UFRRJ
INSTITUTO DE TECNOLOGIA
CURSO DE PÓS-GRADUAÇÃO EM CIÊNCIA E TECNOLOGIA DE ALIMENTOS

DISSERTAÇÃO

OBTENÇÃO E CARACTERIZAÇÃO DE PÃES SEM GLÚTEN VEGANOS FORMULADOS COM FARINHA MISTA DE ARROZ E GRÃO-DE-BICO

Mylena Rafaelle Maciel Guimarães

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RESUMO

GUIMARÃES, Mylena Rafaelle Maciel. **Obtenção e caracterização de pães sem glúten veganos formulados com farinha mista de arroz e grão-de-bico.** 2024. 72p. Dissertação (Mestrado em Ciência e Tecnologia dos Alimentos). Instituto de Tecnologia, Universidade Federal Rural do Rio de Janeiro, Seropédica, RJ, 2024.

A doença celíaca é uma doença crônica de origem autoimune que se desenvolve em indivíduos geneticamente suscetíveis devido à ingestão da proteína formadora de glúten naturalmente presente em grãos como trigo, centeio e cevada. É considerada uma doença inflamatória do intestino delgado, associada à intolerância alimentar, acometendo aproximadamente 1% da população mundial. Portanto, o tratamento mais importante é a adesão rigorosa a uma dieta totalmente isenta de glúten, que garante melhora dos sintomas e normalização dos marcadores sorológicos. A extrusão termoplástica é um processo de tratamento térmico que, através da combinação de calor, umidade e trabalho mecânico, altera as matérias-primas para fornecer novas formas e estruturas com novas propriedades funcionais e nutricionais. O processo de extrusão oferece diversas aplicações, principalmente na indústria alimentícia, produzindo produtos como cereais matinais, snacks, macarrão, farinhas pré-gelatinizadas e amidos para formulação de diversos produtos, como neste caso, para elaboração de pães. Esta pesquisa teve como objetivo caracterizar quanto às propriedades físicas, químicas e tecnológicas os pães sem glúten veganos formulados com farinha mista de arroz e de grão-de-bico cruas e extrusadas na proporção de 10, 20 e 30% de adição de grão-de-bico. Foi determinada a composição centesimal dos pães, realizado as análises de umidade, atividade de água, proteínas, lipídios, cinzas, fibra alimentar, carboidratos, granulometria e cor. Os pães foram elaborados, em seguida, foi verificado o melhor resultado em relação à textura (dureza, elasticidade, coesividade e mastigabilidade), cor (da crosta e do miolo) e volume específico e analisado os seus efeitos sobre os parâmetros texturais, cor, volume específico, atividade de água, composição centesimal e digestibilidade in vitro. Os dados foram submetidos a análise de variância (ANOVA) e as médias comparadas pelo teste de Tukey ($p < 0,05$). Foram obtidos pães isentos de glúten veganos com boa qualidade tecnológica com características semelhantes e teor de fibras superior aos pães sem glúten comercializados.

Palavras-chave: Extrusão, celíacos, panificação, glúten, nutrição.

ABSTRACT

GUIMARÃES, Mylena Rafaelle Maciel. **Obtaining and characterizing vegan gluten-free breads formulated with mixed rice and chickpea flour.** 2024. 72p. Dissertation (Master's in Food Science and Technology). Institute of Technology, Federal Rural University of Rio de Janeiro, Seropédica, RJ, 2024.

Celiac disease is a chronic disease of autoimmune origin that develops in genetically susceptible individuals due to the ingestion of the gluten-forming protein naturally present in grains such as wheat, rye and barley. It is considered an inflammatory disease of the small intestine, associated with food intolerance, affecting approximately 1% of the world population. Therefore, the most important treatment is strict adherence to a completely gluten-free diet, which guarantees improvement in symptoms and normalization of serological markers. Thermoplastic extrusion is a process that, through the combination of heat, moisture and mechanical work, alters raw materials to provide new shapes and structures with new functional and nutritional properties. The extrusion process offers several applications, mainly in the food industry, producing products such as breakfast cereals, snacks, pasta, pre-gelatinized flours and starches for the formulation of various products, as in this case, for making bread. This research aimed to characterize the physical, chemical and technological properties of vegan gluten-free breads formulated with mixed rice and raw chickpea flour and extruded in a proportion of 10, 20 and 30% addition of chickpeas. The proximate composition of the breads was determined, analyzing moisture, water activity, proteins, lipids, ash, dietary fiber, carbohydrates, granulometry and color. The breads were prepared, then the best result was verified in relation to texture (hardness, elasticity, cohesiveness and chewiness), color (crust and crumb) and specific volume and their effects on textural parameters, color, specific volume, water activity, proximate composition and in vitro digestibility. The data were subjected to analysis of variance (ANOVA) and the means were compared using the Tukey test ($p < 0.05$). Vegan gluten-free breads with good technological quality were obtained with similar characteristics and higher fiber content than commercially available gluten-free breads.

Keywords: Extrusion, celiacs, bakery, vegan, nutrition.

SUMÁRIO

1. INTRODUÇÃO GERAL

Nos últimos anos, vem crescendo o número de pessoas diagnosticadas com a doença celíaca em todo o mundo, chegando a um aumento de cinco vezes nas últimas quatro décadas (SINGH *et al*, 2018). Esta doença é de origem genética, considerada crônica, tendo como característica a inflamação do intestino delgado gerada pela intolerância ao glúten, resultando na dificuldade de absorção de nutrientes pelo intestino e provocando sintomas como vômito, falta de apetite, anemia, perda de peso, coceira, entre outros. Diante disso, a principal recomendação dietética aplicada ao paciente diagnosticado com a doença celíaca é a adesão a uma dieta isenta de glúten (MOHAMMADI *et al.*, 2015).

O glúten consiste em uma proteína presente nos cereais como trigo, centeio e cevada, e possui um papel tecnológico importante na elaboração de massas alimentícias e produtos como pães, bolos, biscoitos, atuando no aumento do volume da massa, pela retenção de ar produzido durante a fermentação. Além disso, é responsável pela textura, maciez e flexibilidade do miolo de produtos como pães e bolos (DŁUŻEWSKA; MARCINIAK-LUKASIAK; KUREK, 2015).

A farinha de arroz é a farinha mais utilizada para substituir o trigo na produção de alimentos sem glúten devido a diversos aspectos como a cor clara, sabor suave e principalmente o fato de ser hipoalérgênico. Contudo, o arroz possui baixo teor de proteínas e fibras alimentares. Desta forma, se torna primordial o acréscimo de outros ingredientes que possuam essas características para garantir um melhor valor nutricional ao produto elaborado (BURESOVÁ *et al.*, 2017). O acréscimo de leguminosas, como soja, feijão, grão-de-bico e ervilha podem ser considerados uma alternativa a ser utilizado em produtos sem glúten, principalmente por possuírem alto teor de proteínas, o que facilita tanto na melhoria do valor nutricional quanto auxilia nas propriedades tecnológicas da massa (NAQASH *et al.*, 2017).

Além de adicionar proteínas nas formulações, existem outras técnicas para que os produtos sem glúten alcancem melhores propriedades, resultados sensoriais e nutricionais, melhor textura da massa e maior volume, como adição de amidos pré-gelatinizados, fibras, enzimas e hidrocolóides (NAQASH *et al.*, 2017).

O processo de extrusão tem o potencial de mudar a estrutura, solubilidade e digestibilidade da proteína através de uma combinação de cisalhamento, calor e pressão. De acordo com Ciudad-Mulero *et al*., (2020), o processo de extrusão apresenta efeitos considerados benéficos do ponto de vista nutricional, como por exemplo, a destruição de fatores antinutricionais, a gelatinização do amido, o aumento do conteúdo de fibra e a redução da oxidação lipídica.

O desenvolvimento de produtos especiais que atendam às necessidades dos portadores da doença celíaca é um grande nicho de mercado para a indústria alimentícia. De acordo com Croall *et al*. (2019), não somente pessoas portadoras da doença celíaca se interessam nesse tipo de alimento, mas também pessoas saudáveis, o que torna ainda mais amplo o mercado de consumidores.

Em se comparando a produtos elaborados com farinha de trigo, considera-se difícil a elaboração de produtos sem glúten no ponto de vista tecnológico, principalmente os pães, visto que o glúten é o responsável pela estrutura da massa, proporcionando a elasticidade, viscosidade e capacidade de retenção de gás (MOORE *et al*., 2004). Além destas limitações, pães sem glúten normalmente apresentam baixos valores nutricionais, visto que normalmente são elaborados com cereais (refinados ou não) e gomas, com baixo ou nenhum valor protéico (KUPPER, 2005). O aumento do valor nutricional destes produtos pode ser atingido ao ser utilizado ingredientes com maior valor nutritivo, como farinhas de pseudocereais: amaranto, quinoa, ou também de leguminosas como o grão de bico (ALVAREZ-JUBETE *et al*., 2010).

Desenvolver alternativas alimentares com melhores propriedades nutricionais, tecnológicas e sensorias, utilizando a extrusão como aliada no processo de desenvolvimento destes produtos, se torna fundamental para a oferta de produtos destinados ao mercado consumidor de produtos sem glúten.

Dentro deste contexto, o estudo de formulações com diferentes proporções de grão de bico como substituição a farinha de arroz bem como a análise e caracterização das farinhas e dos pães obtidos permite otimizar o desenvolvimento de produtos cada vez melhores e aceitáveis perante os consumidores.

2. REVISÃO BIBLIOGRÁFICA

2.1 A Doença Celíaca

A doença celíaca é uma doença crônica de origem autoimune que se desenvolve em indivíduos geneticamente suscetíveis devido à ingestão da proteína formadora de glúten naturalmente presente em grãos como trigo, centeio e cevada. É considerada uma doença inflamatória do intestino delgado, associada à intolerância alimentar, acometendo aproximadamente 1% da população mundial (ESCUDERO-HERNÁNDES *et al*., 2017; MALALGODA e SIMSEK, 2017).

A suscetibilidade genética à doença celíaca está relacionada principalmente ao gene HLA (antígeno de histocompatibilidade humana), principalmente a expressão dos alelos HLA-DQA1 e HLA-DQB1, que estão presentes na superfície das células receptoras de antígenos e se ligam com alta afinidade a proteínas do glúten. Esses alelos HLA induzem os linfócitos TCD4 a reconhecer peptídeos derivados da digestão do glúten, desencadeando uma resposta autoimune que por sua vez se expressa pelo aumento de uma citocina pró-inflamatória chamada interleucina 15 (IL15). A IL15 promove a ativação, proliferação e sobrevivência de linfócitos NK (natural killer) intraepiteliais, ocasionando a destruição das células epiteliais e à atrofia das vilosidades, resultando em danos nos tecidos intestinais (ESCUDERO-HERNÁNDES *et al*., 2017; SHANNAHAN e LEFFLER, 2017).

Em adultos, a doença geralmente se apresenta com sintomas gastrointestinais como diarreia, dor e inchaço abdominal, gases, náuseas, vômitos e perda de apetite, mas também pode ocorrer com desgaste do esmalte dentário, osteoporose, artrite, infertilidade, manifestações atípicas como cefaleia, neuropatia periférica, ataxia, depressão, ansiedade e epilepsia. Associados a esses sintomas e devido à má absorção intestinal crônica de nutrientes, os pacientes diagnosticados com doença celíaca também apresentam deficiência de vitaminas do complexo B (B1-tiamina, B2-riboflavina, B3-niacina, B6-piridoxina). doxiciclina, B12-cobalamina e vitamina E. Em bebês e crianças com menos de 11 anos, os sintomas da doença são frequentemente insidiosos, como diarreia, fadiga e inchaço. Durante a puberdade, no entanto, baixa estatura, sintomas neurológicos e anemia podem ocorrer (DeGEORGE *et al*., 2017).

O principal objetivo do tratamento da doença celíaca é garantir o alívio sintomático e a recuperação da histologia intestinal, pois a atrofia e a inflamação das vilosidades permanentes estão associadas ao aumento da morbidade e ao aumento do risco de malignidade. Portanto, o tratamento mais importante é a adesão rigorosa a uma dieta totalmente isenta de glúten, que garante melhora dos sintomas e normalização dos marcadores sorológicos em apenas alguns dias ou semanas, além da recuperação da doença intestinal em cerca de 1 ou 2 anos. (WATKINS e ZAWAHIR, 2017).

No entanto, apesar de sua eficácia no manejo de doenças, a adesão a uma dieta isenta de glúten pode apresentar muitos desafios, principalmente aqueles relacionados às restrições alimentares. Além de insistir no hábito de ler rótulos de produtos industriais, comer fora também é um desafio devido ao risco de contaminação cruzada entre alimentos com e sem glúten, que pode ocorrer simplesmente manipulando esses alimentos usando as mesmas bancadas, equipamentos e utensílios (WATKINS e ZAWAHIR, 2017; LEBWHOL; SANDER e GREEN, 2017).

Após o diagnóstico, é necessário o acompanhamento regular por profissionais como médicos e nutricionistas para avaliar possíveis deficiências nutricionais, manutenção da saúde óssea e intestinal e manifestações da própria doença. O acompanhamento psicológico também é importante, pois o tratamento e o controle da doença precisam ser adaptados aos hábitos alimentares e ao convívio social do paciente (LEBWHOL *et.al*., 2017).

2.2 O papel do glúten na dieta e panificação

As proteínas que constituem os cereais são consideradas heterogêneas e classificadas em diferentes tipos que variam de acordo com a sua solubilidade, aminoácidos e peso molecular. Segundo a solubilidade, podem ser albuminas (água), globulinas (sal), prolaminas (etanol) e gluteninas (ácidos ou alcalinos ou álcool na presença de agente redutor) (HAJAS *et al*., 2017).

O glúten é uma proteína composta por prolamina, que é extensível, porém pouco elástica e glutenina que é responsável pela elasticidade, atua com a função de armazenamento se unindo ao amido no endosperma e representa cerca de 70-80% das proteínas totais de grãos como trigo, centeio e cevada (LEXHALLER; TOMPOS e SCHERF, 2017).

Nos cereais cada espécie possui um grupo prolamínico diferente, sendo gliadina a presente no trigo, hordeína na cevada e secalina no centeio. A glutenina é formada por cadeias de ligações de dissulfureto e podem possuir tanto alto peso molecular como baixo peso molecular (HAJAS *et al*., 2017; BROMILOW *et al*., 2017).

Desse modo, o glúten tem um papel tecnológico primordial, pela sua característica de elasticidade e extensibilidade, no desenvolvimento de massas, pães, bolos, atuando no aumento do volume da massa e retenção de ar, bem como nas demais propriedades. Porém, apesar de sua importância, o seu consumo pode desencadear reações adversas em indivíduos com predisposição genética, principalmente os celíacos (DŁUŻEWSKA; MARCINIAK-LUKASIAK e KUREK, 2015).

2.3 A panificação sem glúten

A indústria de alimentos, na atualidade, aposta na produção de produtos sem glúten para satisfazer esta necessidade crescente de usuários geralmente de celíacos. Entre os produtos sem glúten produzidos, o pão ocupa um lugar especial. A produção de pães sem glúten saborosos e de boa qualidade representa um grande desafio para a indústria de panificação. A receita do pão sem glúten varia dependendo dos ingredientes sem glúten utilizados (PESSANHA *et. al.,* 2021)

No mercado circulam diferentes misturas sem glúten. Os substitutos mais comuns da farinha de trigo para a produção de pão sem glúten são as farinhas de arroz e/ou de milho combinadas com amido de diferentes origens (por exemplo, batata, milho, mandioca). Esses ingredientes são os mais abundantes e menos dispendiosos. As misturas sem glúten são compostas principalmente por carboidratos e carecem de teor de proteínas. Este último não afeta apenas a quantidade diária necessária de proteínas na dieta, mas também afeta amplamente a estrutura e a qualidade do pão. No pão de trigo convencional, a estrutura celular aberta se deve à elasticidade do glúten que, após misturado com água, após amassamento, consegue reter o dióxido de carbono (CO_2) produzido pelas leveduras durante a fermentação na massa levedada, fazendo com que a massa cresça. Embora seja um componente menor do glúten, a fração do macropolímero de glúten (GMP) é considerada a principal contribuinte das propriedades elásticas observadas na massa de trigo, desempenhando um papel importante na panificação (SKENDI, 2021).

A massa sem glúten é um sistema semilíquido muito complexo que contém polissacarídeos e outros componentes formadores de estrutura, substâncias que aumentam a viscosidade e substâncias estabilizadoras da massa. É caracterizado por alta densidade e baixa elasticidade. A massa sem contém glutencontém mais água do que a massa de trigo convencional. A quantidade de água depende da natureza das matérias primas básicas, da sua capacidade de absorção de água e da granulação do as matérias primas. Além disso, o amassamento, a duração e a velocidade são muito importantes. O amassamento prolongado aumenta o volume específico do pão (ŠMÍDOVÁ, 2022). Estes autores fizeram uma compilação das características comparativas de pães feitos com farinha de trigo e elaborados com farinhas sem glúten (Tabela, 1)

Quadro 1. Diferenças generalizadas entre massa de trigo e pão e entre massa sem glúten e pão. Adaptado e modificado de acordo com os autores Šmídová, *et al*, 2022.

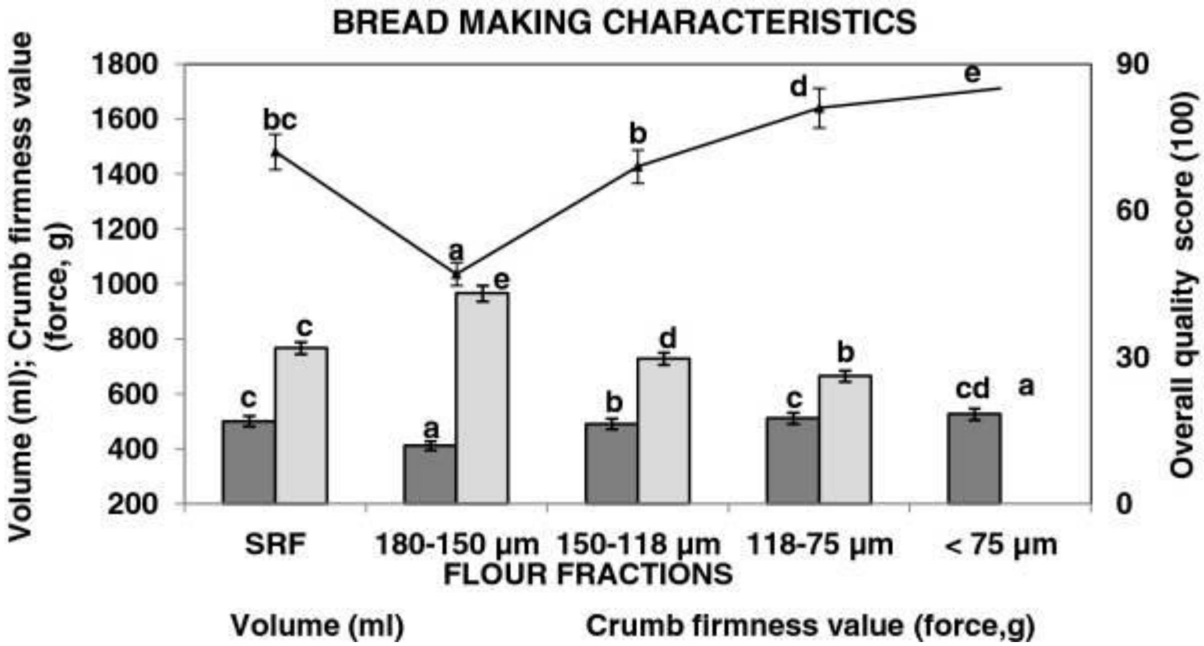
Característica	Farinhas de Trigo	Farinhas Sem Glúten
Matérias Primas		
Inchamento	Bom	Melhor
Massa		
Amassamento repetido pegajosidade.	Sim Pequeno	Não Tipicamente alto
Reometria de oscilação dinâmica		
Módulo de armazenamento G Módulo de perda G Fase superior tg (d)	Baixo Baixo Alto	Alto Alto Baixo
Extensógrafo		
Extensibilidade (E) Resistência à extensibilidade (R) Proporcionalidade R/E Área sob a curva (energia de extensibilidade)	Alto Alto Muito baixa Alta	Ruim Muito baixa Muito baixa Alta
Farinógrafo		
Tempo de desenvolvimento Estabilidade Grau de amolecimento Capacidade de ligação com água	Baixo Alta Não é uma tendência clara Muito baixa	Diferente, de acordo com a matéria-prima Diferente, de acordo com a matéria-prima Não é uma tendência clara

		Muito baixa
Pão		
Volume	Alto	Baixo
Cor da crosta	Obscuro	Suave
Crosta	Crocante	Mais úmido, denso
Elasticidade do miolo	Bom	Baixo
Porosidade	Bom	Baixo
Tamanho dos poros	Grande	Pequeno

2.4 O papel do tamanho de partícula da matéria prima em panificação

Segundo Sakhare *et. al* (2014), a qualidade de panificação das frações da farinha (Fig. 1) mostrou que o volume aumentou com a redução do tamanho das partículas da farinha. O maior volume de pão de 525 ml foi observado para a fração <75 µm e o menor volume de 410 ml para >150 µm. A farinha simples tinha volume de 500 mL. O valor de firmeza do miolo, uma medida de textura, mostrou que os pães das frações mais finas eram mais macios, conforme indicado pelo menor valor de firmeza do miolo quando comparados às frações mais grossas. A avaliação sensorial mostrou que os pães provenientes das frações de farinhas mais finas apresentaram crosta marrom-dourada, melhor formato e simetria da crosta em relação às frações mais espessas e farinha simples. A brancura da cor do miolo aumentou com a redução do tamanho das partículas. Isto pode ser devido à presença de menos partícula de farelo nas frações mais finas, conforme indicado pela diminuição do teor de cinzas de 0,93% para 0,45%. Os pães das frações mais finas apresentaram miolo fino e uniforme com paredes celulares finas, enquanto as frações mais espessas apresentaram miolo denso com paredes celulares espessas. Os pães das frações mais finas apresentaram sensação de limpeza na boca, enquanto a fração mais espessa apresentou leve formação de resíduos durante a mastigação. A qualidade do pão das frações mais finas foi melhor, conforme demonstrado pela pontuação geral de qualidade de 85,81, do que as frações mais grossas.

Figura 1. Características de panificação de farinha simples e frações de farinha



Bread Making Characteristics: Características de Fabricação do Pão; SRF: farinha simples; Flour fractions: Frações de farinha Crumb firmness value: Valor de firmeza do miolo; Overall quality score: Índice de qualidade geral.

Fonte: Sakhare SD, *et al*., (2014).

2.5. Hidrocolóides em panificação sem glúten

Os hidrocolóides são um dos ingredientes mais importantes na panificação sem glúten. Isto também se reflete ao fato de, até agora, os hidrocolóides continuam a ser o principal e mais estudado aditivo na panificação sem glúten. Estes são normalmente utilizados para melhorar o comportamento coesivo e viscoelástico da massa, formando géis. Melhoram a expansão da massa estabilizando as células gasosas e atuam como aglutinantes de água, o que também retarda a retrogradação. O efeito nas propriedades da massa dependerá da quantidade e tipo de hidrocolóide utilizado e da sua interação com outros componentes do alimento, bem como dos parâmetros do processo (temperatura, cisalhamento, pH) estabelecidos durante a fabricação do pão. As propriedades de colagem, gelatinização, inchaço e envelhecimento do amido são significativamente influenciadas pela adição de hidrocolóides (Bender *et.al.*, 2020). Na tabela 2, idealizada pelos autores (Guarda, et al., 2004) pode-se verificar os resultados dos diferentes efeitos dos hidrocoloides utilizados na elaboração de pães sem glúten.

Tabela 1. Propriedades reológicas de massa com hidrocolóides

Hidrocolóide	**Dosagem (%)**	**WA (%)**	**TDM (min)**	**Estabilidade (min)**	***P/L***	***W*** **($\times 10^{-4}$ J)**
Nenhum	–	64,6	2,5	9,3	0,72	366
κ-Carragena	0,1	65,2	3,6	7,0	1,18	374
	0,5	65,6	1,5	4,7	1,49	409
Xantana	0,1	65,2	3,5	8,3	1,40	436
	0,5	67,0	2,0	14,0	2,29	387
HPMC	0,1	66,8	3,5	7,5	1,65	335
	0,5	68,8	2,5	11,0	1,76	377
Alginato	0,1	66,0	3,0	8,5	1,24	364
	0,5	67,8	5,5	10,5	1,40	436

Os valores são a média das duplicatas; WA: absorção de água; TDM: tempo de desenvolvimento da massa; P/L: tenacidade/extensibilidade que descreveu a razão de configuração da curva; W: energia de deformação, HPMC: hidroxipropilmetillcelulose. Fonte: GUARDA, *et al*., 2004.

2.6 Grão-de-bico

O grão-de-bico (*Cicer arietinum* L.) é uma leguminosa que pode crescer em diferentes ambientes, solos e climas e é cultivada em diferentes partes do planeta, incluindo o subcontinente indiano, norte da África, Oriente Médio, sul da Europa, Ásia, Américas e Austrália. As variedades de grão de bico podem ser divididas em dois grupos: *desi* e *kabuli*. As sementes de *Kabuli* são mais utilizadas comercialmente para consumo humano por serem maiores, mais firmes, redondas e uniformes e de cor amarelo claro. As sementes de *Desi* são pequenas, enrugadas, de forma irregular e de cor escura (marrom, preta ou verde) (MOHAMMED *et.al*., 2012).

Os grãos-de-bico possuem alto teor de proteínas, fibras alimentares, carboidratos, vitaminas A, B, C, K e minerais como cálcio, fósforo, ferro e zinco, mas a composição química pode variar de acordo com as condições edafoclimáticas da região de cultivo, com variações sazonais e presença de pragas e doenças que afetam as plantas, sendo assim um alimento de importante valor nutricional (AGUILAR, *et al.*, 2015; ACHARJEE e SARMAH, 2013).

Entre os carboidratos que compõem o grão de bico estão os oligossacarídeos, como a rafinose e a estaquiose, que podem atuar como prebióticos, pois podem modular a composição microbiana intestinal e estimular o crescimento de bifidobactérias no cólon. Em relação aos lipídios, os principais componentes são os ácidos graxos insaturados, como o ácido linoléico (ω-6) e o ácido oléico (ω-9) (DADON, ABBO e REIFEN, 2017).

O grão de bico é rico em proteínas e considerado uma boa fonte de aminoácidos essenciais, especialmente a lisina. Comparando com demais leguminosas como a soja, as proteínas dos grãos possuem maior valor biológico, além de apresentar importantes propriedades funcionais como capacidade emulsificante, capacidade de formação de espuma e alta capacidade de absorção de óleo que o torna um ingrediente adequado para aplicação em produtos de panificação sem glúten, a fim de melhorar a qualidade tecnológica da massa (AGUILAR, *et al.*, 2015; DADON, ABBO e REIFEN, 2017).

2.7 Arroz

Botanicamente, o arroz (*Oryza sativa L.*) é o fruto semente de gramíneas da família *Poaceae*, subfamília *Oryzae* e *Oryza*, chamadas de grãos ou cariopse. Os grãos de arroz são compostos de casca, pele, germe e endosperma amiláceo. De acordo com a diferente submissão e processamento de grãos, o arroz pode ser dividido em: arroz integral, arroz parolizado e arroz polido (BECKER, 2010).

O arroz é considerado um produto economicamente importante, pois constitui o alimento básico para a maioria da população mundial, cultivado e consumido em todos os continentes. É uma cultura que pode se adaptar a diferentes ambientes, diferentes solos e condições climáticas. Ao ser comparado a outras culturas, o arroz fica atrás apenas do trigo (EMBRAPA, 2004).

A camada mais externa do grão contém mais proteínas, lipídios, fibras, minerais e vitaminas, enquanto o centro é composto principalmente de amido. Dessa forma, o processo de polimento do arroz acaba retirando a maior parte dos nutrientes e retendo mais amido. A composição aproximada dos grãos e suas partes também podem variar dependendo do ambiente, manuseio, processamento e condições de armazenamento (GODOY, 2013).

Durante o processo de beneficiamento, os grãos de arroz passam por diversas operações, incluindo a etapa de brunimento, com a função de retirar a epiderme, casca e gérmen, com danos mínimos ao arroz e mantendo sua forma original. O material removido durante esse processo é chamado de farelo. Além disso, o beneficiamento do arroz produz grandes quantidades de grãos quebrados, a maioria dos quais é descartada para uso em cervejas ou na produção de ração animal (GODOY, 2013).

No entanto, o valor nutricional dos grãos quebrados é semelhante ao dos grãos inteiros, o que pode conferir a esse resíduo agroindustrial significativo potencial industrial para uso como matéria-prima para a produção de farinha e amido modificado, além de reduzir custos (GODOY, 2013).

2.8 O Pão

O pão, alimento primordial para grande parte da humanidade, é definido como um produto feito de massa fermentada ou não fermentada, preparada em condições tecnicamente adequadas a partir de farinha de trigo ou outras farinhas que contêm naturalmente proteínas formadoras de glúten e água podendo conter outros ingredientes, seus tipos são denominados a partir de sua forma, ingrediente ou processo de fabricação (BRASIL, 2000). De acordo com Gray e Bemiller (2003), o pão é uma espuma sólida, resistente e instável que contém uma fase contínua que consiste em parte de glúten interligado formando uma rede de proteínas e em parte de moléculas de polímero de amido (principalmente é composto de amilose). Além de atender às definições acima, o pão de boa qualidade também deve ser organolepticamente apto para consumo.

A maioria dos produtos de panificação é composta por ingredientes que desempenham um papel específico na formação da massa. Embora o grau de importância dos ingredientes no processo de fabricação possa variar, todos eles desempenham um papel. (BORGES et. al., 2006).

Os ingredientes básicos de uma receita de pão são: farinha, fermento, sal e água. Se algum desses ingredientes estiver faltando, o produto não é pão (HOSENEY, 1991). No entanto, outros ingredientes também podem ser adicionados para alterar suas propriedades técnicas, como aumento de volume, maciez, mistura de ar ou durabilidade, ou para conferir algumas outras propriedades desejadas ao pão (EL-DASH; CABRAL; GERMANI, 1994).

2.9 Extrusão termoplástica

O processamento de farinhas por extrusão termoplástica é um procedimento que permite a pré-cocção ou cocção total de diferentes derivados de grãos naturais ou refinados, de cereais ou multicereais, pulses, raízes e tubérculos, bem como fibras, proteínas, entre outros. A extrusão é um processo de tratamento térmico que, através da combinação de calor, umidade e trabalho mecânico, altera as matérias-primas para fornecer novas formas e estruturas com novas propriedades funcionais e nutricionais (PATIL & KAUR, 2018). O processo promove a gelatinização do amido, desnaturação e redirecionamento de proteínas, inativação de enzimas, destruição de inibidores de proteases e redução da carga microbiana, resultando em um produto com propriedades físicas e geométricas pré-determinadas (PARK, *et al*, 2014).

O cozimento por extrusão apresenta alguns aspectos peculiares em relação a outros processos térmicos, notadamente porque durante a extrusão o material é submetido a um intenso cisalhamento mecânico capaz de romper as ligações covalentes dos biopolímeros. A forte ruptura estrutural que acompanha a mistura promove reações que de outra forma seriam limitadas pela difusão de reagentes e produtos (MANINGAT, 2022).

A extrusora é composta por cinco partes principais: o mecanismo de alimentação, que pode ser vertical ou horizontal; rosca ou parafuso que transporta a matéria-prima; o cilindro ou canhão utilizado para controlar a temperatura; a matriz que molda o produto na forma desejada, e o mecanismo de corte que é crucial para a formação do produto (VATANSEVER, *et al*., 2020).

O alimentador é onde é introduzida a mistura previamente homogeneizada e umedecida, caso a água de condicionamento, não seja por injecção ou gotejamento, e a alimentação deve ser mantida a uma taxa constante e contínua para manter a homogeneidade no processo. É nesta seção que os materiais são misturados e aquecidos (MAURYA, & SAID, 2014).

Uma rosca ou parafuso pode ser considerado uma parte importante de uma extrusora. Sua função é misturar a massa, conduzir a massa pelo cilindro, aumentar a temperatura e pressão através do atrito com as paredes do cilindro e forçar a massa através do molde (PLATTNER, 2009; AREAS, 1992). A velocidade de rotação, forma e tamanho, o passo das ranhuras do parafuso e a distância entre a rosca e o cano determinam a taxa de cisalhamento a ser produzida no interior do canhão e a sua respectiva transformação dos materiais, influenciando de forma direta as propriedades finais do produto extrudado (MOSIBO, *et al*., 2020).

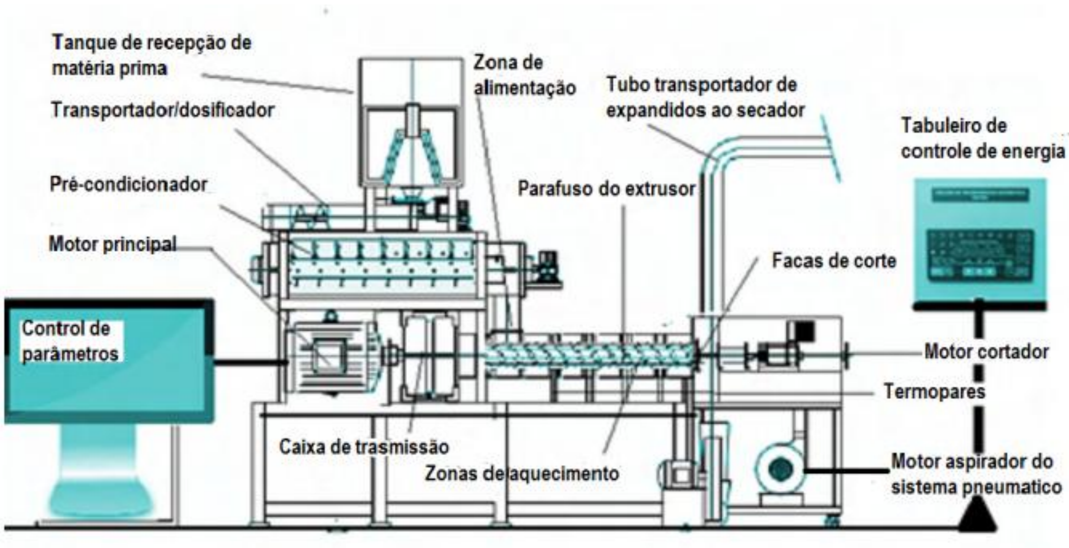
A extrusora pode ser formada de um parafuso (extrusor de parafuso único) ou dois parafusos (extrusor de parafuso duplo) bem encaixados dentro de um barril (Figura 2). As extrusoras de rosca simples são de baixo custo, limitadas a matérias-primas com umidade de 10% a 30% e baixo teor de lipídios, enquanto as extrusoras de rosca dupla são altamente versáteis, pois operam com umidade mais alta (até 90%) para funcionar com maior teor de lipídios, diferentes velocidades de rosca e seus mecanismos de transporte são independentes do atrito (ASCHERI, 2022).

O cilindro na extrusora pode possuir ranhuras, na forma de canais, ou formatos em espiral (sentidos horário ou anti-horário), na parte interna para evitar que a massa escorregue, aumentando assim a taxa de cisalhamento, ou simplesmente ser um tubo liso. Para proporcionar maior flexibilidade e controle do processo, os cilindros são encamisados para permitir a circulação de vapor, óleo quente ou água fria. As matrizes da extrusora podem ter uma variedade de configurações que afetam a forma e a textura do produto extrudado (HARPER, 1981).

Durante o processo de extrusão, devido à alta pressão e alta força de cisalhamento dentro da extrusora, a matéria-prima é convertida em massa fundida em alta temperatura, mantendo assim a água líquida. Quando esta massa fundida passa pela matriz da extrusora, na extremidade do barril ou canhão, a água líquida supersaturada evapora imediatamente devido à mudança de pressão, fazendo com que o produto mude de uma massa fundida para um produto poroso após solidificar à temperatura ambiente (PARK, *et al*., 2014).

O processo de extrusão oferece diversas aplicações, principalmente na indústria alimentícia, produzindo produtos como cereais matinais, snacks, macarrão, cereais ricos em proteínas, bebidas em pó e proteínas texturizadas de soja, farinhas pré-gelatinizadas e amidos para formulação de sopas instantâneas, molhos, doces, etc. Esses produtos geralmente são feitos de grãos, como milho e arroz, e amidos de raízes e tubérculos, como mandioca e batata (ASCHERI, 2022).

Figura. 2 Esquema de um típico sistema de extrusão.



Fonte: Ascheri, 2022

3 CONSIDERAÇÕES FINAIS

O conhecimento da arte da panificação, por milênios, permite a elaboração de uma série de produtos, quando utilizados ingredientes com altor teor de glúten. Porém, ao se tratar de elaborar produtos panificáveis, principalmente, pães sem a presença da farinha de trigo, na elaboração de pães isentos de glúten, os procedimentos de formulação mudam, e se procuram farinhas que permitam fazer uma liga semelhante à proporcionada pelo glúten, entre as mais utilizadas, a farinha de arroz, seguida de hidrocoloides alimentícios, farinhas de raízes e tubérculos (mandioca e batata, principalmente), bem como farinhas pré-cozidas por extrusão, integrais ou não. O resultado dependerá da qualidade intríseca das matérias primas, proporcionalidade dos ingredientes e do manejo da arte de se fazer pães sem glúten. A adição de farinhas de pulses, concentrados protéicos, visa melhorar o valor nutricional do produto, visto que grande parte dos alimentos sem glúten contem baixos valores de proteína total.

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CAPÍTULO I

OBTAINING AND CHARACTERIZING MIXED RICE AND CHICKPEA FLOUR FOR USE IN VEGAN GLUTEN-FREE BAKERY

Artigo submetido à revista Ciência Rural (Qualis Capes A4)

OBTAINING AND CHARACTERIZING MIXED RICE AND CHICKPEA FLOUR FOR USE IN VEGAN GLUTEN-FREE BAKERY

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ABSTRACT

This work aimed to use thermoplastic extrusion technology as a pre-treatment of flour for future use in the production of gluten-free vegan bread. The study characterized the flour according to physical characteristics (particle size analysis and color analysis) and technological characteristics (water absorption index, water solubility index, paste properties and farinography). The results indicate that the technological characterization of the flours allowed us to understand the gel formation behavior, demonstrating the specific characteristics of each one and their probable application to obtain food products.

Keywords: rice, chickpea, extrusion, gluten-free, vegan.

1. INTRODUCTION

In recent years, the number of people diagnosed with celiac disease has been growing around the world. This disease is of genetic origin, considered chronic, and is characterized by inflammation of the small intestine caused by gluten intolerance, resulting in difficulty in absorbing nutrients from the intestine and causing symptoms such as vomiting, lack of appetite, anemia, weight loss, itching, between others. Therefore, the main dietary recommendation applied to patients diagnosed with celiac disease is adherence to a gluten-free diet (MOHAMMADI et al., 2015).

Gluten is the hydrated protein fraction when wheat flour is combined with water, which gives unique viscoelastic properties to the wheat flour during the preparation of doughs for bakery products. In breads, gluten contribute to increase the volume of the dough, by retaining CO2 produced during fermentation. Subsequently, it is responsible for the texture, softness and flexibility of the bread crumb (DŁUŻEWSKA; MARCINIAK-LUKASIAK; KUREK, 2015).

Rice flour is the ingredient most used to replace wheat flour in the production of gluten-free foods due to several aspects such as its light color, mild flavor and mainly the fact that it is hypoallergenic. However, rice has a low protein and dietary fiber content. Therefore, the addition of protein-rich and fiber-rich ingredients become essential to guarantee a better nutritional value to the prepared product. (BURESOVÁ et al., 2017). The addition of pulse wholeflours such as beans, chickpeas and peas, can be considered an alternative to be used in gluten-free products, mainly because they have a high protein content, which facilitates both the improvement of nutritional value and helps with technological properties of dough (NAQASH et al., 2017).

In addition to adding proteins to formulations, there are other techniques for gluten-free products to achieve better properties, sensory and nutritional results, better dough texture and greater volume, such as adding pre-gelatinized starches, fibers, enzymes and hydrocolloids (NAQASH et al., 2017).

The extrusion process has the potential to change the structure, solubility and digestibility of protein through a combination of shear, heat and pressure. According to Mulero et al., (2020), the extrusion process presents effects considered beneficial from a nutritional point of view, such as the destruction of antinutritional factors, the gelatinization of starch, the increase in fiber content and the reduction of lipid oxidation.

The development of special products that meet the needs of people with celiac disease is a large niche market for the food industry. According to Niland et al. (2018), not only people with celiac disease are interested in this type of food, but also healthy people, which makes the consumer market even broader.

It is considered difficult to prepare gluten-free products from a technological point of view, especially breads, since gluten is responsible for the structure of the dough, providing elasticity, viscosity and gas retention capacity (MOORE et al., 2004). In addition to these limitations, gluten-free breads typically have low nutritional values, as they are normally made with cereals (refined or not) and gums, with low or no protein value (KUPPER, 2005). The nutritional increase in these products can be achieved by using ingredients with greater nutritional value, such as pseudocereal flours: amaranth, quinoa, or legumes such as chickpeas (ALVAREZ-JUBETE et al., 2010).

Developing food alternatives with better nutritional, technological and sensorial properties, using extrusion as an ally in the development process of these products, becomes fundamental for the offer of products destined for the gluten-free consumer market.

2 MATERIALS AND METHODS

2.1 Materials

Chickpea grains were supplied by the Granfino company (Nova Iguaçu, RJ, Brazil). Polished rice grains (type 2, long fine class, Palmares), olive oil, and particulate ingredients (potato starch, cassava starch, salt, sugar, dry yeast, xanthan gum, and carboxymethyl cellulose CMC), were acquired from local stores, in the Rio de Janeiro city, Brazil.

2.2 Methods

2.2.1 Treatment and experimental design

The treatments used were a mix of flours with different proportions of chickpeas as a replacement for rice flour and industrial treatments (raw and extruded). It was a design with two replications, 3x2 factorial type, with three proportions of chickpea flour (10%, 20% and 30%) and two industrial treatments (raw and extruded) as shown in Table 1.

Table 1. 3×2 factorial experimental design with 2 replications to formulate vegan gluten-free breads based on mixed rice and chickpea flour.

Test	Coded levels		Real levels	
	x_1	x_2	X_1 (Addition of chickpea flour to the formulation) %	X_2 (Type of flour in the formulation) -
1	-1	-1	10	Raw
2	0	-1	20	Raw
3	1	-1	30	Raw
4	-1	1	10	Precooked
5	0	1	20	Precooked
6	1	1	30	Precooked
7 [1]	-1	-1	10	Raw
8 [2]	0	-1	20	Raw
9 [3]	1	-1	30	Raw
10 [4]	-1	1	10	Precooked
11 [5]	0	1	20	Precooked
12 [6]	1	1	30	Precooked

x_1: quantitative variable

x_2: qualitative variable

Design model:

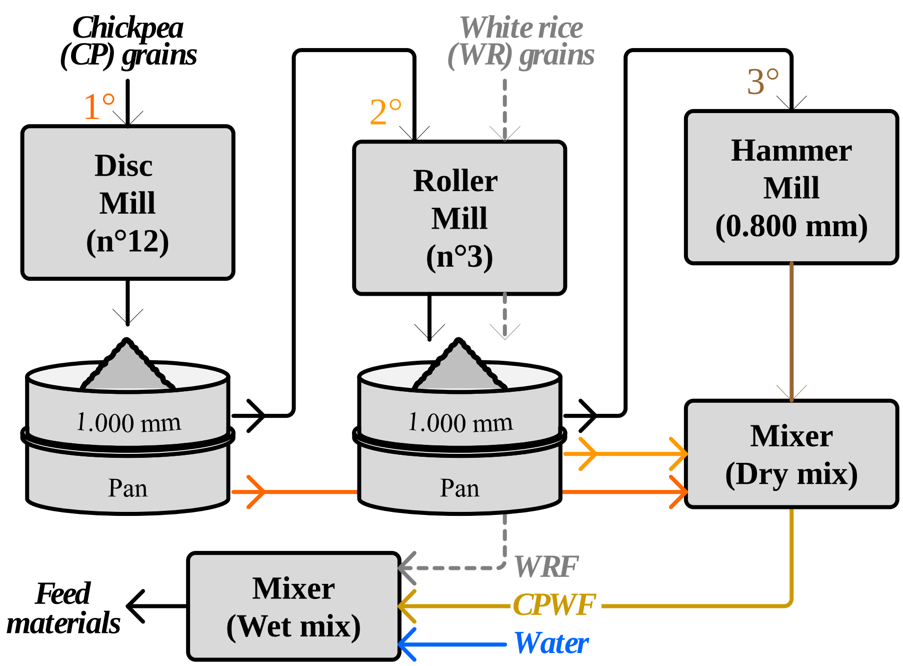
$$Y = \beta_0 + \beta_1 x_1 + \beta_2 x_2 + \beta_{12} x_1 x_2 + \beta_{11} x_1^2 + \beta_{112} x_1^2 x_2 + \epsilon$$

Where Y is the response evaluated in the flour, x_i are the independent variables at coded levels, β_i are the model coefficients that will be obtained by analysis of variance (ANOVA) and regression analysis.

2.2.2 Obtaining rice and chickpea flour

Grains were size reduced up to particles ≤ 1.00 mm using a combination of laboratory millers for chickpea(discs, rollers, and hammers), and a roller mill for white rice, according to Figure 1. All millers' characteristics were described in VARGAS-SOLÓRZANO (2019). The gap between discs was fixed at n°12, the roller millfeeder openingwas positioned at n°3, and the sieve diameter attached to the hammer mill was 0.800 mm. In the chickpea grinding, the three flour streams were mixed in a rotary mixer (MR10L, Chopin technologies, Paris, France). The resultant flours were mixedand moistened in proportions according to a factorial design, wherein eight conditions of feed material were obtained.

Figure 1 - Preparation of chickpea whole flour (CPWF), white rice flour (WRF), and feed materials for extrusion cooking.



2.2.3 Obtaining extrudates from mixed rice and chickpea flours

2.2.3.1 Moisture Conditioning

Mixed rice and chickpea flours were conditioned to 24% moisture. Humidification was carried out with distilled water sprayed with a manual sprayer onto the flour. The amount of water to be added to the samples was calculated considering the amount of flour to be extruded and its initial moisture according to Equation 1:

$$Aw = Hf - Hi / 100 - Hf \times Ms$$

In which:
Aw = amount of water to be added (mL);
Hf = final humidity of the established sample (24%);
Hi = initial sample moisture;
Ms = sample mass (g)

After humidification, the flours were stored in plastic bags at a temperature of 7º C overnight until extrusion to homogenize the moisture.

2.2.3.2 Extrusion

Mixed rice and chickpea flours were extruded at Embrapa Agroindustria de Alimentos, Rio de Janeiro – RJ, using a single screw extruder (Brabender, 19/20 DN, 832500, Duisburg, Germany) with compression ratio 3:1, a 3mm circular die. The temperature profile was set at: 40, 90 and 120°C, the screw speed at 150 rpm and the feed rate at 4 kg/h using a volumetric feeder (Brabender, 625415.394).

For extrusion of rice, it was necessary to change the extrusion parameters, aiming at the desired characteristics (without expansion), when mixing mixed flours it required more force due to the presence of chickpeas in the mixture, whereas with rice there was no need for so much strength. Therefore, a 2:1 compression ratio and a 4 mm circular die were used. The temperature profile was set at: 50, 80 and 120°C, the screw speed at 100 rpm.

After extrusion, the extrudates were collected, dried in an oven (Hauber, Joinville/SC) at 60º overnight and ground in a hammer mill (Lab Mill 3100, Perten Instruments AB, Huddinge, Sweden) then sieved on a sieve with a 0.250mm mesh and frozen, for the preparation of bread and other analyses.

2.3 Physical Characterization

2.3.1 Particle size distribution

The particle size distribution of raw and extruded flours was determined in triplicate using an S3500 series particle size analyzer (Microtrac Inc., Montgomeryville, USA) according to modified method 55–40.01 (AACC, 1999) with deionized water.

2.3.2 Color analysis

The color analysis of rice flour, chickpeas and potato and cassava starches was carried out by reflectance using a Hunterlab Colorquest colorimeter, model XE (Reston, Virginia, USA), CIELAB scale, with an opening of 1 mm in diameter, with D65/10 illuminant. The values of L* (brightness), a* (green - /red +), b* (yellow - /blue +), C* (relationship between the values of a* and b*, where the real color is obtained were measured of the analyzed object) and h (angle formed between a* and b*, indicative of the object's color saturation).

2.4 Technological Characterization

2.4.1 Water absorption index (WAI) and Water solubility índex (WSI)

Raw and extruded flours from treatments (T1, T2 and T3) and potato and cassava starches were used to evaluate the water absorption indexes (WAI) and water solubility

(WSI) in quadruplicate. The preparation of suspensions and the absorption-solubilization process were carried out according to the methodology described by ANDERSON *et al*. (1969), with some modifications indicated in VARGAS-SOLÓRZANO *et al*. (2014). The WAI and WSI calculations were carried out according to the equations described in DOGAN and KARWE (2003).

2.4.2 Pasting properties

The visco-amylographic characteristics of the raw and extruded flour samples were carried out in duplicate using the Rapid Vico Analyzer (RVA) equipment (Perten, RVA 4500, Huddinge, Sweden), using 14% humidity and 25mL of water as standards distilled according to the methodology reported by Ragaee and Abdel-Aal (2006). The values of minimum viscosity, drop in viscosity, final viscosity, tendency to retrogradation, peak temperature and gelatinization temperature were obtained.

2.4.3 Farinograph measurement

The analysis was carried out using the Farinograph© model FD0234H (Brabender, Duisburg, Germany) as described by method 54–21.01 AACC (2000b) with modifications, water was added little by little until the best consistency and hydration of the dough was obtained. The parameters studied were water absorption, dough development time (T.D.M), stability (EST), mixing tolerance index (I.T.M.) and 10 and 20 minutes of falling.

2.5 Statistical Analysis

The results of the analyzes were subjected to analysis of variance (ANOVA) and means comparison test (Tukey at 5%), with the aid of the STATISTICA statistical package version 12.0 (Statsoft Inc., Tulsa, OK, USA).

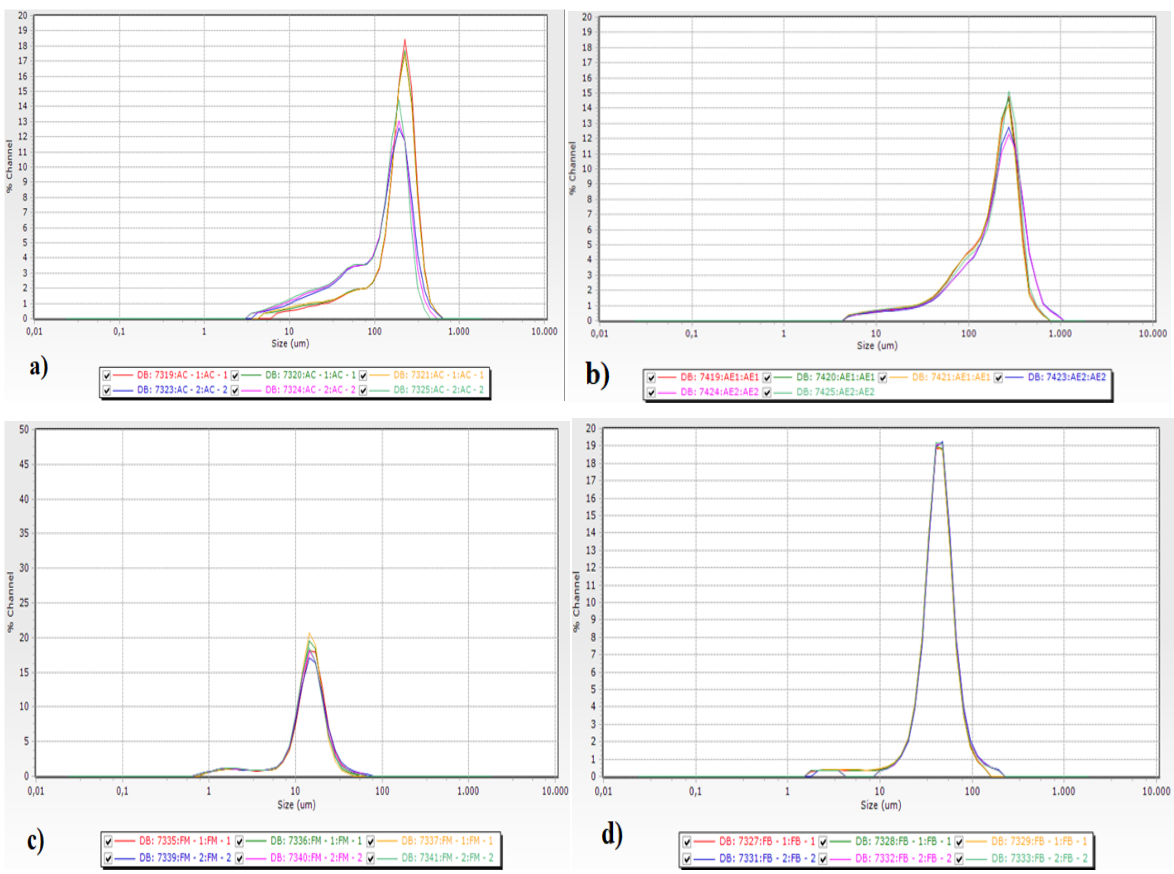
3 RESULTS AND DISCUSSION

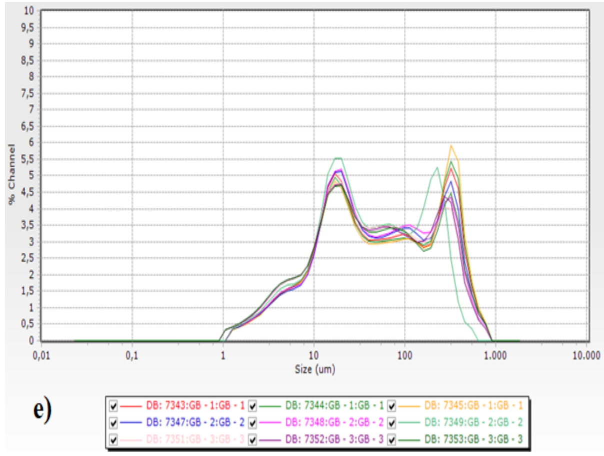
3.1 Physical Characterization

3.1.1 Particle size distribution

The particle size profile of samples RR (raw rice flour), ER (extruded rice flour), CP (chickpea), CS (cassava starch) and PS (potato starch) are shown in figure 2.

Figure 2 - Particle size analysis by laser diffraction, where: a) RR, b) ER, c) CS, d) FB and e) CP.





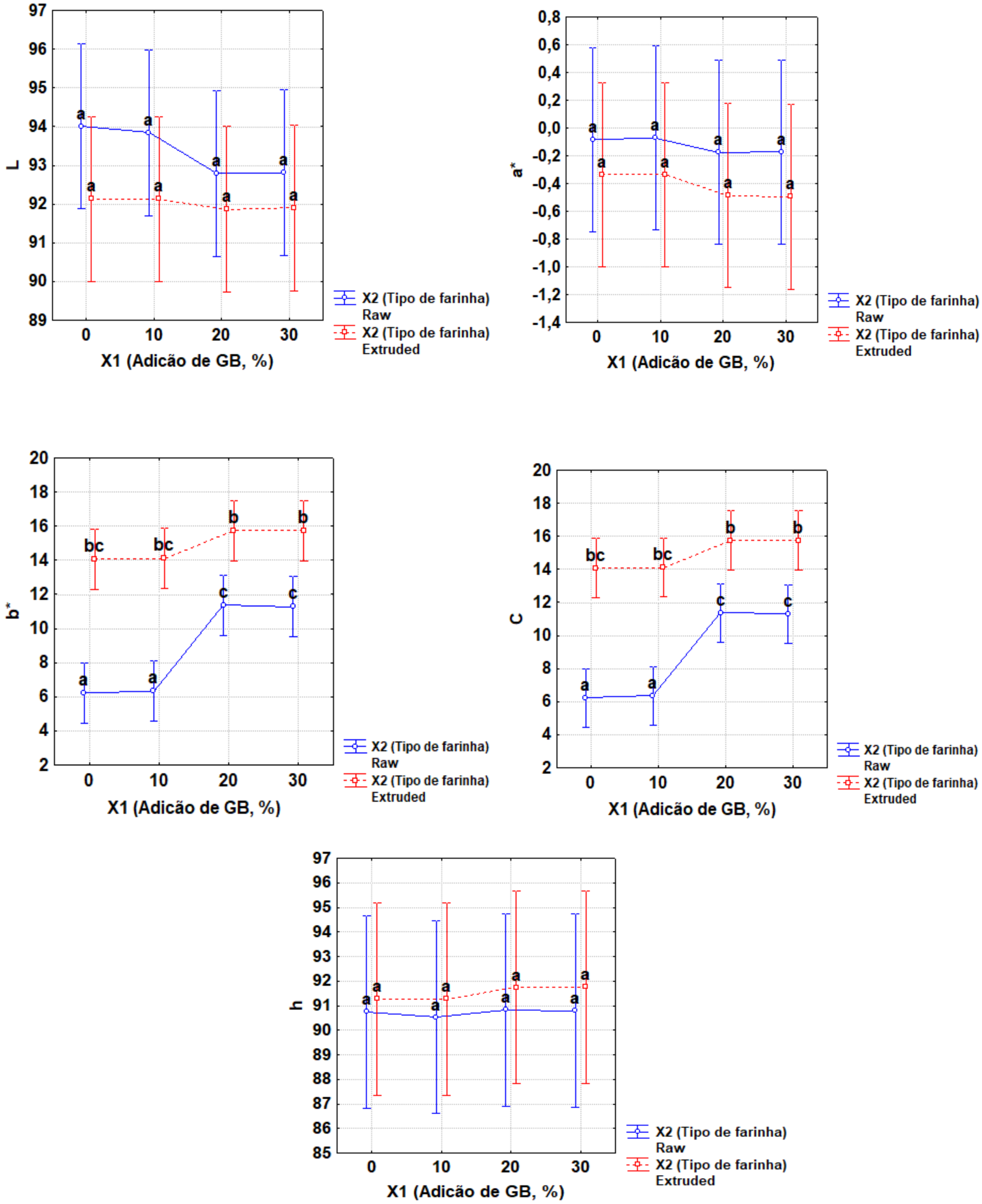
Brazilian legislation establishes specific particle size parameters only for wheat flour, serving as a basis for comparing the flours and starches used in the research. Considering that the granulometry established by Normative Instruction N° 8/2005 is 0.250 mm, rice flour, chickpea flour and potato and cassava starch have this characteristic similar to commercially available wheat flour, since the largest percentage of particles were less than or equal to 0.250 mm (BRASIL, 2005).

This is due to the fact that the flours were sieved through 0.250 mm meshes to standardize the samples for future bread preparation, as, according to Wu *et al*. (2019) in their study, the particle size of the samples influences the result of the bread, as excessive grinding of the grains affects the existing starch granules, thus interfering with the viscosity, volume and elasticity of the dough (SAKHARE, *et al*., 2014).

3.1.2 Color analysis

The results regarding the colorimetry of samples of the formulations with raw and extruded treatments with an addition of 10%, 20% and 30% of chickpeas are shown in figure 3.

Figure 3 - Color analysis results, where: L (brightness), a* (-80 to 0 = green and 0 to +100 = red)), b* (-100 to 0 = blue and 0 to +70 = yellow), C (relationship between the values of a* and b*, where the real color is obtained were measured of the researched object) and h (angle formed between a* and b*, indicative of the object's color saturation).



[1]Different letters differ significantly by Tuckey test (P ≤ 0.05)

The instrumental color parameters L, a*, b*, c and h were analyzed because color is an important characteristic in consumer sensory evaluation and for the food industry, in addition to the fact that extrusion can affect color parameters, thus being important parameters that need to be evaluated.

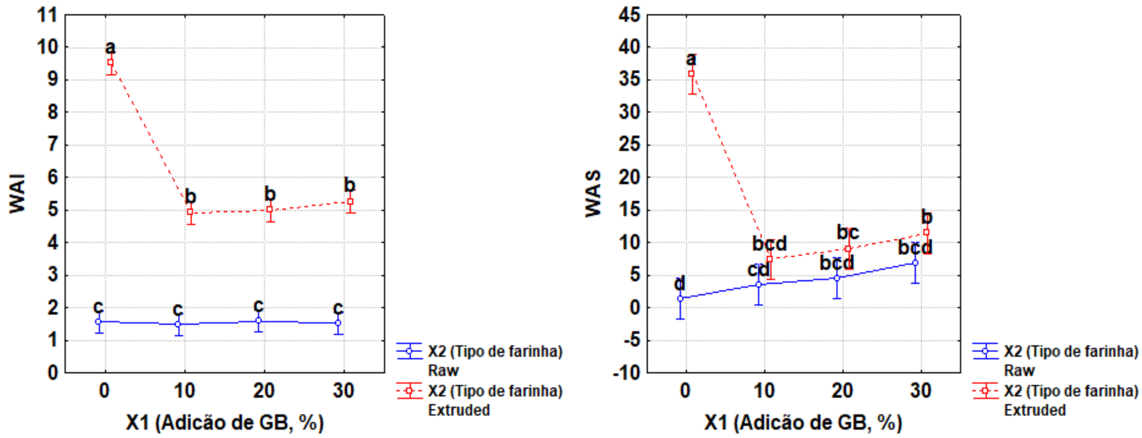
The parameters L, a* and h did not differ significantly between the samples, whereas the chromaticity coordinates b* and c, which vary from - blue and + yellow and which vary the intensity or saturation, varied from 6.21 to 15.74 and 6.22 to 15.75, respectively, showing a tendency to yellow in all the flours analyzed and with greater saturation in the extruded samples, which justifies the fact that they are flours that have undergone a type of cooking, thus differing in color from those of other raw flours.

3.2 Technological characterization

3.2.1 Water absorption index (WAI) and Water solubility índex (WSI)

Water solubility index (WSI) is related to the amount of soluble solids present in a dry sample and allows controlling the degree of intensity of heat treatment, depending on the gelatinization, dextrinization and subsequent solubilization of the starch components of the raw material, such as proteins, lipids and fibers. This index is widely used to measure the solubility of extruded starch in beverages, soups, baby foods and other products. WSI is an important parameter in the characterization of extruded flours for subsequent thawing, such as in soups, as it allows checking the degree of starch cooking and evaluating the thawing conditions in an aqueous medium. To prepare pre-cooked soups, for example, it is necessary that the dissolution of the substance meet the desired organoleptic characteristics, such as the absence of lumpy and dense matter, flavor, among others. The product obtained must guarantee homogeneity due to the complete interaction of the solid components and water. Meanwhile, water absorption index (WAI) is a measurement that reflects the ability of starch granules to absorb water even at room temperature. It reflects the integrity of part of the dough structure after the extrusion process. The water absorption capacity of starchy raw materials generally increases when heat is applied in a higher moisture during the extrusion gelatinization process (ASCHERI, 2009). The water absorption index and water solubility index are shown in figure 4.

Figure 4 - Analysis result of Water Solubility Index (WSI) and Water Absorption Index (WAI).



[1]Different letters differ significantly by Tuckey test (P ≤ 0.05)

T0R (raw with 0% chickpeas) had the lowest WAI (1.48 g/g) along with the other raw samples, as expected for raw flours. The extrusion process generated a four-fold increase in the WAI of raw flours while the greatest increase was observed for T0E (extruded with 0% chickpeas) almost six times higher (9.51 g/g). This discrepancy in the T0E sample is due to the fact that the rice extrusion was carried out with different extrusion parameters than the other samples. According to Carvalho *et al*. (2010) when going through the extrusion process, the starch granules are broken, thus causing a greater water absorption capacity.

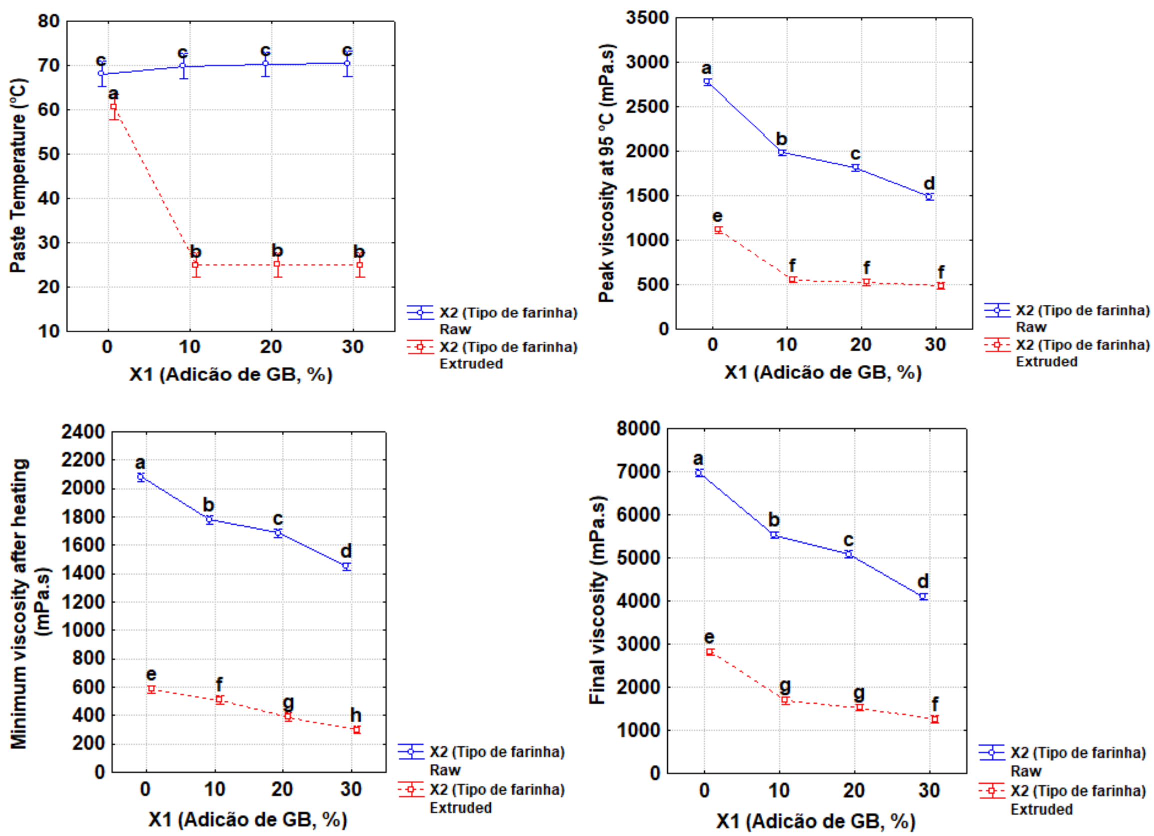
The WSI percentage showed that when comparing raw and extruded flours there was an increase in solubility in the extruded flours and this value increased according to the addition of chickpeas in the formulation. According to Wang *et al*. (2024) the high solubility index can be attributed to the greater presence of water-soluble substances in the chickpea composition.

3.2.2 Pasting properties

The pasting property of starches is influenced by the size of the granules, swelling power, amylose and lipid content, as well as the molecular structure of amylopectin. Hydrothermal extrusion treatment significantly interferes with the viscomilographic properties of starchy materials. After modification, there is a reduction in paste temperature, minimum viscosity, peak viscosity, final viscosity and tendency to retrogradation.

Because of these modifications, is possible expect better applications of pre-cooked materials in the formulation of bread doughs, as there would be better water absorption, and consequent connection with starchy structures, when the dough is developed.

Figure 5 – Result of the analyzed parameters regarding folder properties: PT= Paste temperature; PV= Peak viscosity at 95 °C; MV= Minimum viscosity after heating and FV= Final viscosity.



[1]Different letters differ significantly by Tuckey test ($P \leq 0.05$)

As seen in figure 5, the values of the raw samples were higher than those of the extruded ones. The paste temperature (PT) of the raw samples was similar (~70.0 °C), while the extruded samples had reduced PT (25.0 °C) indicating that during extrusion processing there was a large modification of the starch present in the extruded.

According to Tomar *et al*. (2022) high values when starting starch cooking may indicate greater resistance to water absorption, due to lipid layers present in the grains analyzed.

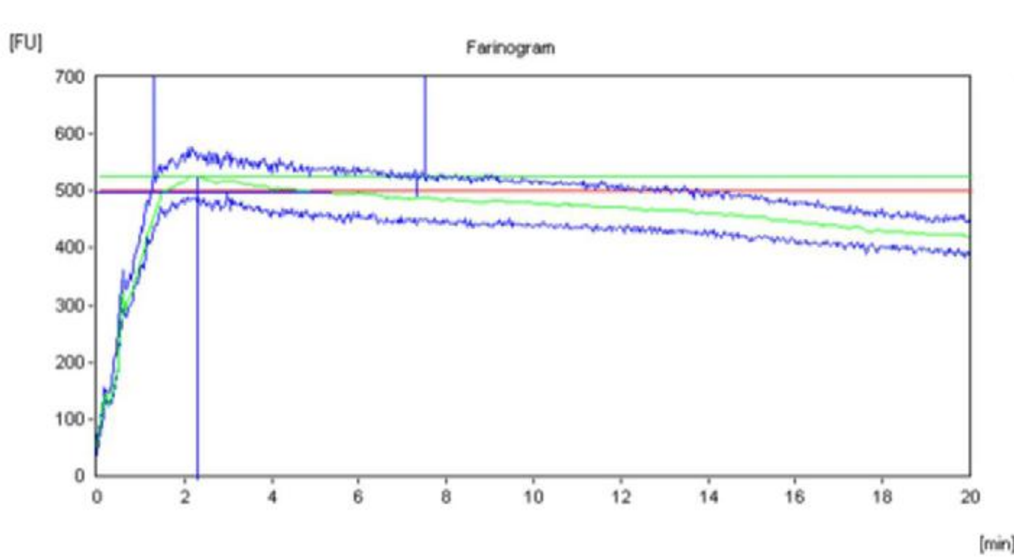
3.2.3 Farinograph measurement

Rheological methods are used to determine the technological quality of wheat and the quality of wheat flour, which is the basic raw material of the mill and bakery industry. The results of these tests have a direct relationship to the quality of the product.

The farinography was carried out in accordance with the experiments during the preparation of the breads and the best breads were chosen for analysis according to the volume and texture parameters.

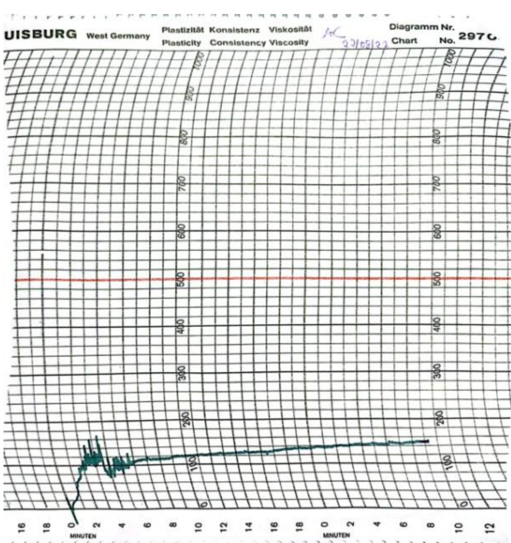
A typical graph resulting from the farinograph analysis with wheat flour is shown in figure 4. According to the composition values, mainly gluten, the trend of the curve follows the characteristics described in figure 6. All parameters indicate a flour with excellent conditions for producing good quality bread. This graph was placed as a way of comparing and verifying the great difference between the flours used to prepare gluten-free breads. Of the four graphs in Figure 7, **a**, **b**, **c**, and **d**, only graph **c** shows a slight behavior in the curvature of the graph, but above the 500 line, that is, around 800 UB, due to the use of extruded flour that allowed greater absorption and better mass formation for this test.

Figure 6 - Typical wheat flour farinogram for baking.

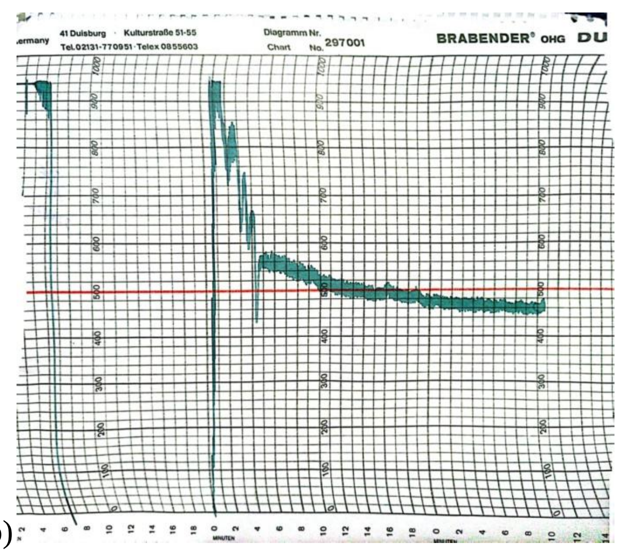


Source: MARTÍNEZ-GIRÓN, 2017

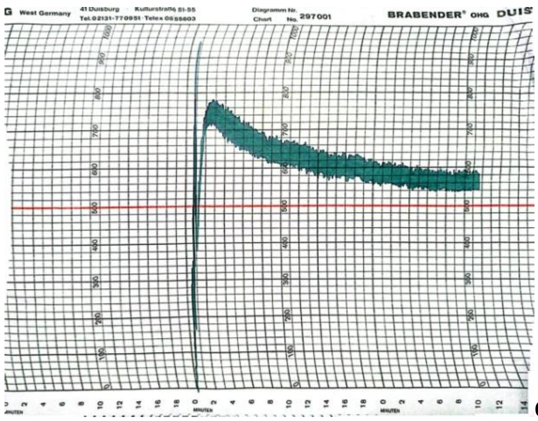
Figure 7 - Farinography results for samples a) T0R (raw with 0% chickpeas), T0E (extruded with 0% chickpeas), T1E (extruded with 10% chickpeas) and T2R (raw with 20% chickpeas beak).

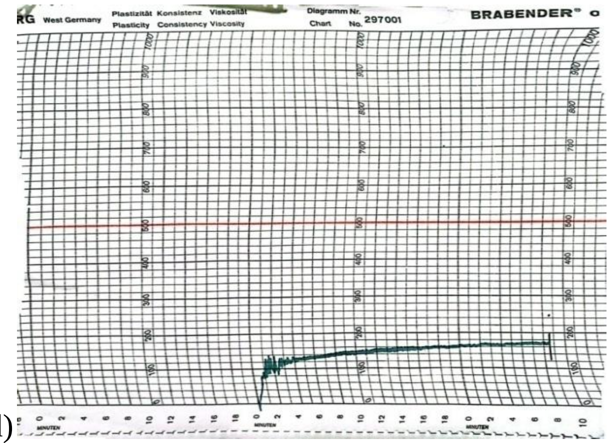


a)



b)

c) 

d) 

According to the results, the raw flours T0R and T2R did not exceed 200 UB (brabender units), while the extruded flours T0E and T1E exceeded 500 UB, which is the reference value for wheat flour, even though the flours do not contain gluten.

According to Ascheri *et al*. (2020), extrusion cooking modifies the polymers and consequently, the parameters related to paste properties and composition. In addition, by quickly reaching DDT it can contribute to reducing the cost and processing time of bread, thus speeding up its manufacturing. Under these conditions, when studying farinography with gluten-free flours, large divergences are expected when compared to wheat flours. Carbohydrates derived from rice, ingredients added to act as binders, cannot show even close results in terms of trends in farinographic curves. This is evident, as absorption levels are related to the type of starch and its previous degree of conversion, as is the case with formulations containing extruded flours. In the case of farinographic curves with wheat flour, they absorb water at the same time as the dough develops, creating a characteristic resistance throughout the test. Gluten-free formulations differ in behavior and even in water absorption levels. All treatments tested (**a**, **b**, **c**, and **d**) showed significantly different curves. In each case related to the types of ingredients in the formulation.

4 CONCLUSION

Pre-gelatinized flours using thermoplastic extrusion can significantly help in the formation of bonds in structures when developing the dough for making gluten-free breads. On the other hand, the use of combinations of raw and pre-cooked rice flour or cassava flour, constitute excellent ingredients in the preparation of bread products when used in the ideal proportion, degree of pre-cooking sufficient to produce binding conditions, forming similar structures to that of wheat flour. It is necessary to consider, apart from the type of ingredient, their particle size may influence the development of the dough. Starting from the principle of water diffusion, smaller particles will absorb more water, leaving larger particles with less water. On the other hand, during grinding, a significant amount of starch is damaged. This mechanical damage can have a significant impact on the properties of the material to be used; in particular, damaged starch has a greater water absorption capacity and is more susceptible to enzymatic hydrolysis. Heating starch suspensions in water above a specific temperature leads to rupture at the molecular level, with consequent loss of crystallinity and irreversible swelling of the granules. This process, called gelatinization, facilitates molecular mobility, the dissociation of amylopectin double helices and the fusion of crystallites. Gelatinization is also related to a reduction in starch solubilization -mainly amylose leaching- which determines an increase in the viscosity of the starch suspension. This phenomenon continues with further heating and, above the gelatinization temperature, a continuous phase of solubilized macromolecules -mainly amylose- and a discontinuous phase of swollen, amorphous starch granules are formed. Consequently, when the dough is formed, there will be greater interrelationships with hydrogen bonds and other bonds caused by the addition of hydrocolloides, forming a network, thus allowing the retention of CO_2, a product of fermentation.

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CAPÍTULO II

CHARACTERIZATION OF VEGAN GLUTEN-FREE BREADS FORMULATED WITH MIXED RICE AND CHICKPEA FLOUR

CHARACTERIZATION OF VEGAN GLUTEN-FREE BREADS FORMULATED WITH MIXED RICE AND CHICKPEA FLOUR

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ABSTRACT

Vegan gluten-free breads (without eggs and milk) are an increasing trend, due to pathologies such as celiac disease, irritable bowel syndrome, among others, as well as individuals who do not have any pathology, but wish to follow a diet based on plants. The objective of this study was to evaluate the effect of adding chickpea flour ($\boldsymbol{X1}$: 10%, 20% and 30%) and using a white rice-based flour blend ($\boldsymbol{X2}$: raw and pre-cooked by extrusion) in the preparation of breads free of gluten, eggs and dairy ingredients. Breads made with raw flour had a higher specific volume, regardless of the addition of chickpea flour. Hardness and gumminess were greater in breads produced with raw flour and $\boldsymbol{X1}$: 20%. On the other hand, breads made with pre-cooked flour and $\boldsymbol{X1}$: 10% were more cohesive and with a higher dietary fiber content. The differences found in the treatments can be attributed to the combined action of the protein fraction subjected to varying degrees of modification during the extrusion cooking process. Bread made with raw flour and $\boldsymbol{X1}$: 20% had a more uniform crumb and higher protein content.

Keywords: breads; gluten-free; vegan; extrusion.

1 INTRODUCTION

The gluten-free food market was estimated at US$6.45 billion in 2022 and is expected to increase at a compound annual growth rate (CAGR) of 9.8% from 2023 to 2030, becoming a global trend in the area of foods, baked goods held the highest percentage (28.5%) of total revenue in 2022. The growth is due to the fact that consumers are increasingly concerned about their health and a healthy lifestyle. As a result, it is expected that gluten-free products will see increasing innovation and influence the growth of the sector in the coming decades (Grand View Research, 2023).

Vegan gluten-free breads (without eggs and milk) are an increasing trend, due to pathologies such as celiac disease, irritable bowel syndrome, among others, as well as individuals who do not have any pathology, but wish to follow a diet based on plants (Guimarães, et.al 2022). The nutritional and technical quality of most gluten-free products on the market is unsatisfactory, since gluten is responsible for the formation of the dough, providing flexibility and gas retention. Furthermore, gluten-free breads tend to have lower nutritional value because they are often made from refined grains, which have a lower protein value (Bernardes et al., 2022).

With the addition of legume flours such as chickpea flour and the use of pre-cooked flours by extrusion at intermediate moisture, breads with acceptable characteristics can be produced. Extrusion cooking of cereal and legume mixtures thermoplasticizes starch granules and thermosetting proteins. Chickpeas have a high protein content and have emulsifying properties, foaming capacity and high oil absorption capacity, making them a suitable ingredient for baking (Santos et al., 2021).

The objective of this study was to evaluate the addition of chickpea flour and the use of raw flour/flour pre-cooked by extrusion (mixture of white rice/chickpea flour) in the preparation of gluten-free breads and vegans and their physical-chemical and technological characteristics.

2 MATERIALS AND METHODS

2.1 Materials

The white rice used in the preparation of the breads was obtained from local stores, in the city of Rio de Janeiro (RJ) in the form of grains (type 2, polished, long fine class) from the Palmares brand (Palmares do Sul/RS). The chickpeas were supplied by the company Granfino (Nova Iguaçu/RJ) and the other ingredients (potato starch, cassava starch, salt, sugar, olive oil, dry yeast, xanthan gum and CMC) were obtained from local stores.

2.2 Methods

2.2.1 Treatment and experimental design

The treatments used were a mix of flours with different proportions of chickpeas as a replacement for rice flour and industrial treatments (raw and extruded). It was a design with two replications, 3x2 factorial type, with three proportions of chickpea flour (10%, 20% and 30%) and two industrial treatments (raw and extruded) as shown in Table 1.

Table 1. 3×2 factorial experimental design with 2 replications to formulate vegan gluten-free breads based on mixed rice and chickpea flour.

Test	Coded levels		Real levels	
	x_1	x_2	X_1 (Addition of chickpea flour to the formulation) %	X_2 (Type of flour in the formulation) -
1	-1	-1	10	Raw
2	0	-1	20	Raw
3	1	-1	30	Raw
4	-1	1	10	Precooked
5	0	1	20	Precooked
6	1	1	30	Precooked
7 [1]	-1	-1	10	Raw
8 [2]	0	-1	20	Raw
9 [3]	1	-1	30	Raw
10 [4]	-1	1	10	Precooked
11 [5]	0	1	20	Precooked
12 [6]	1	1	30	Precooked

x_1: quantitative variable

x_2: qualitative variable

Design model:

$$Y = \beta_0 + \beta_1 x_1 + \beta_2 x_2 + \beta_{12} x_1 x_2 + \beta_{11} x_1^2 + \beta_{112} x_1^2 x_2 + \epsilon$$

Where Y is the response evaluated in the flour, x_i are the independent variables at coded levels, β_i are the model coefficients that will be obtained by analysis of variance (ANOVA) and regression analysis.

2.2.2 Bread formulation

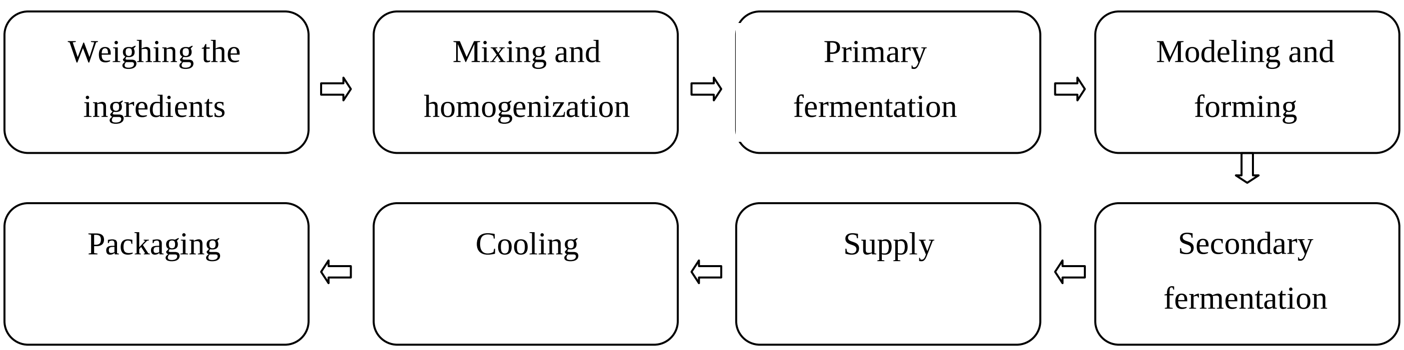
Table 2 presents the materials used in the preparation of gluten-free breads. Chickpea flour was used in three concentrations (10%, 20% and 30%) to replace rice flour, while the other compounds were kept constant.

To prepare the dough, instant yeast (Fleischmann, Pederneiras, Brazil) was previously activated with deionized water (total water in the formulation) at 35 ◦C together with sugar and placed in a fermentation chamber at 85% relative humidity for 30 min for activation. After the time, the oil and salt were added and rested for another 20 minutes. All dry ingredients were homogenized and mixed with the cream. Portions of 100 g were cut, shaped and placed in previously greased steel molds measuring 12cm x 5.5cm. They were subsequently placed in a fermentation chamber at 30 ◦C and 85% relative humidity for 60 min. Finally, they were placed in a convection oven (model FVT5D, Venâncio, Venancio Aires, Brazil) at 150ºC/60 min and then allowed to cool at room temperature. The bread analyzes were carried out after 24 h, using two controls for comparison: the mixture with non-extruded rice flour (T0R) and extruded (T0E).

Table 2. Proportions established for rice and chickpea flour

Samples	Flour type	Rice flour (%)	Chickpea flour (%)
1 - T0R	Raw	100%	-
2 - T0E	Extrused	100%	-
3 - T1R	Raw	90%	10%
4 - T1E	Extrused	90%	10%
5 - T2R	Raw	80%	20%
6 - T2E	Extrused	80%	20%
7 - T3R	Raw	70%	30%
8 - T3E	Extrused	70%	30%

Figure 1 - Steps in the bread production process



Source: Own authorship (2024)

2.3 Physical Characterization

2.3.1 Specific volume

For specific volume analysis, the finished breads at room temperature (25°C) were weighed on an analytical balance. Then, the volume of each loaf was determined in triplicate by displacing the millet seeds, according to the method described by the AACC Method (10-11) (AACC, 2000).

2.3.2 Texture

The texture analysis of the breads was carried out according método 74-09 (AACC, 2000). The analysis was carried out using a texturometer (Stable Micro Sistem, TA XT Plus) with the TPA method (Texture Profile Analysis), considering the attributes of hardness, elasticity, cohesiveness and chewability. These parameters are used due to their relationship with sensory parameters. The tests were carried out simultaneously specific volume measurements in samples of each formulation, in slices of 2 cm.

2.3.3 Color parameter analysis

The color analysis of rice flour, chickpeas and potato and cassava starches was carried out by reflectance using a Hunterlab Colorquest colorimeter, model XE (Reston, Virginia, USA), CIELAB scale, with an opening of 1 mm in diameter, with D65/10 illuminant. The values of L* (brightness), a* (green - /red +), b* (yellow - /blue +), C* (relationship between the values of a* and b*, where the real color is obtained were measured of the analyzed object) and h (angle formed between a* and b*, indicative of the object's color saturation).

2.3.4 Water activity

Water activity was measured using an AquaLab LITE direct-reading digital electronic hygrometer. The readings took place at 25ºC and were carried out in triplicate with the bread crumb samples cooled before determination.

2.4 Chemical Characterization

2.4.1 Centesimal composition

The analyzes to determine the proximate composition were carried out in the physical-chemical analysis laboratories of Embrapa Agroindústria de Alimentos (Rio de Janeiro/Brazil). To determine moisture and ash, the Thermo Gravimetric Analyzer (TGA) Method was used. T= 100°C and 550ºC, N_2 5.0 and O_2 6.0; constant weight and flow of 2L/min, respectively.

For protein analysis, the Kjeldahl Method, POP LFQ 012, rev. 5. F = 4. For analysis of ether extract, the AOCS Am 5-04 method (Automatic Fat Extractor) was used. For the analysis of soluble fiber, insoluble fiber and dietary fiber, the Enzymatic-Gravimetric method was used. POP LFQ 050, rev.2. The carbohydrate content and caloric value were calculated according to Resolution – RDC nº. 360 of December 23, 2003.

2.4.2 Polyacrylamide gel electrophoresis

The polypeptide profile by polyacrylamide gel electrophoresis was determined according to Laemmli (1970), using polyacrylamide electrophoresis gel containing SDS (SDS-PAGE). A discontinuous system was assembled with a separation gel with a concentration of acrylamide/bis-acrylamide at 12% (1.5 mm), with the samples applied to a concentration gel (5%). Samples and molecular weight marker (Cytiva) were incubated with sample buffer for 5 min at 95°C, applied to the gel, and separated at 200 V for 55 min. Then, the gel was stained with 0.05% Comassie Blue R-260 solution, overnight. Excess dye was removed with bleaching solution (methanol: glacial acetic acid: water, 3:1:6, v:v:v) until blue bands appeared against the transparent background of the gel. Then the gel was scanned.

2.4.3 Digestibility in vitro

The raw and extruded breads and flours were analyzed in triplicate in three digestion phases: mouth, stomach and small intestine, using the simulated digestion fluids, respectively, prepared according to the consensual INFOGEST method (Santos, 2023).

In the first stage, the oral phase was carried out by diluting 1g of flour in a total of 5mL containing simulated salivary fluid and amylase with a concentration of 75 U/mL (αamylase Type IX-A, 1000-3000 U/mg of protein, Sigma).

Flour-simulated salivary fluid was incubated while mixing for 2 min, pH 7.0, at 37°C. Then, for the gastric phase, the oral mix bolus was added in bottles simulating gastric fluid (1:1 (vol/vol)) containing pepsin with a concentration of 2,000 U/mL (TS 1:10,000U/mL, Bela Vista®, Brazil) and remained under stirring for 2 h, pH 3.0, 37 °C.

Sequentially, the small intestine phase began by adjusting the chime pH to 7.0 with a simulated intestinal fluid (1:1 (vol/vol)) added 800 U/mL pancreatin solution (porcine pâncreas 8×USP, Sigma®, USA) was kept while mixing for 2 h, pH 7.0, 37 °C.

Then the samples were centrifuged (4,000 x g, 10 min) and the supernatant was frozen until Bradford and Tyrosine analyzes to evaluate the in vitro digestibility of the proteins present in the samples. Bradford was analyzed using the method described by Bradford (1976) and Tyrosine using the method described by Chang-Lee et. al (1989).

2.5 Statistical Analyzes

The results of the analyzes were subjected to analysis of variance (ANOVA) and means comparison test (Tukey at 5%), with the aid of the STATISTICA statistical package version 12.0 (Statsoft Inc., Tulsa, OK, USA).

3 RESULTS AND DISCUSSIONS

3.1 Physical Characterization

3.1.1 Specific volume

Bread volume is considered one of the main desirable characteristics, as it provides a quantitative measurement of breadmaking performance and consumer acceptance. Specific volume and images of the loaves are shown in Figure 2 and 3, respectively.

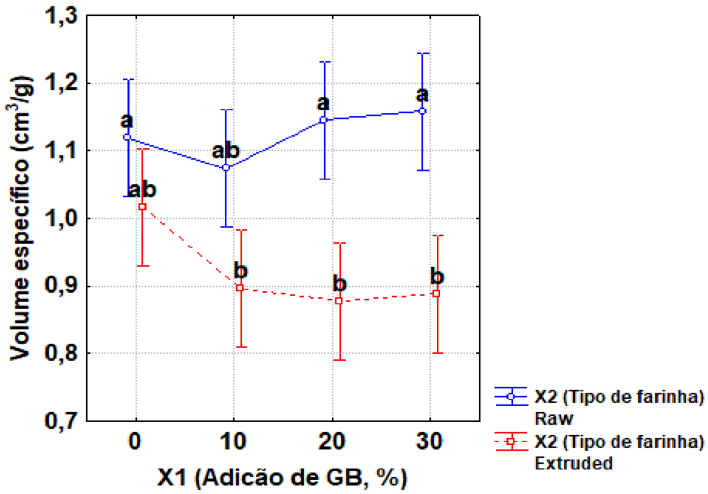
It was found that breads produced with raw flour presented the highest specific volume values (Figure 2), regardless of the addition of chickpea flour (1.15 cm3/g vs. 1.0 cm3/g).

Clerici *et al*. (2009) found higher volumes (1.63 to 2.25 cm3/g) for bread based on extruded rice flour containing lactic acid. The results of the present work were lower than the volumes reported by Sánchez *et al*. (2008) for bread based on extruded rice flour without acidification (3.30 to 4.33 cm3/g) and lower than the results for bread made from quinoa flour without acidification, with values between 1.4 ± 0.02 cm3 /g and 1.51 ± 0.07 cm3/g (Alvarez-Jubete *et al*., 2010; Hager et.al, 2012).

Different results may be due to the use of extruded or non-extruded flours, the addition of lactic acid, different extrusion conditions or different varieties of rice, in addition to the fact that the breads in the present study are gluten-free and vegan, the absence of milk and egg influence the final volume of bread, indicating that research should continue to increase the volume of bread.

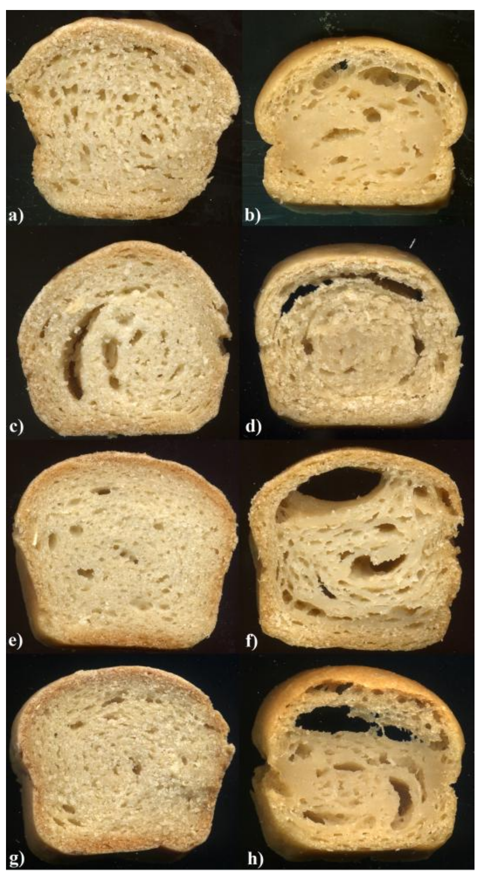
The use of flours other than wheat provides bread with smaller volume and specific volume, as the absence of the gluten network reduces the ability to retain gases generated during fermentation and baking (CAPRILES; ARÊAS, 2012).

Figure 2. Specific volume of breads



Means followed by the same letter in the column do not differ statistically from each other by the Tukey test ($p<0.05$).

Figure 3. Visual aspects of gluten-free bread slices.



a-T0R b-T0E c-T1R d-T1E e-T2R f-T2E g-T3R h-T3E

3.1.2 Texture

According to Aguilar *et al*. (2015), the use of flours with a higher protein content, such as chickpea flour, can influence the improvement of texture parameters and increase the shelf life of gluten-free breads as they delay the process of starch retrogradation by promote the formation of protein-starch complexes and reduce the formation of starch-starch complexes. Furthermore, according to Aguilar *et al*. (2015) and Bashir and Aggarwal (2016), the proteins and lipids present in chickpea flour have important emulsification and foaming properties, making it an alternative commonly used in the preparation of gluten-free breads and cakes. by providing an increase in specific volume and improvement in texture parameters. According to Arslan *et al*. (2019), the emulsification and foaming properties can be facilitated by the greater amount of fiber present in chickpea flour, which promotes greater absorption of water by the dough and provides the chemical bond between water and sugar molecules. protein.

It was possible to observe that the elasticity values increased as the level of chickpea substitution increased. The formulations with 10% and 20% chickpeas presented the highest elasticity values (1.1 and 1.2), although there was no significant difference between the formulations.

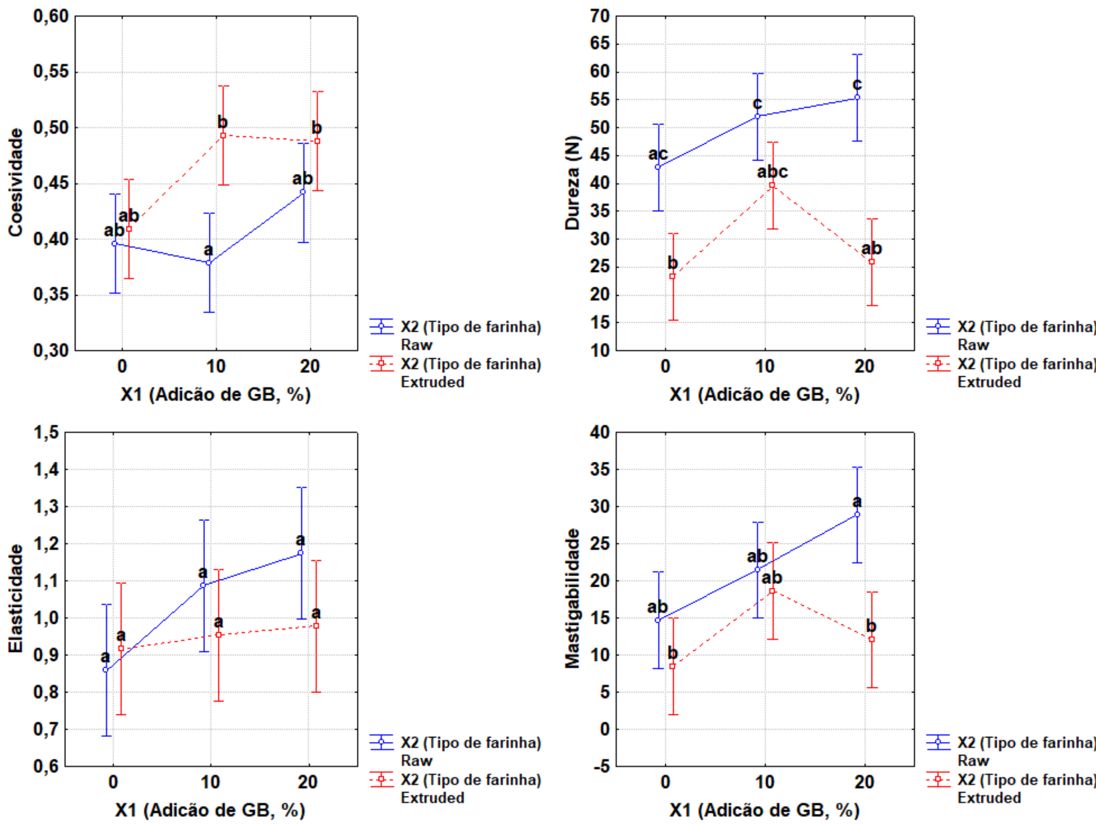
The values for the cohesiveness parameter increased as the level of chickpea substitution was increased, with extruded formulations showing higher values (0.50) in relation to formulations that used raw flour (0.40).

The values for the chewiness parameter also increased as the level of substitution for chickpea flour was increased, but the parameters were higher for breads made with raw flour. Minarro *et al*. (2012) evaluated gluten-free breads formulated with chickpea flour and found a value of 0.55 for cohesiveness, higher than that found in this research (0.49). Buresóva *et al*. (2017) also evaluated gluten-free breads formulated with chickpea flour and found values of 0.58 for cohesiveness and 1.7 for elasticity, also higher than those found in this work. The hardness of breads made with raw flour was greater with the addition of 20% chickpea flour (55 N vs. 25 N).

A high extrusion temperature together with a high feed humidity can produce an extruded material with greater glucose release. This can affect the specific volume and increase the firmness of the crumb (Ortolan *et al*., 2015).

In contrast, the firmness of chickpea breads increased significantly with the addition of chickpeas. Rosell (2009) suggests that excessive addition of chickpeas can have a negative effect, resulting in extensive cross-links between proteins and a very firm texture.

Figure 4. Result of texture parameters.



Means followed by the same letter in the column do not differ statistically from each other by the Tukey test ($p<0.05$).

3.1.3 Color parameter analysis

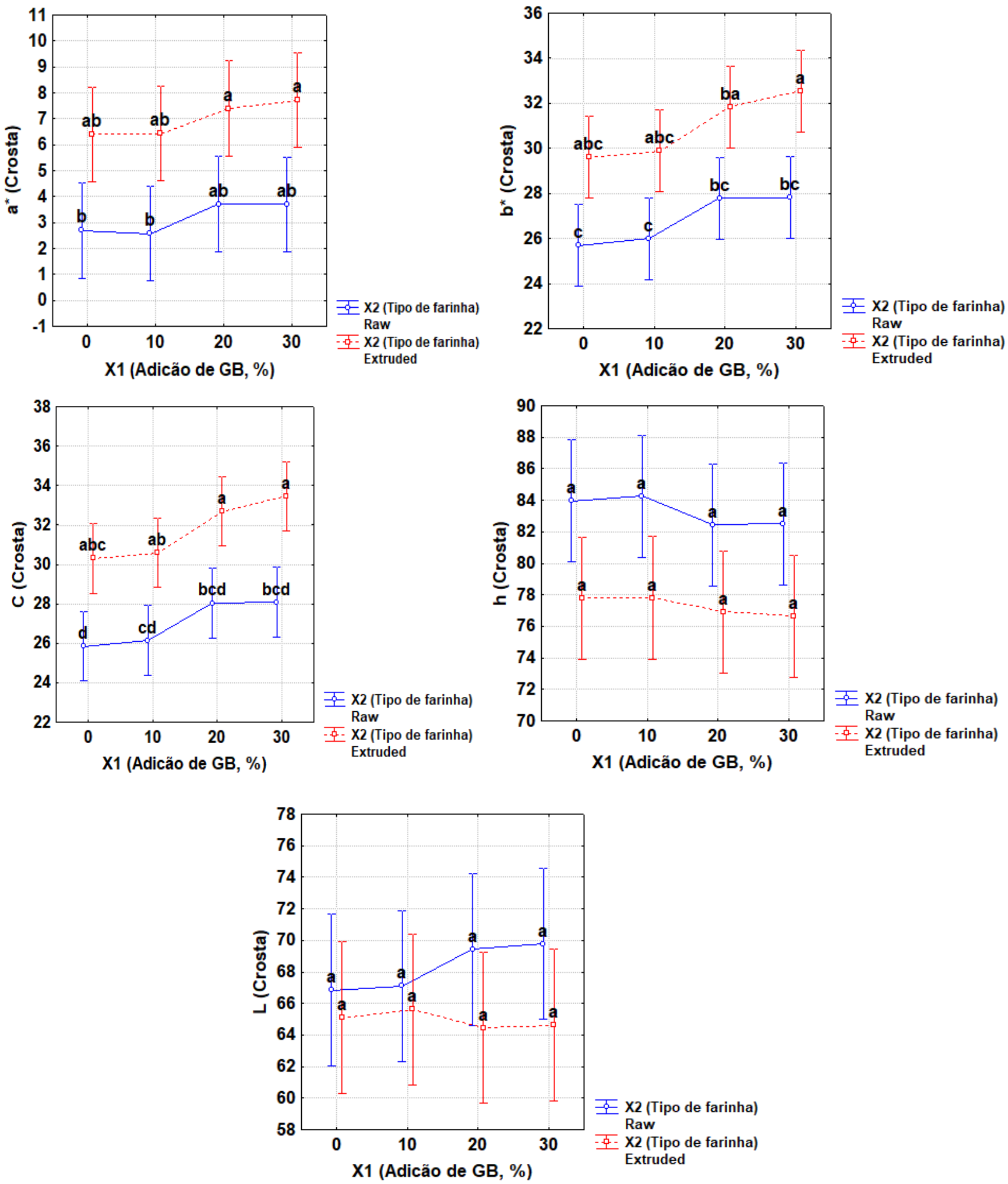
The values of chromas a*, b*, C, h and L did not present significant differences for the analysis carried out on the bread crumbs, according to figure 6. The values of h and L for the parameters referring to the crust of the breads also did not had a significant difference, as shown in figure 5. However, it was possible to observe that the values for the parameters a*, b* and C* increased significantly as the concentration of chickpeas was increased. According to Oliveira et.al (2017) and Dadon et.al (2017), chickpea seeds are naturally yellowish due to their carotenoid content which, according to Shrivastava and Chakraborty (2018), can cause changes in color, especially in the b* parameter, when applied to bakery products.

According to Shrivastava and Chakraborty (2018), the addition of chickpea flour to gluten-free breads promotes browning of the crust due to the reaction of free amino acids with reducing sugars resulting from the hydrolysis, respectively, of proteins and starch during the process. fermentation process, which facilitates the Maillard and caramelization reactions and the consequent formation of pigments, a fact that probably also occurred in this study due to the higher parameters occurring in the extruded samples, which have already undergone prior heat treatment through extrusion.

According to Arslan *et al*. (2019), the color of the crust is one of the factors related to consumer acceptance of baked products, but gluten-free bakery products typically have a lighter and paler color of the crust, making the application of grain flour de-beak becomes an interesting alternative to improve this sensorial characteristic and ensure greater commercial acceptance of products.

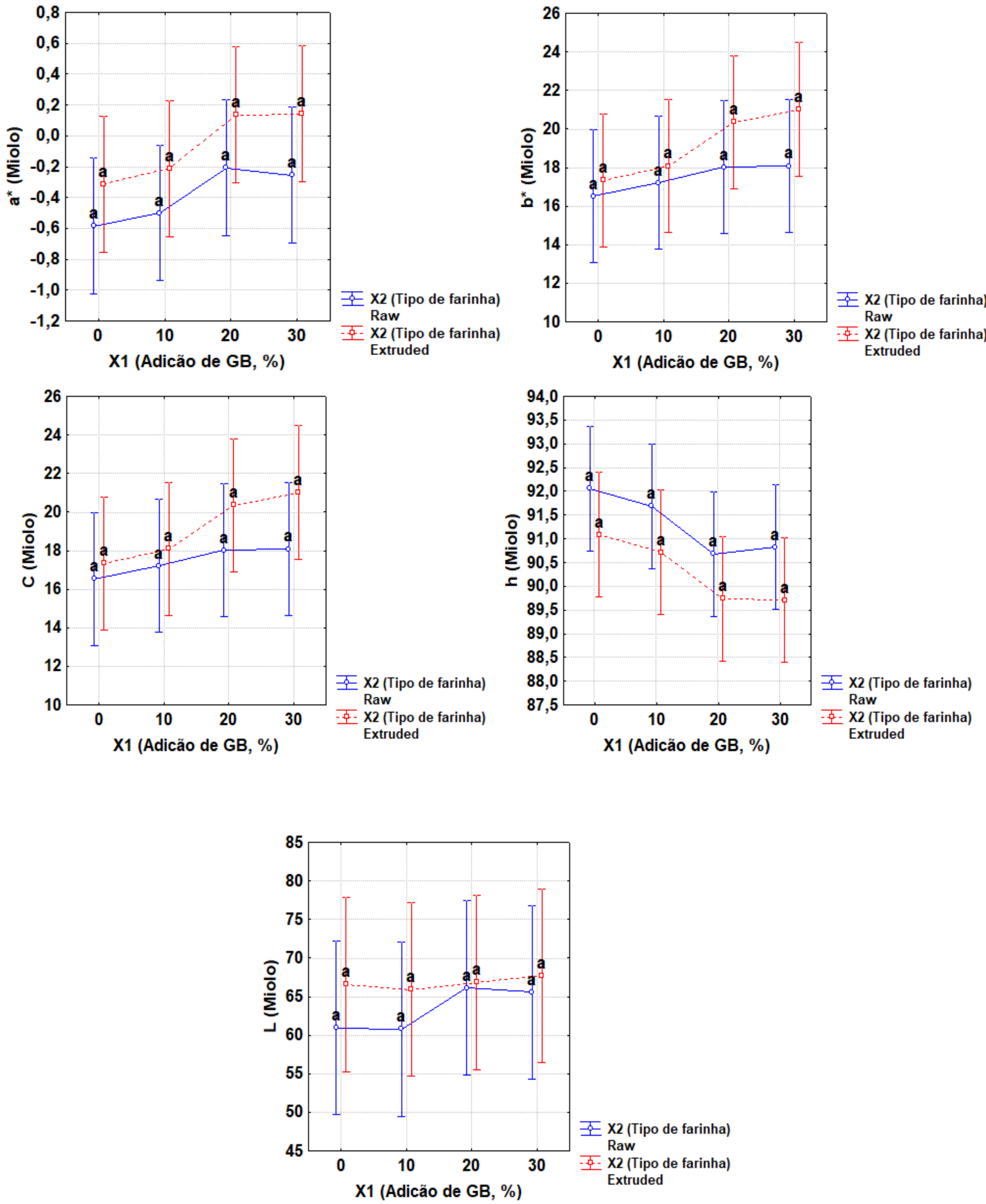
In relation to the b* and C* parameters of the shell, it was possible to observe an increasing trend in values related to the level of chickpea flour used in each formulation. However, according to Arslan *et al*. (2019), the color of the shell can be influenced not only by the composition of the raw material and ingredients used in the product formulation, but also by the conditions related to the baking process, such as temperature and heat transfer mode.

Figure 5. Bread crust color parameters results.



Means followed by the same letter in the column do not differ statistically from each other by the Tukey test ($p<0.05$).

Figure 6. Bread crumb color parameters results.

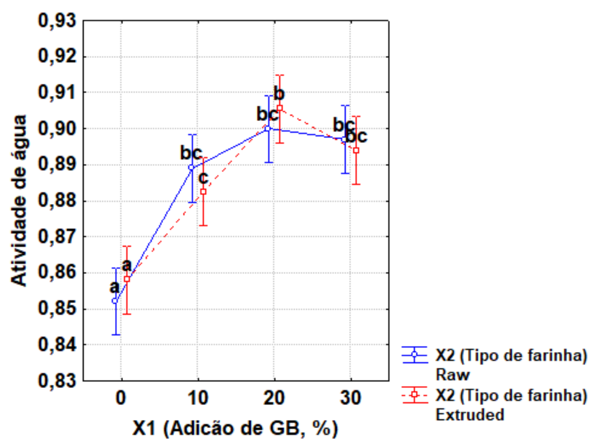


Means followed by the same letter in the column do not differ statistically from each other by the Tukey test ($p<0.05$).

3.1.4 Water activity

The results obtained in the water activity analysis were between 0.852 and 0.905, similar to the values found by César et.at (2006). As chickpeas were added, an increase in the water activity of the sample was observed. According to Dadon et.al (2017), this may occur due to the high solubility index of chickpea flour and the greater presence of water-soluble components. . The analysis of water activity is important considering that the high availability of water (>0.700) in bread can increase the proliferation of microorganisms, therefore, care is needed with the conditioning of the bread and its storage in order to increase the shelf life of the product.

Figure 7. Water activity results.



Means followed by the same letter in the column do not differ statistically from each other by the Tukey test ($p<0.05$).

3.2 Chemical characterization

3.2.1 Centesimal composition

Considering the formulations containing 10% and 20% chickpeas, analyzes of the proximate composition of the samples were carried out in order to quantify the nutritional increase obtained in relation to gluten-free breads made with raw flour and extruded with 100% rice. The data can be viewed in Table 3.

The incorporation of chickpeas contributes to increase the ash and protein levels of the formulations. On the other hand, formulations with 100% raw rice increased the amount of carbohydrates available in bread, since refined cereal flour has a higher content of available carbohydrates (starch) compared to legumes, which are rich in protein and fiber (Bresciani ; Marti, 2019). As for moisture, the highest values were found in formulations that went through the extrusion process (T0E and T1E), due to the higher water absorption index (WAI) in these samples, which reflects the ability of starch granules to absorb water and the integrity of part of the dough structure after the extrusion process. The water absorption capacity of starchy raw materials generally increases when heat is applied at higher humidity during the extrusion gelatinization process (ASCHERI, 2009).

Formulations T2R (80% rice + 20% raw chickpeas) and T1E (90% rice + 10% extruded chickpeas) presented the highest values, respectively, of ash (3.98g/100 g and 3.69 g/100 g), proteins (4.83 g/100 g and 4.60 g/100 g) and total dietary fiber (12g/100 g and 13.3 g/100 g). Formulations T0R and T0E (both developed with 100% rice) present the lowest values, respectively, of ash (3.12 g/100 g and 3.68 g/100 g), proteins (3.97 g/100 g and 3. 73 g/100 g) and dietary fiber (11.11 g/100 g and 11.99 g/100 g).

From these results, it is observed that replacing rice flour with chickpea flour makes it possible to increase the ash, protein and fiber content of bread. It is noted that all formulations may present content claims regarding dietary fiber. According to the current criteria, solid foods that have at least 2.5 g of fiber per serving can be considered sources of fiber, and those that have at least 5.0 g of fiber per serving can use the high content claim of fibers (Brazil, 2020).

Table 3 – Result proximate composition of breads

Sample	T0R	T0E	T2R	T1E
Moisture (g/100g)	4,54	7,81	3,91	4,93
Ashes (g/100g)	3,12	3,68	3,98	3,69
Nitrogen (g/100g)	0,69	0,65	0,84	0,80
Protein (g/100g)	3,97	3,73	4,83	4,60
Ethereal extract (g/100g)	6,68	7,22	9,79	7,56
Soluble fiber (g/100g)	3,48	3,86	3,71	4,78
Insoluble fiber (g/100g)	7,63	8,13	8,29	8,52
Dietary fiber (g/100g)	11,11	11,99	12,00	13,30
Carbohydrate (g/100g)	70,58	65,56	65,49	65,92
Calories (kcal/100g)	358,32	342,18	369,39	350,12

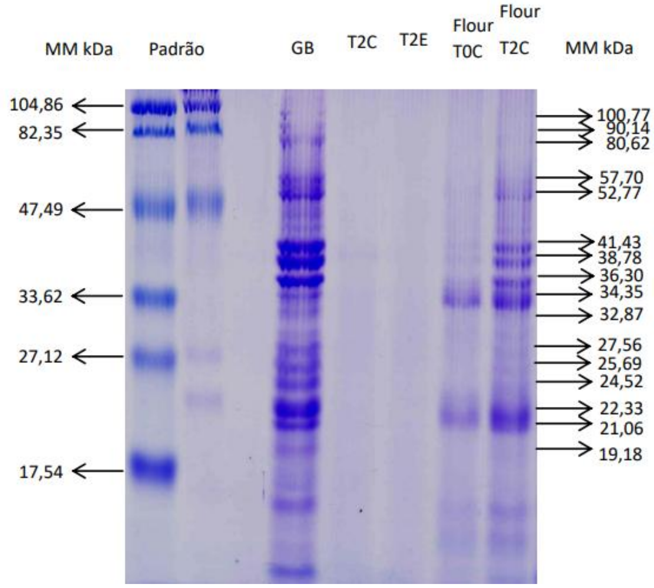
T0= 100% rice:0% chickpea; T1= 90% Rice:10% chickpea; T2= 80%rice:20%chickpea

R=Raw; E=Extruded.

3.2.2 Polyacrylamide gel electrophoresis

The search for an identity pattern of proteins in flour and bread mix(es), through visualization of the polypeptide profile by polyacrylamide gel electrophoresis, proved to be effective only for flour mix(es), without and with chickpeas. The flour mix without chickpeas showed the presence of two polypeptide chains, one of 32.87 kDa and the other of 21.06 kDa, which probably come from rice flour, as the mix also contains potato starch and potato starch. cassava, both with low protein concentration. The flour mix with (20%) chickpeas showed the presence of 6 polypeptide chains: 52.77; 41.43; 38.78; 36, 30; 34.35 and 22.33 kDa. The bread samples, both extruded and non-extruded, did not show polypeptide bands, possibly the method used for solubilization was not adequate to place the proteins in solution. Future studies need to be carried out in order to implement a method of solubilizing denatured proteins by heat (non-extruded bread) and by heat and extrusion (extruded bread).

Figure 8. Results polyacrylamide gel electrophoresis



GB: chickpea flour; **T2C**: bread from raw treatment with 20% chickpeas; **T2E**: bread from extruded treatment with 20% chickpeas; **Flour T0C**: flour from raw treatment with 100% rice flour and **Flour T2C**: flour from raw treatment with 20% chickpeas.

3.2.3 Digestibility in vitro

It can be seen from figures 9 and 10 that the proteins present in the bread were not soluble, as observed in electrophoresis. The protein levels detected by Bradford were very low at time zero, reaching practically zero. Interestingly, during the digestion period the protein levels became soluble, as in all types of bread the soluble protein level went from zero to 1mg/mL. Digestion possibly takes place in two cycles, first releasing soluble protein and perhaps then hydrolysis will occur. The result of free tyrosine production occurred in the same way. There was no increase in tyrosine, which is expected when there is hydrolysis. Again, it can be suggested that the method should be adjusted to allow sufficient time for hydrolysis to occur.

Figura 9 – Determination of soluble protein by the Bradford method

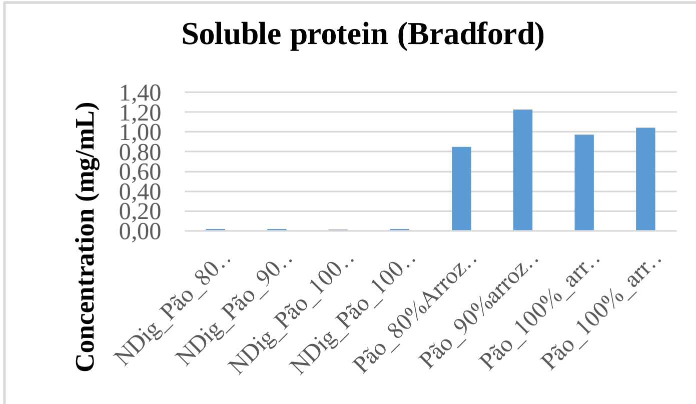
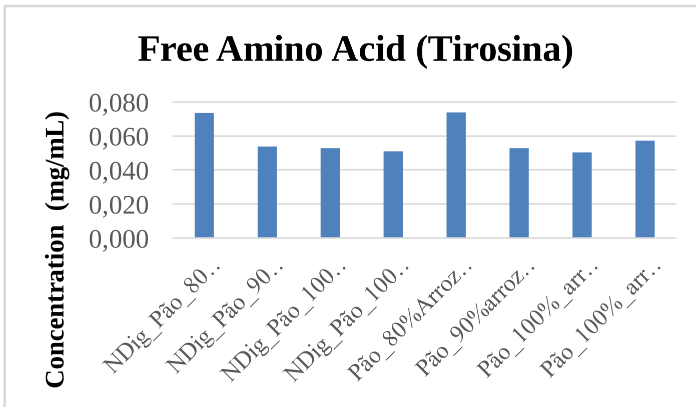


Figure 10 - Determination of amino acids by the Tyrosine method



4 CONCLUSION

The use of chickpea flour in the preparation of gluten-free breads provided better dough behavior in terms of support and structure, resulting in breads in pan and French formats. The T2R formulation (20% raw chickpeas) showed the best results, with better values of specific volume, ash and high fiber content. High humidity and water activity are the main factors that can facilitate bread spoilage. Careful packaging and control of relative air humidity and storage temperature can help maintain the stability of breads during their shelf life, and they can even be sold frozen as an alternative. The breads developed presented an excellent product option to be marketed to individuals with celiac disease and those following a gluten-free and vegan diet. Thus, chickpea flour proved to be a promising alternative for the development and enrichment of gluten-free breads.

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CAPÍTULO III

EFFECT OF THE EXTRUSION PRECOOKING AND CHICKPEA FLOUR ADDITION ON GLUTEN-FREE AND VEGAN BREADS

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EFFECT OF THE EXTRUSION PRECOOKING AND CHICKPEA FLOUR ADDITION ON GLUTEN-FREE AND VEGAN BREADS

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ABSTRACT:

Nowadays, there is a growing market for gluten-free baked goods for vegans. Usually, thedough viscoelasticity to make gluten-free breads is achieved with ingredients derived from animal origin (egg and milk). The objective of this study was to evaluate the effect of chickpea flour addition ($\boldsymbol{X1}$: 10%, 20%, and 30%) and the use of flours' blend based on white rice ($\boldsymbol{X2}$: raw and precooked by extrusion) in the preparation of breads free of gluten, egg and milk ingredients. Breads made with raw flour had a higher specific volume, regardless of chickpea flour addition. Hardness and gumminess were higher in breads produced with raw flour and 20% $\boldsymbol{X1}$. On the other hand, breads made with precooked flour and 10% $\boldsymbol{X1}$ were more cohesive. The differences found in the treatments could be attributed to the combined action of the proteic fraction subjected to various degrees of modification during the extrusion cooking process. The bread made with raw flour and 20% $\boldsymbol{X1}$ presented the most uniform crumb.

Keywords: gluten-free bread, extrusion cooking, bakery product, vegan product.

1. INTRODUCTION

The demand for gluten-free breads without the use of egg and milk as main ingredients, is growing because more and more vegans and lactose intolerant people, as well as people who do not have any celiac disease pathology but wish to follow a plant-based diet. Most gluten-free products on the market have unsatisfactory nutritional and technological quality because gluten is responsible for the dough, providing flexibility and gas retention capacity. In addition, gluten-free breads usually have low nutritional values, as they are usually made with refined or non-refined cereals with low protein value (Bernardes *et al*., 2022).

With the addition of legume flours such as chickpea flour and the use of precooked flours by extrusion at intermediate moisture, breads with acceptable characteristics can be produced. Extrusion cooking of cereal-legume mixtures thermoplasticizes starch granules and thermosets proteins. Chickpea has a high protein content and has emulsifying properties, foaming capacity and high oil absorption capacity, which makes it an ingredient suitable for baking (Santos *et al*., 2021).

The objective of this study was to evaluate the addition of chickpea flour and the use of raw flour/precooked flour by extrusion (flours blend of white rice/chickpea), in the preparation of gluten-free and vegan breads.

2. MATERIALS AND METHODS

2.1 Flour preparation

White rice grains and chickpea grains were acquired from commercial trades at local markets in Rio de Janeiro, Brazil. Both grains were size reduced up to particles ≤0.250 mm (for raw flours) and ≤1 mm (for precooked flours) using a combination of laboratory millers (discs, rollers, and hammers). The resultant flours were blended according to a factorial design 3×2.

2.2 Extrusion cooking process

Each flours blend of white rice/chickpea were moistened at 24% (wet basis) and manually homogenized. Then, they were cooked in a single-screw extruder (Brabender 19/20 DN, coupled with a 3:1 compression ratio screw and a 3 mm frontal die. The temperature profile was fixed at: 40, 90, and 120°C, the screw speed at 150 rpm, and the feed rate of solids at 4 kg/h using a volumetric feeder (Brabender, 625415,394).

2.3 Formulation and bread making procedure

Both, raw and extruded flours were incorporated to a base formulation containing instant yeast, refined sugar, table salt, olive oil, potato starch, cassava starch, xanthan gum, carboxy-methyl cellulose, and water. The dough was manually worked, then separated in portions of 150 g, molded and placed into previously greased steel molds of 150 cm3 capacity, and rested for 60 min in a fermentation cabinet conditioned at 30°C and 85% RH. The dough was baked in a convective oven at 150°C for 120 min. Then, the breads were cooled and stored for 24 h in a dissicator.

2.4 Specific volume analysis

The bread volume was measured according to the 10-05.01 method (AACC, 2000) using standardized diameter millet seeds. The container used to measure the bread apparent volume was a cylinder of plastic material (11 cm diameter, 7 cm height). The specific volume (SV, cm3/g) was calculated as the inverse of the bread apparent density.

2.5 Texture profile analysis (TPA)

It was carried out with a Texture Analyser TA-XT Plus (Stable Micro Systems, Surrey, UK) equipped with a 5 kg load cell. An aluminum probe of cylindrical base (15 mm diameter) was coupled to the texturometer, which deformed the crumb area of bread slices of 25 mm thickness by up to 50%. Then, the following texture parameters were calculated from the force-distance curve: hardness (Ha), cohesiveness (Co), and gumminess (Gu).

2.6 Experimental design and statistical Analysis

The treatments were arranged either in a 3×2 (for SV) or in a 2×2 (for TPA) factorial experiments, with two replicates. The independent variables were $X1$: chickpea flour addition (10, 20, and 30%), and $X2$: flour type incorporated in the dough (raw, and precooked by extrusion). The assessed responses on breads were: specific volume and texture parameters. For each response, factorial ANOVAs following multiple comparison of means by Tukey's test, with α = 0.05, were carried out. Then, response curves were generated.

3. RESULTS AND DISCUSSION

The assessed responses after performing the analysis of variance and Tukey's test are shown in Figure 1. It was verified that breads produced with raw flour showed the highest specific volume values (Figure 1A), regardless of chickpea flour addition (1.15 cm3/g vs. 0.9 cm3/g).

Regarding texture parameters, the hardness of breads produced with raw flour was higher only with 20% chickpea flour addition (55 N vs. 25 N). Bread cohesiveness was higher when precooked flour was used only with 10% chickpea flour addition (0.49 vs. 0.38). With respect to gumminess, it was observed that breads produced with raw flour had greater gumminess only with 20% chickpea flour addition (25 N vs. 12 N).

Figure 1 – Effect of chickpea flour addition and flour type on: A) specific volume,B) Hardness,C) Cohesiveness, and D) Gumminess. Different lowercase letters differ according to the Tukey test.

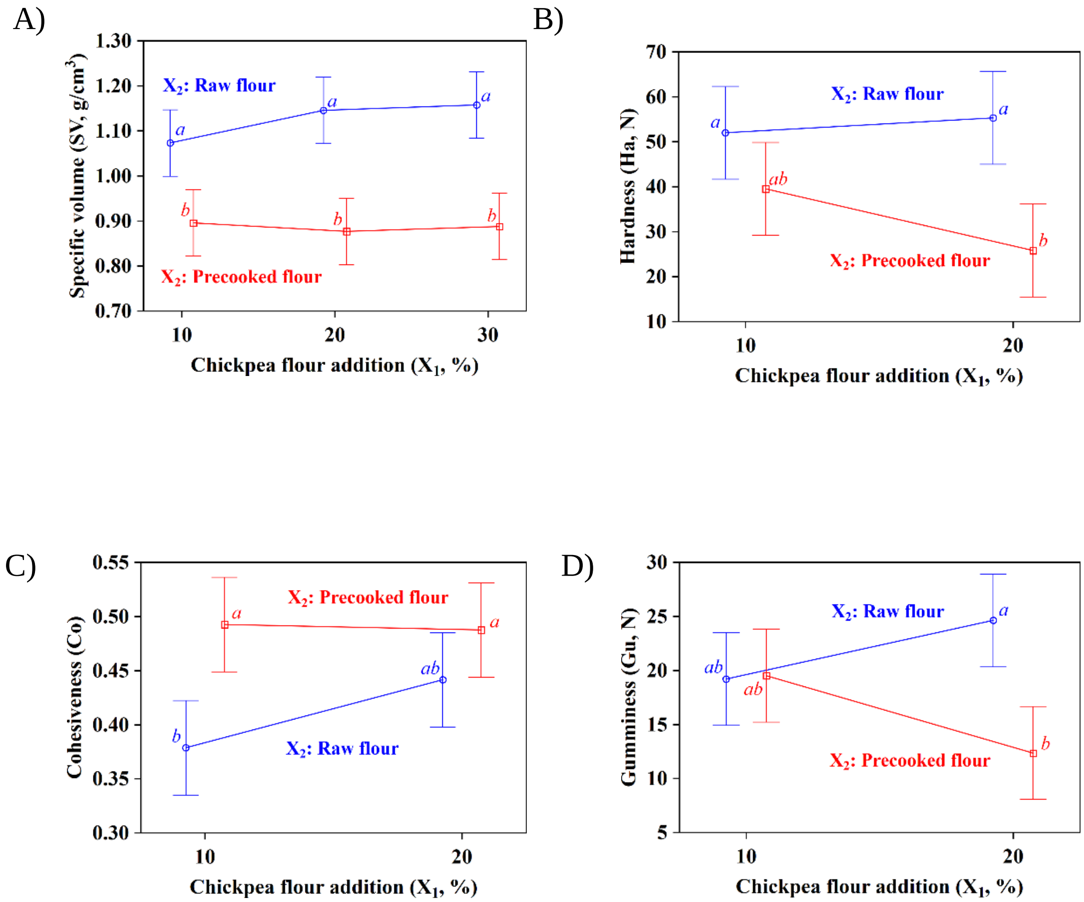
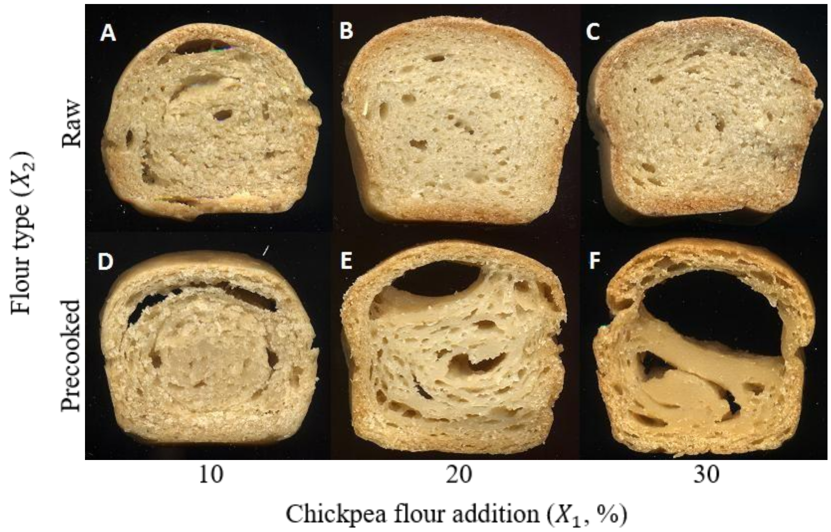


Figure 2 shows the differeces in the breads crumb as a result of chickpea flour addition and the flour type incorporated in the formulation (raw and precooked by extrusion). As the proportion of chickpea flour addition was increased in the precooked flour, a smaller area of the crumb was observed, with the formation of large air bubbles. The bread crumb consistency made with 30% chickpea flour addition and precooked flour (Figure 2F) was more compact than the others. This fact could be associated to a greater water retention by chickpea proteins.

Figure 2 – Visual aspects of bread slices.



4. CONCLUSION

The flour type incorporated in the bread formulation affected the specific volume. Precooked flours decreased the breads' specific volume. Chickpea flour addition in combination with the flour type used affected the breads' texture parameters. Breads produced with raw flour (non extruded) presented uniform crumbs, being better with 20% chickpea flour addition (Figure 2B). These differences could be linked to the combined action of the chickpea proteic fraction under going diverse degrees of modification during the extrusion cooking process. For future works, it is suggested to blend different proportions of raw flour and precooked flour in order to obtain a gluten-free and vegan bread with acceptable technological properties and nutritional characteristics.

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