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The Effect of Fermentation Time and Bimo-CF Starter Concentration on the Chemical Characteristics and Pasting Properties of Corn Flour

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ABSTRACT

Background: Corn flour represents a promising alternative due to the increasing national corn production; however, its utilization in food products is constrained by the absence of gluten, resulting in inferior viscoelastic and textural properties compared to wheat flour. Therefore, the modification of corn flour is essential to enhance its functional characteristics, enabling broader application in value-added products such as breakfast cereals, which may also contribute to improving nutritional quality and addressing stunting in Indonesia.

Methods: Design expert software version 13 was used for statistical analysis. The experiment has been planned in simplex lattice design (SLD) using response surface methodology (RSM) which is an appropriate technic for optimization. The factors tested were the fermentation time (lower limit = 20 hours, upper limit = 60 hours) and the concentrations of starter (lower limit = 0.1%, upper limit = 0.3%). The data obtained were subjected to analysis of variance (ANOVA).

Result: Fermentation time plays a more dominant role than starter concentration in influencing the characteristics of fermented corn flour. Increasing fermentation time increases starch viscosity and digestibility and decreases starch content, gelatinization temperature and water absorption due to changes in starch structure during fermentation. Meanwhile, the setback value and soluble protein remain relatively unchanged. Overall, longer fermentation produces corn flour with better functional properties for food applications.

Key words: BIMO-CF, Corn flour, Fermentation, Starch modification.

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INTRODUCTION

Zea mays L. also known as corn or maize, is an important annual grain crop belonging to the *Poaceae* family (Madan *et al*., 2022). To increase public interest in consuming corn products, it is necessary to diversify corn products in the form of bakery products or other functional foods (Aini *et al.*, 2019). The problem is that the use of corn flour in food products faces obstacles because it cannot form a viscoelastic dough like wheat flour. Wheat flour contains glutenin and gliadin, which when added to water and made into dough will form gluten (Agba *et al*., 2024). Gluten produces a viscoelastic dough that is easy to shape and helps form texture (Ma *et al.*, 2021). If the dough is made from non-wheat flour that cannot form gluten, it cannot form a viscoelastic dough. This results in suboptimal texture formation in food products made from non-wheat flour and in products such as cookies, the texture is hard and not crumbly (Farkas *et al.*, 2021).

Therefore, corn flour needs to be modified so that it has dough-forming properties similar to wheat flour. One possible application of corn flour is in the form of breakfast cereal (Onyekwelu *et al.*, 2022). Breakfast cereal can be an alternative food to prevent stunting, one of the serious nutritional problems that still exists in Indonesia.

MATERIALS AND METHODS

Materials and tools

This study was conducted at the Agricultural Technology Laboratory and the Integrated Academic Building Laboratory at Jenderal Soedirman University from July to August 2025. The materials used in this study were Bisi 18 corn, citric acid “Gajah” brand, BIMO-CF starter and chemicals for analysis. The tools needed for this research are lidded containers, analytical scales, digital scales, flour grinders, 80-mesh sieves and tools for analysis.

Experimental design

Design expert software version 13 was used for statistical analysis. The experiment has been planned in simplex lattice design (SLD) using response surface methodology (RSM) which is an appropriate technic for optimization.

The factors tested were the fermentation time (lower limit = 20 hours, upper limit = 60 hours) and the concentrations of starter (lower limit = 0.1%, upper limit = 0.3%). The data obtained were subjected to analysis of variance (ANOVA).

Corn flour preparation

The preparation of corn flour samples began with washing the cracked corn until clean. Then, it was soaked in 1.5% citric acid for 12 hours. After that, the corn was rinsed until clean and fermented by soaking it in BIMO-CF starter with various concentrations and fermentation times. The factors tested were the fermentation time (lower limit = 20 hours, upper limit = 60 hours) and the concentrations of starter (lower limit = 0.1%, upper limit = 0.3%). There are 13 running in this research. Next, the corn is washed thoroughly, then dried for 12 hours at 60°C in an oven dryer. Then, the corn is ground into flour, then dried again for 4 hours at 60°C in an oven dryer. The dried flour is stored in a closed container.

RESULTS AND DISCUSSION

Peak viscosity

Peak viscosity indicates water absorption and the ability of starch granules to expand when heated with sufficient water. High peak viscosity indicates strong thickening power, which is highly desirable in food applications to create a thick texture (Riaz and Pasha, 2021). Fig 1 presents a 3D model graph with a p-value of 0.0051 ($p<0.05$). Increasing peak viscosity is significantly influenced by fermentation time. The longer the fermentation time, the higher the peak viscosity. However, increasing the starter concentration does not have a significant effect, although it slightly increases peak viscosity. The lowest peak viscosity in this study was at 40 hours fermentation time and 0.2% starter, at 3128 cP, while the highest was 4427 cP in corn flour fermented for 68 hours with a starter concentration of 0.2%.

The maximal viscosity of corn flour and cereal flour is increased by the extended fermentation. This is because amylose is released when starch granules break down, increasing the flour's thickening qualities. These outcomes align with those of Qi *et al*. (2020), who used *Lactobacillus plantarum* Y1 to ferment corn flour. This is due to the fermentation process involving starch hydrolysis, which produces simpler sugars and acids. These compounds interact with starch granules, causing granule expansion and increasing viscosity (Qi *et al.*, 2020).

Breakdown

Breakdown (BD) or viscosity reduction indicates resistance to heat and agitation. The greater the viscosity reduction, the less stable the starch is to heat and agitation. Conversely, if the viscosity reduction value is small or low, it indicates that the starch is more stable to heat (Riaz and Pasha, 2021). With a model p-value of 0.0357 ($p<0.05$), the 3D model graph shown in Fig 2 is appropriate for describing the breakdown reaction.

An increase in fermentation time had a substantial impact on the rise in peak viscosity. Nevertheless, the BD value did not considerably rise with an increase in starting concentration. This indicates that while an increase in starter somewhat increased the drop in viscosity, the longer the fermentation period, the greater the viscosity decrease. The greatest decrease in viscosity at 68 hours fermentation time and 0.2% starter concentration, at 2327 cP, while the smallest decrease in viscosity at 40 hours fermentation and 0.2% starter at 1223 cP.

As fermentation time grew, so did the breakdown value, which measures the paste's stability upon heating. This is because the paste becomes less stable under shear stress due to the partial hydrolysis of starch. Furthermore, the fermentation process damages the starch granules' internal structure, increasing their vulnerability to disintegration after heating.

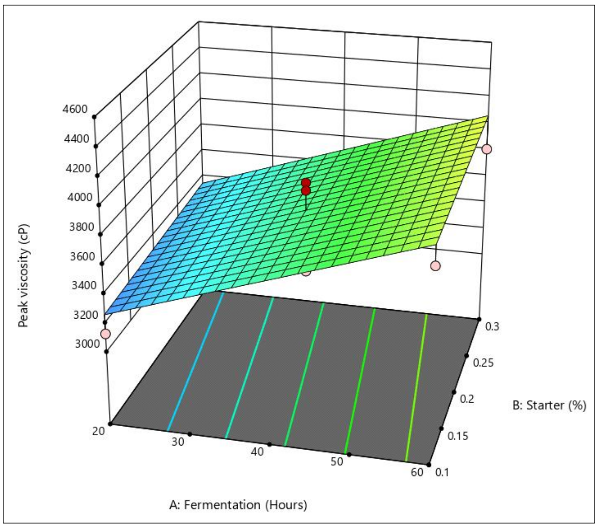

Fig 1: Peak viscosity of fermented corn flour.

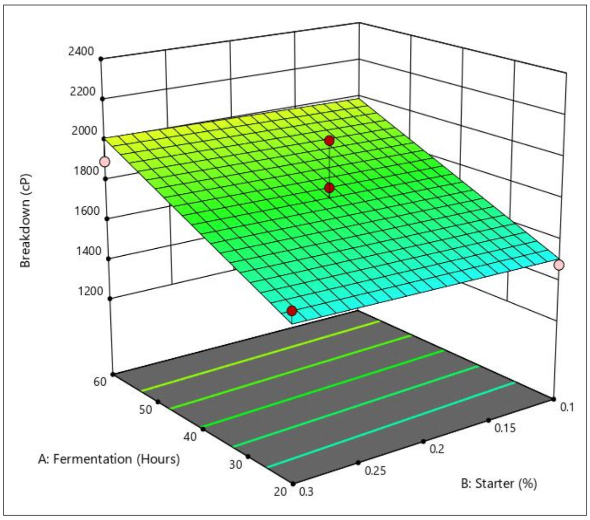

Fig 2: Breakdown (BD) of fermented corn flour.

Setback

Setback indicates an increase in viscosity during cooling or a tendency toward retrogradation. Starches with high setback values have a high tendency to harden during retrogradation. Meanwhile, starch with a low setback value will have a texture that remains soft after undergoing retrogradation (Riaz and Pasha, 2021). The setback value remained comparatively stable throughout all treatment combinations. Changes in the setback value were not significantly impacted by either fermentation duration or starter concentration. The setback value range for fermented corn flour was 2257 to 2727 cP, with no significant difference.

The relatively similar setback values in this study are thought to be due to insufficient fermentation time and too small a difference in starter concentration. Yang and Xiao (2021) show that fermenting corn flour for 3 days can increase setback. Therefore, 12-68 hours fermentation in this study is considered insufficient to have a significant effect on setback values.

Peak time

Peak time indicates how thick the starch becomes when heated with sufficient water. A shorter peak time generally results in a faster processing time in its application (Riaz and Pasha, 2021). The peak time increases with the length of fermentation. Although it marginally lengthens the peak period, the increase in starting concentration has no discernible impact. The lowest peak viscosity produced in this study was at 40 hours fermentation and 0.2% starter at 3128 cP, while the highest was 4427 cP in corn flour fermented for 68 hours with a starter concentration of 0.2%.

This in line with Oloyede *et al.* (2019), which found that the longer the fermentation time, the shorter the time required to reach peak viscosity in fermented corn flour and fermented moringa flour. This is because the pre-digested starch thickens more quickly. The starch granules are broken down by the acids and enzymes created during fermentation, which shortens the time needed for swelling and thickening (Oloyede *et al*., 2019).

Pasting temperature

The temperature at which viscosity starts to rise or starch granules start to swell prior to gelatinization is known as the pasting temperature. In food processing, a lower initial gelatinization temperature is more energy efficient, but a higher initial gelatinization temperature indicates that the starch is more stable against heat (Riaz and Pasha, 2021).

The factor that significantly affected the gelatinization temperature in relation to changes in the initial gelatinization temperature was only the fermentation time, while the starter concentration did not. The pasting temperature ranged from 74.65 to 75.90°C (Fig 3). The starch will expand and gelatinize at a lower temperature when corn flour is fermented for a longer period of time than when it is fermented for a shorter period of time. An increase in starter concentration increases the gelatinization temperature of corn flour, although not significantly or relatively the same. An rise in the pasting temperature following modification suggests that the crystallinity zone grew throughout the modification process (Sarkar, 2016).

The initial gelatinization temperature, at which viscosity begins to increase, decreases with longer fermentation times. This is due to partial starch hydrolysis, which lowers the gelatinization temperature. Additionally, the acid created during fermentation decreases the flour's pH, which lowers the starch's initial gelatinization temperature. The pH of the surrounding environment affects the temperature at which starch gelatinizes. Lower pH levels brought on by acid production typically result in a drop in the starch's gelatinization temperature. This is because an acidic environment can disrupt hydrogen bonds within starch granules, making them more susceptible to gelatinization at lower temperatures (Siroha *et al.*, 2019).

Moisture content

According to Kathuria *et al*. (2021), The moisture content of flour is crucial because microorganisms that are naturally present in flour start to grow at higher moisture levels, resulting in an unpleasant odor and the higher the moisture content, the lower the dry matter content. Starch's gelatinization and retrogradation characteristics, as well as its capacity to absorb water and form specific structures, can all be impacted by moisture concentration (Donmez *et al*., 2021). The fermentation time factor had a significant impact on variations in the moisture content of fermented corn flour, whereas the starter concentration had no significant influence, despite the fact that its rise slightly decreased the moisture content. The lowest moisture content was found in corn flour fermented for 12 hours with a starter concentration of 0.2% at 7.44%; while the highest moisture

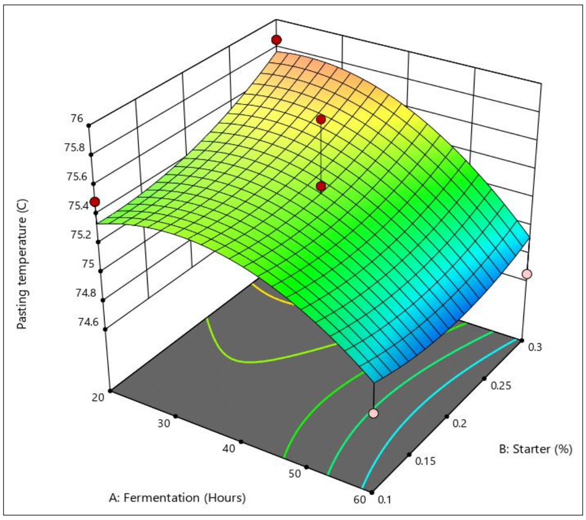

Fig 3: Pasting temperature of fermented corn flour.

content was 14.36% in the 68-hour fermentation with a starter concentration of 0.2% (Fig 4).

These findings are in line with a number of recent investigations, such as the 60-hour fermentation of corn flour with Lactobacillus plantarum Y1, which greatly enhanced water binding capacity (Qi *et al*., 2020). Furthermore, the relative moisture content of corn flour was raised by fermentation using *Rhizopus oryzae* and *Lactobacillus plantarum* (Hu *et al*., 2020).

Soluble protein

Soluble proteins are proteins that can dissolve in aqueous solutions, either in monodisperse form or as soluble aggregates, which are intermediate forms between protein monomers and insoluble gel networks. These proteins are very important for various biological functions and industrial applications due to their stability, activity and functionality (Huang *et al*., 2024; Yousefi *et al*., 2022).

The graph in Fig 5 is statistically valid for explaining changes in soluble protein content, with a model p-value of 0.0306 ($p<0.05$). The highest soluble protein content was found in the 40-hour fermentation with a starter concentration of 0.2% at 0.57%. The lowest soluble protein content was found in the 12-hour fermentation with a starter concentration of 0.2% A1 B3 at 0.4%. There was an increase in soluble protein content as fermentation time increased up to a certain point, followed by a decrease in soluble protein content. Similarly, the starter concentration factor had an increasing effect up to a certain point, followed by a decrease. However, based on the results of the ANOVA, neither factor had a significant effect on changes in soluble protein content.

Beta carotene

As a precursor to vitamin A, beta carotene offers a number of health advantages, such as antioxidant qualities and illness prevention (Akram *et al*., 2021; Ebadi *et al*., 2023). Beta-carotene is used in processed food products as a colorant and nutritional supplement, but its low water solubility and instability to light, heat and air pose challenges in its processing into food products (Zhao *et al*., 2024).

Based on ANOVA, fermentation time significantly affects changes in beta-carotene levels in corn flour after fermentation is, while the highest beta-carotene concentration is found at a fermentation time of 60 hours with a starter concentration of 0.1%. The lowest beta-carotene level was found in fermentation for 12 hours with a starter concentration of 0.2% at 90.422%; while the highest level was found in fermentation for 60 hours with a starter concentration of 0.1% at 193.645% (Fig 6).

The increase in beta-carotene in fermented corn flour is thought to be due to the increase in microbial populations such as bacteria and fungi during fermentation, which contribute to biochemical changes in the substrate. These microbes can produce enzymes that break down complex components into simpler compounds, including beta-carotene precursors (Chowdhury *et al.*, 2019).

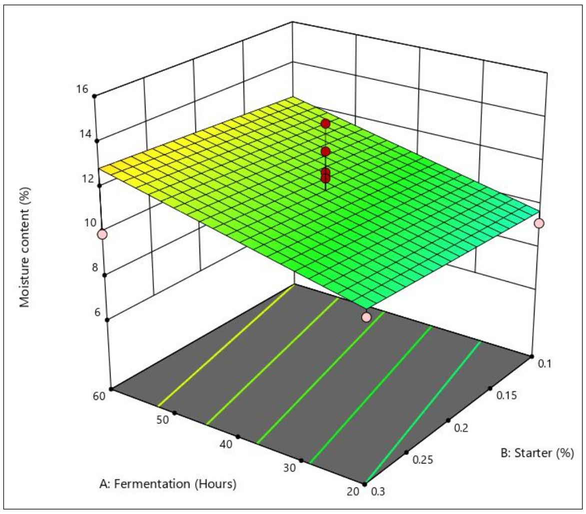

Fig 4: Moisture content of fermented corn flour.

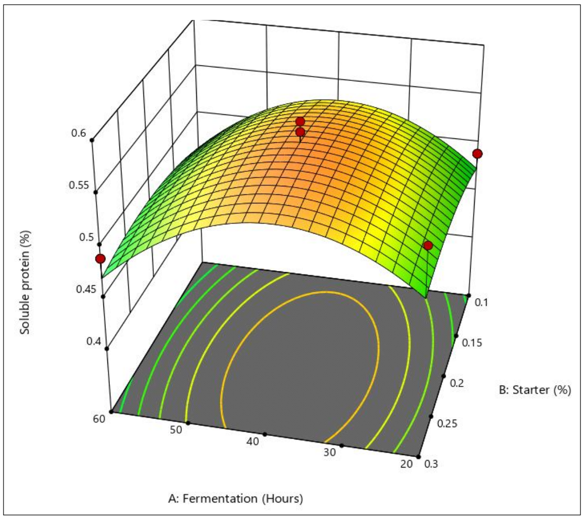

Fig 5: Soluble protein content of fermented corn flour.

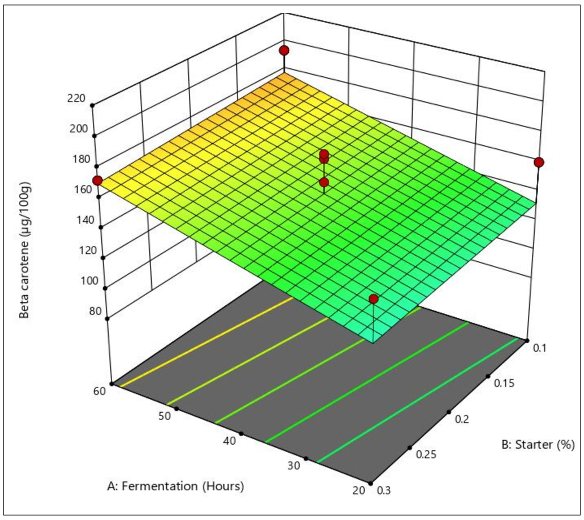

Fig 6: Beta carotene content of fermented corn flour.

Starch content

The amylose and amylopectin fractions which make the food's starch content have a direct impact on the texture, appearance and functional qualities of food products as well as their nutritional value and processing quality (Wei *et al*., 2025). With a p-value <0.001 ($p<0.05$), the graph may accurately depict variations in the starch content of fermented corn flour.

The analysis of variance (ANOVA) results show that changes in the starch content of fermented corn flour are significantly influenced by both fermentation time and starter concentration. The starch content significantly decreases with increasing fermentation duration and starter concentration in corn flour fermentation. Fermented corn flour with a starting concentration of 0.2% and a fermentation time of 12 hours had the maximum starch content (73.571%), while fermentation time of 68 hours with a starter concentration of 0.2% had the lowest value (64.491%).

The decrease in starch content during fermentation in this study is consistent with the results of Yang and Xiao (2021), who found that fermenting corn flour with Cordyceps militaris for 3 days reduced total starch by 11.7%. Additionally, in other cereals, such as sorghum, fermentation for 12 weeks also reduced total starch content by 69.0% (Li *et al.*, 2024).

Starch digestibility

Starch digestibility refers to how easily starch is broken down and absorbed in the digestive system (Thuy *et al.*, 2022). Fig 7 shows a 3D model graph with a p-value of 0.0001 ($p<0.05$), which can be considered suitable for describing the effect of fermentation time and starter concentration on the digestibility of fermented corn flour.

Based on the results of analysis of variance (ANOVA), it can be seen that both fermentation time and starter concentration have a significant effect on changes in corn flour digestibility and fermentation. The graph presented in Fig 7 shows that the longer the fermentation time, the more significantly the starch digestibility increases. Similarly, an increase in starter concentration also significantly increases starch digestibility. The lowest starch digestibility was found in corn flour fermented for 12 hours with a starter concentration of 0.2% at 64.851%, while the highest was found in corn flour fermented for 68 hours with a starter concentration of 0.2% at 73.252%.

These outcomes are in line with those of *Lactobacillus plantarum* Y1 fermentation of corn flour, which likewise shown an improvement in starch digestibility (Qi *et al*., 2020). Fermentation causes erosion or damage to the surface of starch granules, creating a porous structure that makes it easier for enzymes to access and digest starch (You *et al.*, 2025). In addition, the microbes involved in fermentation produce enzymes that can break down the starch structure, making it easier for human digestive enzymes to digest. The fermentation process can also reduce the content of simple sugars such as glucose and fructose, which can be used as substrates by microbes during fermentation, making them easier to digest (Hu *et al.*, 2021).

Water absorption index (WAI)

Water absorption index is an important parameter in evaluating the quality and function of flour, especially in the processing of various food products. The graph in Fig 8 shows that the water absorption capacity of fermented corn flour decreases with increasing fermentation time and starter concentration. The analysis of variance (ANOVA) results, however, indicate that both variables significantly impact variations in the fermented corn flour's water absorption index. Fermentation with a starting concentration of 0.34% for 40 hours had the lowest water absorption index (A3B5), whereas fermentation with a starter concentration of 0.3% for 20 hours had the highest (A2B4).

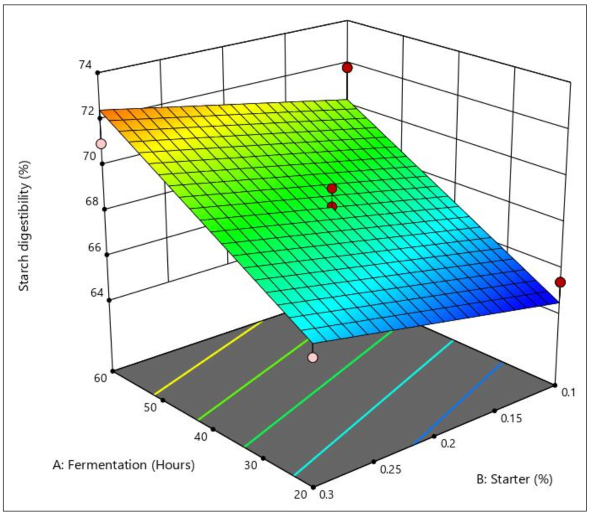

Fig 7: Starch digestibility of fermented corn flour.

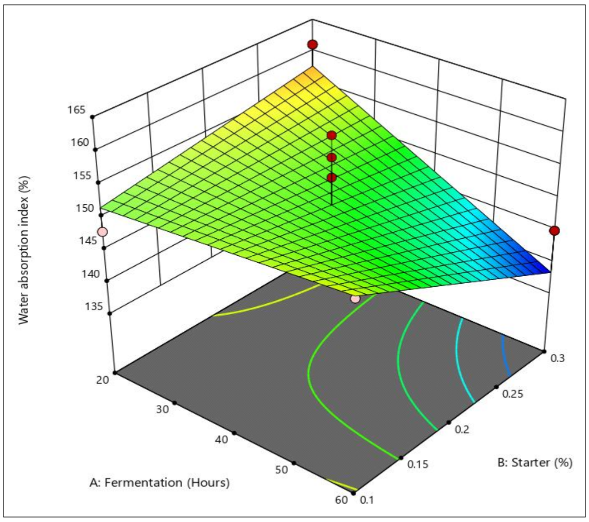

Fig 8: Water absorpsion index (WAI) of fermented corn flour.

These findings are consistent with a number of other investigations that discovered that water absorption decreased with increasing fermentation duration (Yang and Xiao, 2021). During fermentation, a variety of microorganisms produce lactic acid, acetic acid and other metabolites that change the flour's chemical makeup and physical characteristics (Wei *et al*., 2025). The synthesis of organic acids during fermentation causes the pH to drop, which subsequently impacts the stability and quality of fermented flour (Li *et al*., 2024). Furthermore, fermentation alters the structure of starch granules, which lowers the amount of amylose and raises the amount of amylopectin. These changes have an impact on the texture and digestibility of flour (Wei *et al*., 2022).

CONCLUSION

Corn flour fermentation is mainly influenced by fermentation time, which significantly increases peak viscosity, breakdown, peak time, moisture content, β-carotene and starch digestibility, while decreasing gelatinization temperature, starch content and water absorption capacity. Starter concentration generally has no significant effect. These changes occur due to partial starch hydrolysis and granule structure modification during fermentation, which improves the thickening ability and digestibility of the flour. The setback value and soluble protein content remained relatively stable across all treatments, indicating that the fermentation time range and starter concentration used were not sufficient to significantly affect retrogradation and soluble protein fraction. Overall, medium to long fermentation produces corn flour with better functional characteristics and has the potential to be applied to various processed food products.

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Disclaimers

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Informed consent

No animal procedures in this research.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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