



**UNIVERSIDADE FEDERAL RURAL DE PERNAMBUCO**

**PRÓ-REITORIA DE PESQUISA E PÓS-GRADUAÇÃO PROGRAMA DE PÓS-GRADUAÇÃO  
EM RECURSOS PESQUEIROS E AQUICULTURA**

**ELABORAÇÃO E AVALIAÇÃO DE PRODUTOS TECNOLÓGICOS  
UTILIZANDO O SARAMUNETE (*Pseudupeneus maculatus* – Bloch, 1793)**

**Pedro Henrique de Sá Vieira**

Tese apresentada ao Programa de Pós-Graduação em Recursos Pesqueiros e Aquicultura da Universidade Federal Rural de Pernambuco como exigência para obtenção do título de Doutor.

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## **Dedicatória**

Dedicamos este trabalho a ...

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## Resumo

A produção de pescado vem aumentando ao longo das últimas décadas e está entre as principais fontes de proteína animal para o consumo humano. No entanto, no Brasil, o consumo médio aparente de pescado varia de 5-10 kg per capita/ano, o que está abaixo da recomendação da Organização Mundial da Saúde, que é de 12 kg per capita/ano. Esse baixo consumo pode ser atribuído a fatores culturais, alto custo do produto, alta perecibilidade e limitada diversidade de produtos de pescado no mercado, especialmente produtos prontos para consumo. Nesse contexto, a aplicação de novas tecnologias, como o ultrassom, na elaboração de marinados de saramunete (*Pseudopenaeus maculatus*), e o desenvolvimento de produtos inovadores como nhoques de inhame adicionados com concentrado proteico de saramunete, são importantes para diversificar os produtos e estimular o consumo de pescado no Brasil. Foram elaborados marinados de saramunete, avaliou-se a influência das potências de ultrassom (US) (110, 175 e 330 W) e dos tempos de aplicação (1, 3 e 5 min) nas características físico-químicas, cor instrumental e nos aspectos microbiológicos dos filés marinados. Observou-se que o pH independente da potência e do tempo variou de 4,67 a 5,06. Quanto maior o tempo de US, menor a luminosidade ( $L^*$ ). A atividade de água (0,944 e 0,957) e umidade (63,51 e 71,32) não apresentaram muita diferença entre os tratamentos, proteína bruta variou entre 15,89 e 19,52, exceto US (330W/5 min), cujo valor foi de 5,40 e cinzas (2,61 e 3,44), exceto para o tratamento US (110 W/5min) que foi de 4,16. As características microbiológicas estavam de acordo com a legislação brasileira. Em relação a elaboração de nhoque de inhame com concentrado proteico de saramunete, 0% (tratamento controle), 7,5% e 15%, observou-se que o teor de umidade e carboidratos, textura instrumental, luminosidade (valor de  $L^*$ ) e atributos sensoriais diminuíram com a adição de 15% de CPS ao nhoque de inhame. Por outro lado, os conteúdos de proteínas, lipídios, cinzas e valor calórico aumentaram significativamente em comparação ao controle. Portanto, o nhoque de inhame com CPS, melhora a qualidade nutricional do produto, sendo que a formulação com adição de 7,5% de CPS apresentou melhores características físico-químicas, por outro lado, o resultado da avaliação sensorial das formulações teve nota um pouco acima de 5 (nem gostei nem desgostei). Como conclusão geral da tese observa-se que a integração de tecnologias inovadoras como o ultrassom na produção de marinados e o desenvolvimento, de novos produtos, como nhoques enriquecidos com concentrado proteico de pescado, representa um avanço na diversificação e valorização da cadeia de consumo de pescado no Brasil.

**Palavras-chave:** nhoque; peixe marinado; ultrassom; pescado de valor agregado.

## Abstract

Fish production has been increasing over the last decades and is among the main sources of animal protein for human consumption. However, in Brazil, the average apparent consumption of fish varies between 5-10 kg per capita/year, which is below the World Health Organization's recommendation of 12 kg per capita/year. This low consumption can be attributed to cultural factors, the high cost of the product, high perishability, and the limited diversity of fish products in the market, especially ready-to-eat products. In this context, the application of new technologies, such as ultrasound, in the preparation of marinated fish (*Pseudopenaeus maculatus*), and the development of innovative products like yam gnocchi supplemented with fish protein concentrate, are important to diversify products and stimulate fish consumption in Brazil. In the first study, the influence of ultrasound (US) power levels (110, 175, and 330 W) and application times (1, 3, and 5 minutes) on the physicochemical characteristics, color, and microbiological aspects of marinated fillets were evaluated. It was observed that pH tended to average 5.0 as power increased, while values below 4.7 were observed for shorter US times. The longer the US time, the lower the lightness ( $L^*$ ). Water activity and crude protein decreased as US power and time increased. Microbiological characteristics were by Brazilian legislation. The second study evaluated the addition of 0% (control treatment), 7.5%, and 15% fish protein concentrate (FPC) to yam gnocchi. It was observed that moisture content, carbohydrates, instrumental texture, lightness, and sensory attributes decreased with the addition of 15% FPC to yam gnocchi. On the other hand, protein, lipid, ash content, and caloric value increased significantly compared to the control. Therefore, it is recommended to supplement yam gnocchi with FPC as it improves the nutritional quality of the product, with the addition of up to 7.5% FPC being recommended so as not to impair the product's physicochemical and sensory parameters. Therefore, the integration of innovative technologies such as ultrasound in the production of marinated fish and the development of functional products, such as gnocchi enriched with fish protein concentrate, represents a promising advance in the diversification and enhancement of the fish consumption chain in Brazil. The strategic use of ultrasound not only enhances the desired characteristics in marinated products, adjusting essential physical and microbiological parameters for product safety and quality but also paves the way for the development of foods with greater nutritional and sensory appeal. Moreover, the controlled addition of fish protein concentrate to yam gnocchi results in a final product



with high nutritional quality, without compromising consumer sensory acceptance. These advances not only encourage fish consumption, raising levels closer to the WHO recommendation, but also offer practical solutions to the market, making fish more accessible, varied, and appealing to the public.

Keywords: gnocchi; marinated fish; ultrasound; value-added fish.

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## 1- Introdução

### 1.1 - Contextualização da pesquisa

O pescado (peixes, camarões, moluscos, etc.) está entre o grupo de animais mais produzidos no mundo, sendo uma das principais fontes de proteína para a alimentação humana. Desde 1961 até 2019, o consumo global aparente de pescado cresceu a uma taxa média de 3% ao ano, sendo esse crescimento maior que o de todas as outras proteínas animais [carne, laticínios, leite; bovino, caprino e suíno (2,7%)], exceto para carne de aves (4,7%) (FAO, 2024). Esse aumento no consumo de pescado ocorreu devido a um conjunto de fatores, tais como: redução no desperdício, utilização otimizada, crescimento da demanda, aumento da população e crescimento econômico geral dos países (FAO, 2024).

Em 2021, o pescado representou 15% das proteínas animais e 6% de todas as proteínas consumidas globalmente em 2019 (FAO, 2024). Embora esses dados sejam otimistas, no Brasil, onde o consumo médio aparente varia entre 5-10 kg per capita/ano (FAO, 2024), ainda é abaixo do recomendado pela Organização Mundial da Saúde (OMS) que é de 12 kg per capita/ano (FAO, 2016). Isso pode ser resultado de questões culturais, pouca informação sobre os benefícios nutricionais, alto valor e pouca variedade de produtos de pescado (Lopes et al., 2016).

A produção mundial de pescado em 2022 foi de 185 milhões de toneladas, batendo o recorde desde que se começou a ser contabilizado em 1950. Desse total, 90 milhões foram oriundas da pesca extrativa e 88 milhões da aquicultura (FAO, 2024). Para o ano de 2018 a captura de pescado em águas interiores do Brasil foi estimada em 220.000 toneladas, porém dados da produção marinha (FAO, 2022).

O saramunete (*Pseudupeneus maculatus*) é um peixe que apresenta corpo raso, alongado, fusiforme e com cabeça de tamanho médio. A coloração varia de acordo com a atividade, enquanto ativo, apresenta três manchas retangulares escuras ao longo da linha lateral, e quando em repouso, a coloração apresenta-se avermelhada com manchas amareladas nas margens das escamas e linhas diagonais azuladas na cabeça (Cervigón, 1993; Gosline, 1984). Essa espécie ocorre em boa parte da costa oeste do oceano Atlântico, desde New Jersey nos EUA até Santa Catarina no Brasil, habitando áreas de recife de coral, podendo o substrato dessas áreas ser arenoso ou lamoso, em profundidade média que pode chegar a 90 metros (Carvalho-Filho, 1994; Cervigón, 1993).

No Brasil, a captura do saramunete é realizada principalmente por embarcações de pesca artesanal (Campos & Oliveira, 2001). Lessa et al. (2004) comentam que o

saramunete é uma das espécies mais capturadas e de grande importância socioeconômica no estado de Pernambuco, registrando cerca de 78% do total das espécies capturadas.

O saramunete é capturado através de um apetrecho de pesca denominado covo, onde as armadilhas são lançadas entre 18 a 27 metros de profundidade (Campos & Oliveira, 2001) e recolhidas após 48 horas do lançamento (Campos, 2000). Ele é comercializado no mercado externo, principalmente nos Estados Unidos da América e na Europa (Campos, 2000; Marques e Ferreira, 2010). Contudo, o consumo dessa espécie no Brasil restringe-se principalmente na região litorânea. Na área de tecnologia do pescado alguns trabalhos já foram realizados, observando-se possibilidade de agregar valor ao saramunete com o processamento de fishburgers elaborados com farinha de trigo, banana verde e berinjela pelos bons resultados de análises físico-químicas e avaliação sensorial (Silva et al., 2016). Em outro estudo avaliou-se o rendimento corporal de saramunete e verificou-se que a retirada da pele após a remoção do filé aumenta o rendimento, podendo ser realizado com o peixe inteiro ou então sem cabeça e vísceras (Santos et al., 2016). Para o aproveitamento dos resíduos do processamento de saramunete a elaboração de silagem com adição de melaço da cana de açúcar é uma excelente alternativa por proporcionar vida útil de até 4 meses e boa composição centesimal (Jatobá & Oliveira Filho, 2017). Salsichas elaboradas com saramunete mostraram composição química, capacidade de retenção de água, textura instrumental e características microbiológicas adequadas. Além disso, esse produto foi bem aceito de acordo com a análise sensorial (Sá Junior et al., 2021).

O ato de marinar é uma das formas de conservação dos alimentos mais antigas, datando do século VII a.C. (Sallam, 2008; Duyar & Elke, 2009). O termo “marinado de pescado” é usado para definir produtos feitos com pescado fresco, congelado ou salgado, processados com salmoura, ácido orgânico comestível e sal. São considerados produtos em semiconserva e prontos para serem consumidos. Além disso, não é necessário submetê-lo ao tratamento térmico, mas é preciso armazená-los sob refrigeração (Fuentes et al., 2010). O sal e o ácido acético são utilizados com o objetivo de retardar as ações bacterianas e enzimáticas, resultando em maior vida útil e boa aceitação sensorial (Sallam et al., 2007; Yannes & Casales, 2008; Yusop et al., 2011). Geralmente para a elaboração dos marinados de pescado são usadas espécies de pequeno porte e carne escura. Caso sejam comercializadas *in natura* estas espécies apresentam baixo valor comercial, no entanto, caso sejam utilizadas para a elaboração de marinados, podem gerar um produto de alto valor agregado com boas características tecnológicas e sensoriais.

Devido a importância desse tipo de produto, estudos sobre o tema têm sido desenvolvidos. Observou-se que filés marinados de tilápia do Nilo submetidos ao ultrassom ficaram mais ácidos e com baixa atividade de água, indicando características que impedem o desenvolvimento microbiano (da Silva et al., 2022). A tenca (*Tinca tinca*) marinada não foi observada presença de *Salmonella*, *E. coli*, e *S. aureus* durante o período de 6 meses de armazenagem a 4°C, além de boa aceitação sensorial do produto (Ozogul et al., 2008). Marinados de anchovas (*Engraulis encrasicolus*) testando diferentes óleos (semente de girassol, alecrim, coentro, louro e alho) permaneceram frescos e estáveis durante 6 meses de armazenagem refrigerada (Ilpe et al., 2019). Em outro estudo, observou-se que três diferentes tipos de marinados de anchovas, dentre eles um contendo alho, permaneceram frescos, estáveis e aptos para o consumo após 7 meses de armazenagem a 4°C (Ficicilar & Genccelep, 2020).

Observou-se que o óleo essencial de tomilho causa efeito positivo nos atributos químicos e sensoriais dos marinados de filés de truta arco-íris, além de aumentar a vida útil do produto final (Yildiz, 2016). Em outro estudo, marinados de bagre Africano (*Clarias gariepinus*) elaborados com óleo de girassol e óleo de girassol com tomate preservaram as características químicas com 110° e 80° dias de armazenamento refrigerado, respectivamente (Kaya & Basturk, 2018). Esses mesmos autores elaboraram marinados de sargo (*Sparus aurata*) e, durante 200 dias de armazenagem a 4°C, não constatarem diferença na porcentagem de proteína, lipídeos, umidade e cinzas, além dos produtos apresentarem boa aceitação sensorial (Kaya & Basturk, 2015). Diferentes emulsões de azeite e suco de limão (0%, 25%, 35% e 50%) foram adicionadas em marinados de anchova (*Engraulis encrasicolus*), apresentando boa estabilidade química. O tratamento com 35% de suco de limão foi considerado o mais bem aceito, de acordo com a análise sensorial (Topuz et al., 2016). Em outro estudo, analisou-se marinados de filés de truta arco-íris (*Oncorhynchus mykiss*) utilizando quatro compostos antioxidantes (orégano, alecrim, antioxidante sintético e extrato de quilaia) (Fellenberg et al., 2020), sendo o grau de efetividade na seguinte ordem: sintético, alecrim, orégano, quilaia e controle.

Ao avaliar o uso do processamento de alta pressão hidrostática em marinados de arenque, Ucak et al. (2019) observaram que não houve desenvolvimento de bactérias psicrotróficas em amostras com 4% de ácido acético submetidos a 500 MPa, além de não ser detectada formação de histamina e cadaverina durante 3 meses de armazenagem a 4°C.



Tendo em vista que é possível adicionar vários ingredientes e obter um produto estável e diferenciado, o processo de marinar torna-se bastante interessante. Essid et al. (2019) verificaram que os marinados elaborados com extratos de casca de romã e folha de alcachofra apresentaram melhora nos aspectos sensoriais de cor e aparência, estabilidade do produto e concentração de ácidos graxos poli-insaturados. Além disso, os marinados obtiveram índices inferiores ao mínimo para causar sintomas de intoxicação de histamina que é de 100 ppm ou 100 mg/kg no Brasil. Outro estudo mostrou que, durante 240 dias de armazenamento a 4 °C, a adição de extratos de chá verde e louro em marinados de anchova fez a carga microbiológica, N-BVT (nitrogênio das bases voláteis totais) e TBARS (substâncias reativas ao ácido tiobarbitúrico) reduzirem (Fiçilar et al., 2018). No estudo de Kindossi et al. (2015) observou-se que o tempo de marinagem afetou diretamente a quantidade de bactérias viáveis e de ácido lático, enquanto o ácido cítrico afetou significativamente o pH de marinados de corvina (*Pseudolithus senegalensis*).

De acordo com esses estudos, nota-se que os marinados de pescado são produtos bastante estudados, principalmente em países da Europa, Ásia e América do Sul. Contudo, no Brasil, existem poucos estudos, principalmente no enfoque de agregar valor ao pescado de baixo valor comercial, que é um alimento de alta qualidade nutricional, fácil disponibilidade e baixo preço.

A FAO (FAO, 2019) comenta que 10,8% da população mundial encontra-se em situação de vulnerabilidade alimentar. Na América do Sul, esse percentual é de aproximadamente 5,5%. Assim, a importância de estudos sobre o desenvolvimento de alimentos como o marinado de saramunete e a contribuição que esse produto possa ter para as populações mundiais e regionais, principalmente as que se encontram em vulnerabilidade alimentar, é de extrema importância.

Diversas tecnologias inovadoras estão disponíveis para a conservação de peixes, como plasma não térmico, campos elétricos pulsados, luz pulsada, água eletrolisada e ultrassom (US). A utilização de ultrassom na tecnologia de alimentos para processamento, na esterilização, extração, congelamento e filtração, tem proporcionado menor tempo de processamento (Awad et al., 2012) e incremento na aceitação sensorial dos produtos (Leong et al., 2017). O ultrassom pode ser utilizado na indústria do pescado de maneira sinérgica com outras tecnologias, tais como o congelamento (Zhan et al., 2018; Sun et al., 2019b), descongelamento (Li et al., 2019; Li et al., 2019) e secagem (Ismail & Kocabay, 2020; Huang et al., 2020; Kocabay, 2021; Ozyalcin & Kipack., 2022). Além disso,

ultrassom pode ser um pré-tratamento para extração de substâncias através da ruptura da parede celular, que possui componentes essenciais (Armin et al., 2019; Petcharat et al., 2021). Devido a esses benefícios, estudos têm sido desenvolvidos para melhor compreensão dessa tecnologia.

Verificou-se que o uso do ultrassom de baixa frequência (40 kHz) e média intensidade (2,5 W/cm<sup>2</sup>) aumentou a segurança alimentar de asas de frango devido a diminuição de 1 log UFC/cm<sup>2</sup> na contagem de *Escherichia coli* e *Pseudomonas fluorescens* (Kordowska-Wiater & Stasiak, 2011). O uso de ultrassom também tem contribuído para ajudar na diminuição do sódio em produtos cárneos (Inguglia et al., 2017). Marinados de peito de frango contendo 75% e 100% de substituição do sódio por KCl resultou em aumento da captação de sódio e diminuição da oxidação lipídica (Inguglia et al., 2019).

A utilização do ultrassom (40kHz a 110W) em marinados de carne de coelho diminuiu o tempo de imersão no marinado, aumentou a dureza, além de ter melhorado os aspectos da cor, com aumento dos valores de L\* e diminuição de a\* (Gómez-Salazar et al., 2018). Em outro estudo, a intensidade de 200 W/L em filés de sardinha (*Sardina pilchardus*) retardou o desenvolvimento microbiano e aumentou ligeiramente a dureza do produto (Gündüz et al., 2019). O uso de diferentes frequências de ultrassom (250W/L) em marinado de azeitonas (*Engraulis encrasicolus*) determinou a vida útil dos tratamentos entre 3 e 4 meses, sugerindo que quando a intensidade de ultrassom aumenta, a vida de prateleira diminui (Ayvaz et al., 2019).

Ao estudar a influência do ultrassom na estabilidade microbiológica e físico-química em salsichas elaboradas com saramunete, foi verificado que a combinação de ultrassom e tratamento térmico obteve os melhores resultados e aumento de vida de prateleira, quando comparada aos embutidos sem aplicação de ultrassom ou calor (Macedo et al., 2021). Observou-se que filés marinados de tilápia do Nilo submetidos ao ultrassom ficaram mais ácidos e com baixa atividade de água, indicando boas características que impedem o desenvolvimento microbiano (Silva et al., 2022).

O concentrado proteico de pescado (CPP) é um produto desidratado e moído, podendo variar o teor de proteínas e apresentar ou não odor e sabor de pescado (Ordóñez-Peneda, 2005). Ele pode ser elaborado com filé ou carne mecanicamente separada, apresentando na composição final o índice proteico superior ao encontrado na matéria-prima original (Ordóñez-Peneda, 2005). O CPP tem mostrado bons resultados na

suplementação de produtos ricos em carboidratos, tais como pizzas (El-Beltagi et al., 2017) sorvetes (Shaviklo et al. 2011a; Shaviklo et al. 2011b), pães (Adeleke & Odedeji, 2010), biscoitos (Ibrahim, 2009), maionese (Sathivel et al., 2005) e bolachas (Huda et al. 2001b).

Nhoque é uma massa fresca à base de batata, prato típico italiano (Alexander, 2000), que também pode ser feito a partir de outros tubérculos como inhame (*Dioscorea spp*), farinha de trigo, derivados de trigo duro ou derivados de outros cereais, leguminosas, sem fermentação, podendo também ser apresentado seco, fresco, pré-cozido, instantâneo ou pronto para consumo (Brasil, 2022). Nesse contexto o CPP de saramunete seria uma alternativa interessante para ser incluído em massas alimentícias tipo nhoque. Pizzas foram elaboradas com diferentes concentrações (5; 7,5 e 10%) de CPP de carpa comum (*Cyprinus carpio*) em substituição a farinha de trigo. A substituição aumentou o teor proteico, dureza, gomosidade e digestibilidade. Além disso, o produto apresentou boa aceitação sensorial e diminuiu a  $A_w$ , aumentando a segurança do alimento (El-Beltagi et al., 2017). Salgadinhos de milho com adição de 18% de CPP foram bem aceitos por crianças iranianas com idades entre 7 e 12 anos, e se mantiveram estáveis durante 4 meses à temperatura ambiente (Shaviklo et al. 2013).

Pão elaborado com substituição de farinha de trigo por CPP de tilápia (5, 10, 15 e 20%) apresentou boa aceitação sensorial. Notou-se um aumento significativo no teor proteico, passando de 9 para 18%, indicando que o CPP pode ser utilizado para elaborar pão devido a boa aceitação e alto valor nutricional (Adeleke & Odedeji, 2010). Em outro estudo, a inclusão de 5% de CPP foi a melhor porcentagem de suplementação de biscoitos (Ibrahim, 2009), e essa mesma porcentagem mostrou-se ideal para emulsão de maionese (Sathivel et al. 2005).

Observou-se aumento no teor proteico de biscoitos elaborados com diferentes concentrações de CPP de sargo japonês (*Nemipterus japonicus*) (0, 5, 10 e 20%), passando de 0,6% para 11,8%, sendo que os biscoitos com adição de 10% de CPP foram mais bem aceitos (Huda et al. 2001b). Em alfajores elaborados com 15% de CPP (10% de salmão e 90% de tilápia), observou-se que essa inclusão teve boa aceitação sensorial (notas entre 7,30 e 7,96), melhorou a composição nutricional e manteve a carga microbiológica dentro dos parâmetros legais brasileiros (Kimura et al., 2017).

De acordo com Thorkelsson et al. (2009), a demanda por proteína de pescado como suplemento nutricional de alimentos com alegação funcional e produtos tipo *read-to-eat*,

tem aumentado. Assim, afirma-se que o concentrado proteico de pescado é uma excelente fonte de aminoácidos de alta qualidade e pode competir com outras fontes de proteína, como a da soja e a albumina do ovo, indicando que o uso de espécies com baixo valor comercial e subutilizadas na indústria são uma boa possibilidade para o enriquecimento proteico e nutricional de alimentos, além de estimular o consumo de pescado (Shaviklo, 2015). Duas formulações de biscoitos foram elaboradas e analisadas, uma com adição de farinha concentrado proteico de pescado (CPP) e outra sem adição de CPP. O teor proteico do CPP foi alto (85,16%), indicando um grande potencial para enriquecimento alimentar. Além disso, a adição 51% de CPP ao biscoito resultou em uma proporção de 35g de proteínas de pescado para cada 100g de biscoito, além de apresentar boa aceitabilidade (Rebouças et al., 2012).

Ao analisar CPP obtida de carne mecanicamente separada (CMS) de resíduos de filetagem de tilápia (carcaças, nadadeiras peitorais e aparas da toalete de filé), observou-se que o produto final possui maior concentração proteica em relação à CMS (85,26% e 17,48%, respectivamente), menor umidade (4,85%) e carga microbiológica dentro dos padrões permitidos pela legislação para o consumo humano (Rebouças et al., 2012). Em outro estudo realizado com CPP de CMS de resíduos de tilápia do Nilo, o nível de umidade (1,38%) também foi baixo e o de proteína (62,39%) alto quando comparada à matéria-prima (77,55% e 9,6%, respectivamente), além disso, o CPP foi bem aceito, indicando possibilidade de uso do CPP para alimentação humana (Vidal et al., 2011).

Massas frescas enriquecidas com concentrado proteico de tilápia (0%, 10%, 20% e 30%) obtiveram alto valor nutricional e boa aceitação sensorial, sendo a concentração sugerida pelos autores de 20% (Goés et al., 2016). Em outro estudo, pode-se notar que a adição de concentrado proteico de carpa comum (5% e 10%) em biscoitos melhorou os aspectos nutritivos, reológicos e sensoriais do produto final, sendo a concentração sugerida como ideal de 5% do CPP (Abul-Fadel et al., 2018). Na Indonésia, pessoas consomem a espécie de peixe cabeça de cobra (*Channa striata*) em situações de pós-operatório, com o intuito de auxiliar na recuperação devido ao fato dessa carne (ter boa qualidade nutricional). Por sua vez, concentrado proteico dessa espécie foi elaborado e utilizado na formulação de cookies, os quais obtiveram alto teor proteico, melhora na textura e na aparência dos produtos elaborados (Ikasari et al., 2020). Crianças entre 3 e 6 anos avaliaram *gabit cake*, comida tradicional da Indonésia adicionados com concentrado

proteico de *Decapterus russelli*. O produto final foi bem aceito pelas crianças, sendo o CPP responsável por aumentar o teor proteico (45,38%) (Kurniawan et al., 2020).

A quantidade de pessoas desnutridas no mundo voltou a aumentar desde 2015. Segundo a FAO, 821,6 milhões de pessoas no mundo estão desnutridas (10,8% da população mundial), e destas, 42,5 milhões encontram-se na América Latina e Caribe e 23,7 milhões na América do Sul (FAO, 2019). O CPP poderia se tornar uma ferramenta importantíssima por ser fonte de proteína de alta qualidade nutricional, barato, prático, além de poder ser adicionado no preparo de qualquer alimento.

De acordo com a FAO (FAO, 2020), o Brasil produziu cerca de 251.489 toneladas de inhame em 2018. O inhame, *Dioscorea* sp., é uma das plantas cultivadas pela humanidade mais antigas. Este tubérculo, embora seja originário da Ásia, foi com escravos africanos que chegou ao Brasil (Mesquita, 2002), sendo que na região Nordeste, com os estados de Alagoas, Bahia, Pernambuco, Paraíba, Sergipe e Maranhão, onde apresentou maior destaque (Brito et al., 2011). Isso ocorreu devido às características edafoclimáticas (Santos, 2002), além de forte influência da cultura local. A hortalica possui alto valor nutricional, pois são ricas em vitaminas A, D, C e do complexo B, amido, baixa percentagem de gordura e bom perfil de aminoácidos essenciais (Mesquita, 2002; Oliveira et al., 2011; Oliveira et al., 2006).

As pastas são alimentos muito conhecidos pelos consumidores do mundo todo. Dentre os principais tipos de pastas encontra-se o nhoque que é bastante apreciado no Brasil e no mundo. Mishra et al. (2012) propõe que a composição da estrutura da massa de nhoque seja densa, possua amido polimérico disponível, sofra gelatinização incompleta e grânulos de amido de oclusão de proteínas. Geralmente ele é elaborado com batata, embora vários estudos têm demonstrado a viabilidade de produzi-lo com outras matérias-primas, sendo o inhame uma ótima alternativa, tendo em vista o alto valor nutricional, abundância, baixo custo e alta aceitabilidade.

Assim, estudos têm sido desenvolvidos sobre esse tema. Um deles mostrou que nhoques de inhame obtiveram boa aceitação sensorial com índice de aceitabilidade acima de 90% (Carolino et al., 2007). A adição de 10% de inhame na elaboração de pães não interferiu nas características sensoriais, sugerindo que a farinha de trigo pode ser substituída pela de inhame, agregando valor nutritivo ao produto (Maziero et al., 2009). Em outro estudo, a adição de polpa de batata-doce em nhoque aumentou o tempo de cocção e a concentração de fibras (Silva et al., 2016).

A elaboração de marinados com aplicação de ultrassom e nhoque de inhame enriquecido com concentrado proteico de saramunete (*Pseudopenus maculatus*) é uma alternativa para diversificar a oferta de produtos de pescado, podendo contribuir para o aumento do consumo.

## 1.2. Referências bibliográficas da introdução

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### 1.3- Objetivos do trabalho

#### Geral

Avaliar o saramunete (*Pseudupeneus maculatus*) como matéria-prima para o desenvolvimento de produtos tecnológicos.

#### Específicos

1. Avaliar aspectos físico-químicos, nutricionais, microbiológicos e sensoriais de marinados de filés de saramunete utilizando a força ultrassônica.
2. Avaliar aspectos físico-químicos, nutricionais, microbiológicos e sensoriais de nhoques de inhame suplementados com concentrado proteico de saramunete

## 2- Artigo Científico I

### **Ultrasonic potency and exposure times on the quality of marinated spotted goatfish (*Pseudupeneus maculatus*)**

#### **ABSTRACT**

In Brazil, the average apparent fish consumption ranges from 5-10 kg per capita per year, which is below the 12 kg per capita per year recommendation by the World Health Organization. This modest consumption can be attributed to cultural factors, high cost, high perishability, and limited diversity of fish products in the market, especially ready-to-eat products. New technologies were used to obtain good-quality marinated spotted goatfish fillets in this context. The influence of ultrasound (US) potencies (110, 175, and 330 W) and application times (1, 3, and 5 min) on the physico-chemical and color characteristics and the microbiological aspects of the marinated fish were investigated. The pH tended to average 5.0 as the potency increased, whereas values below 4.7 were observed for shorter US times. The longer the US, the lower the luminosity ( $L^*$ ). The water activity and crude protein decreased, as the US potency and time increased. Microbial characteristics complied with legislation. The use of high-potency US for short times maintained a more acidic pH (below 4.5). Crude protein and yellow intensity ( $b^*$  value) stood out as the most strongly influenced by US, offering the possibility of controlling these parameters according to the market preference.

**Keywords:** marinated fish; ultrasound; value-added; new product; sustainability.

#### **INTRODUCTION**

The global apparent fish consumption grew at an average rate of 3% per year, from 1961 up to 2019, which is higher than the growth observed for other animal proteins (2.7%), except poultry meat (4.7%) (FAO 2024). The increase in fish consumption can be attributed to consumer awareness of the nutritional properties of fish (protein content and quality; polyunsaturated fatty acids - PUFA, omega-3 and 6, DHA, and EPA) and its health

benefits. However, in Brazil, the average apparent consumption ranged from 5-10 kg per capita per year (FAO 2024), which is below the recommendation of the World Health Organization (WHO) of 12 kg per capita per year (FAO 2016). This modest consumption can be attributed to cultural factors, limited information about the nutritional benefits, high cost, high perishability, and limited diversity of fish products in the market, especially ready-to-eat (Lopes et al. 2016).

Spotted goatfish is found along the western coast of the Atlantic Ocean, from Northeastern United States of America (USA) to Southeastern Brazil, inhabiting coral reef areas with sandy or muddy substrates at an average depth of 90 m (Cervigón 1993; Carvalho-Filho 1994). This fish is commercialized worldwide, mainly in the USA and Europe (Marques and Ferreira 2010). However, the consumption of this species in Brazil is limited to the coastal region, where it is sold fresh in open-air and municipal markets.

Regarding spotted goatfish technology, some added value products have been developed successfully, such as fish burgers (Silva et al. 2016) and sausage (Macedo et al. 2021). During the processing of spotted goatfish burgers, other ingredients were used (wheat flour, green banana, and eggplant) improving the quality from the physico-chemical, nutritional, and sensory points of view (Silva et al. 2016). Spotted goatfish sausages, when prepared using a combination of high temperature (80 °C/10 min) and ultrasound (US) (frequency of 37 kHz/10 min), had improved shelf life (22 days) when stored at 6 °C (Macedo et al., 2021). The removal of the skin after filleting spotted goatfish increased the yield (Santos et al., 2016). In addition, the processing of waste in the production of spotted goatfish's silage is an alternative for the whole use of the fish (Jatobá and Oliveira Filho 2017), bringing sustainable benefits.

Marinating is one of the oldest food preservation methods, dating back to the 7th century BC (Duyar and Elke 2009). The term "fish marinade" is used to define products made with fresh fish processed with brine, edible organic acid, and salt. These products are considered semi-preserved and are ready for consumption. Furthermore, they do not require thermal treatment (Fuentes et al. 2010). The preservation is through osmotic treatment, preventing and retarding undesirable bacterial and enzymatic activities, resulting in longer shelf life. In addition, it can affect the meat's texture, flavor, and structural properties, which is appreciated as confirmed by sensory evaluation (Sallam et al. 2007; Yeannes and Casales 2008).



Several innovative technologies are available for fish conservation such as non-thermal plasma, pulsed electric fields, pulsed light, electrolyzed water, and ultrasound - US (Speranza et al. 2021). The use of US in food is efficient for sterilization, freezing, and filtration of food, providing shorter processing time (Awad et al. 2012) and increased sensory acceptance of products (Leong et al. 2017). US can be used in the fish industry concomitantly with other technologies such as freezing (Sun et al. 2019), thawing (Li et al. 2019), and drying (Ismail and Kocabay 2020; Huang et al. 2020; Kocabay 2021; Ozyalcin and Kipack 2022). Therefore, the objective of this study was to investigate the nutritional, physico-chemical, and microbiological aspects of marinated spotted goatfish fillets using different times and potency of US.

## MATERIALS AND METHODS

### *Material*

#### *Reagents and ingredients*

All reagents used were of analytical grade. The ingredients used in the marinade were food-grade and were purchased from supermarkets: sunflower oil, sodium chloride, garlic powder, and black pepper.

Commercial kits from Compact Dry® (Nissui Pharmaceutical Co. Ltd., Tokyo, Japan) were used for microbiological analysis, including psychrotrophic aerobes total counts (Compact Dry® TC), *Escherichia coli* (Compact Dry® EC), coagulase-positive *Staphylococcus* (Compact Dry® XSA), and *Salmonella* sp (Compact Dry® SL).

### *Raw Materials*

Chilled, scaleless, and eviscerated spotted goatfish (*Pseudupeneus maculatus*) were obtained from the municipal markets of Recife, PE, Brazil. The fish were transported in thermal boxes with crushed ice to the Fish Technology Laboratory of the Department of Fisheries and Aquaculture at UFRPE, Recife, PE, Brazil. In the laboratory, the fish were washed with chlorinated water (5 ppm) to remove surface mucus, the heads and fins were removed, and finally, they were filleted, keeping the skin intact.

### *Methods*

#### *Marination*

After the cleaning and filleting processes, the fillets were immersed into a 3% NaCl brine solution (w/v) for 1 hour at 6 °C, using a fish-to-brine ratio of 1:1 (Capaccioni et al. 2011). Then, the fillets were removed from the brine, and placed in another container, and a marinade solution composed of 30% acetic acid (mL) and 10% salt (g) (Capaccioni et al. 2011) was added, with a marinade-to-fish ratio of 1.5:1, respectively (Sallam et al. 2007; Ozogul et al. 2009; Szymczak and Kolakowski 2012; Kaya and Basturk 2015). The fish were kept in this solution for 72 h at 6 °C. Afterward, the fillets were transferred to sterilized glass jars containing 98.7% sunflower oil (mL), 0.2% dehydrated garlic (g), 0.1% powdered black pepper (g), and 1% salt (g). After 48 hours of marinated storage, the influence of US on fish quality and safety was investigated.

### *Experimental design*

The marinated fillets were submitted to treatments with two variables, each one at three different levels using a 20 kHz probe, QR550 Ultronique® sonicator (Indaiatuba, São Paulo, Brazil). The first variable was the US potencies, which differed among treatments (110, 175, and 330 W), and the second factor was the exposure times to US, which were 1, 3, and 5 minutes. Afterward, the jars were stored at 6 °C for 24 h for stabilization of the marinade. After this period, the samples were submitted to physico-chemical and microbiological analysis.

A factorial experimental design was used (Rodrigues and Iemma 2005) with two explanatory variables (US potency and exposure time) at three different levels each, and triplicate analyses at the central point (Table 1).

<Table 1, near here>

### *Methods of analysis*

#### *Proximate composition*

The proximate composition of the marinated fish was estimated according to the AOAC official methods of analysis (AOAC 2016). First, the moisture content was determined by gravimetry in an oven at 105 °C with air circulation until constant weight. Then, the dry matter was subjected to further analysis. Crude protein was estimated using the Kjeldahl method (N x 6.25), the fat was extracted with petroleum ether using a Soxhlet

apparatus, and the ash was determined by incineration in a muffle furnace at 550 °C for 5 h.

#### *Instrumental color*

To measure the instrumental color of the samples, a portable colorimeter (Konica Minolta®, model CR-400, Tokyo, Japan) was used, and it was previously calibrated with a white standard before each analysis. It operated with a xenon lamp as the light source, illuminant C ( $Y=92.78$ ;  $x=0.3139$ ;  $y=0.3200$ ), an observation angle of 2°, and a measurement area of 8 mm diameter at 3 points on three fillets from each treatment. The color was expressed using the CIELAB system:  $L^*$  [lightness (+) lighter to (-) darker],  $a^*$  [redness (+) to greenness (-)], and  $b^*$  [yellowness (+) to blueness (-)].

#### *pH*

The pH was measured using a pH meter with an immersion electrode (R-TEC-7-MP, Tecnal®, Piracicaba, São Paulo, Brazil) in a sample of 10 g of fillets which were previously homogenized with 40 mL distilled water (Oliveira Filho et al. 2012).

#### *Total volatile basic nitrogen*

The total volatile basic nitrogen (TVBN) was determined according to Howgate (1976). Approximately 10 g of ground and homogenized fillet samples were mixed with 60 mL of 10% trichloroacetic acid (TCA) solution for 1 minute and it was allowed to rest for 2 hours. Then, the sample was filtered through a quantitative filtration paper (Unifil ø150 mm, Brazil), and pipetted (25 mL of the filtrate + 1 g of magnesium oxide) into a nitrogen Kjeldahl distillation flask. The distillate was received into a flask containing 15 mL of mixed indicator (methyl red and bromocresol green compound) and titrated with 0.02 N HCl.

#### *Water activity*

Water activity was measured at a temperature of 25 °C using an Aqualab CX-2 instrument (Decagon Devices, Pullman, WA, USA).

#### *Microbiological analysis*

Microbiological analyses were performed for *Salmonella* sp (Compact Dry SL<sup>®</sup>), *Escherichia coli* (Compact Dry EC<sup>®</sup>), and psychrotrophic aerobic count (Compact Dry TC<sup>®</sup>) using commercial kits (Nissui Pharmaceutical Co. Ltd., Tokyo, Japan). The results were compared to Brazilian food regulations (RDC 331 and IN 60) (Brasil 2019a; Brasil 2019b) to ensure compliance.

### *Statistical analysis*

The relationship between the explanatory variables and the results of the proximate composition, instrumental color, and physico-chemical characteristics were evaluated using a linear model adjustment ( $\mu = X\beta$ ), where  $\mu$  is the mean of the response variable,  $X$  is the matrix of the explanatory variable, and  $\beta$  is the parameter vector to be estimated. The explanatory variables were treated quantitatively (covariables) and were considered in the original scale and squared to consider the possibility of a non-linear association with the response variables. To select the relevant explanatory variables, the Akaike Information Criterion (Akaike 1974) was used. It was assumed that the response variables followed a normal distribution, verified by the normality of the residues by the Quantis and Shapiro-Wilk test. The analyses were performed with the aid of the free open-source R statistical software (R Core Team 2023).

## **RESULTS AND DISCUSSION**

### *Proximate composition*

The moisture contents of the control and samples following the different treatments ranged from 63.51 to 71.32 g/100 g (Table 2). There was no significant influence ( $p = 0.314$ ) of the treatments on the moisture content of the marinated spotted goatfish fillets. Therefore, this explanatory variable was not selected for regression estimation.

Different from our results, increases in moisture content have been reported in the literature during the marination of other fish species. *Cancer pagurus* meat subjected to constant US and cooking had an increase of 14 g/100 g in moisture content (Abanto et al. 2018). Similarly, hake fillets subjected to US (30 kHz) showed an increase of 8 g/100 g in the moisture content (Garrido et al. 2017), and frozen *Tenualosa ilisha* fillets treated with US (20 kHz for 10 min) exhibited an increase of 3 g/100 g in moisture content (Sarwar et al. 2019). An increase in moisture content in tilapia fillets (*Oreochromis niloticus*) marinated with US fillets (64.41 g/100 g) compared to the control (62 g/100 g) was also

observed (Silva et al. 2022). According to Barekat and Soltanizadeh (2018), changes in the moisture content can result from the cavitation motion of US which can change microstructures, fragmenting the z-lines of the myofibrils and increasing the inter-myofibrillar spaces, contributing to mass transfer. It is well-known that the moisture content can influence juiciness, texture, and stability of the product, which will affect acceptability of the food (Humaid et al. 2019). Therefore, the maintenance of the moisture content is a positive result for the marinated fish.

<Table 2, near here>

<Table 3, near here>

The crude protein of the treatments and control varied from 5.40 g/100 g up to 19.21 g/100 g (Table 2). When compared to the control, US caused a loss in crude protein, e.g., the US potency and application time affected significantly ( $p < 0.05$ ) protein levels (Table 3). As reported by Szymczak and Kołakowski (2012), during marination, there can be protein loss in the fish by lixiviation to the brine (. Although the isolated effects of potency and time were positive, the interaction was strong and negative. Therefore, there was a strong tendency for the content of crude protein to decrease as US potency and application time increased, with a reduction of approximately 18 g/100 g for low power and reduced exposure time, up to 5 g/100 g (high potency and extended exposure time) (Table 3 and Figure 1a). One hypothesis is that nitrogen protein extraction from the muscle towards the brine occurred with the application of US.

<Figure 1, near here>

Based on the results, combinations of high potency and prolonged time should be avoided to offer a product with small or no protein losses. The combined effect of 330 W with an exposure time of 5 minutes was particularly detrimental in the case of spotted goatfish marinades.

The contents of lipids of the control treatment and assays ranged from 6.08 to 10.62 g/100 g (Table 2). There was no significant influence ( $p = 0.074$ ) of US potency and exposure time on the lipid content in spotted goatfish marinades. These results are similar

to those obtained by Silva et al. (2022) with marinated tilapia fillets (*Oreochromis niloticus*). However, Ayvaz et al. (2019) observed a significant effect of US potency and exposure time on lipid contents in marinated anchovies (*Engraulis encrasicolus*), increasing from  $4.12 \pm 0.03$  g/100 g (without US) up to  $6.36 \pm 0.04$  g/100 g (500 W /2 min). These results suggest that the power, time, and equipment used can directly influence the lipid content. Therefore, fish consumers choose fish for its high protein content and lipid profile.

The mean ash contents of the control treatment and assays ranged from 2.61 up to 4.16 g/100 g (Table 2) and it was not affected ( $p = 0.082$ ) by the treatments. Similar results were found for marinated tilapia fillets (*Oreochromis niloticus*) subjected to US (100 W/10 min) (Silva et al. 2022). On the other hand, Ayvaz et al. (2019) observed that in vacuum-packaged marinated anchovies, the ash content initially decreased considerably from  $4.86 \pm 0.01$  g/100 g (control) to  $4.05 \pm 0.04$  g/100 g (250W /2 min) but then increased to  $4.96 \pm 0.05$  g/100 g (500W) and  $5.39 \pm 0.01$  g/100 g (750W). The potency used by these authors was higher than those from the present study, and it can explain the difference in results. Perhaps, in fish marinades, increasing the US potency leads to an increase in ash content, but it is essential to consider what has already been reported for lipids and proteins.

#### *Instrumental color*

The average lightness ( $L^*$  value) ranged from approximately 62 to 68 between the control treatment and assays (Table 2). The lightness of the marinades was significantly affected ( $p < 0.05$ ) by the US application time, but US potency did not have a marked influence (Table 3). The effect of US application time was negative, meaning that the longer the exposure, the lower the brightness of the marinades (Figure 1b). It is possible that the US exposure time, combined with cavitation, facilitated the transfer of mass (brine, acetic acid, and sunflower oil) to the fillets, making them darker. In addition, the cavitation movement may have caused the breaking of muscle and cellular walls in the fillets, aiding the penetration of oil into the fillets. Silva et al. (2022) also observed a decrease in  $L^*$  values in marinated tilapia fillets after US application (10 minutes). The authors attributed this change to the movement of acetic acid and salt into the meat due to the cavitation motion generated by US, which generates longitudinal waves, causing contractions and expansions of the meat tissue. At the beginning of storage (day 0), it was observed that spotted goatfish sausages subjected to US (10 minutes of exposure) became

significantly darker, while no such change was observed in the other treatments (control, heat, and heat + ultrasound) (Macedo et al. 2021).

The redness ( $a^*$  value) of spotted goatfish marinades ranged from  $-1.39 \pm 0.06$  up to  $0.18 \pm 0.96$  (Table 2) and US potency and application time did not affect it ( $p < 0.432$ ). Estimates of  $a^*$  in marinated tilapia fillets (*Oreochromis niloticus*) also did not vary significantly after US application (100 W/10 min), with an average value of 2.31 (Silva et al. 2022). No significant changes were also observed in redness for spotted goatfish sausages subjected to US (10 min) compared to control, with an average value of 8.43 (Macedo et al. 2021). It was observed that marinated anchovy (*Engraulis encrasicolus*) subjected to US potency (250 up to 750 W /2 min) resulted in similar  $a^*$  values although differing from the control (Ayvaz et al. 2019).

The yellowness scale ( $b^*$ ) of spotted goatfish marinades by control treatment was 3.45, and the values of the assays ranged from less than 4 (high US (20 a 100 kHz ) potency and long exposure time) to more than 7 (high US potency and reduced exposure time) (Table 2 and Figure 1c). Both US potency and exposure time had a significant negative effect on yellowness ( $p < 0.05$ ) (Table 3). This situation shows that it is possible to use US to achieve a color that is preferred by consumers. Additionally, US application of 330 W facilitated the penetration of marinating (acetic acid and salt) and storage liquids (sunflower oil, black pepper, salt, and garlic) into spotted goatfish fillets, making the fillets yellowish. On the other hand, in marinated tilapia fillets with US application at 110 W/10 min, it was observed that these fillets had a mean  $b^*$  value of 5.91 (Silva et al. 2022), similar to that obtained with spotted goatfish with 3-minute exposure times. One factor that may have influenced the lower value found by Silva et al. (2022) compared to the present study is the frequency used (37 kHz), which was higher than the one used in this study (20 kHz).

### *pH*

The pH values of the spotted goatfish marinade in all experiments (control and assays) were around 5.0 (Table 2). The pH was significantly influenced ( $p < 0.05$ ) by US potency and application time (Table 3). There were isolated effects and interactions between the explanatory variables. When evaluating the predictions of the different combinations of US power and application time, the pH tended to average 5.0 or higher if the power increased, whereas values below 4.7 were observed for shorter times (Figure

1 day). According to Normative Instruction No. 60 (Brasil 2019b), a food is considered acidic or acidified when the pH is below 4.4, in which it becomes safe to be consumed. Furthermore, such low pH associated with water activity lower than or equal to 0.94, the ready-to-eat food can be exempt from *Listeria monocytogenes* testing.

It was observed that the pH of sausages made with spotted goatfish and subjected to different treatments differed significantly, with higher values (pH 6.92) for the control, intermediate values in treatments subjected only to US (pH 6.87) or US plus heat (pH 6.79), and lower pH (6.76) when subjected to high temperatures (Macedo et al. 2021). In marinated tilapia fillets (*Oreochromis niloticus*) subjected to US (100 W/10 min), there was also no significant difference compared to the control, although they maintained a pH below 4.5, thus inhibiting bacterial growth (Silva et al. 2022). Since it is important to maintain a more acidic pH in marinades, and if this is a major criterion in selecting a processing protocol, the recommendation would be to apply high-potency (20 a 100 kHz) ultrasound for short periods. The cavitation generated by ultrasound allows greater mass transfer, making the transfer of acetic acid in the marinating liquid and salt more efficient.

#### *Total volatile basic nitrogen (TVBN)*

The mean values for TVBN of the spotted goatfish marinades of the control treatment was 5.01 mg N/100 g and the different treatments ranged from 4.6 (110 W/1 min) up to 6 mg N/100 g (175 W/3 min) (Table 2). These values are by the Brazilian legislation, which limits TVBN in fish to 30 mg N/100 g (Brasil 2017). There was no significant influence ( $p=0.090$ ) of US potency and application time on TVBN. Similarly, US did not influence TVBN in marinated tilapia fillets (9.83 to 12.85 mg N/100 g) (Silva et al. 2022). However, Ayvaz et al. (2019) found that US-marinated anchovy fillets (*Engraulis encrasicolus*) had higher TVBN (9.18 mg/100 mg) compared to control (2.05 mg/100 mg). They also found that the higher the US potency, the higher the TVBN. Macedo et al. (2021) also observed higher TVBN values in spotted goatfish meat submitted to US and heat treatments.

#### *Water activity*

The mean values of water activity ranged from 0.944 to 0.957 among treatments (Table 2). Only the US application time had an effect ( $p<0.05$ ) on water activity (Table 3), with a tendency for the water activity to decrease as the US exposure time increased



(Figure 1e). Similarly, Gündüz et al. (2019) observed that the US application potency (200, 300, and 500 W/2 min) did not affect the water activity of sardine fillets (*Sardina pilchardus*). The same situation was reported by Ayvaz et al. (2019) for marinated anchovy (*Engraulis encrasicolus*). It is important to emphasize that spotted goatfish fillets are approximately 78% moisture (Sá Junior et al. 2020), and the water activity can be affected due to water loss, gain, or migration (Amit et al. 2017). Furthermore, a portion of the bound water is associated with tissues, while the remaining portion acts as a solvent, referred to as free water, which is available for the multiplication of microorganisms and biochemical reactions that degrade the fish muscle tissue (Tadapaneni et al. 2017). The latter can be used as a growth medium for undesirable microorganisms (*Salmonella*, *E. coli*, *Staphylococcus aureus*, *vibrio*, and *Listeria*), which affect the quality and safety of the product, as well as consumer health. Jatobá and Oliveira Filho (2017) reported that water activity values between 0.98 and 0.88 are more conducive to the development of these microorganisms in fish. Therefore, based on the water activity observed in the marinated spotted goatfish, it is characterized as a product where the growth of spoilage microorganisms can occur.

### *Microbiological analyses*

Microbiological analyses were performed at the beginning of the experiment and after 30 and 60 days of storage at 6 °C of the marinated spotted goatfish. In both storage periods, *Salmonella* sp. was absent. After 30 and 60 days, the results for *Escherichia coli* were < 2 log UFC/g, and psychrotrophic bacteria were < 2 log UFC/g indicating levels by the Brazilian legislation (Brasil 2019a; Brasil 2019b).

The use of US has been reported to inhibit microbial growth. Carp fillets (*Ctenopharyngodon idella*) treated with US significantly lower microbial counts compared to the control group (Shi et al. 2019). The inactivation of microbial growth or the reduction in microbial load is one of the objectives of US use in food (Speranza et al. 2021). Therefore, the marinated spotted goatfish was stable, showing microbiological characteristics within the legislation.

## **CONCLUSIONS**

The use of high-potency US for short times was beneficial in the production of marinated spotted goatfish fillets, as it maintained a more acidic pH (below 4.5). The

protein content and yellow intensity ( $b^*$  value) stood out as the most strongly influenced by US, offering the possibility of controlling these parameters according to the market preference.

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Table 1. Complete experimental design  $2^2$  with 3 central points to investigate the influence of ultrasound potencies and application times in spotted goatfish marinades.

| <b>Assay</b> | <b>Ultrasound potency<br/>(W)</b> | <b>Application time<br/>(min)</b> |
|--------------|-----------------------------------|-----------------------------------|
| 1            | 110                               | 1                                 |
| 2            | 110                               | 5                                 |
| 3            | 330                               | 1                                 |
| 4            | 330                               | 5                                 |
| 5*           | 175                               | 3                                 |
| 6*           | 175                               | 3                                 |
| 7*           | 175                               | 3                                 |

\*Central points.

Table 2. Values of proximate composition, instrumental color and physico-chemical characteristics (mean  $\pm$  standard deviation) of spotted goatfish marinades submitted to ultrasound potencies (110, 175, and 330 W) and application times (1, 3, and 5 min).

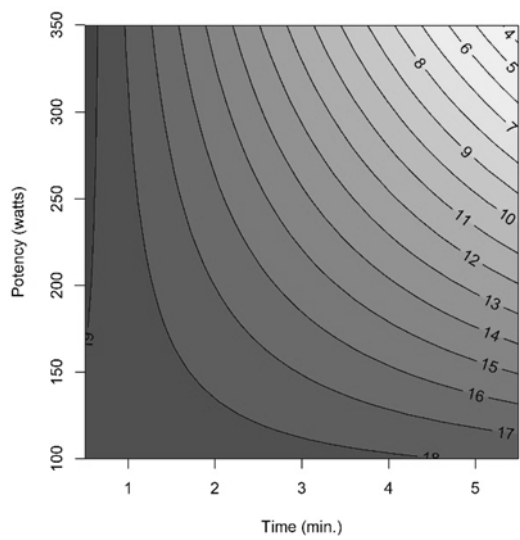
| Characteristics                                    | Values per assay <sup>1</sup> |                   |                   |                   |                   |                   |                   |                   |
|----------------------------------------------------|-------------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
|                                                    | Control                       | 1                 | 2                 | 3                 | 4                 | 5 <sup>3</sup>    | 6 <sup>3</sup>    | 7 <sup>3</sup>    |
| <i>Proximate composition (g/100 g)<sup>2</sup></i> |                               |                   |                   |                   |                   |                   |                   |                   |
| Moisture                                           | 68.22 $\pm$ 4.07              | 71.06 $\pm$ 2.32  | 68.72 $\pm$ 2.29  | 69.27 $\pm$ 2.40  | 68.61 $\pm$ 4.48  | 63.51 $\pm$ 1.89  | 71.32 $\pm$ 1.88  | 67.45 $\pm$ 2.58  |
| Protein                                            | 17.74 $\pm$ 1.95              | 17.18 $\pm$ 0.08  | 15.89 $\pm$ 1.38  | 17.30 $\pm$ 1.23  | 5.40 $\pm$ 3.08   | 19.52 $\pm$ 3.61  | 16.30 $\pm$ 1.81  | 17.32 $\pm$ 2.07  |
| Lipids                                             | 6.73 $\pm$ 1.51               | 7.92 $\pm$ 2.92   | 6.85 $\pm$ 1.72   | 6.55 $\pm$ 0.59   | 6.08 $\pm$ 2.37   | 10.62 $\pm$ 1.75  | 7.73 $\pm$ 3.78   | 7.31 $\pm$ 2.55   |
| Ash                                                | 2.85 $\pm$ 1.19               | 2.63 $\pm$ 2.01   | 4.16 $\pm$ 0.31   | 2.61 $\pm$ .55    | 3.05 $\pm$ 0.38   | 3.44 $\pm$ 0.14   | 2.96 $\pm$ 0.34   | 2.85 $\pm$ 0.10   |
| <i>Instrumental color</i>                          |                               |                   |                   |                   |                   |                   |                   |                   |
| L*                                                 | 66.15 $\pm$ 3.41              | 67.97 $\pm$ 0.30  | 65.51 $\pm$ 1.25  | 68.11 $\pm$ 2.48  | 62.43 $\pm$ 2.88  | 66.16 $\pm$ 1.11  | 65.17 $\pm$ 2.29  | 66.04 $\pm$ 4.37  |
| a*                                                 | -0.874 $\pm$ 0.87             | -0.44 $\pm$ 0.06  | -1.01 $\pm$ 0.08  | -0.86 $\pm$ 2.19  | 0.18 $\pm$ 0.96   | -1.39 $\pm$ 0.06  | -0.13 $\pm$ 0.49  | -0.39 $\pm$ 0.53  |
| b*                                                 | 3.45 $\pm$ 1.78               | 6.14 $\pm$ 1.34   | 5.32 $\pm$ 1.54   | 7.31 $\pm$ 0.56   | 3.25 $\pm$ 1.30   | 5.35 $\pm$ 1.11   | 6.34 $\pm$ 0.94   | 5.03 $\pm$ 2.29   |
| <i>Physico-chemical characteristics</i>            |                               |                   |                   |                   |                   |                   |                   |                   |
| pH                                                 | 4.82 $\pm$ 0.01               | 4.70 $\pm$ 0.03   | 4.90 $\pm$ 0.09   | 4.67 $\pm$ 0.01   | 5.06 $\pm$ 0.04   | 4.94 $\pm$ 0.01   | 4.84 $\pm$ 0.11   | 4.79 $\pm$ 0.05   |
| TVBN <sup>4</sup> (mg N/100 g)                     | 5.01 $\pm$ 0.01               | 4.60 $\pm$ 1.36   | 5.96 $\pm$ 0.63   | 5.05 $\pm$ 0.84   | 5.55 $\pm$ 1.59   | 6.02 $\pm$ 0.69   | 5.26 $\pm$ 0.73   | 5.25 $\pm$ 0.60   |
| Aw                                                 | 0.953 $\pm$ 0.01              | 0.954 $\pm$ 0.001 | 0.947 $\pm$ 0.011 | 0.956 $\pm$ 0.002 | 0.949 $\pm$ 0.006 | 0.944 $\pm$ 0.004 | 0.957 $\pm$ 0.001 | 0.956 $\pm$ 0.003 |

<sup>1</sup>(1 = 110 W / 1 min, 2 = 110 W / 5 min, 3 = 330 W / 1 min, 4 = 330 W / 5 min, 5, 6, and 7 = 175W / 3 min).<sup>2</sup>Values expressed on a wet basis.<sup>3</sup>Central points.<sup>4</sup>Total volatile basic nitrogen

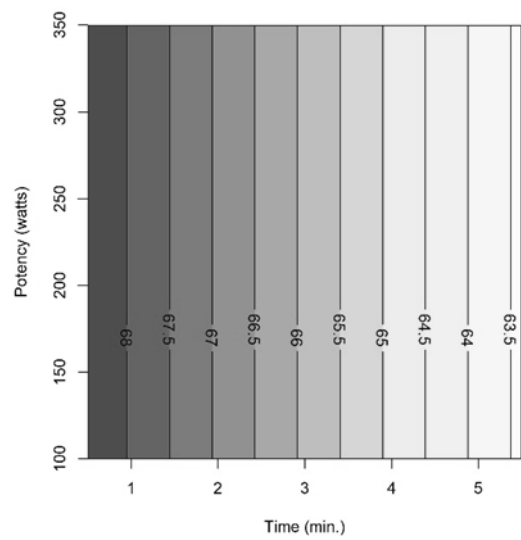


Table 3. Estimates of the adjusted models, adjusted determination coefficients ( $R^2$  - adj), and p-values.

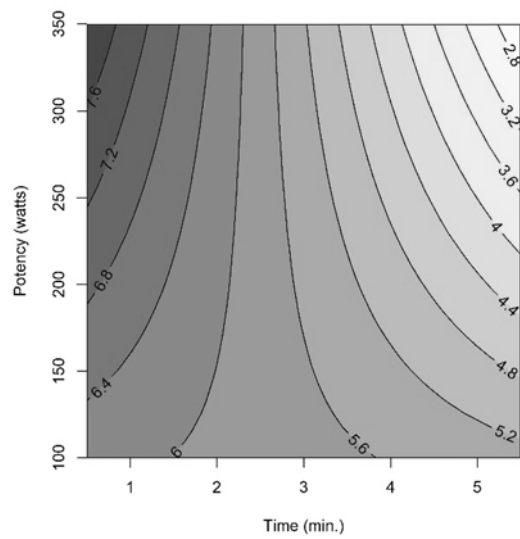
| Responses                               | Adjusted models                                                                                                         | $R^2$ -adj | p-values              |
|-----------------------------------------|-------------------------------------------------------------------------------------------------------------------------|------------|-----------------------|
| <i>Proximate composition (g/100 g)</i>  |                                                                                                                         |            |                       |
| Protein                                 | $18.098 + 1.000\text{time} + 0.008\text{potency} - 0.012*\text{time: potency}$                                          | 0.692      | $3.43 \times 10^{-5}$ |
| <i>Instrumental color</i>               |                                                                                                                         |            |                       |
| L*                                      | $68.965 - 1.018\text{time}$                                                                                             | 0.298      | 0.006                 |
| b*                                      | $5.338 + 0.201\text{time} + 0.009\text{potency} - 0.004\text{time: potency}$                                            | 0.376      | 0.011                 |
| <i>Physico-chemical characteristics</i> |                                                                                                                         |            |                       |
| pH                                      | $4.729 + 2.290 \times 10^{-2}\text{time} + 4.45 \times 10^{-4}\text{potency} + 2.24 \times 10^{-4}\text{time: potency}$ | 0.702      | $2.60 \times 10^{-5}$ |
| Aw                                      | $0.958 - 0.002\text{time}$                                                                                              | 0.147      | 0.048                 |



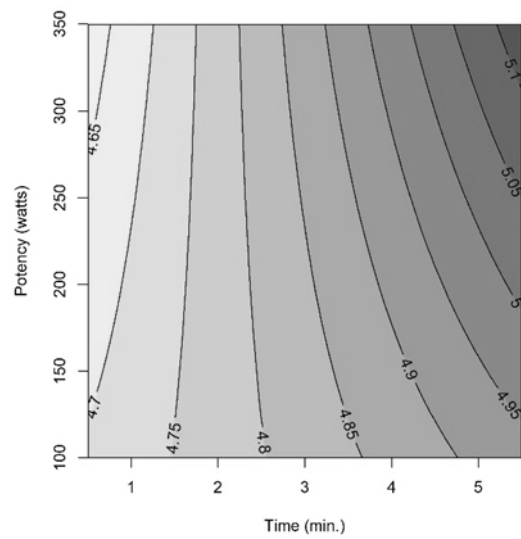
(a)



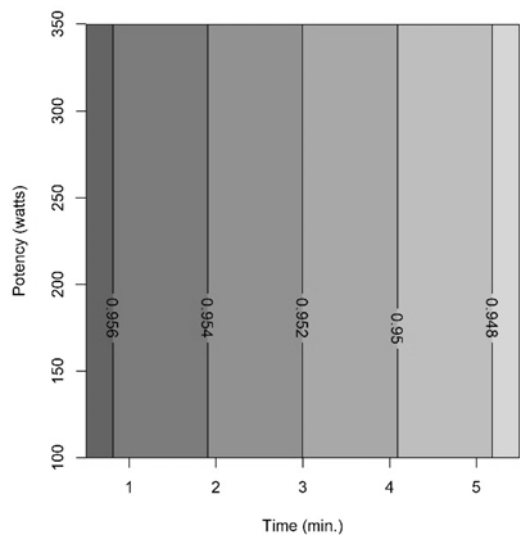
(b)



(c)



(d)



(e)

Figure 1. Response surfaces for the parameters (a) crude protein (g/100 g), (b) L\* color, (c) b\* color, (d) pH, and (e) Aw of spotted goatfish marinades submitted to different ultrasound potencies (110, 175, and 330 W) and application times (1, 3, and 5 min).

### 3- Título do Artigo Científico 2

## **Preparation and evaluation of yam gnocchi (*Dioscorea* spp.) supplemented with spotted goatfish (*Pseudupeneus maculatus*) protein concentrate**

### **ABSTRACT**

The apparent fish consumption in Brazil, currently between 5 and 10 kg/per capita/year, is still lower than the minimum recommended by the World Health Organization (12 kg/per capita/year). A viable alternative to increasing fish consumption would be producing added-value products, such as yam gnocchi added with spotted goatfish (*Pseudupeneus maculatus*) protein concentrate. This study aimed to evaluate the physicochemical, nutritional, microbiological, and sensory aspects of yam gnocchi supplemented with spotted goatfish protein concentrate (0, 7.5, and 15%). Moisture and carbohydrate content, instrumental texture, luminosity, and sensory attributes decreased with the addition of 15% SPC to yam gnocchi. On the other hand, the contents of proteins, lipids, ash, and caloric value increased significantly when compared to the control. Therefore, supplementing yam gnocchi with SPC is recommended as it improves the nutritional quality of the product, and the recommendation is the addition of up to 7.5% SPC so as not to harm the physicochemical and sensory parameters of the product.

**Keywords:** alternative consumption, fish, protein concentrate, gnocchi, pasta.

### **INTRODUCTION**

Between 1961 and 2019, apparent global fish consumption grew at an average rate of 3% per year (FAO, 2022). This increase in consumption was due to a set of factors, such as reduction in waste, optimized use, growth in demand, increase in population, economic growth of countries, and consumer awareness about the nutritional properties of fish (polyunsaturated fatty acid - PUFA, omega 3/6, docosahexaenoic fatty acid - DHA and eicosapentaenoic fatty acid - EPA), which are beneficial to health (FAO, 2022). In 2017, fish represented 17% of animal proteins; however, in Brazil, the average apparent consumption varied between 5 and 10 kg/per capita/year (FAO, 2024), values lower than those recommended by the World Health Organization (WHO), which is 12 kg/per capita/year (FAO, 2016). This modest consumption and below the dietary

recommendation can be attributed to cultural issues, little information about nutritional benefits, high price, high perishability, and low fish diversity in the market (Lopes et al., 2016), especially ready-to-eat products.

Spotted goatfish (*Pseudupeneus maculatus*) occurs on the west coast of the Atlantic Ocean, from the northeastern USA to southern Brazil, inhabiting coral reef areas, at an average depth of up to 90 meters (Carvalho-Filho, 1994; Cervigón, 1993). The species is sold on the international market, mainly in the United States of America and Europe (Campos, 2000; Marques and Ferreira, 2010), with consumption in Brazil restricted mainly to the coastal region.

In the area of fish technology, studies have already been carried out using spotted goatfish meat. spotted goatfish fishburger made with wheat flour, green banana, and eggplant resulted in a good quality product according to physicochemical analyses and sensory evaluation (Silva et al., 2016). To use waste from spotted goatfish processing, the preparation of silage with the addition of molasses is an excellent alternative as it provides a shelf life of up to 4 months and good chemical composition (Jatobá & Oliveira Filho, 2017). Spotted goatfish sausages, when submitted to the combination of high temperature (80°C/10 min) and ultrasound (37kHz/10 min), had a shelf life of 22 days when stored at 6°C (Macedo et al., 2021). In another study, sausages made with spotted goatfish were shown to have acceptable chemical composition, water-holding capacity, instrumental texture, and microbiological characteristics. Furthermore, the product was well accepted according to sensory analysis, obtaining average scores above 7 in all attributes (color, odor, texture, flavor) (Sá Junior et al., 2021).

Per capita, pasta consumption in Brazil was 5.2 kg in 2019, with the country exporting 12,625,328 kg and importing 27,613,346 kg in 2022 (ABIMAPI, 2020, 2023). In addition, pasta is a product that can be nutritionally enriched, increasing biological value, protein content, and digestibility. Gnocchi is a fresh potato-based pasta, a typical Italian dish (Alexander, 2000), which can also be made from other tubers such as yam (*Dioscorea* spp), wheat flour, durum wheat derivatives or derivatives of other cereals, legumes, without fermentation, and can also be presented dry, fresh, pre-cooked, instant or ready to eat (Brazil, 2022).

The *Dioscoreaceae* family constitutes an important food source and is distributed in tropical and subtropical regions throughout the world. In Brazil, cultivation and consumption are concentrated on family production and the surplus is for direct sale to

consumers or intermediaries for sale in urban centers (Ferreira et al., 2020). According to Shinohara et al. (2017), *Dioscorea bulbifera* L. represents an alternative food in the diet of vulnerable populations in periods of scarcity, in rural areas of the state of Pernambuco. In this research, 86% of cooked *D. bulbifera* L was used; 12.7% wheat flour, and 1.3% supporting ingredients for the gnocchi preparation.

Due to the demand for healthy, nutritious, and easy-to-prepare products, studies on pasta enriched with fish protein concentrate (FPC) have been carried out. Pizzas made with common carp flour (*Cyprinus carpio*), replacing wheat flour, achieved improvements in chemical, sensory, and texture aspects (El-Beltagi et al., 2017). The addition of FPC from snakehead fish (*Channa striata*) to cookies increased the protein, fat, and moisture content, in addition to improving the appearance, crunchiness, and texture (Ikasari et al., 2020). Pasta enriched with seabass FPC (*Dicentrarchus labrax*) resulted in an increase in protein and omega-3 polyunsaturated fatty acids. Furthermore, these products remained stable during frozen storage and were well accepted in sensory analyses, especially those in which rosemary extract was added (Ainsa et al., 2021). These data indicate the potential of FPC and the possibility of using it in pasta formulations, adding value and contributing to the increase in fish protein consumption.

The use of spotted goatfish meat, a species of fish with great technological potential, as a protein supplement in pasta has not yet been investigated. Therefore, the present study aimed to evaluate the physicochemical, nutritional, microbiological, and sensory aspects of yam gnocchi (*Dioscorea* spp.) supplemented with spotted goatfish protein concentrate.

## MATERIAL AND METHODS

### *Material*

#### *Reagents and ingredients*

All reagents were of analytical grade. The ingredients used to make the gnocchi were all food ingredients: spotted goatfish protein concentrate (SPC), yam, corn starch, whole milk, refined salt, and artisanal butter.

#### *Raw materials*

Freshly caught spotted goatfish (*P. maculatus*) were used, being purchased in fishmongers of Recife/PE, northeast of Brazil. Fish were placed in thermal boxes with flaked ice and transported to the Laboratory of Fish Technology of the Department of

Fishery and Aquaculture at UFRPE, Recife, PE. In the laboratory, fish were washed with chlorinated water (5 ppm) to remove surface mucus, and the scales, viscera, fillets, skin, and fins were removed, and the spines were reserved, which were kept cooled at 6°C until obtaining the mechanically separated meat (MSM).

### *Methods*

#### *Obtaining MSM*

To obtain MSM, spotted goatfish spines were processed by a mechanical deboning machine (PV® 150) and the MSM obtained was packaged in 500g bags and stored in the freezer (-20°C) until the process of obtaining the spotted goatfish protein concentrate (SPC), which was carried out according to Amaral et al. (2021).

#### *Gnocchi preparation*

Gnocchi was prepared according to formulations used by Malucelli et al. (2009) and Carolino et al. (2007) with modifications (Table 1). Three treatments were tested to supplement gnocchi with SPC to replace cooked yam (0%, 7.5% and 15%).

Table 1. The formulation for making 1000g of gnocchi with spotted goatfish protein concentrate (SPC) supplementation (0%, 7.5%, and 15%).

| Ingredients (g)   | Addition of SPC (%) |             |             |
|-------------------|---------------------|-------------|-------------|
|                   | 0                   | 7.5         | 15.0        |
| SPC               | -                   | 75          | 150         |
| Yam               | 869                 | 794         | 719         |
| Cornstarch (66%)  | 66                  | 66          | 66          |
| Whole milk (5.5%) | 55                  | 55          | 55          |
| Refined salt (1%) | 10                  | 10          | 10          |
| <i>Total</i>      | <i>1000</i>         | <i>1000</i> | <i>1000</i> |

Yams were washed with chlorinated water (200 ppm), peeled, and cooked in boiling water for 30-40 minutes until they reached a soft puree consistency. Subsequently, they were kneaded and manually mixed with the other ingredients according to the treatment. After mixing, 100g portions were separated and manually shaped into rolls, cut every 2cm, forming gnocchi. Gnocchi was cooked in boiling water until they floated, cooled in ice water, dried, packaged in polyethylene bags, and kept frozen (-20°C) until physicochemical, microbiological, and sensory analyses.

### *Analysis methods*

#### *Proximate composition*

The proximate composition of gnocchi was determined according to the official AOAC methodology (2016). Moisture content was determined by gravimetry in an oven with air circulation at 105°C until constant weight. After analysis, dry matter was submitted to other proximate composition analyses. Crude protein was determined using the Kjeldahl method ( $N \times 6.25$ ), fat was extracted with petroleum ether in a Soxhlet extractor, and ash content was determined by incineration in a muffle furnace at 550°C for 5 hours. The carbohydrate percentage of gnocchi was determined by subtracting from 100% the moisture, protein, fat, and ash percentage, and the caloric value was determined by multiplying the protein and carbohydrate percentage by 4 and fat by 9 (Zenebon et al., 2008).

#### *Water activity*

Water activity was determined in gnocchi pre-homogenized in a food processor at a temperature of 25°C in the Aqualab CX-2 apparatus (Decagon Devices).

#### *Instrumental texture profile (TPA) determination*

The instrumental texture profile (TPA) was determined using a texturometer (CT3 Texture Analyzer Brookfield®). Gnocchi was compressed to 50% of the total thickness with pre-test, test, and post-test speeds of 2 mm/s at a temperature of 25°C according to Bourne (2002). The parameters analyzed were: hardness (g), cohesiveness (admission), and elasticity (mm). Hardness is the force necessary to produce a certain deformation to the product, demonstrated by the force peak during the first compression. Elasticity is the ability of the sample to recover its original height after removal of the compressive force. Cohesiveness is the extent to which a material can be deformed before rupture (Bourne, 2002).

#### *Instrumental color*

To determine the instrumental color of gnocchi, a portable colorimeter (Konica Minolta®, model CR – 400) was used, which was previously calibrated with white standard before each analysis, using a xenon lamp, illuminant C ( $Y=92.78$ ;  $x=0.3139$ ;  $y=0.3200$ ), observation angle of 2° and measurement area of 8 mm in diameter at 3 points



of three gnocchi from each treatment. The color was expressed using the color standards of the CIELAB system: L\* [lightness (+) brighter to (-) darker], a\* [color intensity from red (+) to green (-)], and b\* [color intensity from yellow (+) to blue (-)].

#### *Microbiological analyses*

Microbiological analyses of *Staphylococcus aureus*, *Salmonella* sp, *Escherichia coli*, and psychrotrophic aerobes were carried out using compact Dry® kits. The results obtained were compared with RDC 331 and IN 60 (Brazil, 2019).

#### *Sensory evaluation*

Sensory evaluation was carried out in the Sensory Analysis Laboratory of the Department of Consumer Sciences - UFRPE, provided with individual cabins with white fluorescent light. Gnocchi were cooked in water and a sample of each treatment was monadically served in random order, along with tomato sauce. To clean the taste buds between one sample and another, water and cream-cracker cookies were served. Affective acceptance tests were carried out by 80 untrained tasters, randomly recruited among UFRPE students, staff, and professors, using the methodology described by Meilgaard et al. (2006). The sensory attributes evaluated were: color, odor, texture, flavor, and overall acceptance, using a 9-point hedonic scale (9 - I liked it very much, 8 - I liked it much, 7 - I liked it moderately, 6 - I liked it slightly, 5 - I neither liked nor disliked, 4 - I disliked it slightly, 3 - I disliked it moderately, 2 - I disliked it much, 1 - I disliked it very much).

#### *Experimental design and statistical analysis*

The experimental design used was completely randomized with 3 treatments (0, 7.5%, and 15% of spotted goatfish protein concentrate), with three replicates each. The results were initially evaluated for normality (Shapiro-Wilk test), and homogeneity of variances (Bartlett test). When prerequisites were met, one-way analysis of variance (One-Way ANOVA) was used and the mean comparison post-test (Tukey test) was subsequently applied using a 5% significance level ( $P < 0.05$ ). Pearson's linear correlation analysis was also performed between variables under study and SPC concentrations. Analyses were carried out using the Jamovi 2.2.5® statistical software.

## RESULTS AND DISCUSSION

### *Proximate composition*

The average moisture content of gnocchi enriched with spotted goatfish protein concentrate (SPC) varied significantly between treatments (Table 2). The lowest moisture percentage occurred in treatment with 15% SPC. This may have occurred because SPC is a dry product with high hydration capacity, absorbing moisture from the other ingredients. Lower humidity may result in greater product stability, which would be a desirable characteristic. In cookies made with different fish protein concentrate proportions, it was observed that increasing the addition from 5 to 10% also decreased the moisture percentage from 5.6 to 5.1% (Ikasari et al., 2020). The lower moisture percentage in cookies to gnocchi in the present study may be related to the fact that cookies are dry products and the gnocchi was analyzed after cooking. However, pasta enriched with different Nile tilapia protein concentrate proportions (10, 15, and 20%) did not show significant variations in moisture content ( $37.3\% \pm 1.22$ ) (Goes et al. 2016). These products may have presented lower moisture percentages because the authors analyzed the product before cooking. In a study with fusilli-type pasta in which seabass PC was added, Calanche et al. (2019) observed that the product had around 10% moisture content. The moisture percentage in gnocchi showed a significant ( $p < 0.05$ ) negative correlation with the lipid percentage ( $r = -0.772$ ). It is important to take into account that the moisture content of products is affected by the fish species, type of pasta, and nature of the material under analysis, whether fresh, dried, cooked, or frozen.

Table 2. Moisture, ash, protein, lipid, carbohydrate contents (mean  $\pm$  standard deviation), caloric value, and water activity of gnocchi prepared with different spotted goatfish protein concentrate (SPC) percentages (0% – control; 7.5%, and 15%).

| Physicochemical analyses  | Treatments (%)     |                    |                    |
|---------------------------|--------------------|--------------------|--------------------|
|                           | 0                  | 7.5                | 15.0               |
| Moisture (g/100g)         | 69.16 $\pm$ 0.57b  | 70.14 $\pm$ 0.12a  | 65.85 $\pm$ 0.32c  |
| Protein (g/100g)          | 1.04 $\pm$ 0.04c   | 6.34 $\pm$ 0.18b   | 10.15 $\pm$ 0.52a  |
| Lipids (g/100g)           | 0.20 $\pm$ 0.05c   | 1.02 $\pm$ 0.14b   | 2.16 $\pm$ 0.17a   |
| Ashes (g/100g)            | 0.80 $\pm$ 0.05b   | 0.84 $\pm$ 0.03b   | 1.47 $\pm$ 0.02a   |
| Carbohydrates (g/100g)    | 28.8 $\pm$ 0.56a   | 21.7 $\pm$ 0.12b   | 20.4 $\pm$ 0.82b   |
| Caloric value (kcal/100g) | 121.2 $\pm$ 2.02b  | 121.2 $\pm$ 1.19b  | 141.5 $\pm$ 0.87a  |
| Water activity            | 0.956 $\pm$ 0.006a | 0.948 $\pm$ 0.007a | 0.945 $\pm$ 0.003a |

<sup>1</sup>Different letters on the same row indicate significant differences according to the Tukey test ( $p < 0.05$ ).

The protein content of gnocchi increased ( $p < 0.05$ ) with the addition of SPC (Table 2). Other authors also found that the addition of FPC considerably increased the protein content, as observed in cookies (from 7.36 to 11.4 g/100 g), pasta (from 9.7 to 18.28 g/100 g), pizzas (from 11.4 to 15.34 g/100 g), breads (from 9.73 to 19.26 g/100 g) and fusilli (from 13 to 17.5 g/100 g) (Ikasari et al., 2020; Goes et al., 2015; El-Beltagi et al., 2017; Cercel et al., 2016; Calanche et al., 2019). Therefore, the addition of fish protein concentrate to pasta provides high-quality protein rich in essential amino acids (Ryu et al., 2021; Sikorski et al., 2020).

The lipid percentage in gnocchi increased as SPC increased (Table 2). Other studies also reported an increase in lipid content in products with increase in the FPC percentage, such as that observed in cookies (from 0.49 to 1.33 g/100 g), pizzas (from 3.93 to 4.31 g/100 g), pasta (from 1.11 to 1.43 g/100 g) and fusilli (from 2.83 to 5.95 g/100 g) (Ikasari et al., 2020; El-Beltagi et al., 2017; Goes et al., 2015; Calanche et al., 2019). Furthermore, seabass protein concentrate supplementation in fusilli provided an increase of approximately 24% in polyunsaturated fatty acids compared to the standard formulation. Among fatty acids, it is important to highlight the incorporation of eicosapentaenoic (EPA) and docosahexaenoic (DHA) fatty acids (Calanche et al., 2019). These results show that the addition of FPC to food products can considerably increase the levels of fatty

acids, which are beneficial to human health, with anti-inflammatory and cardiovascular system protection properties.

The addition of 15% SPC caused an increase in the ash content of gnocchi compared to the other treatments (Table 2). Furthermore, the ash percentage showed a high positive correlation ( $p < 0.05$ ) with the percentage of lipids ( $r = 0.923$ ) and proteins ( $r = 0.844$ ). This indicates that the addition of SPC improves the nutritional aspects of gnocchi. In other studies, with different types of masses, an increase in the ash percentage was also observed by increasing FPC levels. For example, pizzas added 5-10% common carp flour showed an increase in the ash percentage from 0.89 to 0.93 g/100g (El-Beltagi et al., 2017). Goes et al. (2015) found that the addition of FPC (0, 10, 20 and 30%) to fresh pasta considerably increased the ash content (1.20 g/100 g; 2.90 g/100 g; 4.39 g/ 100 g; and 6.62 g/100 g, respectively). The ash content of fusilli enriched with protein concentrate from seabass fillets (0.98 g/100 g) was higher than that of the standard formulation (0.51 g/100 g) (Calanche et al., 2019). These values are close to those found in gnocchi in this study.

The carbohydrate content decreased significantly ( $p < 0.05$ ) with the addition of up to 7.5% SPC and remained constant ( $p > 0.05$ ) with the addition of 15% SPC (Table 2). Thus, as in the present study, fresh pasta added of 0, 10, 20 and 30% Nile tilapia PC considerably reduced the carbohydrate percentage (50.92; 46.18; 39.99 and 37.63 g/100g, respectively) (Goes et al., 2015). This same trend was observed in pizzas made with common carp flour (5; 7.5 and 10%), where a decrease in total carbohydrates was observed (81.83; 80.69, and 79.42 g/100g, respectively), according to the addition of fishmeal (El-Beltagi et al., 2017). In another study, the carbohydrate content of fusilli decreased when 20% of seabass protein concentrate (62.71 g/100 g) was added to the basic formulation (73.00 g/100 g) (Calanche et al., 2019). These data indicate that the decrease in carbohydrate concentration is directly related to the replacement of traditional flour, which is rich in carbohydrates, by fish protein concentrate, which contains low levels of this compound. Furthermore, it was observed that the yam gnocchi in the present study had lower carbohydrate concentrations compared to the fresh pasta, pizza, and fusilli analyzed in the studies mentioned above. This difference is related to the tuber used to make the dough and the fish species used to develop the protein concentrate.

The caloric value of gnocchi increased ( $p < 0.05$ ) with the addition of 15% SPC (Table 2). The caloric value showed a positive correlation with the concentration of lipids

( $r = 0.897$ ) and proteins ( $r = 0.803$ ), which are important caloric components for the product. In pizzas made with 0, 5, and 7.5% common carp PC, as in the present study, an increase in the caloric value was observed with the increase in the FPC percentage (El-Betagi et al., 2017). Fresh pasta made with wheat flour and 3% seabass PC had 352.95 kcal/100 g of energy (Ainsa et al., 2021). In another study, it was observed that fusilli prepared with the addition of 10% seabass PC (*D. labrax*) resulted in 367 kcal/100 g (Calanche et al., 2019). These values were higher when compared to those of the present study. This may be due to the variation between different types of fish and the PC percentage added to products.

The water activity ( $a_w$ ) of gnocchi did not vary significantly ( $p > 0.05$ ) with the addition of spotted goatfish protein concentrate (SPC) (Table 2). Differently, in pizzas,  $a_w$  decreased with the increase in the carp PC percentage in the dough: 0.987 (0%), 0.908 (5%), and 0.888 (10%) (El-Betagi et al., 2017). In fusilli pasta, there was no significant difference in water activity when the protein concentrate came from seabass fillet or skin (Ainsa et al., 2021a). The  $a_w$  values observed by the authors were low (approximately 0.385) (Table 2), probably because the analysis was performed with dry mass. The variations in concentrations found in the different results mentioned above may be related to the different types of products under study, whether the dough is fresh or dry, the type of protein product (flour or FPC), in addition to variation between fish species.

#### *Instrumental texture profile (TPA)*

The hardness (N) of gnocchi samples decreased significantly ( $p < 0.05$ ) in response to the increased SPC proportion in the dough (Table 3). The same trend was also observed in other products made with PC from different fish species: fresh pasta without (36.54 N) and with the addition of 10% *Dicentrarchus labrax* PC (31.44 N) (Calanche et al., 2019) and fusilli without (2.45 N) and with 3% *Thunnus obesus* PC (1.47 N) (Ainsa et al., 2021). These results indicate that the addition of fish protein concentrate makes the product softer, regardless of fish species.

The instrumental texture profile (TPA) of foods can be changed by physical and chemical factors. In the case of yam gnocchi with the addition of SPC, a strong negative correlation ( $p < 0.05$ ) was observed between hardness and the percentage of proteins ( $r = -0.941$ ) and lipids ( $r = -0.947$ ). Washing in the process of obtaining the protein concentrate results in the loss of stromal (collagen and elastin) and myofibrillar (actin and myosin)

proteins, which provide consistency and firmness to the cell structure. This new protein configuration can hydrate more easily when the product is cooked, resulting in lower hardness.

The cohesiveness and adhesiveness of gnocchi samples varied with the addition of SPC, but these changes were only significant ( $p < 0.05$ ) when the addition was at the highest treatment (15% CPS) (Table 3). Furthermore, it was possible to verify that there is a strong negative correlation between adhesiveness and concentration of proteins ( $r = -0.918$ ) and lipids ( $r = -0.929$ ), which comes from the addition of SPC. However, in pizzas made with the addition of 5; 7.5, and 10% common carp PC, no variation in cohesiveness was observed (El-Beltagi et al., 2017). This difference between results may originate from the amount of SPC added to formulations, which in the present study was higher, up to 15%, or from the differences between the concentrations and compositions of proteins and lipids from spotted goatfish and common carp. However, in general, the results indicate that PC additions equal to or less than 10% would not result in a significant change in cohesiveness or adhesiveness.

The elasticity of gnocchi samples decreased as the SPC proportion in the gnocchi dough increased (Table 3). A negative correlation ( $r = -0.778$ ) was observed between elasticity and protein percentage, resulting from the addition of SPC. The same pattern of relationship between elasticity and PC addition was observed in pizzas, with a reduction in elasticity when 10% PC was added (El-Beltagi et al., 2017). Given the negative relationship between elasticity and protein, the loss of stromal proteins (collagen and elastin) during the washing processes to obtain PC, and the replacement of yam by SPC makes the product less elastic.

Table 3. Results (mean  $\pm$  standard deviation) of the instrumental texture (hardness, cohesiveness, elasticity, stickiness) of gnocchi samples prepared with different spotted goatfish protein concentrate (SPC) percentages (0% – control; 7.5%, and 15%).

| Instrumental<br>Texture | Treatments (%)    |                    |                   |
|-------------------------|-------------------|--------------------|-------------------|
|                         | 0                 | 7.5                | 15.0              |
| Hardness (N)            | 1.23 $\pm$ 0.01a  | 0.97 $\pm$ 0.14b   | 0.53 $\pm$ 0.01c  |
| Cohesiveness            | 0.67 $\pm$ 0.01a  | 0.66 $\pm$ 0.01a   | 0.61 $\pm$ 0.12b  |
| Elasticity (mm)         | 14.60 $\pm$ 0.34a | 14.31 $\pm$ 0.29ab | 13.74 $\pm$ 0.28b |
| Adhesiveness            | 5.38 $\pm$ 0.53a  | 3.97 $\pm$ 0.92a   | 1.88 $\pm$ 0.30b  |

<sup>1</sup>Letters on the same row indicate significant differences according to the Tukey test ( $p < 0.05$ ).

#### *Instrumental color*

Lightness ( $L^*$ ) was lower ( $p < 0.05$ ) when SPC was added, both 7.5 and 15% SPC (Table 4). SPC is darker in color than yam dough, which would cause the darkening of enriched gnocchi samples. Ainsa et al. (2021) found that the luminosity of pasta added with tuna PC was lower than that of non-enriched pasta, or pasta enriched with seabass PC and that there was no significant difference in lightness between the latter two pasta samples. The results presented above indicate that the lightness of protein concentrate is influenced by the fish species used, as it depends on the muscle properties.

The increase in the SPC percentage is associated with a significant increase ( $p < 0.05$ ) in the redness ( $a^*$ ) of yam gnocchi samples (Table 4). Thus, it was observed that with the addition of SPC, lightness decreases and redness increases significantly ( $r = -0.937$ ). This may be related to the fact that the spotted goatfish meat is dark and reddish, so with greater addition of SPC, gnocchi became less luminous and redder. Pasta made with tuna protein concentrate also obtained significantly higher  $a^*$  values compared to formulations without PC addition (Ainsa et al., 2021b).

The yellowness ( $b^*$ ) differed significantly ( $p < 0.05$ ) between the control treatment and treatments with additions of 7.5% or 15% SPC, which presented yellowness greater than in the control (Table 4). As with redness ( $a^*$ ), the correlation between yellowness ( $b^*$ ) and lightness ( $r = -0.987$ ) was also negative. On the other hand, fusilli prepared with

*Dicentrarchus labrax* and *Thunnus obesus* PC showed  $b^*$  values similar to each other and lower than treatment without the addition of protein concentrate (Ainsa et al, 2021).

Therefore, it is clear that the addition of fish protein concentrate tends to change the color of pasta and derived products. Understanding the PC properties of different species and the degree of modification caused to products, if well managed, can add value according to the preferences of different consumer markets.

Table 4. Results (mean  $\pm$  standard deviation) of instrumental color  $L^*$ ,  $a^*$ , and  $b^*$  of gnocchi samples prepared with different spotted goatfish protein concentrate (SPC) percentages (0% – control; 7.5% and 15%).

| Instrumental Color | Treatments (%)    |                   |                   |
|--------------------|-------------------|-------------------|-------------------|
|                    | 0                 | 7.5               | 15.0              |
| $L^*$              | 64.09 $\pm$ 0.16a | 57.89 $\pm$ 1.32b | 57.45 $\pm$ 0.86b |
| $a^*$              | -2.83 $\pm$ 0.15c | -0.47 $\pm$ 0.33b | 0.58 $\pm$ 0.26a  |
| $b^*$              | 3.75 $\pm$ 0.15b  | 14.91 $\pm$ 1.54a | 15.99 $\pm$ 0.67a |

<sup>1</sup>Letters on the same row indicate significant differences according to the Tukey test ( $p < 0.05$ ).

#### Microbiological analyses

Cooked yam gnocchi with different SPC percentages was by Normative Instruction No. 60 (Brazil, 2019), with counts of psychrotrophic aerobes, *E. coli*, and *Staphylococcus aureus* being less than 2 CFU/g, and absence of *Salmonella*, molds, yeasts, and *Bacillus cereus*. Therefore, it was possible to verify that the product is safe in microbiological terms and suitable for human consumption. In another study, the microbiological analysis of pasta enriched with different tilapia PC levels (0, 10, 20, and 30%) also showed the absence of *Salmonella* sp., *Coliform*, and *Staphylococcus* spp. according to specific legislation ( $< 3$  MPN/g and  $< 1 \times 10^2$  CFU/g, respectively) (Goes et al., 2015). The addition of protein concentrate does not affect the microbiological quality. It is important to take into account the fish origin, especially the place of capture, storage, and handling by fishermen/middlemen, because if the quality of the raw material is compromised, the quality of the final product will also be compromised.

#### Sensory evaluation



The sensory evaluation of the color attribute of gnocchi samples varied significantly ( $p < 0.05$ ) between treatments (Table 5). Gnocchi from the control treatment (0% SPC) showed better color acceptance, receiving scores equivalent to “I liked it slightly”, while gnocchi made with the inclusion of 7.5 and 15% SPC received scores equivalent to “I neither liked nor disliked”. This shows that the acceptance of gnocchi color decreased when SPC was added. Furthermore, the sensory color showed a highly significant positive correlation ( $p < 0.05$ ) with lightness,  $L^*$  value ( $r = 0.925$ ) and negative correlation with redness,  $a^*$  value ( $r = -0.809$ ) and yellowness,  $b^*$  value ( $r = -0.889$ ), showing that the color acceptance of yam gnocchi samples improves when the product is brighter, less red and yellow. This is probably because these characteristics are close to those found in traditional gnocchi, which are made from potatoes and wheat flour. In another study; however, it was observed that the acceptance of the color of noodles prepared with different tilapia PC percentages (0, 10, 20, and 30%) remained unchanged, receiving score equivalent to “I liked it slightly” ( $6.74 \pm 0.28$ ) (Goes et al., 2015). This result may have been influenced by the mild color of the tilapia fillet (white), making it more difficult for the taster to detect the presence of PC in the product. On the other hand, the spotted goatfish meat is reddish and oily, generating protein concentrate with a darker color, which may have been noticed and partially disapproved by tasters.

Odor acceptance was greater in the control treatment (0% SPC) and decreased with the inclusion of up to 7.5% or 15% SPC (Table 5). This attribute showed a negative correlation ( $p < 0.05$ ) ( $r = -0.679$ ) with protein percentage. One way to improve the gnocchi odor could be to carry out more washing procedures when obtaining SPC.

The texture of yam gnocchi was the sensory attribute in which the greatest variation was observed regarding the inclusion of SPC. Texture acceptance was greater in yam gnocchi without SPC, which received an average score equivalent to “I liked it slightly”, changing to “I neither liked nor disliked”, in gnocchi made with the addition of 7.5% SPC and “I disliked slightly”, with the addition of 15% SPC (Table 5). This sensory attribute showed a negative correlation ( $p < 0.05$ ) with the percentage of proteins ( $r = -0.886$ ) and lipids ( $r = -0.947$ ). This may indicate that the increase in protein and fat from SPC decreased the texture acceptance of gnocchi samples. Furthermore, texture acceptance showed a positive correlation ( $p < 0.05$ ) with hardness ( $r = 0.853$ ), elasticity ( $r = 0.689$ ), and adhesiveness ( $r = 0.835$ ), showing that tasters showed greater texture acceptance in harder, more elastic, and adhesive gnocchi samples, characteristics that decrease as SPC

is added, causing the product to fall apart in the mouth. The texture of noodles made from wheat flour and supplemented with 20% Nile tilapia protein concentrate obtained a higher score (7.95) to control (7.05), 10% (6.55), and 30 % (5.98) of PC inclusion (Goes et al., 2015). The addition of 5 and 10% FPC improved the texture aspects of cookies to the control treatment, where tasters reported that products had a softer texture (Ikasari et al., 2020). However, in noodles made with sago flour (control treatment) and sago flour with the addition of skipjack tuna flour (8, 10, 12, and 14%), the texture acceptance was close to 5 points (“I neither liked nor disliked”), with no significant difference between treatments (Litaay et al., 2022).

The flavor acceptance of gnocchi decreased from the equivalent of “I liked it slightly” in the control treatment to the equivalent of “I neither liked nor disliked” for treatments with 7.5 and 15% SPC (Table 5). These results suggest that the addition of SPC concentrations lower than 7.5% to yam gnocchi can provide a better flavor for the product. Ikasari et al. (2020) observed that evaluators disliked the taste of cookies made with PC, reporting that they were more bitter compared to the control treatment (without the addition of PC).

The overall acceptance of gnocchi decreased with the inclusion of 7.5% or 15% SPC (Table 5). One of the factors that influence the purchasing of food is the difference between the idealized image and the product itself. Consumers are used to consuming traditional gnocchi, made from potatoes, whose color is yellowish-white, soft in texture, and mild in flavor. A change to this pattern is received with resistance. If the attributes of spotted goatfish gnocchi were closer to traditional ones, it is likely that the product would be better accepted and could even gain space, opening up the opportunity for possible industrial production.

Table 5. Acceptance (mean  $\pm$  standard deviation) of gnocchi samples prepared with different spotted goatfish protein concentrate (SPC) percentages (0% – control; 7.5%, and 15%).

| Attributes         | Treatments (%)   |                   |                  |
|--------------------|------------------|-------------------|------------------|
|                    | 0                | 7.5               | 15.0             |
| Color              | 6.67 $\pm$ 1.90a | 5.50 $\pm$ 2.09b  | 5.51 $\pm$ 1.98b |
| Odor               | 6.35 $\pm$ 1.83a | 5.03 $\pm$ 2.25b  | 5.39 $\pm$ 2.11b |
| Texture            | 6.39 $\pm$ 1.89a | 5.55 $\pm$ 2.01b  | 4.59 $\pm$ 2.29c |
| Flavor             | 6.13 $\pm$ 1.83a | 5.47 $\pm$ 2.26ab | 5.24 $\pm$ 2.37b |
| Overall acceptance | 6.41 $\pm$ 1.71a | 5.55 $\pm$ 1.83b  | 5.15 $\pm$ 2.00b |

<sup>1</sup>Letters on the same row indicate significant differences according to the Tukey test ( $p < 0.05$ ).

<sup>2</sup>9-point hedonic scale: 9 - I liked it very much, 8 - I liked it much, 7 - I liked it moderately, 6 - I liked it slightly, 5 - I neither liked nor disliked, 4 - I disliked it slightly, 3 - I disliked it moderately, 2 - I disliked it much, 1 - I disliked it very much.

## CONCLUSION

The addition of spotted goatfish protein concentrate (SPC) increases the protein, lipid, and ash (mineral) content in yam gnocchi, adding nutritional value to the product. The color and instrumental texture of gnocchi are influenced by the addition of SPC. The addition of up to 15% SPC does not harm the microbiological aspects of yam gnocchi, being considered suitable for human consumption. However, the sensory acceptance of gnocchi supplemented with more than 7.5% SPC was equivalent to a neutral rating (“neither liked nor disliked”), but lower than control (0% SPC). Therefore, supplementing yam gnocchi with SPC is recommended, as it improves the nutritional quality of the product, in addition to adding value and encouraging fish consumption in Brazil.

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#### 4- Considerações finais

Os estudos apresentados nesta tese demonstram o potencial uso do saramunete (*Pseudupeneus maculatus*) na elaboração de produtos tecnológicos inovadores. A aplicação do ultrassom nos filés marinados e o desenvolvimento de nhoques enriquecidos com concentrado proteico de saramunete revelaram avanços importantes na diversificação e valorização do pescado.

Os marinados de saramunete submetidos ao ultrassom mostraram que o controle das potências e dos tempos de aplicação pode melhorar aspectos físico-químicos e microbiológicos, resultando em produtos de alta qualidade e maior segurança alimentar.

Esses resultados destacam o ultrassom como uma ferramenta eficaz para a indústria de alimentos, especialmente na produção de marinados de pescado com características otimizadas para o mercado.

Além disso, a adição de concentrado proteico de saramunete aos nhoques de inhame resultou em um produto nutricionalmente superior. Isso sugere que a incorporação de ingredientes de pescado em produtos tradicionais pode ser uma estratégia viável para aumentar o valor nutricional dos alimentos e, ao mesmo tempo, estimular o consumo de pescado, especialmente em países como o Brasil, onde o consumo per capita ainda está abaixo das recomendações da Organização Mundial da Saúde.

Em suma, a elaboração de produtos tecnológicos a partir do saramunete não só contribui para a diversificação da oferta de alimentos, mas também promove o uso sustentável dos recursos pesqueiros. Esses avanços tecnológicos podem desempenhar um papel crucial no aumento do consumo de pescado, oferecendo ao mercado opções de produtos mais variados, acessíveis e nutricionalmente ricos, contribuindo para a segurança alimentar e o desenvolvimento da indústria de pescado no Brasil.

## ANEXOS

Fotos dos nhoques com diferentes adições de concentrado proteico de saramunete (CPS).



a) Nhoques sem adição de concentrado proteico de saramunete (CPS); b) nhoques com adição de 7,5% de CPS; c) nhoques com adição de 15% de CPS.