santa Maria, Rio Grande do Sul, Brazil
[b] Facultad de Ingeniería Agroindustrial, Universidad Nacional de Moquegua (UNAM), Moquegua, Peru
[c] Centro Tecnológico de la Carne de Galicia, Parque Tecnológico de Galicia, San Cibrán das Viñas, Rúa Galicia N 4, Ourense, Spain

ARTICLE INFO

Keywords:
Microwave hydrodiffusion and gravity
Jabuticaba peel extract
Healthier meat products
Hydrogelled emulsion
Chia oil
Linseed oil

ABSTRACT

Hydrogelled emulsions (HE) from chia and linseed oils (1:1) were made with different concentrations (0, 6, 8, and 10%) of jabuticaba peel extract (JPE) obtained by microwave hydrodiffusion and gravity (MHG) extraction. Burgers (20% fat) were produced with the replacement of 60% of fat by HEs. The oxidative profile and the sensory quality of raw and cooked burgers were evaluated for 120 days (−18 °C). The JPE exhibited 1.72 mg/mL of phenolic compounds and 57,741.67 μmol TE/mL of antioxidant capacity. In addition, the MHG extraction eliminated the mesophilic bacteria from the jabuticaba peel. The burgers made with HE and without the addition of JPE showed a 5-fold increase in TBARS values when compared to the control. On the other hand, the addition of 10% JPE to HE was effective to maintain the lipid oxidation similar to the control until the 60th day of storage. Besides, the incorporation of JPE into HE reduced the sensory defects caused by the lipid reformulation.

1. Introduction

One of the major challenges for the production of healthier meat products is the high concentrations of saturated fatty acids (SFA) in animal fat. The partial replacement of animal fat by oils with high n-3 PUFAs levels, such as chia and linseed oils, can confer healthier characteristics to meat products, thus reducing the risk of chronic diseases to consumers (Bodoira, Penci, Ribotta, & Martínez, 2017; Chaudhary et al., 2016; Rubilar et al., 2012).

However, despite the potential health benefits, some concerns including the oil release and depreciation of texture, aroma, and flavor have been reported in studies using chia and linseed oils as an animal fat substitute in cooked meat products (Gómez-Estaca et al., 2019; Heck et al., 2017). Hydrogelling of oils has proven to be an effective alternative for reducing these technological and sensory problems. In this technique, the oil phase is homogenized with the aqueous phase containing a gelling agent. Afterward, the mixture is heated to allow gelation, and then cooled to room temperature and stored under refrigeration. In previous studies, our research group replaced 0, 20, 40, 60, 80, and 100% pork back fat by HE from chia and linseed oils in beef burgers. In addition to reducing fat, a healthier lipid profile was observed, allowing burgers to be labeled with health claims. The sensory and technological quality was not affected by up to 60% replacement of pork back fat by HE. However, higher lipid oxidation was observed with the increase in the HE replacement level (Heck et al., 2019). Thus, improvements in the oxidative stability of HE from chia and linseed oils are required so that this ingredient can be used on a large scale in the meat industry. In this context, the incorporation of natural extracts from agroindustrial waste has stood out (Menegali et al., 2020; Saldaña, Serrano-León, Selani, & Contreras-Castillo, 2019).

Jabuticaba (*Myrciaria cauliflora*) has a large amount of bioactive compounds, which are mainly concentrated in the fruit peel, such as ellagic acid, quercetin, and anthocyanins (Quatrin et al., 2019). Studies have shown that jabuticaba peel extracts (JPE) obtained by simple extraction methods (using water or ethanol) were effective in reducing the lipid oxidation of meat products during storage (Almeida et al.,

* Corresponding author.
E-mail address: paulo.campagnol@ufsm.br (P.C.B. Campagnol).

https://doi.org/10.1016/j.meatsci.2020.108230
Received 28 January 2020; Received in revised form 21 May 2020; Accepted 22 June 2020
Available online 06 July 2020
0309-1740/

2015; Baldin et al., 2016). Thus, the jabuticaba peel can be considered a promising alternative to control the increase in lipid oxidation after the addition of oils with high n-3 PUFAs levels to meat products. However, further studies on the extraction techniques to obtain a greater amount of bioactive compounds from jabuticaba peel are required.

The microwave by hydrodiffusion and gravity (MHG) extraction is a process designed for extraction bioactive compounds in vegetable material using the gravity force to accelerate the extraction process (Kusuma, Putri, Triesty, & Mahfud, 2019). In general, the MHG extraction consists of heating the plant material in a microwave oven, which will lead to a disruption of the cells that contain the bioactive compounds. MHG extraction is known as an environmentally friendly method, as it can reduce the waste generation and energy consumption and eliminate or reduce the use of toxic solvents (Chemat et al., 2017).

So far, the incorporation of bioactive compounds extracted by MHG in oils rich in n-3 PUFAs has been little explored. Thus, this study investigated the extraction of bioactive compounds from jabuticaba peel by MHG and the incorporation at different concentrations (0, 6, 8, and 10%) into a 1:1 blend containing chia and linseed oils. The blend of healthier oils was hydrogelled and used to replace 60% fat in burger formulations. The effect of the lipid reformulation on the oxidative and sensory quality of raw and cooked burgers was evaluated during their shelf life (120 days at −18 °C).

2. Materials and methods

2.1. Extraction of JPE by MHG

Jabuticaba of the variety *Myrciaria cauliflora* was harvested in November 2018 in the city of Alecrim (Southern Brazil). The peels were separated from the pulp and cut into small pieces. The peels were cleaned in water and sanitized by immersion in sodium hypochlorite solution (200 mg/kg for 10 min) and kept at 4 °C before extraction. The extraction procedure was carried out using a 2.45 GHz multimode microwave oven (NEOS-GR, Milestone, Bergamo, Italy), at atmospheric pressure under 400 W of power for 20 min. During the extraction process, no solvent was used. The extract was drained by gravity, condensed, and collected in a container outside the microwave cavity. The extraction procedure was repeated three times on different days. For each extraction, 200 g of fresh jabuticaba peels were used.

2.2. Characterization of jabuticaba peel and JPE obtained by MHG

2.2.1. Total phenolic contents (TPC) and antioxidant capacity

The total phenolic contents (TPC) and the antioxidant capacity of jabuticaba peel and the JPE obtained by MHG were evaluated in triplicate by the Folin-Ciocalteu (Chandra & Mejia, 2004) and ORAC assays (Oxygen Radical Absorbance Capacity) (Ou, Hampsch-Woodill, & Prior, 2001), respectively. The TPC was expressed as milligrams of gallic acid equivalent per 100 g of sample (mg GAE/100 g) and the antioxidant capacity was expressed as μmol of Trolox equivalent to 1 g of vegetable oil.

2.2.2. Microbiological characterization

The microbiological characterization of the jabuticaba peel and the JPE obtained by MHG were carried out in triplicate. For the enumeration of mesophilic bacteria, PCA (Plate Count Agar) culture medium was used, with incubation at 36 °C for 48 h. For molds and yeast counts, BDA (Potato Dextrose Agar) was used with incubation at 25 °C for 7 days (ISO- International Organization for Standardization, 2013).

2.3. Elaboration of the hydrogelled emulsion with the addition of JPE and use in the burger formulations

The hydrogelled emulsion (HE) and the burger formulations were made in triplicate, as described by Heck et al. (2019). HEs containing 0, 6, 8, and 10% JPE were prepared, calculated in relation to the total weight of HE, and used in the formulations before HE gelatinization. The control burger was made with 20% pork back fat. The reformulated burgers were made with a 60% replacement of pork back fat by HEs containing 0, 6, 8, and 10% JPE, which corresponded to a concentration of 0 (CHE), 0.72 (T6), 0.96 (T8), and 1.2% (T10) of JPE in the product. The burgers were individually packed in high-density polyethylene bags (20 × 14 cm, 50 μm thick) and stored for 120 days at −18 °C. The oxygen transmission rate and the water vapor transmission rate of the bags were 1434 cm^3/m^2/day and 0.6 g/m^2/day, respectively.

2.4. Characterization of the reformulated burgers

The burgers were thawed at 4 °C for 12 h before analysis. The analyses were performed in both raw and cooked burgers. The burgers were cooked on an electric grill (Multi Grill, Britânia, Brazil) preheated to 150 °C until reaching the internal temperature of 72 °C. A skewer-type thermometer (HM-600, Highmed, Brazil) was inserted in the center of each burger to determine the internal cooking temperature.

2.4.1. Determination of pH and redox potential (Eh)

The pH and the redox potential (Eh) were determined in triplicate during storage (0, 30, 60, 90, and 120 days). For that, 5 g of sample was homogenized with 50 mL of distilled water, and both the pH and Eh were measured using a digital pH meter (DM-23-DC, Digimed, Brazil).

2.4.2. Color measurements

The color was measured on the first day of processing, and after 30, 60, 90, and 120 days of storage. A Minolta CR-400 colorimeter (Minolta Sensing Inc. Konica, Japan) was used in the CIE system (L*, a*, b*), using spectral reflectance included as a calibration mode, D65 illuminant, viewing angle of 10° and a round aperture size of 8 mm. The color variables were measured at six points on each side of three samples per treatment. The values of lightness (L*), redness (a*) and yellowness (b*) were determined (Faria et al., 2015).

2.4.3. TBARS assay

The lipid oxidation was determined in triplicate at days 0, 30, 60, 90, and 120 of storage, and expressed as milligrams of malonaldehyde per kg of sample, as described by Bruna, Ordonez, Fernández, Herranz, and Hoz (2001).

2.4.4. Sensory evaluation

The sensory tests were carried out in individual booths at the sensory analysis laboratory of the Federal University of Santa Maria, according to the protocol approved by their Human Research Ethics Committee (CAAE: 57433316.8.0000.5346). The overall liking and the Check-All-That-Apply (CATA) questionnaire was answered by 100 regulars (at least once a month) consumers of beef burgers (58% female, 42% male, aged 18–60). The cooked burgers were cut into 4 × 4 × 2.5 cm pieces, individually wrapped in aluminum foil, and kept at 60 °C in a conventional oven until the time of analysis. The samples were served in a sequential monadic way, encoded with three-random digits following a Latin square design (Macfie, Bratchell, Greenhoff, & Vallis, 1989). First, consumers rated the overall liking of beef burgers using the 9-point hedonic scale (1 = dislike extremely and 9 = like extremely) and then they answered the CATA questionnaire, selecting all the attributes that characterized each sample (Heck et al., 2019; Saldaña et al., 2019) using 18 sensory descriptors selected according to a preliminary study (Alves et al., 2016). The descriptive descriptors were related to the appearance (ideal color, pale color, strange appearance, and oily aspect), aroma (fruity, rancid, acidic, and mild), flavor (rancid, sweet, pleasant, bitter, unpleasant, and seasoning in the right amount) and texture (juicy, ideal, rubbery, and dry).

2.5. Statistical analysis

The entire experiment was replicated three times ($n = 3$). The physicochemical data were evaluated using a general linear model ANOVA considering treatments and storage period and their interaction as fixed factors, and the replicates as random factor. When the fixed factors were significant, Tukey's test was applied for a post-hoc comparison of the means at a 5% significance level.

For the sensory data, a general linear model ANOVA was also used to evaluate the overall liking scores, considering treatment as a fixed factor and consumer as a random factor. When appropriate, Tukey's test was used for pairwise comparison at a 5% significance level. The correspondence analysis (CA) was used to depict the CATA data. Confidence ellipses were constructed using bootstrapping at 95% confidence to show the stability of the samples in the sensory map (Rios-Mera et al., 2019). In addition, a cluster analysis was performed to find groups of samples with similar sensory characteristics. All statistical analyses were performed using R environment.

3. Results and discussion

3.1. Characterization of jabuticaba peel and JPE

The fresh jabuticaba peel exhibited 0.95 mg/g of phenolic compounds and antioxidant capacity of 29.64 μmol TE/g. More details about the characterization and quantification of phenolic compounds from jabuticaba peel can be consulted in the paper of Quatrin et al. (2019). The alternative process MHG was effective to produce an extract enriched with bioactive compounds. JPE exhibited 1.72 mg/mL phenolic compounds and antioxidant capacity of 57,741.67 μmol TE/mL (Table 1). The high contents of bioactive compounds in the jabuticaba peel are due to the heating caused by MHG, which led to a decrease in water viscosity, allowing a better mass transfer rate and penetration into the matrix (Plaza & Turner, 2015). In addition to obtaining an extract concentrated in bioactive compounds, the advantages of MHG include the low extraction time (20 min), the low energy expenditure, the non-use of toxic solvents, and high yield (52.5 ± 0.8%) (data not shown). The MHG eliminated the mesophilic bacteria present in the fresh jabuticaba peel, thus allowing the production of a microbiologically safe extract (Table 1). This reduction of microbial contamination is due to cell lysis caused by microwaves and the thermocoagulation of cytoplasmic proteins by microorganisms (Djordjević et al., 2019).

3.2. Characterization of the burgers

3.2.1. pH, Eh, and instrumental color

A significant interaction ($P < .001$) between the treatments and the storage time was observed for the parameter pH. Raw burgers had pH values between 5.62 and 5.96 (Fig. 1A) while the cooked burgers exhibited pH values ranging from 5.88 to 6.08 (Fig. 1B) during the 120 days of storage. In general, a slight increase in pH during storage was observed mainly in raw samples, probably due to the release of alkaline compounds generated during protein degradation (Lu, Zhang, Liu, Wang, & Ding, 2011). However, despite the statistical differences, the pH variation during storage was within the expected for this type of product for all treatments (Comi, Tirloni, Andyanto, Manzano, & Iacumin, 2015; Kryževičūtė, Jaime, Diez, Rovira, & Venskutonis, 2017; Selani et al., 2016). Thus, the lipid reformulation proposed in this study did not have a major impact on the pH of burgers.

The redox potential (Eh) of the samples was affected by the interaction between the treatments and the storage time ($P < .001$), which ranged from 142.50 to 216.16 and from 101.83 to 233.33 for raw and cooked burgers, respectively, during the storage (Fig. 2A and B). In general, an increase in Eh values was observed during the storage of the burgers (raw and cooked), with the most pronounced increase at the end of storage (day 120). This result is expected and can be due to the substrate's ability to gain or lose electrons, that is, the oxidation or reduction capacity, respectively. Thus, the higher the Eh, the more oxidized the substrate. At the end of the storage, the CHE samples (raw and cooked) showed higher Eh values ($P < .001$), indicating a greater tendency to undergo oxidative reactions (Latoch & Stasiak, 2015). On the other hand, the addition of JPE to HE was effective to keep the Eh values close to the control throughout the storage for both raw and cooked burgers.

A significant interaction ($P < .001$) between the treatments and the storage time was found for the color parameters (Fig. 3). No changes in L*, a*, and b* values were observed in raw and cooked burgers with the addition of HE (Figs. 3A-F). In general, a decrease in a * values was observed for all treatments during the storage, which was more pronounced in raw burgers probably due to the oxidative reactions (Pignitter et al., 2014).

3.2.2. Lipid oxidation - TBARS

A significant ($P < .001$) interaction between the treatments and the storage time was observed for TBARS values. As expected, TBARS values increased for all treatments (raw and cooked) during the storage (Fig. 4A and B). In general, the treatment CHE showed the highest ($P < .001$) TBARS values throughout the storage, which was evidenced mainly after cooking, once the TBARS values of CHE were about 5 times higher when compared to the control for some periods of storage. These results demonstrate the difficulty of incorporating oils rich in n-3 PUFAs in cooked meat products, due to their high sensitivity to oxidation reactions upon heating (Barros et al., 2020). In general, the JPE treatments showed lower ($P < .001$) TBARS values when compared to CHE during the entire storage period, probably due to the high content of phenolic compounds and the high antioxidant capacity of JPE (Table 1). Several recent studies have also shown that extracts rich in phenolic compounds decrease the lipid oxidation of meat products (Al-Juhaimi et al., 2019; Prommachart et al., 2020; Rodrigues et al., 2020). This fact occurs due to the inhibition of free radical formation caused by donate hydrogen from the phenolic compounds, causing the blocking of the oxidation process (Balasundram, Sundram, & Samman, 2006; Lee, Umano, Shibamoto, & Lee, 2005). No incorporation level of JPE was effective to maintain the oxidative profile similar to the control sample throughout the storage. However, the addition of 10% JPE to HE (T10) was effective to maintain similar lipid oxidation when compared to the control until the 60th day of storage. In addition, this percentage of JPE reduced the TBARS values of cooked burgers when compared to CHE by about 27, 30, 38, 40, and 50% after 0, 30, 60, 90, and 120 days of storage, respectively. A similar trend was observed by Almeida et al. (2015) and Baldin et al. (2016) who reported that the addition of JPE reduced the lipid oxidation of emulsified meat products during the refrigerated storage.

3.2.3. Sensory analysis

The CATA results indicated that consumers were able to discriminate the low-fat burger samples from the burgers with the addition of JPE (Fig. 5). The CA explained 90% of the original data using the first two dimensions, accounting 77.97% and 12.93% in the first and second dimensions, respectively. According to the cluster analysis, four clusters of burger samples distributed in the sensory space were found (Fig. S1).

Table 1
Characterization of jabuticaba peel and extract obtained by MHG.

	Jabuticaba peel	JPE
Phenolic compounds	0.95 mg/g	1.72 mg/mL
ORAC	29.64 μmol ET/g	57,741.67 μmol ET/mL
Microbiological analysis (log CFU/g)		
Mesophilic bacteria	3.14 ± 0.2	0.0
Molds and yeast	2.17 ± 0.1	2.17 ± 0.1

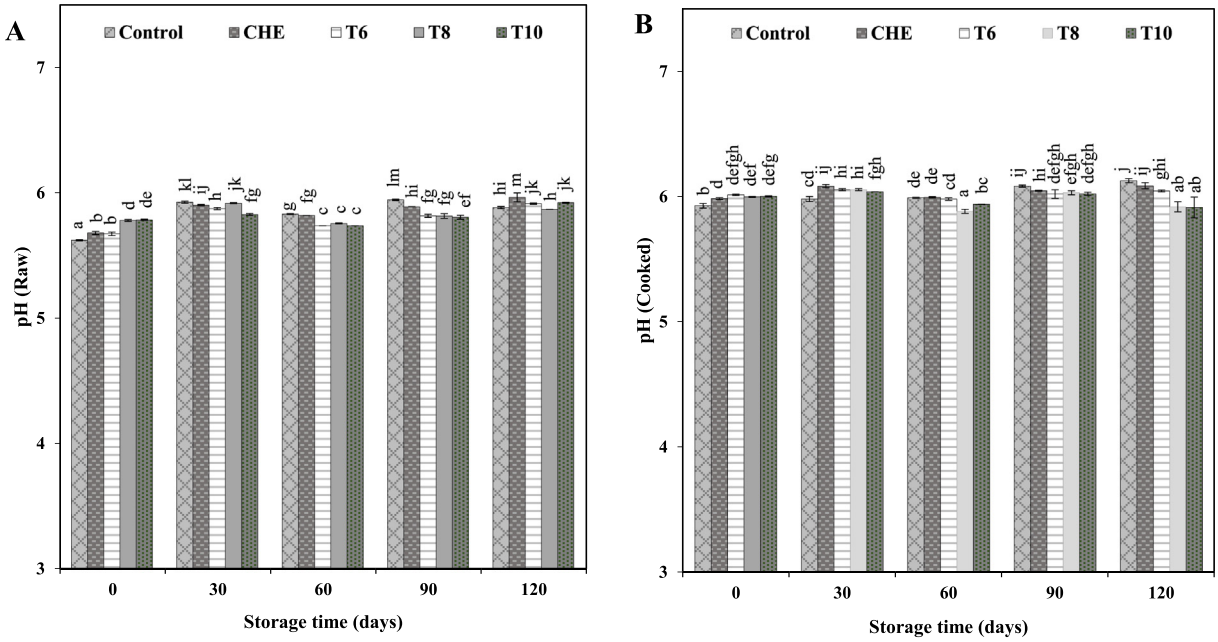


Fig. 1. Overall effects of the partial replacement of pork back fat by HE and storage time on the pH parameters of the beef burgers.
[a–m] Averages followed by the same letter did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

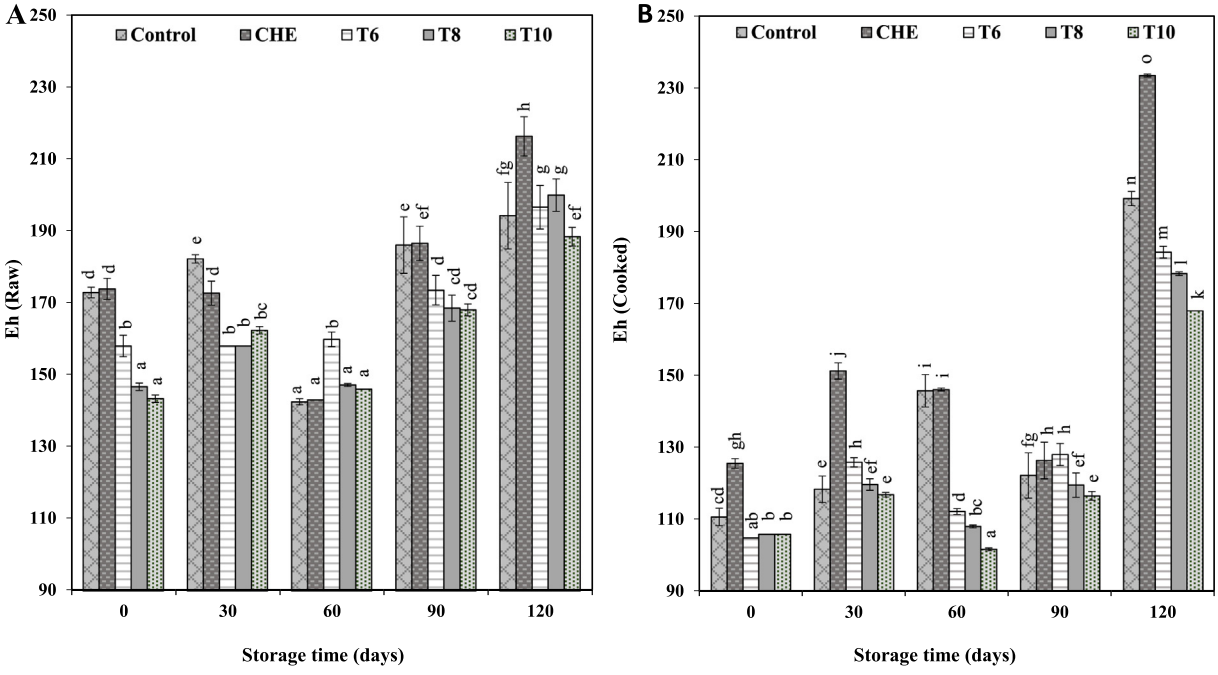


Fig. 2. Overall effects of the partial replacement of pork back fat by HE and storage time on the Eh parameters of the beef burgers.
[a–o] Averages followed by the same letter did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

As shown in Fig. 5, the first cluster contains the sample CHE located in the first quadrant of the sensory map, characterized by negative sensory attributes such as pale color, unpleasant taste, bitter taste, rancid taste, rancid aroma, strange appearance, and rubbery. A similar result was previously reported by Saldaña et al. (2018). According to those authors, mortadella made with healthy pre-emulsions was the least accepted for presenting strange taste and rubbery texture, due to the incorporation of n-3 fatty acids, which are also susceptible to lipid oxidation. The changes in texture were mainly due to a destabilization of the meat matrix due to the protein/water imbalance in the meat batter. In the present study, the burger with 6% JPE was located close to the first cluster (fourth quadrant of the sensory map) that composes the second cluster and presented the following attributes: fruity aroma, sweet taste, dry, and oily appearance. The little incorporation of JPE was enough to considerably improve the sensory profile of the burgers. The samples with the addition of 8% and 10% of JPE (T8 and T10) formed the third cluster and were characterized by the descriptors appreciated by consumers as mild aroma, pleasant taste, and seasoning in the right amount. Finally, the control sample (fourth cluster), which was positioned close to the third cluster, was perceived by consumers as

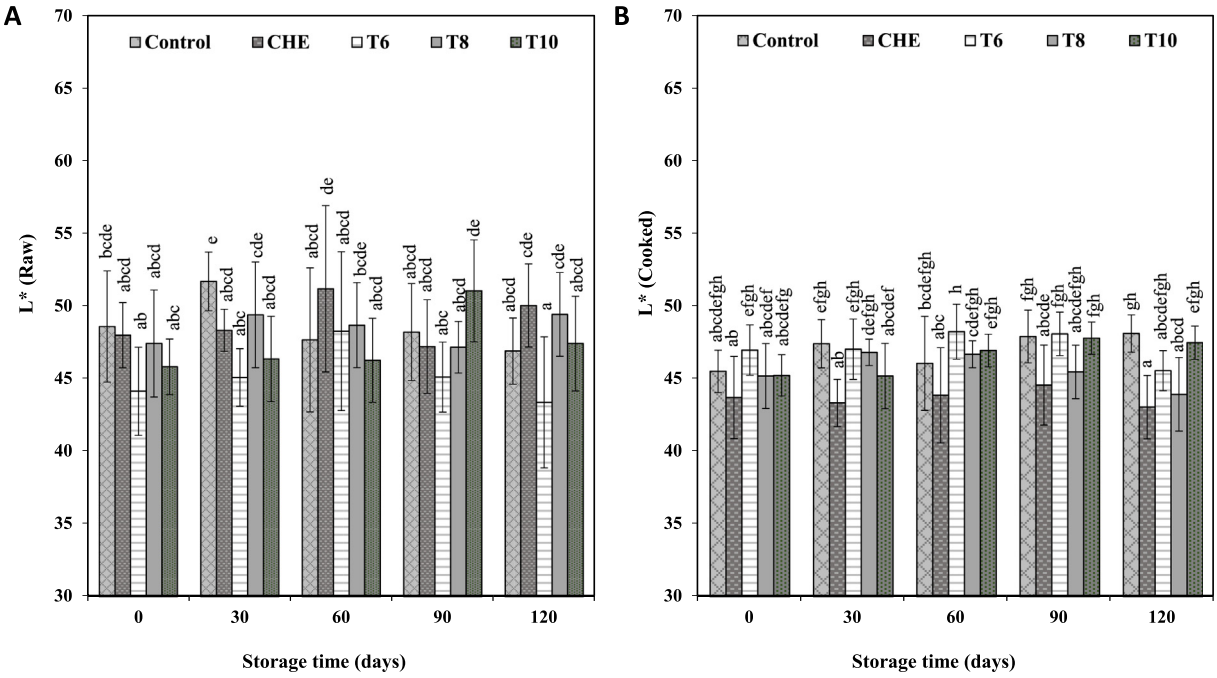


Fig. 3. Overall effects of the partial replacement of pork back fat by HE and storage time on the color parameters of the beef burgers.
[a–j] Averages followed by the same letter did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

ideal color, acidic taste, ideal texture, and succulent. These two clusters exhibited better performance, thus showing that the addition of 8% and 10% JPE was able to inhibit the unpleasant sensory attributes that come mainly from lipid oxidation.

The overall liking of the burgers revealed 3 groups of samples (Fig. 6). The first group, consisting only of the control sample, presented the highest liking, which was expected since it did not undergo any reformulation. The second group with the best liking was the burgers made with the addition of 8% and 10% JPE (T8 and T10), showing that the incorporation of JPE preserved both the oxidative stability and the sensory liking. The last group of samples, with the lowest scores, was composed of the sample without the addition of JPE (CHE) and with the addition of 6% of JPE to HE (T6). These results show the sensory potential of the incorporation of JPE to HE, especially for concentrations greater than 8%.

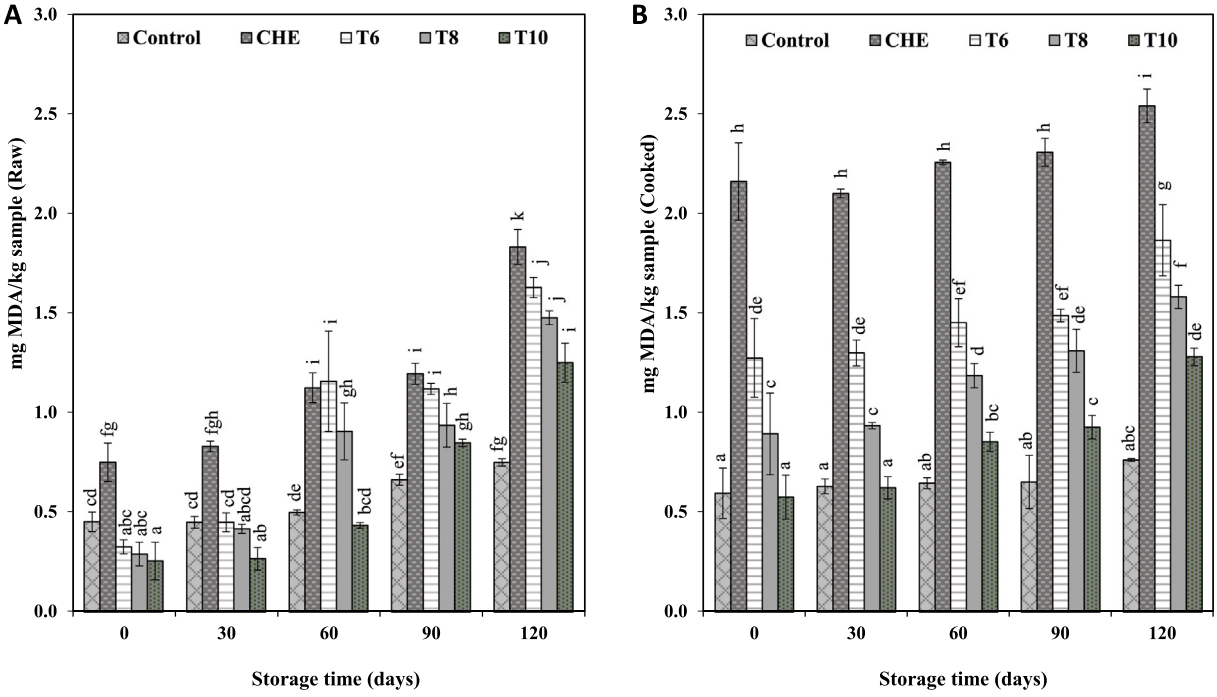


Fig. 4. Overall effects of the partial replacement of pork back fat by HE and storage time on the TBARS parameters of the beef burgers.
[a–k] Averages followed by the same letter did not show any significant difference ($P > .05$) by Tukey test. Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

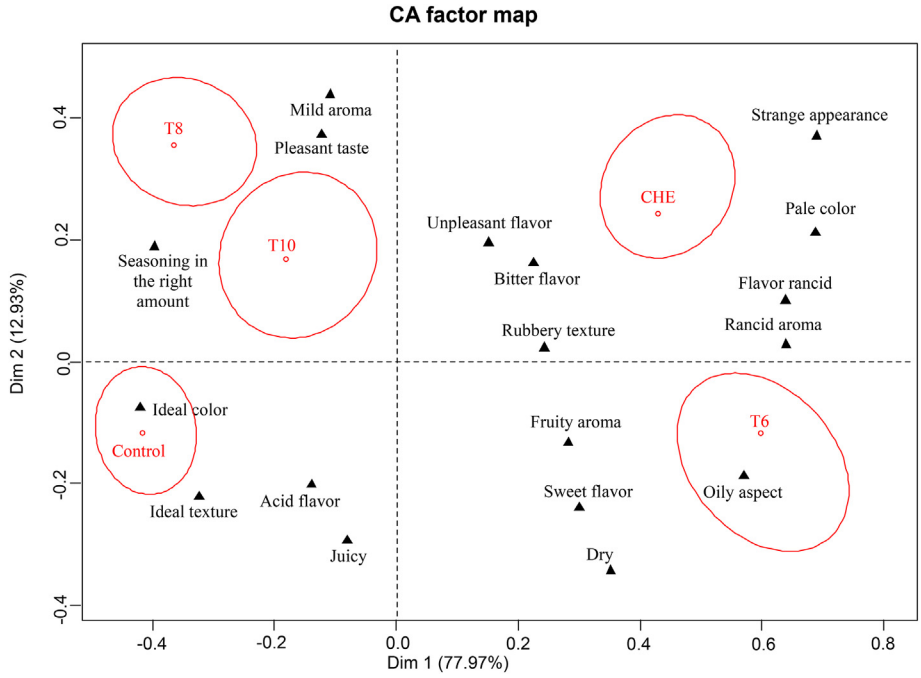


Fig. 5. Representation of the samples and the terms in the first and second dimensions of correspondence analysis performed on CATA results.
Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

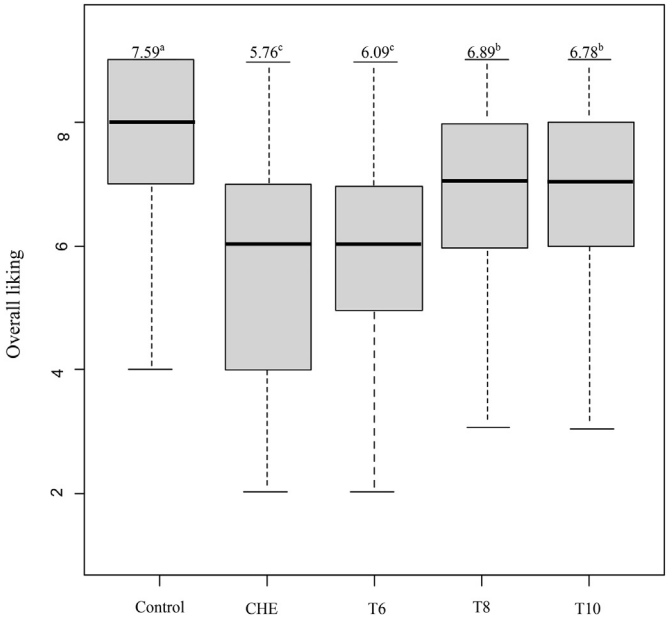


Fig. 6. Averages of the overall liking scores of the beef burgers using a nine-point hedonic scale. Averages followed by the same letter did not show any significant difference ($P > .05$) by Tukey test.
Batches: Control: 20% pork back fat; Reformulated burgers: 60% replacement of pork back fat by HEs containing 0 (CHE), 6 (T6), 8 (T8), and 10% (T10) JPE.

4. Conclusion

The JPE obtained by the MHG extraction was used for the enrichment of HE from chia and linseed oils with bioactive compounds, which was used as a fat replacer in burgers. The MHG extraction was effective to produce JPE rich in phenolic compounds and microbiologically safe. However, further studies are needed to characterize and quantify the phenolic compounds in the JPE.

In addition, the results proved the difficulty of the addition of healthier oils to meat products, once both the oxidative and sensory quality of the burgers were severely affected by the replacement of 60% of pork back fat by HE without the addition of JPE. The addition of 10% JPE to HE minimized these problems as it allowed the production of burgers with good sensory liking and oxidative stability similar to the traditional products during 60 days of storage.

Declaration of Competing Interest

The author have no conflicts of interest for disclose to paper entitled "Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: a green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils".

Acknowledgements

National Council for Scientific and Technological Development (CNPq) (grant number 425577/2018-0), and Coordination for the Improvement of Higher Education Personnel (CAPES finance code 001) have supported this work. P.C·B Campagnol and J.M. Lorenzo are members of the HealthyMeat network, funded by CYTED (ref. 119RT0568).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.meatsci.2020.108230.

References

Al-Juhaimi, F., Babtain, I. A., Ahmed, I. A. M., Alsawmahi, O. N., Ghafoor, K., Adiamo, O. Q., & Babiker, E. E. (2019). Assessment of oxidative stability and physicochemical, microbiological, and sensory properties of beef patties formulated with baobab seed (*Adansonia digitata*) extract. *Meat Science, 162*, Article 108044.

Almeida, P. L., Lima, S. N., Costa, L. L., Oliveira, C. C., Damasceno, K. A., Santos, B. A., & Campagnol, P. C. (2015). Effect of jabuticaba peel extract on lipid oxidation, microbial stability and sensory properties of Bologna-type sausages during refrigerated storage. *Meat Science, 110*, 9–14.

Alves, L. A. A. S., Lorenzo, J. M., Gonçalves, C. A. A., Santos, B. A., Heck, R. T., Cichoski, A. J., & Campagnol, P. C. B. (2016). Production of healthier bologna type sausages

using pork skin and green banana flour as a fat replacers. *Meat Science, 121*, 73–78.
Balasundram, N., Sundram, K., & Samman, S. (2006). Phenolic compounds in plants and Agri-industrial by-products: Antioxidant activity, occurrence, and potential uses. *Food Chemistry, 99*(1), 191–203.
Baldin, J. C., Michelin, E. C., Polizer, Y. J., Rodrigues, I., Godoy, S. H., Fregonesi, R. P., ... Trindade, M. A. (2016). Microencapsulated jabuticaba (Myrciaria cauliflora) extract added to fresh sausage as natural dye with antioxidant and antimicrobial activity. *Meat Science, 118*, 15–21.
Barros, J. C., Munekata, P. E. S., Carvalho, F. A. L., Pateiro, M., Barba, F. J., Domínguez, R., ... Lorenzo, J. M. (2020). Use of tiger nut (Cyperus esculentus L.) oil emulsion as animal fat replacement in beef burgers. *Foods, 9*, 44.
Bodoira, R. M., Penci, M. C., Ribotta, P. D., & Martínez, M. L. (2017). Chia (Salvia hispanica L.) oil stability: Study of the effect of natural antioxidants. *LWT - Food Science and Technology, 75*, 107–113.
Bruna, J. M., Ordonez, J. A., Fernández, M., Herranz, B., & Hoz, L. (2001). Microbial and physico-chemical changes during the ripening of dry fermented sausages superficially inoculated with or having added an intracellular cell-free extract of Penicillium aurantiogriseum. *Meat Science, 59*, 87–96.
Chandra, S., & Mejia, E. G. (2004). Polyphenolic compounds, antioxidant capacity, and quinone reductase activity of an aqueous extract of ardisia compressa in comparison to mate (Ilex paraguariensis) and green (Camellia sinensis) teas. *Journal of Agricultural and Food Chemistry, 52*(11), 3583–3589.
Chaudhary, R., Saadin, K., Bliden, K. P., Harris, W. S., Dinh, B., Sharma, T., & Gurbel, P. A. (2016). Risk factors associated with plasma omega-3 fatty acid levels in patients with suspected coronary artery disease. *Prostaglandins, Leukotrienes and Essential Fatty Acids, 113*, 40–45.
Chemat, F., Rombaut, N., Meullemiestre, A., Turk, M., Perino, S., Fabiano-Tixier, A. S., & Abert-Vian, M. (2017). Review of green food processing techniques. Preservation, transformation, and extraction. *Innovative Food Science and Emerging Technologies, 41*, 357–377.
Comi, G., Tirloni, E., Andyanto, D., Manzano, M., & Iacumin, L. (2015). Use of bio-protective cultures to improve the shelf-life and the sensorial characteristics of commercial hamburgers. *LWT - Food Science and Technology, 62*(2), 1198–1202.
Djordjević, M., Sterniša, M., Možina, S. S., Beszédes, S., Simović, D. S., Hodúr, C., ... Šereš, Z. (2019). Black pepper (Piper nigrum L.) bacterial decontamination by sterilization and microwave treatments. *Review of Faculty of Engineering Analecta Technica Szegedinensia, 13*(2), 1–5.
Faria, M. O., Cipriano, T. M., Cruz, A. G., Santos, B. A., Pollonio, M. A. P., & Campagnol, P. C. B. (2015). Properties of bologna-type sausages with pork back-fat replaced with pork skin and amorphous cellulose. *Meat Science, 104*, 44–51.
Gómez-Estaca, J., Herrero, A. M., Herranz, B., Álvarez, M. D., Jiménez-Colmenero, F., & Cofrades, S. (2019). Characterization of ethyl cellulose and beeswax oleogels and their suitability as fat replacers in healthier lipid pâtés development. *Food Hydrocolloids, 87*, 960–969.
Heck, R. T., Saldaña, E., Lorenzo, J. M., Correa, L. P., Fagundes, M. B., Cichoski, A. J., ... Campagnol, P. C. B. (2019). Hydrogelled emulsion from chia and linseed oils: A promising strategy to produce low-fat burgers with a healthier lipid profile. *Meat Science, 156*, 174–182.
Heck, R. T., Vendruscolo, R. G., Etchepare, M. A., Cichoski, A. J., Menezes, C. R., Barin, J. S., ... Campagnol, P. C. B. (2017). Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science, 130*, 16–25.
ISO- International Organization for Standardization (2013). *Microbiology of food and animal feeding stuffs—Horizontal method for the enumeration of microorganisms. Part 1: Colony count at 30 degrees C by the pour plate technique.* Geneva: International Organization for Standardization (9 p. (ISO 4833-1:2013).
Kryževičūtė, N., Jaime, I., Diez, A. M., Rovira, J., & Venskutonis, P. R. (2017). Effect of raspberry pomace extracts isolated by high pressure extraction on the quality and shelf-life of beef burgers. *International Journal of Food Science and Technology, 52*(8), 1852–1861.
Kusuma, H. S., Putri, D. K. Y., Triesty, I., & Mahfud, M. (2019). Comparison of microwave hydrodistillation and solvent-free microwave extraction for extraction of agarwood oil. *Chiang Mai Journal of Science, 46*(4), 741–755.
Latoch, A., & Stasiak, D. M. (2015). Effect of Mentha piperita on oxidative stability and sensory characteristics of cooked pork sausage. *Journal of Food Processing and Preservation, 39*, 1566–1573.
Lee, S. J., Umano, K., Shibamoto, T., & Lee, K. G. (2005). Identification of volatile components in basil (*Ocimum basilicum L.*) and thyme leaves (*Thymus vulgaris L.*) and their antioxidant properties. *Food Chemistry, 91*(1), 131–137.
Lu, F., Zhang, J. Y., Liu, S. L., Wang, Y., & Ding, Y. T. (2011). Chemical, microbiological and sensory changes of dried Acetes chinensis during accelerated storage. *Food Chemistry, 127*(1), 159–168.
Macfie, H. J., Bratchell, N., Greenhoff, K., & Vallis, L. (1989). Designs to balance the effect of order of presentation and first-order carry-over effects in hall tests. *Journal of Sensory Studies, 4*(2), 129–148.
Menegali, B. S., Selani, M. M., Saldaña, E., Patinho, I., Diniz, J. P., Melo, P. S., ... Contreras-Castillo, C. J. (2020). Pink pepper extract as a natural antioxidant in chicken burger: Effects on oxidative stability and dynamic sensory profile using temporal dominance of sensations. *LWT- Food Science and Technology, 121*, Article 108986.
Ou, B., Hampsch-Woodill, M., & Prior, R. L. (2001). Development and validation of an improved oxygen radical absorbance capacity assay using fluorescein as the fluorescent probe. *Journal Agriculture Food Chemistry, 49*, 4619–4926.
Pignitter, M., Stolze, K., Gartner, S., Dumhart, B., Stoll, C., Steiger, G., & Somoza, V. (2014). Cold fluorescent light as major inducer of lipid oxidation in soybean oil stored at household conditions for eight weeks. *Journal of Agricultural and Food Chemistry, 62*(10), 297–305.
Plaza, M., & Turner, C. (2015). Pressurized hot water extraction of bioactives. *TrAC, Trends in Analytical Chemistry, 71*, 39–54.
Prommachart, R., Belem, T. S., Uriyapongson, S., Rayas-Duarte, P., Uriyapongson, J., & Ramanathan, R. (2020). The effect of black rice water extract on surface color, lipid oxidation, microbial growth, and antioxidant activity of beef patties during chilled storage. *Meat Science, 164*, Article 108091.
Quatrin, A., Pauletto, R., Maurer, L. H., Minuzzi, N., Nichelle, S. M., Carvalho, J. F. C., & Emanuelli, T. (2019). Characterization and quantification of tannins, flavonols, anthocyanins and matrix-bound polyphenols from Jaboticaba fruit peel: A comparison between Myrciaria trunciflora and M. Jaboticaba. *Journal of Food Composition and Analysis, 78*, 59–74.
Rios-Mera, J. D., Saldaña, E., Cruzado-Bravo, M. L. M., Patinho, I., Selani, M. M., Valentin, D., & Contreras-Castillo, C. J. (2019). Reducing the sodium content without modifying the quality of beef burgers by adding micronized salt. *Food Research International, 121*, 288–295.
Rodrigues, A. S., Kubota, E. H., Silva, C. G., Alves, J. S., Hautrive, T. P., Rodrigues, G. S., & Campagnol, P. C. B. (2020). Banana inflorescences: A cheap raw material with great potential to be used as a natural antioxidant in meat products. *Meat Science, 161*, Article 107991.
Rubilar, M., Morales, E., Contreras, K., Ceballos, C., Acevedo, F., Villarroel, M., & Shene, C. (2012). Development of a soup powder enriched with microencapsulated linseed oil as a source of omega-3 fatty acids. *European Journal of Lipid Science and Technology, 114*(4), 423–433.
Saldaña, E., Garcia, A. O., Selani, M. M., Haguiwara, M. M. H., Almeida, M. A., Siche, R., & Contreras-Castillo, C. J. (2018). A sensometric approach to the development of mortadella with healthier fats. *Meat Science, 137*, 176–190.
Saldaña, E., Serrano-León, J., Selani, M. M., & Contreras-Castillo, C. J. (2019). Sensory and hedonic impact of the replacement of synthetic antioxidant for pink pepper residue extract in chicken burger. *Journal of Food Science and Technology, 11*, 1–11.
Selani, M. M., Shirado, G. A. N., Margiotta, G. B., Saldaña, E., Spada, F. P., Piedade, S. M. S., ... Canniatti-Brazaca, S. G. (2016). Effects of pineapple byproduct and canola oil as fat replacers on physicochemical and sensory qualities of low-fat beef burger. *Meat Science, 112*, 69–76.

10. CAPÍTULO 7- LIPID OXIDATION AND SENSORY QUALITY OF OMEGA-3 RICH BUFFALO BURGERS ENRICHED WITH CHLOROGENIC ACIDS FROM THE MATE (ILEX PARAGUARIENSIS) TREE HARVESTING RESIDUES

Rosane Teresinha Heck, Jordana Lima da Rosa, Raquel Guidetti Vendruscolo, Alexandre José Cichoski, Adriana Dillenburg Meinhart, Alexandre Lorini, Bruna Trindade Paim, Vanessa Galli, Silvino Sasso Robalo, Bibiana Alves Dos Santos, Luis Fernando Vilani de Pellegrin, Cristiano Ragagnin de Menezes, Roger Wagner, Paulo Cezar Bastianello Campagnol

Meat Science
Vol. 179, p. 108534, 2021
https://doi.org/10.1016/j.meatsci.2021.108534

Meat Science 179 (2021) 108534

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Lipid oxidation and sensory characterization of Omega-3 rich buffalo burgers enriched with chlorogenic acids from the mate (*Ilex paraguariensis*) tree harvesting residues

Rosane Teresinha Heck [a], Jordana Lima da Rosa [a], Raquel Guidetti Vendruscolo [a], Alexandre José Cichoski [a], Adriana Dillenburg Meinhart [b], Alexandre Lorini [b], Bruna Trindade Paim [b], Vanessa Galli [b], Silvino Sasso Robalo [a], Bibiana Alves Dos Santos [a], Luis Fernando Vilani de Pellegrin [a], Cristiano Ragagnin de Menezes [a], Roger Wagner [a], Paulo Cezar Bastianello Campagnol [a,*]

[a] Universidade Federal de Santa Maria, CEP 97105-900 Santa Maria, Rio Grande do Sul, Brazil
[b] Universidade Federal de Pelotas, CEP 96160-000 Capão do Leão, Rio Grande do Sul, Brazil

ARTICLE INFO

Keywords:
Extract from the bark of mate branches
Natural antioxidant
Healthier meat products
Hydrogelled emulsion
Chia oil

ABSTRACT

A freeze-dried extract from the bark of mate branches (BMBE) containing high chlorogenic acids (CGA) content (30 g 100 g^{-1}) was produced. Then, chia oil was mixed with 7.5% BMBE and sonicated for 0, 10, and 20 min. Chia oil with or without the addition of BMBE was hydrogelled and used to produce buffalo burgers with 50% reduction in animal fat. CGA levels and the nutritional, oxidative, and sensory properties of the burgers were analyzed. A reduction of ~30% fat and an increase above 60% PUFA/SFA ratio was observed for the reformulated raw and cooked burgers. In addition, the Omega-6/Omega-3 PUFA ratio of the burgers decreased from 20.8 (raw) and 31.9 (cooked) to values lower than 2. The addition of BMBE enriched the burgers with CGA, preventing an increase in lipid oxidation caused by chia oil. The addition of BMBE-enriched hydrogelled chia oil not subjected to sonication did not affect the sensory properties of the burgers.

1. Introduction

Buffalo meat can be an interesting alternative for people looking for a healthier diet, as it contains low fat, cholesterol, and saturated fatty acids (SFA) levels, in addition to being rich in high biological value proteins, iron, zinc, and monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids (Kandeepan, Mendiratta, Shukla, & Vishnuraj, 2013; Tamburrano et al., 2019). Therefore, the use of this raw material to produce highly consumed meat products, such as burgers, can bring nutritional benefits to the population.

The partial substitution of saturated fat for healthy oils is another interesting approach to make healthier burgers. In a previous study, our research group used a hydrogelled emulsion (HE) rich in Omega-3 PUFAs to produce burgers with 50% fat reduction. The strategy was effective in reducing fat and improving the lipid profile of burgers without impairing their sensory and technological quality (Heck et al., 2019). However, the hydrogelling process was not effective to fully protect Omega-3 PUFAs from the oxidative reactions. To solve this problem, our team evaluated the incorporation of bioactive compounds from the jaboticaba bark into HE. This approach was useful to keep the TBARS values of the burgers below the sensory threshold during 90 days of frozen storage (−18 °C) (Heck et al., 2020).

Thus, those results showed that the combination of Omega-3 PUFAs and bioactive compounds may be an efficient approach for the production of healthier and safer meat products. However, further studies are necessary to find low-cost and abundant sources of natural antioxidants. In this context, knowledge of the antioxidant effect of the residues from harvesting of plants with high bioactive compounds levels may be an interesting alternative.

Yerba mate or mate (*Ilex paraguariensis A. St. Hil.*) is a plant of great socio-economic importance in several countries, including Brazil, Argentina, Uruguay, and Paraguay. Its cultivation covers approximately 540,000 km^2, which corresponds to about 3% of the territory of South America (Jacques, Santos, Dariva, Oliveira, & Caramarão, 2007). Mate

* Corresponding author.
E-mail address: paulo.campagnol@ufsm.br (P.C.B. Campagnol).

https://doi.org/10.1016/j.meatsci.2021.108534
Received 3 February 2021; Received in revised form 22 March 2021; Accepted 3 May 2021
Available online 6 May 2021

leaves are the main product obtained in the cultivation of yerba mate, as they are used for the preparation of regional beverages such as “mate tea”, “chimarrão”, and “tererê”. However, approximately 5 tons/ha of solid waste is improperly discarded in the soil during harvesting, which is mainly made up of mate branches over 10 mm in diameter (Mendes & Carlini, 2007).

It is well documented in the literature that the yerba mate leaves have high contents of methylxanthines and polyphenols (Esmelindro, Girardi, Mossi, Jacques, & Dariva, 2004; Prediger et al., 2008). In addition, the beneficial antioxidant effect of the extracts from yerba mate leaves has already been proven in meat products (Campos et al., 2007; Racanicci, Danielsen, & Skibsted, 2008). However, studies evaluating the antioxidant potential of extracts of residues from the yerba mate harvest are scarce. Pagliosa et al. (2010) analyzed bark of mate branches extracts and reported a high antioxidant activity and a higher chlorogenic acid (CGA) concentration when compared with extracts from yerba mate leaves.

Given the above, we hypothesized that the extracts from the bark of mate branches (BMBE) may be a low-cost strategy to control the lipid oxidation in Omega-3 rich buffalo burgers. In addition, according to our previous results (Heck et al., 2018; Heck et al., 2019), we also hypothesized that ultrasound-assisted extraction (UAE) may be useful to increase the incorporation of bioactive compounds from BMBE into chia oil. Thus, in this study, chia oil was mixed with a lyophilized BMBE and the mixture was sonicated for 0, 10, and 20 min in an ultrasonic bath. The BMBE rich oil was hydrogelled and used to produce buffalo burgers with 50% reduction in animal fat. The CGA levels, nutritional profile, oxidative stability and sensory properties of buffalo burgers were analyzed.

2. Materials and methods

2.1. Elaboration of the bark of mate branches extract (BMBE)

Branches from the residue from yerba mate harvesting (*Ilex paraguariensis*, cultivar Cambona 4, planted in full sun) were kindly donated by *Associação de Produtores de Erva Mate de Machadinho*, RS (Brazil). Branches were collected in August 2019, in the municipality of Machadinho/RS (27°32′35.5” S and 51°39′46.9” W). After being manually removed from the branches, the bark tissue was subjected to drying in an oven at 135 °C for 40 min and ground to 20 mesh. The dry bark was vacuum-packed and stored frozen (−18 °C) until the extraction procedures. To produce the extract, 75 g of ground bark and 1500 mL of ethanol: water (41:59) were placed in a double-wall reactor coupled to a thermostatic bath with water circulation at 85 °C for 34 min, with a magnetic stirrer. Then, the BMBE was cooled (4 °C) and vacuum filtered using a PVDF membrane filter with 0.22 μm. The extract was freeze-dried, and the powder was stored under vacuum at −18 °C until use. For verification purposes, the botanical identification of the samples was carried out through deposit in a herbarium at the Institute of Biology of the Federal University of Pelotas, under number 26,978.

2.2. Determination of CGA in BMBE

The freeze-dried BMBE (100 mg) and 10 mL of ethanol: water (43:57) were mixed, vortexed, appropriately diluted, filtered, and injected into the chromatograph. The determination of CGA in BMBE was carried out in triplicate according to the methodology of Meinhart et al. (2017). The analyses were performed in a high-performance liquid chromatography with a diode array detector equipped with a quaternary pump and an automatic injector (UFLC-DAD, Shimadzu, Japan) operating at 325 nm. A Cogent 2.0 Bidentate C18 column (MicroSolv Technology Corp., Leland, NC, USA) 2.2 μm × 100 mm × 2.1 mm ID was used.

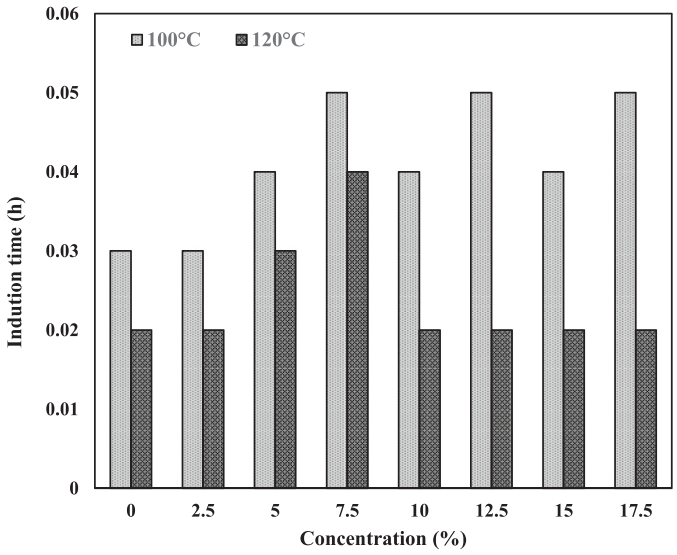


Fig. 1. Induction time of chia oil enriched with bark of mate branches extract (BMBE).

2.3. Accelerated oxidation test in chia oil with the addition of BMBE

BMBE was added to chia oil in the concentrations of 2.5, 5, 7.5, 10, 12.5, 15, and 17.5%. The induction time was measured in triplicate using a Rancimat (model 892, Metrohm, Herisau, Switzerland) operating at 120 °C, and with an airflow rate of 20 L/h. As can be seen in Fig. 1, a longer induction time was observed for the concentration starting from 7.5% of BMBE. Thus, the concentration of 7.5% BMBE was selected to be incorporated into chia oil, which was later hydrogelled and used in burger formulations.

2.4. Ultrasound application in chia oil with the addition of BMBE and preparation of hydrogelled emulsions

Chia oil (100 g) and freeze-dried BMBE (7.5 g) were mixed vigorously with a glass stirring rod for 30 s. Then, the mixtures were sonicated for 0, 10, and 20 min in an ultrasonic bath (TI-H-10, Elma, Germany) operating at 1 W cm^{-2}, sweep mode, 25 kHz, temperature 30 ± 1 °C, and 100% amplitude. The chia oil enriched with BMBE was hydrogelled using the technique developed by Heck, Saldaña, et al. (2019). A hydrogelled emulsion (HE) with chia oil without the incorporation of BMBE was also prepared as a control.

2.5. Buffalo burger formulations

The Control was made with buffalo meat (78.4%), pork back fat (20%), salt (1.5%), and garlic (0.1%). CHE (Control hydrogelled emulsion) treatment was produced with 50% replacement of pork back fat by HE made with chia oil without the incorporation of BMBE. T1, T2, and T3 treatments were produced with 50% replacement of pork back fat by HE made with chia oil enriched with 7.5% BMBE and sonicated for 0, 10, and 20 min, respectively. The burger manufacture was performed according to procedures described by Heck, Saldaña, et al. (2019). A high-density polyethylene bags (20 × 14 cm) with 50 μm thick was used to packed the raw burgers that were stored at −18 °C for 120 days. After 1, 30, 60, 90, and 120 days of storage, the raw burgers were thawed at 4 °C for 12 h and cooked until the internal temperature of 72 °C in an electric grill (Multi Grill, Britânia, Brazil) preheated to 150 °C.

Table 1
Effect of the 50% replacement of pork back fat by hydrogelled emulsion from chia oil on the chemical composition of the raw and cooked buffalo burgers.

(%)	Raw							Cooked						
	Control	CHE	T1	T2	T3	SEM	Sig.	Control	CHE	T1	T2	T3	SEM	Sig.
Moisture	63.01^{b}	68.22^{a}	66.0^{ab}	67.3^{a}	66.02^{ab}	0.48	*	52.49^{d}	56.55^{ab}	58.42^{a}	54.62^{c}	54.68^{bc}	0.38	***
Protein	19.03^{a}	18.14^{a}	18.0^{a}	18.6^{a}	18.67^{a}	0.18	n.s.	24.74^{a}	25.33^{a}	24.73^{a}	24.28^{a}	25.37^{a}	0.23	n.s.
Fat	19.85^{a}	12.29^{b}	12.4^{b}	12.8^{b}	12.30^{b}	0.51	***	15.07^{a}	10.48^{b}	10.49^{b}	10.61^{b}	10.48^{b}	0.34	***
Ash	1.98^{a}	2.15^{a}	2.01^{a}	2.23^{a}	2.58^{a}	0.09	n.s.	2.82^{a}	2.16^{a}	2.27^{a}	2.46^{a}	2.25^{a}	0.08	n.s.

$^{a-d}$Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < 0.05$) by Tukey test.
Batches: Control: 20% pork back fat; CHE: 50% replacement of pork back fat by hydrogelled emulsion from chia oil; T1, T2 and T3: 50% replacement of pork back fat by hydrogelled emulsion from chia oil sonicated with 7.5% bark of mate branches extract during 0, 10 and 20 min, respectively.
SEM: standard error of the mean
Sig.: significance: *** ($P < 0.001$). ** ($P < 0.01$). * ($P < 0.05$). n.s. (not significant).

2.6. Characterization of the reformulated burgers

2.6.1. Chemical composition and fatty acid profile

The chemical composition and the fatty acid profile were determined in the raw material, raw burgers, and cooked burgers at the beginning of storage (day 1), in triplicate. The moisture, protein, and ash contents were quantified in triplicate using the methodologies of AOAC (2005). The lipids of the burger samples were extracted according to the methodology of Bligh and Dyer (1959), in triplicate. Part of the organic fraction was used to determine the fat content by gravimetric analysis, after solvent evaporation in an air circulation oven at 105 °C. The rest of the organic fraction was derivatized as suggested by Hartman and Lago (1973) to obtain Fatty Acid Methyl Esters (FAME). The FAMEs were analyzed in triplicate using the methodology described by Heck et al. (2017). A Gas Chromatograph equipped with Flame Ionization Detector (GC-FID) (Varian, Star 3600, USA) and autosampler (Varian, 8200, USA) was used. The % of the total area taking into account the carbon chain length of FAME and the conversion factor of methyl ester to fatty acid for correction of FID response was used to express the results (Visentainer, 2012).

2.6.2. Quantification of CGA in burgers

The chlorogenic acids contents of the burgers were determined soon after the manufacture and at the end of storage (day 120), in triplicate. The sample was prepared using the methodology of Ballus et al. (2014), with adaptations. For that, sample (1.5 g) was weighed in Falcon tubes of 50 mL capacity, and 10 mL of ethanol: water (43:57), 10 mL of hexane, and 50 μL of saturated trichloroacetic acid were added. After being vortexed for 30 s, the tubes were centrifuged at 5000 rpm for 20 min at 20 °C. The polar fraction was collected, filtered with a 0.22 μm PVDF membrane filter and the CGA content was analyzed as described in Section 2.2.

2.6.3. TBARS assay

TBARS values (mg of malonaldehyde/kg of sample) were determined in raw and cooked burgers after 1, 30, 60, 90, and 120 days of storage, in triplicate, as described by Bruna, Ordonez, Fernández, Herranz, and Hoz (2001).

2.6.4. Determination of pH

Five grams of sample were mixed with deionized water (50 mL) for 60 s using a glass stirring rod. The pH values of the burgers (raw and cooked) were determined in triplicate during the frozen storage (1, 30, 60, 90, and 120 days) using a digital pH meter (DM23-DC, Digimed, Brazil).

2.6.5. Color measurements

The instrumental color (L*, a* and b*) of the burgers (raw and cooked) was determined during the frozen storage (1, 30, 60, 90, and 120 days) using a colorimeter (CR-400 Model, Minolta Sensing Inc. Konica, Japan) with a D65 illuminant and spectral reflectance included as calibration mode. A 10° observation angle and the CIE system (L* a* b*) were used in the color analysis. L*, a* and b* values were measured at six points on each side of three samples per treatment.

2.7. Sensory evaluation

Sensory evaluation was performed in individual booths at the sensory analysis laboratory at the Federal University of Santa Maria (RS, Brazil). The samples were encoded with 3 random numbers and served to the assessors in sequential monadic form, as described by Macfie, Bratchell, Greenhoff, and Vallis (1989). After cooking the burgers (Section 2.5), the samples were cut (4 × 4 × 2.5 cm) and wrapped in aluminum foil. The samples were kept at 60 °C in a conventional oven until served to consumers. One hundred regular burger consumers (61% female and 39% male, aged between 18 and 55 years) participated in the sensory evaluation.

The sensory tests were performed right after the manufacture of the burgers (day 1 of storage). For the acceptance test, consumers were asked to rate their acceptance in relation to color, aroma, flavor, texture, and liking parameters (Meilgaard, Carr, & Civille, 2006) using a nine-point hedonic scale (1: disliked very much; 9: liked very much) (Stone, Bleibaum, & Thomas, 2012). For the CATA questionnaire, 21 previously selected descriptors were evaluated (Da Silva et al., 2019; Heck, Fagundes, et al., 2019). For the appearance of the samples, the descriptors pale color, ideal color, oily aspect, and strange appearance were evaluated. For aroma, the descriptors were rancid aroma, mild aroma, herbal aroma, fish aroma, pleasant aroma, and fat aroma. The flavor-related descriptors were rancid taste, pleasant taste, bitter taste, acid taste, unpleasant taste, and fish flavor. Finally, the texture descriptors were ideal texture, juicy, rubbery, hard, and dry texture.

2.8. Statistical analysis

We replicated the entire experiment three times ($n = 3$). A generalized linear model (ANOVA) was used to analyze the physicochemical results. The variables "treatments" and "storage time", and the interaction "treatments*storage time" were considered as a fixed effect, and repetitions were considered as a random effect. Tukey's test ($P < 0.05$) was used for posthoc comparison of the averages.

The sensory acceptance data were analyzed using a mixed linear model. The treatments were considered as a fixed effect and the consumers as a random effect. Tukey's test ($P < 0.05$) was used for posthoc comparison of the averages. CATA results were analyzed using a correspondence analysis and the correlation between the CATA descriptors and the liking scores was evaluated using a principal coordinate analysis. The instrumental and sensory data were analyzed using the XLStat statistical program.

Table 2
Effect of the 50% replacement of pork back fat by hydrogelled emulsion from chia oil on the fatty acids profile (expressed as g/100 g of fatty acids) of the raw and cooked buffalo burgers.

	Raw							Cooked						
	Control	CHE	T1	T2	T3	SEM	Sig.	Control	CHE	T1	T2	T3	SEM	Sig.
C10:0	0.014^{a}	0.012^{bc}	0.011^{c}	0.013^{ab}	0.012^{bc}	0.00	***	0.008^{ab}	0.007^{ab}	0.002^{b}	0.012^{a}	0.010^{a}	0.00	**
C12:0	0.019^{a}	0.013^{d}	0.014^{cd}	0.016^{b}	0.016^{bc}	0.00	***	0.008^{a}	0.012^{a}	0.008^{a}	0.010^{a}	0.011^{a}	0.00	n.s
C14:0	0.84^{a}	0.57^{c}	0.73^{b}	0.65^{bc}	0.66^{bc}	0.02	***	0.77^{a}	0.52^{c}	0.68^{b}	0.69^{b}	0.65^{b}	0.01	***
C15:0	0.045^{b}	0.047^{b}	0.067^{a}	0.058^{ab}	0.061^{a}	0.00	***	0.017^{a}	0.025^{a}	0.019^{a}	0.031^{a}	0.027^{a}	0.00	n.s
C16:0	21.50^{a}	18.84^{c}	19.79^{b}	18.89^{c}	19.42^{bc}	0.17	***	22.36^{a}	21.54^{ab}	21.31^{b}	21.31^{b}	21.39^{ab}	0.12	*
C16:1	1.57^{a}	1.01^{b}	1.06^{b}	1.12^{b}	1.10^{b}	0.03	***	1.57^{a}	0.94^{c}	1.15^{b}	1.14^{b}	1.21^{b}	0.03	***
C17:0	0.310^{b}	0.277^{b}	0.369^{a}	0.327^{ab}	0.326^{ab}	0.01	***	0.169^{a}	0.177^{a}	0.173^{a}	0.182^{a}	0.192^{a}	0.01	n.s
C18:0	15.32^{ab}	13.80^{c}	16.13^{a}	14.49^{bc}	15.06^{abc}	0.20	***	13.99^{ab}	13.43^{ab}	14.11^{a}	13.96^{ab}	13.31^{b}	0.09	*
C18:1	0.30^{d}	0.66^{c}	1.16^{a}	0.92^{b}	0.93^{b}	0.05	***	0.09^{b}	0.22^{a}	0.14^{b}	0.21^{a}	0.15^{ab}	0.01	***
C18:1n9c	43.93^{a}	37.27^{b}	37.31^{b}	37.96^{b}	37.96^{b}	0.39	***	46.05^{a}	39.87^{b}	40.58^{b}	40.59^{b}	40.18^{b}	0.37	***
C18:1t	2.32^{a}	1.72^{cd}	1.58^{d}	1.86^{b}	1.76^{bc}	0.04	***	1.99^{a}	1.59^{c}	1.58^{c}	1.63^{bc}	1.71^{b}	0.03	***
C18:2n6t	0.029^{c}	0.032^{bc}	0.047^{a}	0.037^{abc}	0.041^{ab}	0.00	***	0.011^{a}	0.017^{a}	0.015^{a}	0.030^{a}	0.014^{a}	0.00	n.s.
C18:2n6c	11.80^{b}	12.89^{a}	11.47^{b}	12.38^{ab}	11.77^{b}	0.14	***	11.81^{b}	12.14^{a}	12.06^{ab}	11.89^{ab}	11.81^{b}	0.04	**
C18:3n6	0.13^{ab}	0.12^{bc}	0.10^{c}	0.14^{a}	0.10^{c}	0.00	***	0.04^{a}	0.04^{a}	0.04^{a}	0.06^{a}	0.06^{a}	0.01	n.s
C18:3n3	0.47^{c}	11.62^{a}	9.15^{b}	9.89^{b}	9.67^{b}	0.61	***	0.33^{d}	9.06^{a}	7.72^{c}	7.53^{c}	8.61^{b}	0.49	***
C20:1n9	0.54^{a}	0.35^{c}	0.33^{c}	0.43^{b}	0.36^{c}	0.01	***	0.37^{a}	0.18^{b}	0.23^{b}	0.27^{ab}	0.27^{ab}	0.02	**
CLA (9c11t)	0.067^{bc}	0.061^{c}	0.078^{ab}	0.087^{a}	0.079^{ab}	0.00	***	0.015^{a}	0.016^{a}	0.010^{a}	0.029^{a}	0.019^{a}	0.00	n.s.
CLA (8t10c)	0.016^{a}	0.010^{b}	0.011^{b}	0.014^{ab}	0.016^{a}	0.00	***	0.007^{b}	0.009^{ab}	0.005^{b}	0.032^{a}	0.009^{ab}	0.00	*
CLA (8t10t + 9t11t + 10t12t)	0.022^{b}	0.029^{ab}	0.026^{ab}	0.032^{a}	0.028^{ab}	0.00	***	0.016^{a}	0.023^{a}	0.021^{a}	0.039^{a}	0.032^{a}	0.00	n.s.
C20:2n6	0.32^{a}	0.19^{c}	0.18^{c}	0.24^{b}	0.19^{c}	0.01	***	0.16^{a}	0.06^{b}	0.07^{b}	0.11^{ab}	0.09^{ab}	0.01	*
C20:3n6	0.05^{a}	0.06^{a}	0.04^{b}	0.07^{a}	0.06^{a}	0.00	***	0.024^{a}	0.022^{a}	0.022^{a}	0.030^{a}	0.024^{a}	0.00	n.s.
C20:3n3	0.037^{a}	0.032^{ab}	0.027^{b}	0.036^{a}	0.035^{a}	0.00	***	0.015^{ab}	0.016^{ab}	0.008^{b}	0.031^{a}	0.014^{ab}	0.00	*
C20:4n6	0.171^{ab}	0.187^{a}	0.130^{c}	0.170^{ab}	0.156^{bc}	0.00	***	0.086^{a}	0.043^{a}	0.038^{a}	0.064^{a}	0.084^{a}	0.01	n.s.
C24:1n9	0.051^{a}	0.038^{b}	0.032^{c}	0.038^{b}	0.035^{bc}	0.00	***	0.019^{a}	0.020^{a}	0.021^{a}	0.048^{a}	0.019^{a}	0.01	n.s.
C22:5n3	0.081^{b}	0.092^{ab}	0.089^{ab}	0.102^{a}	0.090^{ab}	0.00	***	0.025^{a}	0.020^{a}	0.018^{a}	0.050^{a}	0.033^{a}	0.00	n.s.
∑SFA	38.06^{a}	33.57^{d}	37.12^{ab}	34.45^{cd}	35.57^{bc}	0.33	***	37.33^{a}	35.72^{bc}	36.31^{b}	36.20^{bc}	35.60^{c}	0.11	***
∑MUFA	48.90^{a}	41.26^{c}	41.63^{bc}	42.51^{b}	42.32^{b}	0.44	***	50.19^{a}	42.87^{c}	43.73^{bc}	43.97^{b}	43.66^{bc}	0.41	***
∑PUFA	13.03^{c}	25.16^{a}	21.25^{b}	23.03^{ab}	22.11^{b}	0.68	***	12.47^{c}	21.40^{a}	19.94^{b}	19.82^{b}	20.74^{a}	0.50	***
PUFA/SFA	0.34^{c}	0.75^{a}	0.57^{b}	0.67^{ab}	0.62^{b}	0.02	***	0.34^{c}	0.59^{a}	0.55^{b}	0.55^{b}	0.58^{a}	0.01	***
n-3	0.59^{c}	11.75^{a}	9.28^{b}	10.03^{b}	9.80^{b}	0.61	***	0.37^{d}	9.08^{a}	7.74^{c}	7.61^{c}	8.66^{b}	0.49	***
n-6	12.34^{ab}	13.31^{a}	11.86^{b}	12.87^{ab}	12.19^{b}	0.14	***	12.05^{a}	12.27^{a}	12.18^{a}	12.13^{a}	12.01^{a}	0.04	n.s.
n-6/n-3	20.85^{a}	1.14^{b}	1.28^{b}	1.28^{b}	1.25^{b}	1.18	***	31.91^{a}	1.35^{b}	1.57^{b}	1.59^{b}	1.39^{b}	1.84	***
AI	0.40^{a}	0.31^{d}	0.36^{b}	0.33^{cd}	0.34^{bc}	0.01	***	0.40^{a}	0.36^{b}	0.37^{b}	0.37^{b}	0.37^{b}	0.00	***
TI	1.16^{a}	0.52^{d}	0.66^{b}	0.58^{cd}	0.61^{bc}	0.04	***	1.15^{a}	0.63^{c}	0.69^{b}	0.69^{b}	0.65^{c}	0.03	***

[a-d] Mean values in the same row (corresponding to the same parameter) not followed by a common letter differ significantly ($P < 0.05$) by Tukey test.
Batches: Described in Table 1.
SEM: standard error of the mean
Sig.: significance: *** ($P < 0.001$). ** ($P < 0.01$). * ($P < 0.05$). n.s. (not significant).
SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; n-6 = omega-6; n-3 = omega-3. AI: atherogenic index; TI: thrombogenic index.

3. Results and discussion

3.1. Chlorogenic acids (CGA) content of BMBE

Five CGA compounds were quantified in the BMBE, as follows: 5-CQA, 3-CQA + 4-CQA, 3,4-DQA, 3,5-DQA, and 4,5-DQA (Table 3). The total CGA content of BMBE was 30 ± 0.75 g 100 g^{-1}. The CQA and DQA isomers in BMBE corresponded to 61.2% and 38.8% of total CGA, respectively. A similar outcome was reported by Marques and Farah (2009), who reported that CQA isomers were the majority CGA in green and toasted leaves of yerba mate. Regarding the individual CQA isomers, 3-CQA + 4-CQA were the major compounds (about 61% of total CQA and 37% of total CGA). As reported by Lima, Farah, King, De Paulis, & Martin, 2016, the content of these compounds increases when subjected to heat due to isomerization reactions. Concerning the DQA isomers, 3,5-DQA and 4,5-DQA were the major compounds (about 46 and 45% of total DQA and 18 and 17% of total CGA, respectively), followed by 3,4-DQA (about 8% of total CQA and 3% of total CGA). These results are in accordance with Filip, López, Giberti, Coussio, and Ferraro (2001); Filip, Lotito, Ferraro, and Fraga (2000) who reported similar behavior in leaves of *Ilex paraguariensis*.

Table 3
Chlorogenic acids (CGA) content of the lyophilized bark of mate branches extract.

Chlorogenic acids	(g 100 g^{-1})	Retention time (min)
5-CQA (5-caffeoylquinic acid)	7.16 ± 0.12	8.59
3-CQA + 4-CQA (3-caffeoylquinic acid +4-caffeoylquinic acid)	11.19 ± 0.38	10.06
3,4-DQA (3,4-dicaffeoylquinic acid)	1.00 ± 0.02	13.66
3,5-DQA (3,5-dicaffeoylquinic acid)	5.39 ± 0.13	13.93
4,5-DQA (4,5-dicaffeoylquinic acid)	5.25 ± 0.12	14.4
Total CGA	30.00	

Results are shown as mean values ± standard deviation (n = 3).

3.2. Nutritional quality of burgers

The chemical composition values (moisture, protein, fat, and ash) of burgers are shown in Table 1. The lipid reformulation did not affect the protein and ash contents of raw and cooked burgers ($P > 0.05$). The raw burgers from HE treatments (CHE, T1, T2, and T3) showed a decrease of about 37% fat in relation to the Control, which was close to 30% after cooking. In addition, raw and cooked burgers from HE treatments showed higher (Raw: $P < 0.05$; Cooked: $P < 0.001$) moisture content than the Control (except T1 and raw T3).

Table 4
Results of consumer study of buffalo burger formulations.

	Batches					SEM	SIG
	Control	CHE	T1	T2	T3		
Color	6.42^{ab}	6.72^{a}	5.84^{b}	5.84^{b}	5.82^{b}	0.09	***
Aroma	6.72^{a}	6.53^{ab}	6.09^{abc}	5.88^{bc}	5.47^{c}	0.09	***
Flavor	7.26^{a}	6.75^{ab}	6.75^{ab}	6.31^{bc}	5.76^{c}	0.09	***
Texture	7.08^{a}	7.23^{a}	7.15^{a}	6.73^{a}	6.81^{a}	0.07	n.s.
Liking	7.11^{a}	6.96^{ab}	6.86^{ab}	6.46^{bc}	6.03^{c}	0.07	***

$^{a-c}$ Mean values in the same row not followed by a common letter differ significantly ($P < 0.05$).
Batches: Described in Table 1.
SEM: standard error of the mean
Sig.: significance: *** ($P < 0.001$), n.s. (not significant).

The fatty acid profile of burgers is presented in Table 2. Palmitic acid (16:0) and stearic acid (18:0) were the SFAs found in greater amounts in the lipid fraction of burgers. Oleic acid (C18:1n9c) and linoleic acid (C18:2n6c) were the main MUFAs and PUFAs, respectively. The Control showed higher SFAs ($P < 0.001$) and MUFAs ($P < 0.001$) levels and the reformulated burgers presented higher PUFAs levels ($P < 0.001$). As expected, the pork back fat replacement by HE from chia oil increased the Omega-3 PUFAs of the lipid fraction of the burgers ($P < 0.001$). In addition to increasing the PUFA/SFA ratio, the lipid reformulation reduced the Omega-6/Omega-3 ratio, and the thrombogenicity and atherogenicity indices, improving the healthiness of the product (Chaudhary et al., 2016; Simopoulos, 2011).

The results of the chemical composition and the fatty acid profile confirmed that the lipid reformulation gave buffalo burgers healthier characteristics, allowing to label the products with health claims, such as “high in protein”, “reduced in fat” and “high in omega-3 fatty acids” (European Parliament, 2006).

3.3. Chlorogenic acids (CGA) content of burgers

The interaction between the treatments and the storage time significantly affected the CGA levels of burgers ($P < 0.001$) (Fig. 2). As expected, CGA was not detected in the Control and CHE. The treatments T1, T2, and T3 showed the same CGA compounds identified in BMBE. The total CGA levels of T1, T2, and T3 at the beginning of storage (day 1) were 19.84, 23.64, and 21.96 mg.100 g^{-1} in raw burgers, and 22.14, 22.97, and 21.59 mg.100 g^{-1} in cooked burgers. At the end of storage (120 days), the total CGA levels decreased by 35, 33, and 35% in raw burgers and 19, 15, and 9% in cooked burgers from the treatments T1, T2, and T3, respectively. CGA is greatly recognized as an effective free radical scavenger (Clifford, 1999). Thus, the reduction during the storage was probably due to the fact that CGA reacted with free radicals forming compounds of low-energy that do not rapidly promote the oxidation of MUFAs and PUFAs (Sasaki et al., 2010). A decrease in total CGA levels during the storage of sausages enriched with carrot powder has also been reported by Alvarado-Ramírez et al. (2018).

In addition to CGA, the addition of BMBE can also add other compounds that are present in the plant, such as methylxanthines (caffeine, for example), flavonoids and saponins (Mateos, Baeza, Sarriá, & Bravo, 2018; Puangpraphant, Berhow, & de Mejia, 2011). Therefore, for BMBE to be used in commercial burgers, it is necessary to quantify these biological components to inform the consumers.

3.4. TBARS assay

TBARS values of burgers (raw and cooked) were significantly ($P < 0.001$) influenced by interaction between the treatments and storage period (Fig. 3A and B). All treatments showed an increase in TBARS values during storage ($P < 0.001$). The lowest TBARS values during the storage period were found in the Control samples, probably due to the lower PUFA/SFA ratio (about 50% lower) of the Control when compared to the reformulated burgers (Table 2). The CHE samples showed the highest TBARS values during storage ($P < 0.001$). A similar trend was reported by other authors (Barros et al., 2020; Domínguez, Pateiro, Munekata, Campagnol, & Lorenzo, 2016) and demonstrates the difficulty of the incorporation of healthy oils in meat products due to their high susceptibility to lipid oxidation. The addition of BMBE was efficient to increase the oxidative stability of the burgers rich in Omega-3 PUFAs. When comparing the treatments with the addition of BMBE, T1 had lower TBARS values than T2 and T3, probably due to the decrease in fat globules of chia oil caused by US cavitation, which increased the interaction between free radicals and MUFAs and PUFAs (Cichoski et al., 2021). Further, an increase in the number of free radicals during the US treatment may have occurred. As reported by Da Silva et al. (2020), this increase depends on the food composition and the US processing conditions. Thus, more studies should be carried out to find the better conditions of US application to enrich chia oil with CGA from BMBE without impairing its oxidative stability.

T1 showed in relation to CHE a reduction in TBARS values of 77, 48, 59, 40, and 27% in raw burgers and 53, 43, 45, 39, and 36% in cooked burgers, on days 1, 30, 60, 90 and 120 of storage, respectively. This reduction is well correlated with the CGA levels of the burgers (Fig. 2), as previously reported by Santana-Gálvez, Cisneros-Zevallos, and Jacobo-Velázquez (2017), this compound has high antioxidant activity in foods. According to those authors, CGA delays lipid oxidation through the scavenging of free radicals and metal chelation in the lipid and aqueous phases, respectively. This antioxidant effect provided by BMBE allowed cooked burgers from T1 to reach a sensory detectable TBARS value (2 mg MDA/Kg sample) (Wood et al., 2004) only after 60 days of storage. This result is very expressive since the TBARS values of the cooked burgers of the treatment CHE were below 2 mg MDA/Kg only on the 1st day of storage.

3.5. pH and instrumental color

The pH values were influenced ($P < 0.001$) by the interaction between the treatments and the storage period (Fig. 3C-D). The pH values of raw and cooked burgers ranged from 5.5 to 6.0 and 5.6 to 6.2, respectively, during 120 days of storage. This result is in accordance with the findings of Malik and Sharma (2011) in buffalo burgers. In general, the reformulated burgers had a pH behavior during storage similar to the Control, which shows that the addition of HE of chia oil enriched with BMBE as animal fat replacer does not have a great impact on the pH values of the products.

The instrumental color parameters (L*, a* and b*) were influenced ($P < 0.001$) by the interaction between the treatments and the storage time (Fig. 4). The evolution of L*, a*, and b* values of the burgers (raw and cooked) from the Control and CHE was statistically similar throughout the storage. The only exception occurred at day 120 of storage, once the cooked burgers of the treatment CHE had higher ($P < 0.01$) b* values than the Control samples. The addition of HE of chia oil enriched with BMBE did not cause great impact in the color of the burgers during storage. For the raw burgers, a reduction of a* values during the storage was observed for most treatments. This trend was also reported by Heck et al. (2020) and can be attributed to the oxidation of the myoglobin to metmyoglobin (Mancini & Hunt, 2005).

3.6. Consumer study

The acceptance test was performed at the beginning of storage (day 1) and the results are shown in Table 4. Control and CHE samples had similar ($P > 0.05$) scores in all evaluated sensory attributes (color, aroma, flavor, texture, and liking). This result was expected and agrees with our previous study (Heck et al., 2020). However, the high degree of lipid oxidation during the storage (Fig. 3A and B) impairs the commercialization of the treatment CHE. Control and T1 also had

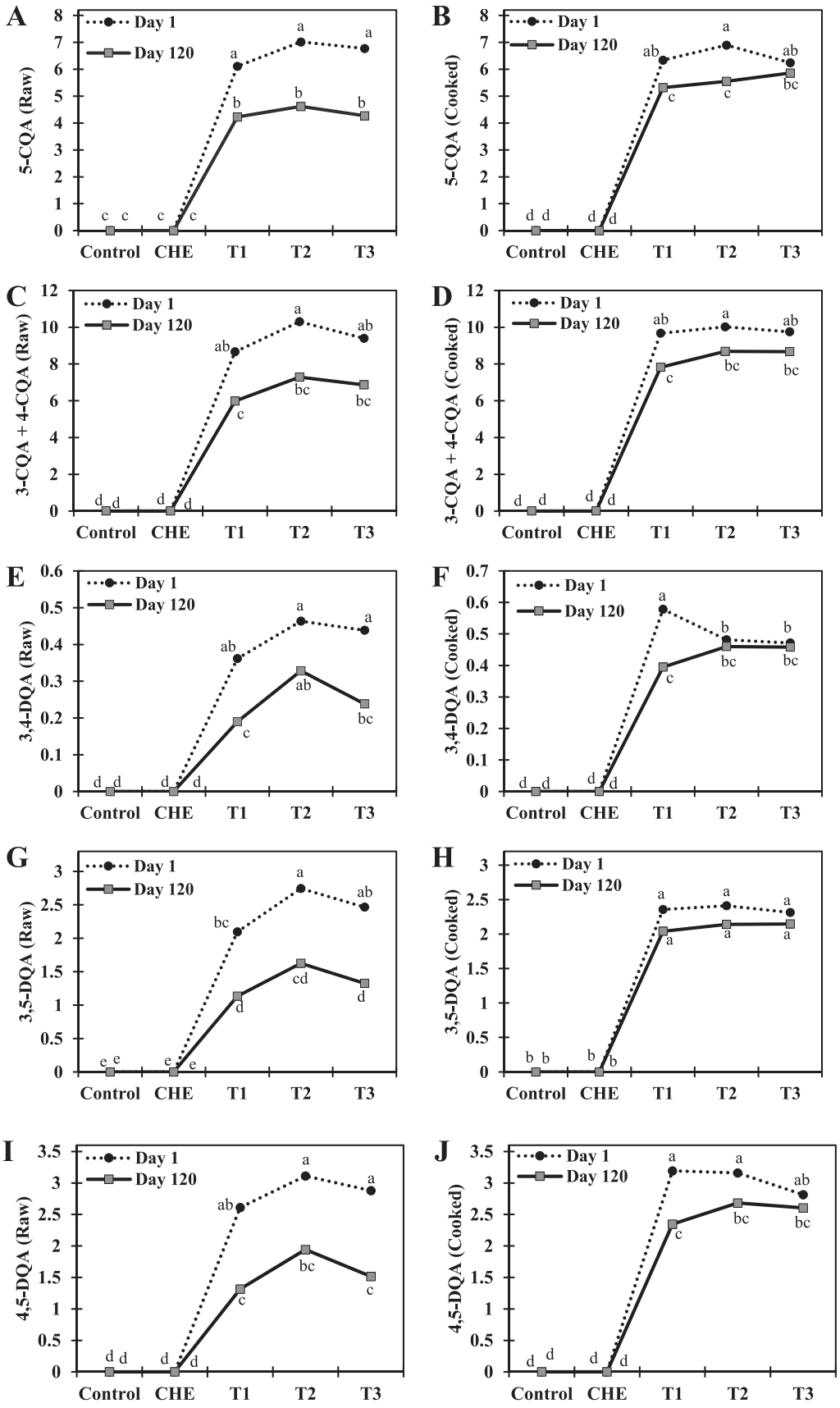


Fig. 2. Overall effects of the treatments and storage period on the chlorogenic acids content (mg 100 g^{-1}) of the raw and cooked buffalo burgers.
[a–e] Averages followed by the same letter did not show any significant difference ($P > 0.05$) by Tukey test. Treatments: Described in Table 1.

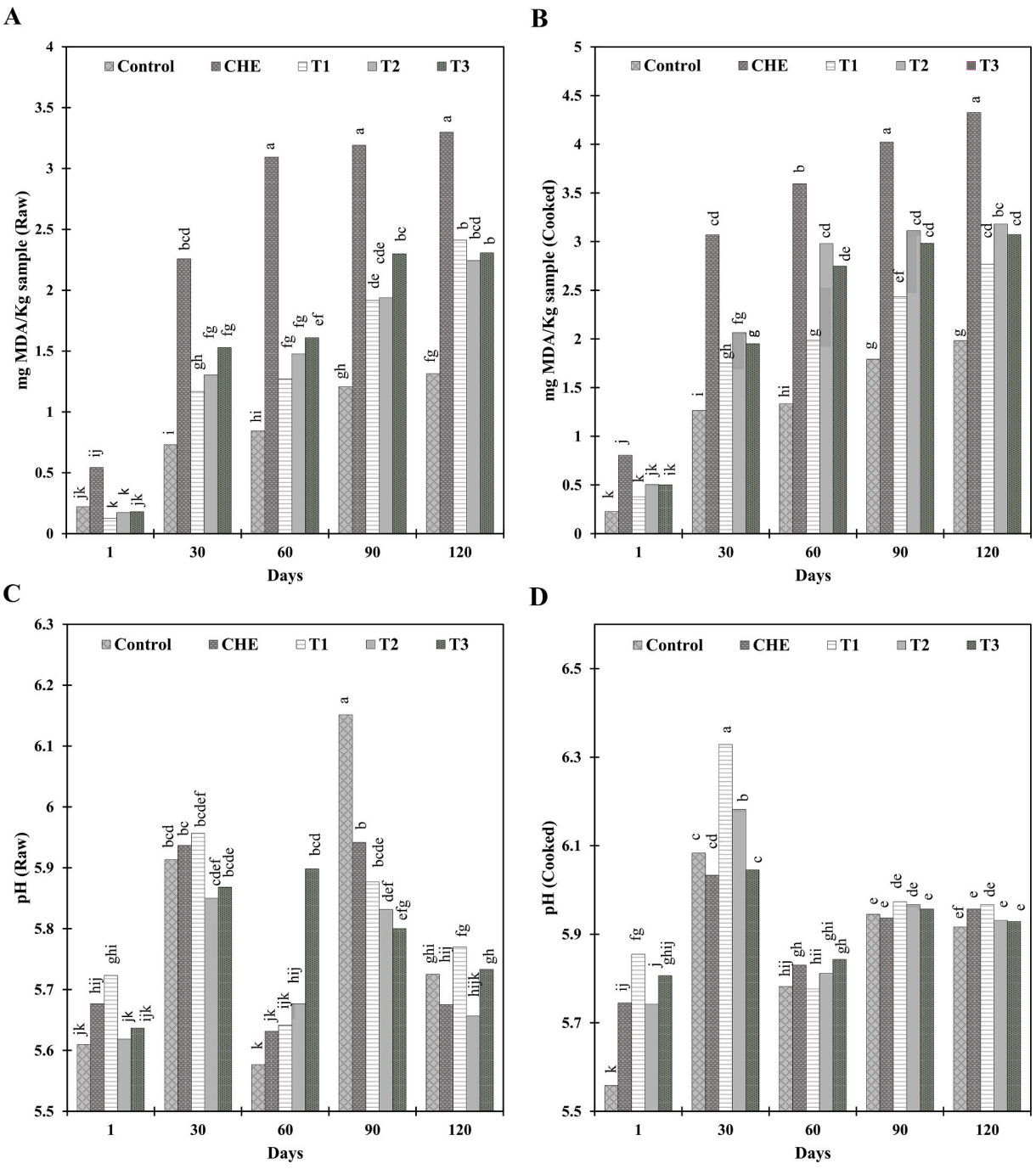


Fig. 3. Overall effects of the treatments and storage period on the TBARS (A–B) and pH (C–D) values of the raw and cooked buffalo burgers.
[a–n] Averages followed by the same letter did not show any significant difference ($P > 0.05$) by Tukey test. Treatments: Described in Table 1.
SEM (Standard error of the mean): TBARS: raw (0.077) and cooked (0.094), pH: raw (0.012) and cooked (0.013).

similar scores ($P > 0.05$) in all the sensory attributes analyzed. However, the treatments T2 and T3 had lower ($P < 0.001$) scores than the Control for the attributes aroma, flavor, and liking.

The correspondence analysis (CA) used to analyze the CATA questionnaire is shown in Fig. 5A. The CA explained 93.64% of the total variation of the data (F1: 83.02% and F2: 10.62%). The samples were divided into two groups in F1. The treatments T2 and T3 were allocated in the positive quadrant of F1, being characterized by the attributes strange appearance, herbs aroma, unpleasant taste, fish aroma, and fish taste. These attributes were negatively correlated with the liking scores (Fig. 5B). The Control, CHE, and T1 were allocated in the negative quadrant of F1 and were characterized by the attributes positively correlated with the liking scores (Fig. 5B), including the attributes juicy, pleasant aroma, mild aroma, pleasant taste, ideal color, and ideal texture.

4. Conclusion

This study analyzed for the first time the antioxidant potential of a natural extract from the bark of mate branches in meat products, which is a low-cost and abundant residue from the yerba mate cultivation. BMBE presented a high CGA level (30 g 100 g^{-1}), showing great potential for application as a natural antioxidant in foods. In addition, this study demonstrated that the application of oils rich in Omega-3 PUFAs

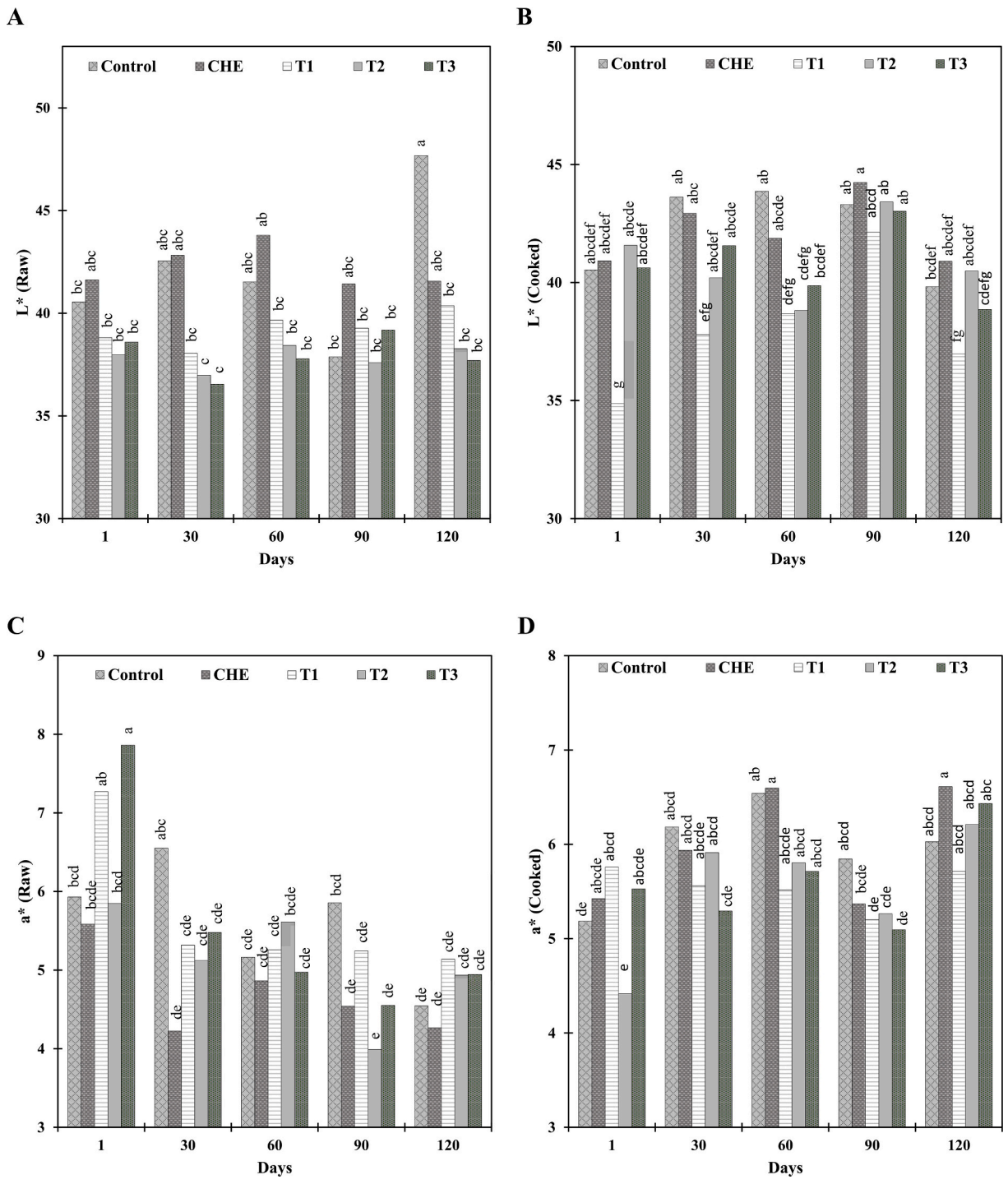


Fig. 4. Overall effects of the treatments and storage period on the L* (A–B), a* (C–D) and b* (4E–F) values of the raw and cooked buffalo burgers. [a–h] Averages followed by the same letter did not show any significant difference ($P > 0.05$) by Tukey test. Treatments: Described in Table 1. SEM (Standard error of the mean): L*: raw (0.274) and cooked (0.082); a* raw (0.106) and cooked (0.201); b* raw (0.054) and cooked (0.098).

in burgers, even in hydrogelled form, should be performed with caution, due to the intensification of the lipid oxidation reactions, especially after cooking. The sensory quality of the low-fat burgers was not affected by addition of HE from chia oil enriched with BMBE without sonication. In addition, the lipid reformulation enriched the products with CGA and partially impaired the increase in lipid oxidation caused by the increase in PUFA / SFA ratio.

Declaration of Competing Interest

The author have no conflicts of interest for disclose to paper entitled "Lipid oxidation and sensory characterization of Omega-3 rich buffalo burgers enriched with chlorogenic acids from the mate (*Ilex paraguariensis*) tree harvesting residues".

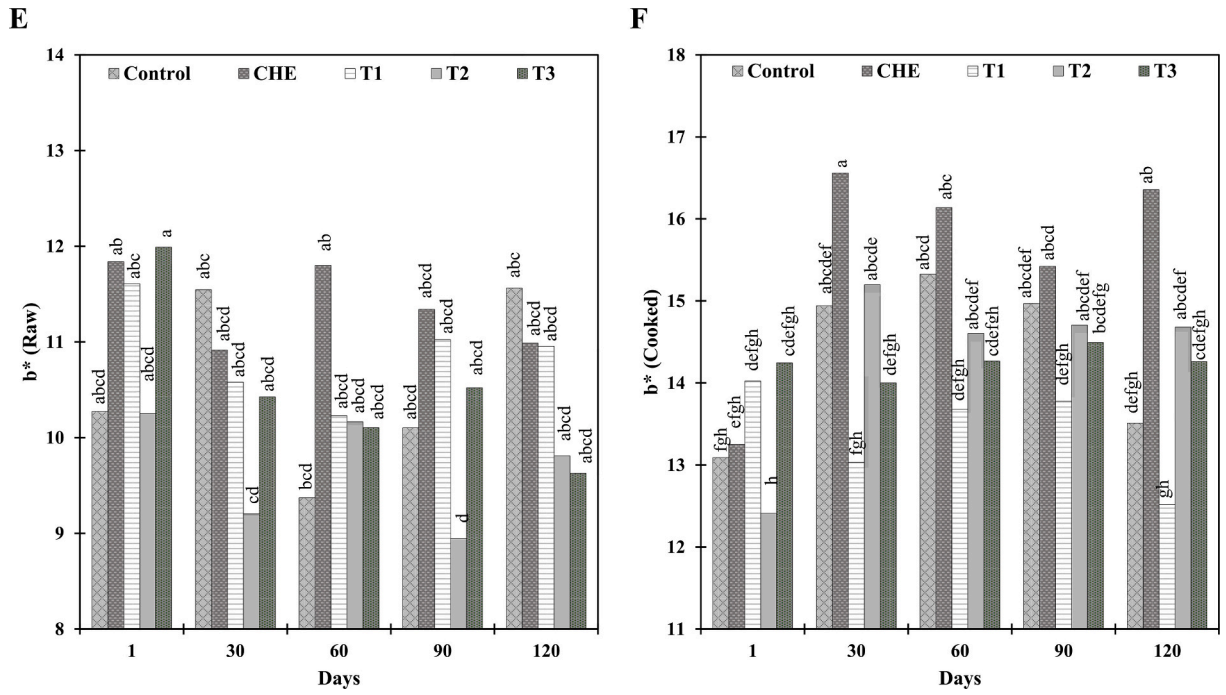


Fig. 4. (*continued*).

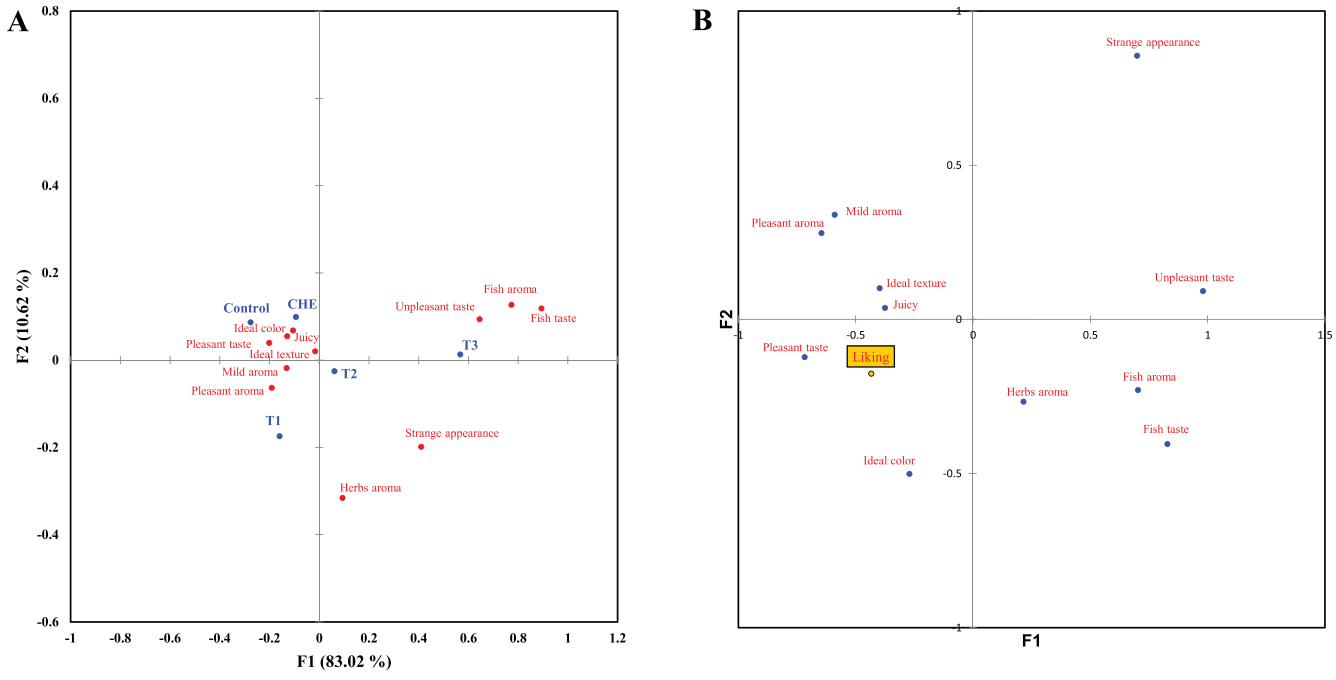


Fig. 5. A: Correspondence analysis used to analyze the CATA questionnaire. B: Principal coordinate analysis used to analyze the correlation between the liking scores and CATA descriptors. Treatments: Described in Table 1.

Acknowledgments

Fellowships from National Council for Scientific and Technological Development (CNPq), financial support from CNPq (grant number 425577/2018-0), and Coordination for the Improvement of Higher Education Personnel (CAPES finance code 001) have supported this work. he authors thank the contribution of *Associação de Produtores de Erva Mate of Machadinho*, RS, and *Barão Indústria de Erva Mate* for donating the samples of bark of mate branches.

References

Alvarado-Ramírez, M., Santana-Gálvez, J., Santacruz, A., Carranza-Montealvo, L. D., Ortega-Hernández, E., Tirado-Escobosa, J., … Jacobo-Velázquez, D. A. (2018). Using a functional carrot powder ingredient to produce sausages with high levels of nutraceuticals. *Journal of Food Science, 83*(9), 2351–2361.

AOAC (Association of Official Analytical Chemistry). (2005). *Official methods of analysis of AOAC international* (17th ed.) (Maryland: USA).

Ballus, C. A., Meinhart, A. D., Campos, F. A. S., Da Silva, F. O., De Oliveira, A. F., & Godoy, H. T. (2014). A quantitative study on the phenolic compound, tocopherol and fatty acid contents of monovarietal virgin olive oils produced in the southeast region of Brazil. *Food Research International, 62*, 74–83.

Barros, J. C., Munekata, P. E. S., Carvalho, F. A. L., Pateiro, M., Barba, F. J., Domínguez, R., & Lorenzo, J. M. (2020). Use of tiger nut (*Cyperus esculentus* L.) oil emulsion as animal fat replacement in beef burgers. *Foods, 9*, 44.

Bligh, E. G., & Dyer, W. J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology, 37*, 911–917.

Bruna, J. M., Ordonez, J. A., Fernández, M., Herranz, B., & Hoz, L. (2001). Microbial and physico-chemical changesduring theripening ofdryfermented sausagessuperficially inoculated with or having added an intracellular cell-free extract of Penicillium aurantiogriseum. *Meat Science, 59*, 87–96.

Campos, R. M. L., Hierro, E., Ordónez, J. A., Bertol, T. M., Terra, N. N., & Hoz, L. (2007). Fatty acid and volatile compounds from salami manufactured with yerba mate (Ilex parguariensis) extract and pork back fat and meat from pigs fed on diets with partial replacement of maize with rice bran. *Food Chemistry, 103*, 1159–1167.

Chaudhary, R., Saadin, K., Bliden, K. P., Harris, W. S., Dinh, B., Sharma, T., & Gurbel, P. A. (2016). Risk factors associated with plasma omega-3 fatty acid levels in patients with suspected coronary artery disease. *Prostaglandins, Leukotrienes and Essential Fatty Acids, 113*, 40–45.

Cichoski, A. J., Da Silva, J. S., Leães, Y. S. V., Robalo, S. S., Dos Santos, B. A., Reis, S. R., … Campagnol, P. C. B. (2021). Effects of ultrasonic-assisted cooking on the volatile compounds, oxidative stability, and sensory quality of mortadella. *Ultrasonics Sonochemistry, 72*, 105443.

Clifford, M. N. (1999). Chlorogenic acids and other cinnamates nature, occurrence and dietary burden. *Journal of the Science of Food and Agriculture, 79*, 362–372.

Da Silva, J. S., Voss, M., Menezes, C. R., Barin, J. S., Wagner, R., Campagnol, P. C. B., & Cichoski, A. J. (2020). Is it possible to reduce the cooking time of mortadellas using ultrasound without affecting their oxidative and microbiological quality? *Meat Science, 159*, 107947.

Da Silva, S. L., Amaral, J. T., Ribeiro, M., Sebastião, E. E., Vargas, C., De Lima, F. F., & Campagnol, P. C. B. (2019). Fat replacement by oleogel rich in oleic acid and its impact on the technological, nutritional, oxidative, and sensory properties of Bologna-type sausages. *Meat Science, 149*, 141–148.

Domínguez, R., Pateiro, M., Munekata, P. E. S., Campagnol, P. C. B., & Lorenzo, J. M. (2016). Influence of partial pork backfat replacement by fish oil on nutritional and technological properties of liver pâté. *European Journal of Lipid Science and Technology, 118*, 1–7.

Esmelindro, A. A., Girardi, J. S., Mossi, A., Jacques, R. A., & Dariva, C. (2004). Influence of agronomic variables on the composition of mate tea leaves (*Ilex paraguariensis*) extracts obtained from CO2 extraction at 30 °C and 175 bar. *Journal of Agricultural and Food Chemistry, 52*, 1990–1995.

European Parliament. (2006). Regulation (EC) no 1924/2006 of the European Parliament and of the council of 20 December 2006 on nutrition and health claims made on foods. *Official Journal of the European Union, L404*, 9–25.

Filip, R., López, P., Giberti, G., Coussio, J., & Ferraro, G. (2001). Phenolic compounds in seven 579 south American Ilex species. *Fitoterapia, 72*, 774–778.

Filip, R., Lotito, S. B., Ferraro, G. E., & Fraga, C. G. (2000). Antioxidant activity of Ilex 603 paraguariensis and related species. *Nutrition Research, 20*, 1437–1444.

Hartman, L., & Lago, R. C. (1973). Rapid preparation of fatty acid methyl esters from lipids. *Laboratory Practice, 22*, 475–476.

Heck, R. T., Fagundes, M. B., Cichoski, A. J., Menezes, C. R., Barin, J. S., Lorenzo, J. M., … Campagnol, P. C. B. (2019). Volatile compounds and sensory profile of burgers with 50% fat replacement by microparticles of chia oil enriched with rosemary. *Meat Science, 148*, 164–170.

Heck, R. T., Ferreira, D. F., Fagundes, M. B., Dos Santos, B. A., Cichoski, A. J., Saldaña, E., … Campagnol, P. C. B. (2020). Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: A green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils. *Meat Science, 170*, 108230.

Heck, R. T., Lucas, B. N., Dos Santos, D. J. P., Pinton, M. B., Fagundes, M. B., Etchepare, M. A., … Campagnol, P. C. B. (2018). Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques. *Meat Science, 146*, 147–153.

Heck, R. T., Saldaña, E., Lorenzo, J. M., Correa, L. P., Fagundes, M. B., Cichoski, A. J., … Campagnol, P. C. B. (2019). Hydrogelled emulsion from chia and linseed oils: A promising strategy to produce low-fat burgers with a healthier lipid profile. *Meat Science, 156*, 174–182.

Heck, R. T., Vendruscolo, R. G., De Araújo, M. E., Cichoski, A. J., Menezes, C. R., Barin, J. S., … Campagnol, P. C. B. (2017). Is it possible to produce a low-fat burger with a healthy n − 6/n − 3 PUFA ratio without affecting the technological and sensory properties? *Meat Science, 130*, 16–25.

Jacques, R. A., Santos, J. G., Dariva, C., Oliveira, J. V., & Caramarão, E. B. (2007). GC/MS characterization of mate tea leaves extracts obtained from high-pressure CO2 extraction. *Journal of Supercritical Fluids, 40*, 354–359.

Kandeepan, G., Mendiratta, S. K., Shukla, V., & Vishnuraj, M. R. (2013). Processing characteristics of buffalo meat- A review. *Journal of Meat Science and Technology, 1*, 01–11.

Lima, J. P., Farah, A., King, B., De Paulis, T., & Martin, P. R. (2016). Distribution of major chlorogenic acids and related compounds in Brazilian green and toasted *Ilex paraguariensis* (maté) leaves. *Journal of Agricultural and Food Chemistry, 64*(11), 2361–2370.

Macfie, H. J., Bratchell, N., Greenhoff, K., & Vallis, L. (1989). Designs to balance the effect of order of presentation and first-order carry-over effects in hall tests. *Journal of Sensory Studies, 4*(2), 129–148.

Malik, A. H., & Sharma, B. D. (2011). Shelf life study of hurdle treated ready-to-eat spiced buffalo meat product stored at 30±3 °C for 7 weeks under vacuum and aerobic packaging. *Journal of Food Science and Technology, Mysore, 51*(5), 832–844.

Mancini, R. A., & Hunt, M. C. (2005). Current research in meat color. *Meat Science, 77*(1), 100–121.

Marques, V., & Farah, A. (2009). Chlorogenic acids and related compounds in medicinal plants and 531 infusions. *Food Chemistry, 13*, 1370–1376.

Mateos, R., Baeza, G., Sarriá, B., & Bravo, L. (2018). Improved LC-MSn characterization of hydroxycinnamic acid derivatives and flavonols in different commercial mate (*Ilex paraguariensis*) brands. Quantification of polyphenols, methylxanthines, and antioxidant activity. *Food Chemistry, 241*, 232–241.

Meilgaard, M. C., Carr, T., & Civille, G. V. (2006). *Sensory evaluation techniques* (4th ed.). Boca Raton: CRC Press.

Meinhart, A. D., Damin, F. M., Caldeirão, L., Ferreira, S. T., Filho, J. T., & Godoy, H. T. (2017). Chlorogenic acid isomer contents in 100 plants commercialized in Brazil. *Food Research International, 1*, 522–530.

Mendes, R. F., & Carlini, E. A. (2007). Brazilian plants as possible adaptogens: An ethnopharmacological survey of books edited in Brazil. *Journal of Ethnopharmacology, 109*, 493–500.

Pagliosa, C. M., Vieira, M. A., Podestá, R., Maraschin, M., Zeni, A. L. B., Amante, E. R., & Amboni, R. D. M. C. (2010). Methylxanthines, phenolic composition, and antioxidant activity of bark from residues from mate tree harvesting (*Ilex paraguariensis* A. St. Hil.). *Food Chemistry, 122*(1), 173–178.

Prediger, R. D. S., Fernandes, M. S., Wopereis, D. R. S., Pereira, V. S., Bosse, T. S., Da Silva, C. B., … Costa-Campos, L. (2008). Effects of acute administration of the hydroalcoholic extract of mate tea leaves (*Ilex paraguariensis*) in animal models of learning and memory. *Journal of Ethnopharmacology, 120*, 465–473.

Puangpraphant, S., Berhow, M. A., & de Mejia, E. G. (2011). Mate (*Ilex paraguariensis* St. Hilaire) saponins induce caspase-3-dependent apoptosis in human colon cancer cells in vitro. *Food Chemistry, 125*(4), 1171–1178.

Racanicci, A. M. C., Danielsen, B., & Skibsted, L. H. (2008). Mate (*Ilex paraguariensis*) as a source of water extractable antioxidant for use in chicken meat. *Journal European Food Research And Technology, 27*(1), 255–260.

Santana-Gálvez, J., Cisneros-Zevallos, L., & Jacobo-Velázquez, D. A. (2017). Chlorogenic acid: Recent advances on its dual role as a food additive and a nutraceutical against metabolic syndrome. *Molecules, 22*, 358.

Sasaki, K., Alamed, J., Weiss, J., Villeneuve, P., Giraldo, L. J. L., Lecomte, J., … Decker, E. A. (2010). Relationship between the physical properties of chlorogenic acid esters and their ability to inhibit lipid oxidation in oil-in-water emulsions. *Food Chemistry, 118*(3), 830–835.

Simopoulos, A. O. (2011). Evolutionary aspects of diet: The Omega-6/Omega-3 ratio and the brain. *Molecular Neurobiology, 44*, 203–215.

Stone, H., Bleibaum, R. N., & Thomas, H. A. (2012). *Sensory evaluation practices* (4th ed.). New York: Academic Press.

Tamburrano, A., Tavazzi, B., Callà, C. A. M., Amorini, A. M., Lazzarino, G., Vincenti, S., … Laurenti, P. (2019). Biochemical and nutritional characteristics of buffalo meat and potential implications on human health for a personalized nutrition. *Italian Journal of Food Safety, 8*(3), 8317.

Visentainer, J. V. (2012). Aspectos analíticos da resposta do detector de ionização em chama para ésteres de ácidos graxos em biodiesel e alimentos. *Química Nova, 19*, 380–385.

Wood, J. G., Rogina, B., Lavu, S., Ilowitz, K., Helfand, S. L., Tatar, M., & Sinclair, D. (2004). Sirtuin activators mimic caloric restriction and delay ageing in metazoans. *Nature, 430*, 686–689.

11. DISCUSSÃO GERAL

A substituição de gordura animal por óleos saudáveis não é uma tarefa simples de ser realizada, pois importantes atributos tecnológicos, de textura e sensoriais dos produtos cárneos podem ser prejudicados. Levando em consideração, aspectos relacionados a saudabilidade, a redução de gordura animal dos produtos cárneos pode reduzir o risco do surgimento de doenças crônicas. Neste contexto, o objetivo deste estudo foi avaliar o efeito na qualidade de hamburgueres da substituição de toucinho por óleos vegetais saudáveis microcapsulados e hidrogelificado.

A primeira etapa deste estudo foi apresentada no Capítulo 3, sob o titulo “Oxidative stability of burgers containing chia oil microparticles enriched with rosemary by green-extraction techniques”. Nesta etapa, foram investigados os efeitos da substituição de 50 % de toucinho por microcápsulas de óleo de chia contendo compostos bioativos de alecrim extraídos por maceração direta atraves do auxilio de ultrassom (UAE) e aplicados em hamburgueres. Durante o processamento, observou-se que os hambúrgueres produzidos com microcápsulas de óleo de chia enriquecidas com alecrim por UAE apresentaram maior estabilidade oxidativa que os demais tratamentos, principalmente após o cozimento. Além disso, foi observada uma redução dos defeitos sensoriais causados pela reformulação lipídica nos tratamentos adicionados de antioxidantes.

A aceitação sensorial apresenta-se como um dos maiores desafios quando se fala em reformulação lipídica de produtos cárneos. Neste sentido, no capítulo 4 “Volatile compounds and sensory profile of burgers with 50 % fat replacement by microparticles of chia oil enriched with rosemary” foram avaliados os compostos voláteis e o perfil sensorial dos hamburgueres adicionados de microcápsulas de óleo de chia enriquecidas com compostos bioativos do alecrim por UAE e CME. Os resultados dos compostos voláteis indicaramm uma diminuição dos

voláteis provenientes da oxidação lipídica e proteica e um aumento dos terpenos no início (dia 1) e no final do armazenamento (dia 120), antes e após o cozimento nos tratamentos adicionados de antioxidantes. Quanto aos descritores sensoriais, a incorporação direta de compostos bioativos do alecrim ao óleo de chia por técnicas UAE ou CME antes do processo de microencapsulação foi eficaz para resolver os impactos sensoriais.

A emulsão hidrogelificada (HE) de óleos de chia e linhaça foi estudada no Capítulo 5 "Hydrogelled emulsion from chia and linseed oils: a promising strategy to produce low-fat burgers with a healthier lipid profile" através de análises físico-químicas, tecnológicas e sensoriais. A substituição do toucinho por HE não afetou as propriedades tecnológicas dos hambúrgueres. A reformulação lipídica reduziu o conteúdo lipídico e melhorou o perfil de ácidos graxos dos hambúrgueres, sem grandes alterações no perfil lipídico dos hambúrgueres após o cozimento. A substituição de apenas 20 % de toucinho por HE levou a melhorias nutricionais significativas. O perfil sensorial de aceitação demonstrou que é possível substituir até 60 % do toucinho pelo HE produzido neste estudo, porém se fizeram necessárias melhorias na estabilidade oxidativa do HE dos óleos de chia e linhaça. Desta forma, nas outras etapas buscou-se melhorar a estabilidade oxidativa da HE através da adição de antioxidantes naturais.

No Capítulo 6 "Jabuticaba peel extract obtained by microwave hydrodiffusion and gravity extraction: a green strategy to improve the oxidative and sensory stability of beef burgers produced with healthier oils", o extrato da casca de jabuticaba (JPE) obtido pela extração de MHG foi utilizado para o enriquecimento de HE a partir de óleos de chia e linhaça com compostos bioativos, que foi utilizado como substituto de gordura em hambúrgueres. A extração de MHG foi eficaz para produzir JPE rico em compostos fenólicos e microbiologicamente seguro. A adição de 10 % de JPE ao HE minimizou os problemas de oxidação lipídica, pois permitiu a produção de hambúrgueres com elevada aceitação sensorial

e com uma estabilidade oxidativa semelhante aos produtos tradicionais durante 60 dias de armazenamento.

No Capítulo 7 “Lipid oxidation and sensory quality of omega-3 rich buffalo burgers enriched with chlorogenic acids from the mate (*ilex paraguariensis*) tree harvesting residues”, avaliou-se pela primeira vez o potencial antioxidante de um extrato natural da casca de ramos de erva-mate (BMBE) em produtos cárneos. O BMBE apresentou alto teor de CGA (30 g 100 g^{-1}), apresentando grande potencial para aplicação como antioxidante natural em alimentos. A substituição de 50 % de toucinho por HE de óleo de chia enriquecido com BMBE não afetou a qualidade sensorial dos hambúrgueres. Além disso, a reformulação lipídica reduziu parcialmente o aumento da oxidação lipídica causado pelo aumento da razão PUFA / SFA.

12. CONCLUSÃO GERAL

As principais conclusões deste estudo são apresentadas abaixo:

- Os hambúrgueres produzidos com micropartículas de óleo de chia enriquecidas com alecrim por UAE apresentaram maior estabilidade oxidativa, principalmente após o cozimento. Além disso, a incorporação de compostos bioativos do alecrim no óleo de chia reduziu os defeitos sensoriais causados pela reformulação lipídica.
- A substituição do toucinho por HE de óleos de chia e linhaça conferiu um perfil de ácidos graxos mais saudável, permitindo que os hambúrgueres fossem rotulados com alegações nutricionais saudáveis. Os testes sensoriais (aceitação e Check-All-That-Apply) indicaram que foi possível substituir até 60 % do toucinho por HE, com indicação da necessidade de maiores estudos sobre a oxidação lipídica.
- A capacidade antioxidante do extrato de casca de jabuticaba (JPE) ficou evidenciada nos hambúrgueres reformulados com HE adicionados com 10 % de JPE, onde observou-se a oxidação lipídica semelhante ao controle até o 60º dia de armazenamento sob congelamento. Além disso, a incorporação do JPE no HE reduziu os defeitos sensoriais causados pela reformulação lipídica.
- A adição de BMBE enriqueceu os hambúrgueres com CGA, evitando o aumento da oxidação lipídica e a perda da qualidade sensorial causada pela HE de óleo de chia.

13. CONSIDERAÇÕES FINAIS

Este estudo demonstrou a viabilidade tecnológica da utilização de óleos vegetais microencapsulados e hidrogelificados como substitutos de gordura animal em produtos cárneos que sofrem tratamento térmico intenso. Os resultados desta pesquisa podem ser úteis para a sociedade, pois podem ser utilizados industrialmente para enriquecer diversos alimentos com ácidos graxos ômega 3. Desta forma, este estudo poderá contribuir para aumentar a ingestão deste nutriente pela população e assim, reduzir os fatores de risco relacionados ao surgimento de doenças cardiovasculares. Considerando que as doenças cardiovasculares são responsáveis por mais de 1100 mortes por dia no Brasil, este estudo poderá contribuir significativamente para melhorar a qualidade e a expectativa de vida da população brasileira.Considerando a dificuldade de transferência dos resultados obtidos em estudos acadêmicos para a sociedade, a partir desta tese foi criada a *startup* "w3 Special Ingredients", cujo desafio é justamente trazer a estratégia tecnológica desenvolvida na academia para ser instrumento de saudabilidade junto ao mercado. A empresa atualmente está incubada na Agência de Transferência de Tecnologia da UFSM (Agittec). A solução que a w3 propõe é a produção de um ingrediente rico em ômega 3 através de uma técnica inovadora de microencapsulação. As microcápsulas possuem como diferencial a alta resistência térmica, o que significa que o ômega 3 não é degradado quando aquecido. Isto possibilita que sejam aplicadas em uma grande variedade de produtos alimentícios. Além disso, por serem 100 % de origem vegetal, as microcápsulas podem ser utilizadas para enriquecer produtos veganos. Outro diferencial é que as microcápsulas somente liberam o ômega-3 no final do intestino humano, possibilitando sua total absorção pelo organismo e por serem produzidas com fontes renováveis e biodegradáveis tem baixo impacto sobre o meio ambiente.