CARPATHIAN JOURNAL OF FOOD SCIENCE AND TECHNOLOGY

journal homepage: **http://chimie-biologie.ubm.ro/carpathian_journal/index.html**

EVALUATING THE IMPACT OF NON-GLUTEN MODIFICATIONS ON BREAD QUALITY: A STRUCTURAL EQUATION MODELING APPROACH

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https://doi.org/10.34302/crpjfst/2024.16.3.17

Article history:
Received:
September 3rd, 2024
Accepted:
October 1st , 2024

Keywords:
Acylation;
Glycosylation;
Innovation Process;
The Maillard Reaction;
Phosphorylation.

ABSTRACT

While modifications for low-gluten bread production have been extensively reported, including the utilization of Non-Gluten Components (NGC), Natural Hydrocolloids (NH), and Chemical Modification Reaction (CMR). Implementing this modification remains challenging and has yet to meet the expectations for bread quality (BQ). This research aims to propose a measurement model for assessing the effectiveness of modifications (NGC, NH, and CMR) on BQ. This research involved 45 bread companies in Indonesia, and the three modifications were attempted for application. Structural Equation Modeling (SEM), with the Partial Least Square (PLS) approach, was employed for analysis. The findings indicate that the modified (NGC, NH, and CMR) variables did not directly contribute to a positive effect on BQ (T stat. <1.65; p >0.05). However, when mediated by Research and Development (R&D) the three modifications showed a positive impact on BQ, with respective contributions of 19.2% (NGC), 14.6% (NH), and 12.8% (CMR). R&D had a fairly strong influence (f^2>0.35), and 28.2% (R^2) of its indicators were understood by NGC, NH, and CMR. The model's suitability was deemed satisfactory, with SRMR<0.07; GFI>0.36; and NFI>0.9. The original contributions of this research lie in providing practical recommendations for the widespread application of modified variables and proposing conceptual a framework for gluten-free bread modification with R&D mediation.

1.Introduction

Gluten is a complex compound characterized by high allelic polymorphism that gives Specific protein codes for glutenin and gliadin. The nature of gluten is stable in binding heat so that it helps in improving the texture, taste and roasting (Biesiekierski, 2016). Gluten is one of the most common causes of gastrointestinal symptoms and triggers a potentially autoimmune disease *Celiac disease, Glutenataxia, Dermatitis herpetiformis* (Biesiekierski, 2016). These conditions encourage research related to modifications of gluten substitutes that do not cause disease. A healthy lifestyle will encourage the manufacture of gluten-free bread with characteristics similar to bread in general (high gluten bread).

Efforts to make gluten-free bread include the use of Non-Gluten Component ingredients (NGC), Natural Hydrocolloid (NH) and Chemical Modifications Reaction (CMR) by involving Research & Development (R&D) in the innovation process. There is a lot of previous literature related to the manufacture of gluten-free bread with a variety of methods and analysis. Referring to the previous literature review, the use of NGC, NH, and CMR is only a recommendation in the modification of gluten-free bread. There is no study that has tested the effect (NGC, NH, and CMR) on bread quality (BQ).

The aims of this study were: a) to measure the effect of the modified variables (NGC, NH, and CMR) on BQ in the bakery industry. b) provide practical recommendations so that modification efforts can be widely accepted and applied in industrial areas.

1.1. Non-Gluten Component (NGC)

The gluten character Elastic has similarities to starch in the form of thick liquid and is very suitable for textural expansion (Balaghi et al., 2011). Starch is a polysaccharide derived from seeds, fruit, plant extracts, seaweed and microorganisms (Meybodi et al., 2015). Starch in the bread formula as a texture softener, and appearance. Substitution of rice flour is more acceptable as a substitute for wheat flour (Navarro & Araya, 2015)

Dietary fiber in seeds, fruit, and vegetables can be an alternative to gluten in wheat flour (Gan et al., 1989). Dietary fiber can replace the function of gluten based on dietary studies using gluten-free bread when compared to normal bread (Grehn et al., 2001). The addition of dietary fiber can help the stability of the dough so as to produce an even crumb, smooth texture, and sharp and even brown coloring (Gallagher et al., 2004).

Whey protein has a mesoscopic character that helps in retaining CO_2 gas produced by fermentation in bread, slows the movement of water in the bread so that the crumb is soft (Lazaridou et al., 2007). The addition of whey protein can increase water absorption thereby helping in the stretch setting of the dough. Which affects the Milllard browning reaction and preferred caramelization over other additives. N-ethyl maleimide (NEM) Whey protein can block thiol agents resulting in nearly eight times the volume of bread and an increase in rheological properties (van Riemsdijk et al., 2011).

The combination of flour and fermented water is the solution to get gluten-free bread which is rich in vitamins, iron, folate and dietary fiber (Zannini et al., 2012). Sourdough is a combination of flour and water that has undergone a fermentation process with lactic acid bacteria (LAB) and yeast (Gobbetti et al., 2018). The gluten network plays a role in slowing water transfer and maintaining CO_2 gas produced during the fermentation process, although the use of fermentation delays the starch retrogradation process and the cessation of gluten-free bread (Sabanis & Tzia, 2011). Biological acidification, amylolytic and proteolytic activity of cultures are the main mechanisms involved in delayed retrogradation (Rojas et al., 1999). Some LAB do not have amylolytic activity, so to get gluten-free bread with a long shelf life, it is recommended to use sprouted grains (Rojas et al., 1999). The addition of sourdough culture to gluten-free bread can improve the celiac immune system by producing peptides containing proline/glycine through proteolytic activity (Rollán et al., 2005). These conditions make gluten-free bread a functional food because of the ability of LAB to produce exopolysaccharides, especially fructooligosaccharides (Schwab et al., 2008)

Referring to the description above, the research hypotheses are: **H1**: NGC has a direct positive effect on BQ; **H2**: NGC has a direct positive effect on R&D

1.2. Modification of gluten protein chemical reaction (CMR)

The structural and functional linkages seen in food and non-food systems are the main areas of chemical alteration of gluten proteins. Protein side chain chemical modification can enhance: a) nutritional quality as a result of necessary amino acid availability, denaturation, and protein matrix degradation. b) functional characteristics (amount of emulsifier, foaming) c) physical state (texture). Abedi & Pourmohammadi (2020) made careful observations regarding: (i) when an essential amino acid (lysine) is derivatized, the Maillard reaction and base degradation are carried out to produce lysinoalanine compounds. (ii) the presence of chemical residues from the modification results (the amount of lysine is quite low) for the modification reaction (de Jongh & Broerse, 2012). This allows the optimal use of gluten for various purposes in the food

and non-food industries. The hypotheses formed are: **H3**: CMR has a direct positive effect on BQ; **H4**: CMR has a direct positive effect on R&D.

The indicators used for the CMR variable are:

Phosphorylation : An alcohol and a carboxylic acid react to produce the esterification reaction, which requires heat as a source of energy and the acid as a catalyst (Robertson et al., 2014). Phosphorylation is an esterification process brought on by urea and phosphoric acid treatment at temperatures higher than 130^{o}C. Phosphorylated gluten can absorb around 200 times its own weight in water because urea can limit hydrogen bonding and decrease the stability of secondary connections (Robertson et al., 2014). It is believed that esters derived from the hydroxyl groups in the gluten amino acids, tyrosine, serine, and threonine, are responsible for the increased absorptivity of water. Because the esterification step requires dehydration events, the negatively charged phosphate group makes the protein polymer chains oppose one another, increasing water absorption (Robertson et al., 2014).

Glycosylation: Glycation, which is based on glycoconjugates between polysaccharides and proteins, is also known as the Maillard reaction. Protein NH2 residues are reduced to sugar or polysaccharide reducing groups by perlecovalent bonds during glycolysis (Abedi & Pourmohammadi, 2020). The protein molecule's negative charge rises and the base level falls as a result of this circumstance. The glycated protein's isoelectric pH subsequently shifts in the direction of a higher pH. Carbohydrates with more hydroxyl groups and negative charge have altered solubility, water retention, foaming characteristics, antioxidant activity, and stability in hot environments(J. Liu et al., 2012).

Deamidation: Through the process of first conversion, the amide groups present in glutamine and asparagine are transformed into carboxylic groups, resulting in the formation of glutamate and aspartic acid. Carboxylic acids are preferable for protein deamidation due to their superior suitability. Carboxylic acid reactions are preferable for deamidation as they effectively hydrolyze peptide bonds (Liao et al., 2016). The deamidation of carboxylic acids allows for a controlled level of protein hydrolysis and enhances the features and organoleptic properties (Y. Liu et al., 2018). (iii) HCL produces an uncontrolled degree of protein hydrolysis and a large number of isomerization of amino acids and dichloropropanol (Liao et al., 2010) (iv) antioxidant activity accelerates the deamidation of gluten (Qiu et al., 2013) (v) improved nutrition due to digestibility and conversion power to low molecular weight hydroxylates (Cui et al., 2013). (vi) the total amount of essential amino acids and lysine in digestion increases. (vii) carboxylic acid (acetic acid) can be efficient due to increased solubility of functional characteristics (Liao et al., 2010; Qiu et al., 2013) (viii) Deamidated gluten is effective in reducing celiac (Qiu et al., 2013)

Acylation is a chemical modification of the amino group turn into amide (Majzoobi & Beparva, 2014) because acylation groups react with -amino groups (arginine and lysine), aliphatic hydroxyl groups (serine and threonine), and nucleophilic groups, such as phenolic (tyrosine). However, the reactivity of lysine -amino group is higher than others (Majzoobi et al., 2017). Functional features of acylated proteins have increased (water absorption, foaming capacity, emulsion properties, water holding capacity due to: breaking of hydrogen bonds, electrostatic repulsion, dissociation of high molecular weight proteins, less protein-protein interactions, but more protein-water and reduce the content of high molecular weight glutenin, gliadin and glutin (Abedi & Pourmohammadi, 2020).

1.3. Natural Hydrocolloids (NH)

The use of materials that have elastic characteristics such as gluten. Salehi (2019) succeeded in classifying NH based on the findings of previous studies. (i) *Xanthan gum* is widely found in bread making because it increases the volume, and carboxyl methyl cellulose is used as a gluten substitute. (ii) *Guar gum* is a natural latex added to bread dough with a certain level of texture and crumb. (iii)

Carrageenan Gum is a seaweed extraction with low viscosity so it is used for texture improver (Rosell et al., 2001) carrageenan function to reduce firmness and increase the volume of baked bread. (iv) *Methylcellulose* (MC) is widely used in the bakery industry because of its properties (soluble in water, high molecular weight, forms a thickener in water systems) (Sanz et al., 2005). (v) *Carboxy methyl cellulose gum* is a natural hydrocolloid for long shelf life through moisture retention and prevention of syneresis (Ozkoc & Seyhun, 2015), this type is able to provide a barrier layer during heating which causes water and oil loss (vi) *methyl propyl hydroxyl / cellulose gum* is used to improve taste, thickener, and gives a melt sensation in the mouth (Turabi et al., 2008). (vii) *Locust Bean Gum* is added to minimize breakage due to brittle texture. The addition of ingredients ranging from 1-3% can add volume, the crumb is softer (Angioloni et al., 2008). Various kinds of natural hydrocolloids can be used such as balangu seed gum, wild sage seed gum, basil seed gum and crispy seed gum.

The description helps in making research hypotheses, **H5**: NH has a direct positive effect on BQ and **H6**: NH has a direct positive effect on R&D

2. Materials and methods

2.1.Materials

The data were taken from 45 bakery companies spread throughout Indonesia. Observations were made for 15 months (March 2022 – June 2023). Respondents were represented by employees of the Research & Development (R&D) division because the research data was in the form of trial results of NGC, MRC, and NH raw materials in the bread-making process and required data analysis. Research data in the form of qualitative analysis (organoleptic) and quantitative analysis (results of laboratory analysis, such as: gluten content, water content, viscosity, etc.).

We gathered information from respondents regarding the ease of application of ingredients for bread making. The scale we used was as follows: 1. Very difficult to apply; 2. Difficult to apply; 3. Neutral; 4. Easy to apply; 5. Very easy to apply.

Qur R&D variable used the following assessment: 1. Very unexpected; 2. Unexpected; 3. Neutral; 4. expected; 5. Highly expected.

Our "BQ" variable refers to: 1. Very dislike; 2. Dislike; 3. Neutral; 4. Liked; 5. Very liked. However, for gluten content, we used laboratory analysis results with the following criteria: 1. (>3%); **2**.(2.1-3%); **3**.(1.1-2.1%); 4. (0.1-1.1%), and 5 (0%).

2.2.Methods

2.2.1. Determination of Variable Indicators

The selection of indicators using Principal Component Analysis (PCA) with SPSS software is carried out to determine indicators of various activity items to be carried out evaluated. The function of PCA is basically to reduce several variables into new variables or new dimensions which are the result of indicator extraction (Budianto et al., 2022). The choice of PCA was because the indicators used were relatively new so there was no reference regarding the indicators used in the variables (budianto; Kusmardini, 2021).

We focus on the equations related to identifying and selecting the principal components that explain most of the variance in the data. The following are the key steps along with the corresponding equations:

$$X = \begin{bmatrix} x_{11} & x_{12} & \cdots & x_{1p} \\ x_{21} & x_{22} & \dots & x_{2p} \\ \vdots & \vdots & \ddots & \vdots \\ x_{n1} & x_{n2} & \cdots & x_{np} \end{bmatrix} \quad \text{.....(1)}$$

X is the original data matrix with dimensions $n \times p$, where: n is the number of observations (rows in the matrix), and p is the number of variables or indicators measured (columns in the matrix). x_{ij} is an element of the X matrix, which represents the value of the j-th variable (column) for the i-th observation (row). Thus, x_{ij} represents the observed data for variable j in the i-th observation.

Covariance indicates the direction of the relationship between variables, whether they move together (positive), in opposite directions

(negative), or have no linear relationship (values close to zero)

$$C = \frac{1}{n-1} X^T X \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

C: Covariance Matrix; X is the original standardized data matrix; X^T: the transpose of the X matrix.

Eigen decomposition to obtain eigenvalues and eigenvectors

$$Cv = \lambda v \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3)$$

v: Eigenvector, representing the linear combination of the original variables that define the principal component. λ: Eigenvalue, representing the variance explained by the principal component.

The percentage of variance explained by each principal component:

$$\mathrm{VE}_k = \frac{\lambda_k}{\sum_{j=1}^{p} \lambda_j} \text{ X } 100\% \ldots\ldots\ldots\ldots\ldots\ldots\ldots (4)$$

VEk: Explained Variance k; λk : Eigenvalue of the k-th principal component; *p*: Total number of principal components.

New indicators based on factor score values are calculated based on the contribution of each variable in the main component, and then used as new indicators.

$$Factor\ Score_i = \sum_{k=1}^{m} PC_k \text{ x } W_k \ldots (5)$$

Factor Score$_i$: The factor score for the i-th observation, used as a new indicator; PC_k: The *k*-th principal component that has been calculated; W_k: The weight or coefficient for the k-th principal component used in the formation of the factor score; m: The number of principal components considered (usually those with the largest eigenvalues

2.2.2. Statistical Analysis

Test the analysis to see how much influence the relationship between variables. Testing the effect of variables using Structural Equation Modeling (SEM), with Partial Least Square (PLS) approach with Smart PLS software version 6.0. Validity test using cross loading value > 0.6 and the value of Square Root of Average Variance Extracted (AVE) > 0.50.

$$AVE = \frac{\sum_1^n \lambda_i^2}{n} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (6)$$

λ_i is the factor loading of the i-th indicator, and *n* is the number of indicators. An AVE greater than 0.5 is generally considered to indicate good convergent validity

Reliability test with Cronbach's Alpha value > 0.6, Composite Reliability > 0.7. Structural model testing by accommodating all construct variables formulated in hypothesis testing. All standard parameters refer to Hair et al. (2011)

$$CR = \frac{\sum_1^n \lambda_i^2}{\sum_1^n \lambda_i^2 + \sum_{i=1}^n \theta \mathrm{i}} \ldots\ldots\ldots\ldots\ldots\ldots (7)$$

CR:composite reliability, Where θi is the error variance of the i-th indicator. Additionally, Cronbach's Alpha is also used to measure reliability, which is formulated as follows:

$$\alpha = \frac{n}{n-1} (1 - \frac{\sum_{i=1}^n \sigma_{\epsilon i}^2}{\sigma_X^2} \ldots\ldots\ldots\ldots\ldots\ldots (8)$$

$\sigma_{\epsilon i}^2$ is the error variance of the *i*-th indicator, and σ_X^2 is total variance of construct X

Coefficients Q^2 and f^2

The Q^2 coefficient is used to measure the predictive relevance of the model, which is formulated as follows:

$$\mathrm{Q}^2 = 1 - \frac{SSE}{SST} \ldots\ldots\ldots\ldots\ldots\ldots\ldots (9)$$

SSE:Sum of Squared Errors, and SST: Total Sum of Squares.

Meanwhile, the f^2 coefficient measures the effect size of the exogenous variable on the endogenous variable

$$\mathrm{f}^2 = \frac{\mathrm{R}^2_{\mathrm{included}} - \mathrm{R}^2_{\mathrm{excluded}}}{1 - \mathrm{R}^2_{\mathrm{included}}} \ldots\ldots\ldots\ldots\ldots\ldots (10)$$

2.2.3. Model testing

Model testing was conducted using SEM-PLS with Smart PLS software to assess the influence of variables and test hypotheses. Model adequacy was assessed based on Hair et al.(2011) for SRMR (<0.07), Chi-Square, NFI (>0.9), and Goodness of Fit index (>0.36) values.

$$SRMR = \frac{1}{p(p+1)/2} \sum_{i<j}(\sum_{ij} - S_{ij} \ldots (11)$$

$$\text{Chi} - \text{Square Test} = (N-1)(\sum - \sum(\theta))^{T}((\sum - \sum(\theta))^{-1} \ (12)$$

$$GFI = 1 - \frac{\sum(S_{ij} - \sum_{ij})^2}{\sum S_{ij}^2} \ldots\ldots\ldots\ldots\ldots (13)$$

2.2.4. Research Framework

The research framework can be seen in Fig. 1, the variables NGC, MRC, and NH as independent variables, the R&D variable as the mediating/intervening variable and BQ as the dependent variable.

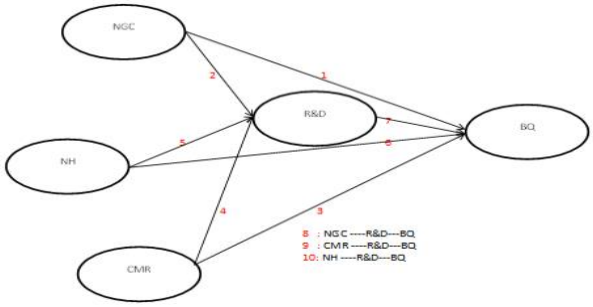


Figure1. Research Framework

3. Results and discussions

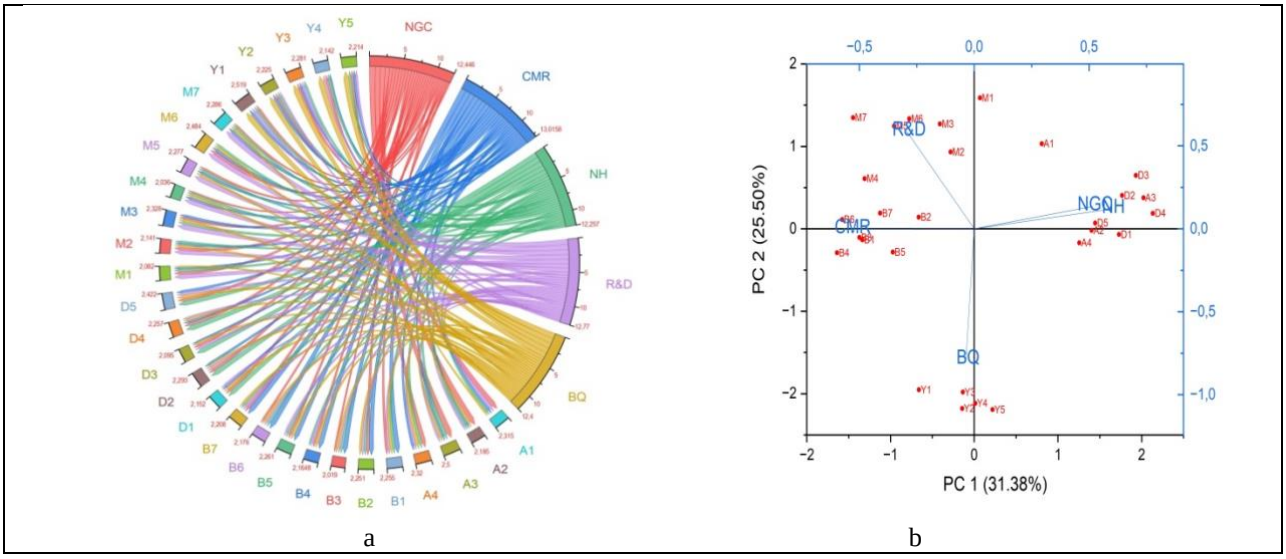
3.1. Principal Component Analysis (PCA)

Data categorization was performed using numerical representations (Table 1) and visualizations (Fig.2) based on the chord diagram and PCA. Referring to Table 1, the indicators of NGC were: A1, A2, A3, and A4. CMR indicators include: B1, B2, B3, B4, B5, B6, and B7. NH indicators consist of D1, D2, D3, D4 and D5. Indicators for the R&D variable include: M1, M2, M3, M4, M5, M6, and M7. Meanwhile, indicators for BQ comprise: Y1, Y2, Y3, Y4, and Y5.

Table 1. Determination of indicators using Principal Component Analysis (PCA)

Indicator	Code	Rotation Method: Varimax with Kaiser Normalization VARIABLE (* significant p<0.05)				
		NGC (1)	**CMR** (2)	**NH** (3)	**R&D** (4)	**BQ** (5)
Starch	A1	**0,754**	0,432	0,356	0,487	0,286
Food fiber	A2	**0,778**	0,375	0,358	0,298	0,376
Whey protein	A3	**0,873**	0,398	0,475	0,369	0,385
Fermentation (flour + water)	A4	**0,634**	0,365	0,472	0,382	0,467
Phosphorylation	B1	0,347	**0,743**	0,389	0,387	0,389
glycosylation	B2	0,452	**0,658**	0,396	0,391	0,354
Glycosylation (gluten+glucose)	B3	0,257	**0,672**	0,385	0,361	0,344
Glycosylation (Gluten + lactose)	B4	0,367	**0,753**	0,298	0,358	0,388
Glutamic acid deamidation	B5	0,392	**0,685**	0,389	0,374	0,421
Deamidation of carboxylic acids	B6	0,298	**0,779**	0,395	0,382	0,322
acylation	B7	0,438	**0,743**	0,352	0,364	0,311
Xanthan gum gum	D1	0,472	0,342	**0,658**	0,298	0,382
Guar Gum	D2	0,435	0,352	**0,745**	0,397	0,364
Carrageenan Gum	D3	0,389	0,267	**0,749**	0,392	0,298
Methylcellulose (MC)	D4	0,482	0,367	**0,763**	0,298	0,347
Methyl carboxy cellulose gum	D5	0,473	0,392	**0,684**	0,421	0,452

Existing product innovation	M1	0,481	0,298	0,374	**0,672**	0,257
Existing product evaluation	M2	0,459	0,338	0,334	**0,643**	0,367
Material composition evaluation	M3	0,378	0,372	0,443	**0,753**	0,382
Low price orientation	M4	0,238	0,435	0,344	**0,632**	0,387
Quality standards based on consumers	M5	0,349	0,389	0,364	**0,784**	0,391
Product innovation based on existing machines	M6	0,487	0,482	0,361	**0,768**	0,386
Material composition trial	M7	0,298	0,473	0,358	**0,793**	0,364
Gluten content	Y1	0,369	0,481	0,374	0,452	**0,843**
Form	Y2	0,382	0,459	0,382	0,257	**0,745**
Color	Y3	0,387	0,378	0,364	0,367	**0,785**
Smell	Y4	0,391	0,238	0,298	0,392	**0,823**
Taste	Y5	0,386	0,349	0,397	0,298	**0,784**



a b

Figure 2. Determination of indicators from the variables based on the correlation between indicators using the Chord Diagram (a), and the correlation of indicators based on PCA testing (b).

The chord diagram (Fig.2a) showed consistent findings, dividing the indicator item into 5 variables with nearly identical bandwidth (12.5). Additionally, we illustrated the indicator mapping using the biplot method with PCI (31.38%) and PC2(25.5%), which revealed identical indicators across the five tested variables.

3.2. Descriptive Statistics

The mean of the NGC variable was good (3.5-4) although it was found that A2=3.44 (table 2). The order of using NGC that was in great demand was A3, A1, A4 and the lowest was A2. The role of A2 was categorized as "adequate" in the process of making gluten-free bread, while other indicators (A3. A1, and A4) were categorized as good (widely used, easy to apply).

The mean of the CMR variable was bad (<3), it can be seen from B5 (2.6), B7 (2.8) and B6 (2.9) although found B1 (3.40), B3 (3.32), B2 (3.2) and B4 (3.20) The order of using CMR based on respondents' interests was B1, B3, B2, B4 B6, B7 and the lowest was B5 and B7 categorized as bad.

Table 2. Descriptive Statistics of Respondent Test Results.

Descriptive Statistics					
SIZE SCALE					
1. Very difficult to apply	2. Difficult to apply	3, Neutral	4. Easy to apply	5. Very easy to apply	
Variable	Statistic	Bootstrap[a]			
		Std. Deviation	Std. Error	95% Confidence Interval	
				Lower	Upper
Non-Gluten Component (NGC)					
Strach (A1)	3.7600	.52281	.1023	3.5200	3.9200
Food fiber (A2)	3.4400	.71181	.1398	3.1600	3.6800
Whey protein (A3)	3.8000	.40825	.0814	3.6400	3.9600
Fermentation (A4)	3.5200	.65320	.1227	3.2800	3.7600
Average	**3.6300**				
Modification of gluten protein chemical reaction (CMR)					
Phosphorylation (B1)	3.4000	.86603	.1662	3.0800	3.6800
Glycosylation (B2)	3.2000	.81650	.1642	2.8800	3.5200
Glycosylation (gluten+glucose) (B3)	3.3200	.85245	.1672	3.0000	3.6400
Glycosylation (Gluten + lactose)(B4)	3.2000	.81650	.1593	2.8800	3.4800
Glutamic acid deamidation(B5)	2.6000	.57735	.1126	2.4000	2.8400
Deamidation of carboxylic acids(B6)	2.9600	.61101	.1220	2.7200	3.2000
Acylation (B7)	2.8800	.60000	.1159	2.6400	3.1200
Average	**2.9100**				
Natural Hydrocolloids (NH)					
Xanthan gum gum (D1)	3.5600	.76811	.1526	3.2400	3.8400
Guar Gum (D2)	3.3600	.81035	.1570	3.0400	3.6400
Carrageenan Gum (D3)	3.6400	.63770	.1225	3.4000	3.8790
Methylcellulose (MC) (D4)	3.5600	.58310	.1205	3.3200	3.8000
Methyl carboxy cellulose gum (D5)	3.0800	.64031	.1234	2.8400	3.3590
Average	**3.4100**	0	0	105	105
SIZE SCALE					
1. Very unexpected	2. Unexpected	3, Neutral	4. Expected	5. Highly expected	
Research & Development (R&D)	Statistic	Bootstrap[a]			
		Std. Deviation	Std. Error	95% Confidence Interval	
				Lower	Upper
Existing product innovation (M1)	3.8000	.40825	.0832	3.6000	3.9600
Existing product evaluation (M2)	3.8400	.37417	.0716	3.6800	3.9600
Material composition evaluation (M3)	3.8400	.37417	.0730	3.6800	3.9600
Low price orientation (M4)	3.8000	.50000	.1010	3.5600	3.9600
Quality standards based on consumers (M5)	3.9200	.27689	.0550	3.8000	4.0000
Product innovation based on existing machines (M6)	3.7600	.59722	.1170	3.4800	3.9600
Material composition trial (M7)	3.8400	.37417	.0716	3.6800	3.9600
Average	**3.8300**				
SIZE SCALE					
1. Very dislike	2. Dislike	3. Neutral	4.Liked	5.Very liked	
1. gluten > 3%	2. gluten 2.1-3%	3. gluten 1.1-2.1%	4. gluten 0.1-1.1%	5. gluten 0%	
Bread Quality (BQ)	Statistic	Bootstrap[a]			
		Std. Deviation	Std. Error	95% Confidence Interval	
				Lower	Upper
Gluten content (Y1)	3.7600	.43589	.0833	3.6000	3.9200
Form (Y2)	3.8400	.37417	.0729	3.6800	3.9600
Color (Y3)	3.7200	.45826	.0896	3.5210	3.8800
Smell (Y4)	3.7200	.54160	.1084	3.4800	3.9200
Taste (Y5)	3.7200	.45826	.0896	3.5200	3.8800
Average	**3.7500**				

The mean of the NH variable was sufficient (3-3.5) it can be seen from, D2, D6) although there were some indicators that were worth >3 (D1, D3, D4) The order of using NH based on the respondents' interests was D3, D1, D4, D2 and the lowest was D5. The roles of D1, D3, D4 were categorized as "good" in the process of making gluten-free bread, while other indicators (D2, D5) are categorized as adequate.

The average R&D variable was good (3.5-4) it can be seen from, M1, M2, M3, M4, M5, M6, and M7. The order of the use of NH based on the respondent's interest was M5, M7, M2, M1, M4, M3 and the lowest was M6. The role of all indicators was categorized as "good" in the mediation process for making gluten-free bread.

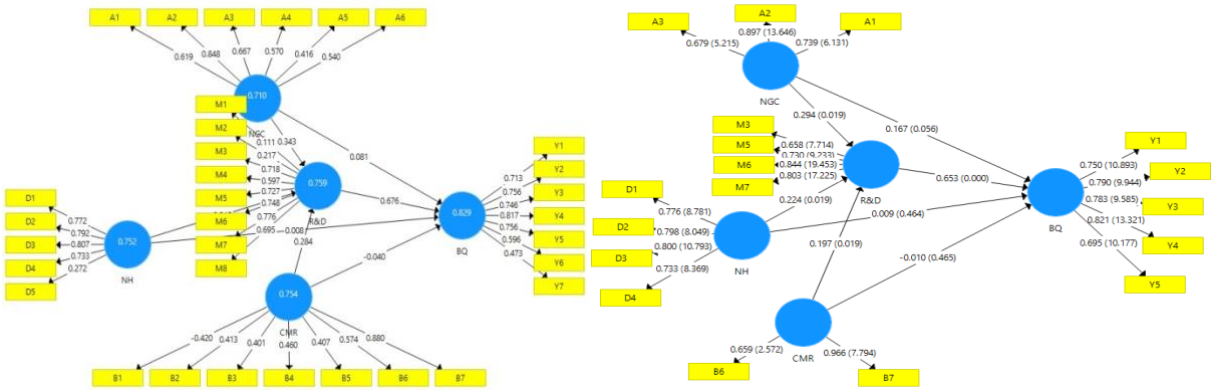
The mean of BQ variable was good (3.5-4). The order of the NH indicators based on the research results was Y2, Y3, Y4, Y5, and the lowest was Y1. The role of all indicators was categorized as “good” in the results of making gluten-free bread. Y1 scale determination was based on the gluten content that was still detected in the bread-making process (table 2).

3.3. SEM_PLS analysis

Determination of indicators using PCA helps in mapping based on the variables, although the indicator will be partially deleted by SEM PLS if the outer loading value is <0.6. The following are the results of the analysis using SEM PLS.

All indicator and their variables were subjected to bootstrap testing, and the loading factors of each indicator are depicted (Fig.3a). To ensure valid and reliable data, indicator loading factors > 0.55 were selected, while those below 0.55 had to be removed. The analysis outcomes are presented in Fig.3b.

Based on Fig.3b, the validity and reliability of the research model can be seen in table 3. Convergent validity refers to the extent to which indicators are able to measure the same variable, in this case, all indicators were able to measure their variables based on outer loading, AVE, and commonality.



a b

Figure 3. SEM_PLS analysis. Loading factor for all indicators (a), and Loading factor > 0.55 (b)

Table 3. Test the Validity and Reliability of the Research Model

Test	Parameter	Standard	Results
Convergent Validity	Factor loading (outer loading)	>0.55	0.659 -0.966
	AVE	>0.5	0.581– 0.684
	Commonality	>0.5	0.581 – 0.684
Discriminant Validity	Root Square AVE and Correlation latent variables	Root Square AVE > Discriminant validity	Root Square AVE> Discriminant Validity
	Cross Loading	>0.6	0.726 – 0.906
Reliability	Cronbach's Alpha	>0.6	0.614 – 0.827
	Composite Reliability	>0.7	0.807 – 0.878

Discriminant validity, which explained that the three variables in the model could be distinguished from one another, was demonstrated in this research model. The model's reliability also met the requirements based on Cronbach's Alpha, and Composite Reliability. All validity and reliability tests were met, so that further tests (variable influence tests) could be carried out.

Based on table 4, the accepted hypotheses were: H2, H4, H6, H7, H8, and H9. While the rejected hypotheses include: H1, H3, and H10. Exogenous variables (NGC, CMR, and NH) all did not have a direct positive impact on BQ, but were able to have a positive effect on BQ if mediated by R&D, this was because R&D had a strong effect of 69.2% (f^2) on BQ. Only H3 had a direct negative impact on BQ. The R&D variable was able to explain the exogenous variable by 28.2% (R2), while the BQ variable was 55.7%.

Table 4. The Effect of Variable Paths and model fit

Hypothesis	Paths	Coefficient (β)	T statistics >1.65	p Value < 0.05	f^2	Remark
1	NGC—BQ	0.167	1,541	0.062	0.039	(+) Not significant
2	NGC—R&D	0.294	2.104	0.018	0.079	(+) significant
3	CMR—BQ	-0.010	0.088	0.465	0.000	(-) not significant
4	CMR—R&D.	0.197	2.274	0.012	0.052	(+) significant
5	NH—BQ	0.009	0.086	0.466	0.000	(+) not significant
6	NH.—R&D	0.224	2.115	0.017	0.046	(+) significant
7	R&D---BQ	0.653	7.286	0.000	0.692	(+) significant
8	NGC—R&D--BQ	0.192	2012	0.022		(+) significant
9	CMR—R&D--BQ	0.128	2,153	0.016		(+) significant
10	NH—R&D--BQ	0.146	2002	0.023		(+) Not significant
f^2 : 0.02- 0.15 Weak Effect; f^2 : 0.15-0.35 Sufficient Effect ; f^2 : 0.35 Strong Effect						
R^2: R&D 0.282……BQ: 0.557						

Model fit	Standard	Saturated model	Estimated model
SRMR	< 0.07	0.054	0.054
Chi-Square	<60.76	51.261	51.261
NFI	> 0.9	0.908	0.908
GFI	>0.36	0.365	0.365

3.4. Model Fit

Throughout our analysis process, we ensured that the model we proposed satisfies several model fit indices.

We chose SRMR to assess how well our research model fits the patterns of relationships among variables in the data. A low SRMR value indicates that the model fits the data well. In this study, the SRMR met the criterion (<0.07).

Although Chi-Square is rarely used as an evaluation measure in PLS models, we employ this analysis to assess the quality of the model built from observational data. In this study, the model also met the criteria for the Chi-Square value (<60.76).

The NFI assesses the extent to which this research model fits the baseline model and lacks correlation between variables. It helps gauge the model's fit with the most basic patterns. In this study, the NFI of this research model was quite high (> 0.9).

The main focus of this research is to measure the effect of the modified variable (NGC, NH, and CMR) to BQ. If you look at the one-way analysis, the three variables were unable to exert a positive impact on BQ, two variables showed a not significant positive effect (NGC and NH), while one variable (CMR) had a negative impact. Referring to the value of one-way analysis, NGC was superior to NH and CMR. Looking at the results of the two-way analysis/mediation role, two variables were able to have a positive effect on BQ (H8 and H9) while H10 has not had a positive effect on BQ. These conditions indicate that the NGC variable was more influential than CMR and NH.

R&D has succeeded in carrying out its function as an intervening (mediation) variable. R&D activities were able to encourage activities in CNG, CMR, and NH through the evaluation of material composition (M3) for easy application and there was no overlapping of modification functions. R&D explores consumer desires for quality reference (M5), this will facilitate the innovation of bakery products based on consumer tastes. Innovations were carried out by R&D by utilizing the condition of the machine (M6) so that the flexibility of the production process would be achieved. Material composition trial (M7) was the main key along with the trial frequency, it will be easy to understand the character of each ingredient.

The next goal of this research was to recommend is practical so that modifications can have a positive effect on BQ. Digging up failure information in the application then evaluating and activating R&D practices in problem solving. The following are the constraints and the role of mediation (R&D) in this study.

NGC did not have a direct positive effect on BQ, because (i) the number of uses was very varied to the standard of friability, and the results of research trials showed that the use of 10-20% could increase the volume 8 times, this strengthens the findings Nunes et al. (2009). (ii) the addition of starch and dietary fiber causes a decrease in vitamins, iron and folate, this was anticipated by the addition of water then fermented with lactic acid bacteria, and the results of the study were in line with the findings Schwab et al. (2008).

CMR had a negative effect on BQ, it shows that there were still many obstacles in its application if it was not mediated. The constraints and the role of mediation were as follows: (i) Solubility in phosphorylation was unstable, the results of research trials showed protein solubility at pH>6. (ii) unstable molecular weight so the role of R&D must be to maintain hydrophilic substitution and the formation of non-disulfide cross-links between protein sub-units. (iii) the strength of the gel was not strong so it must control the reaction of the carbamide formation step, this causes the modified gel to become firm. The findings are supported by Robertson et al. (2014). (iv) understanding the digestibility of gliadin takes a long time, this understanding is broken by the results of R&D analysis through a simulation of stomach and intestinal digestion for 2 hours. This finding strengthens previous research (Xue et al., 2019). (v) protein solubility was very slow, so R&D made efforts to conjugate gluten hydroxylate with glucosamine and transglutamine. (vi) in the deamidation process, there was susceptibility to the addition of alkali, this condition cannot be avoided so efforts are made to use alkali for the generation of fibrous microstructures. This finding is in line with previous research (Li et al., 2018).

Table 5. Constraints and their solutions

Variable	Indicator	Constraints	The role of R&D
Non-Gluten Component (NGC)	Starch	- Addition of starch and dietary fibre leads to a decrease in vitamins. iron, and folate - Loss of dough elasticity - Dough is very crumbly - Dough is not sticky	- Addition of water for lactic acid bacteria fermentation process - Addition of 1-3% Xantan Gum - Addition of 3-5% egg - Addition of 5-10% corn starch
	Food Fiber	- Decreases the volume of the bread - Produces a rough texture - Other odours and flavours emerge	- Addition of 2-3% additives such as hydrocolloids - Improving texture with the addition of 1-2% binding agent (Xantan Gum) - Offsetting flavour and aroma with the addition of dietary fibre.

	Whey protein	- Coagulation during dough mixing - Shorter product life	- Addition of 1-4% emulsifier - Addition of 2-5% natural preservatives
Modification of gluten protein chemical reaction	Phosporylation	- Potential to alter the functional properties of the protein - Inconsistent texture	- Use of phosphate concentration of 0.2-0.5 mol/L - Reaction temperature 25^{o}C and neutral pH
	Glycosylation	- Not all reaction results are safe for consumption - Addition of sugar significantly affects texture and flavour	- Choosing ingredients that are gluten and allergen free - Reducing the amount of sugar in the ingredients
	Glycosylation (gluten+glucose)	- Increases the sugar content of the product - Unstable molecular weight	- Replacing glucose with plant-based sweeteners - hydrophilic substitution and formation of non-disulfide crosslinks between protein sub units
	Glycosylation (Gluten + lactose)	- Lactose allergy - Slow glycoprotein solubility	- Addition of almond milk or soya milk. - Conjugation of gluten hydroxylate with glucosamine
	Glutamic acid deamidation	Bitter taste of glutamate	The addition of 1% CH_3COOH can prevent excessive deamidation.
	Deamidation of carboxylic acids	- Damage to protein structure - Unhealthy trans fat content - susceptible to alkali addition	- Use of transglutaminase enzyme - Addition of vegetable fat (olive oil) - Use of alkali for the generation of fibrous microstructures
Natural hydrocolloids	Xantan Gum	- Organoleptic of bread changes depending on the type of NH - Has a high viscosity - Produces a thick texture in bread dough	- Addition of pectin - Reducing viscosity with the use of xantam gum only in the range of <10% - Addition of water <5%
	Guar gum	- Unstable at low pH - Unstable at high temperatures	Avoid manufacturing process at low pH and high temperature
	Carrageenan gum	- Has low viscosity at low temperatures	- Carragenan gum is preheated before mixing into the dough
	Methylcellulose	- Hydration takes a long time - Coagulation occurs	Longer stirring for homogeneity
	Metyl carboxy cellulose gum	- Degradation at low pH - Low dough binding capacity - Brittle if heated at high temperature (>160^{o}C)	- Mixing is done at neutral pH - Mixing with other binders

NH did not have a positive effect on BQ if it was not mediated by R&D. This was because (a) the organoleptic changes in bread, so it was necessary to add pectin to maintain the organoleptic. The findings of this study are in line with the findings of Roman et al. (2019). (b) the viscosity of the carrageenan extract was low, R&D offered another function as a texture improver and reduced firmness and to increase the specific volume of the baked product although it did not help maintain the general structure of the bread. Product diversification by utilizing the natural properties of carrageenan is also carried out by Rosell et al. (2001).

Table 5, maps the problem of gluten-free bread modification and practical recommendations for research. Practical recommendations refer to the problem of modifying gluten-free bread in the Indonesian bakery industry. This study also provides a conceptual framework related to the application model of gluten-free bread modification with a mediation.

Practical recommendations include re-evaluating the modification process with an emphasis on optimizing the use of NGC and NH and improving the solubility of CMR. Continuous efforts in research and development are needed to overcome the challenges faced, such as solubility and gel strength in CMR. This study provides a foundation for the development of higher-quality and innovative gluten-free bread products.

4. Conclusions

This study proposes a measurement model to evaluate the effectiveness of modification (NGC, NH, and CMR) on bread quality (BQ). The findings indicate that NGC and NH showed a non-significant positive impact on BQ, while CMR had a negative impact. R&D played a crucial role as a mediator, facilitating modification activities and addressing constraints to achieve the desired positive impact on bread quality.

Practical recommendations include re-evaluating the modification process with an emphasis on optimizing the use of NGC and NH and improving the solubility of CMR. Continuous efforts in research and development are needed to overcome the challenges faced, such as solubility and gel strength in CMR. This study provides a foundation for the development of higher-quality and innovative gluten-free bread products.

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Acknowledgment

I would like to thank all the respondents (R&D) in the food company who helped in the modification test of gluten-free bread making.