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Effect of freezing and heating white bread on the glycemic response of healthy individuals

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Abstract

Background: Freezing starchy food causes the starch structure to break down, resulting in resistant starch that behaves like fiber. This leads to a slower rise in blood sugar, which may reduce the risk of type 2 diabetes and weight gain.

Objectives: This study aimed to investigate the impact of reheated white bread and frozen versions on the glycemic responses of healthy individuals.

Methods: A randomized controlled trial was conducted on 32 healthy participants (14 males and 18 females) aged 18–50 years with a body mass index (BMI) of 18.5-29 kg/m^2 and fasting blood sugar levels of 75–100 mg/dl. The participants were given 100 grams of fresh white bread made from Turkish flour (Al-Haramain type), and their blood sugar levels were measured at 30, 60, 90, and 120 minutes. This process was repeated after feeding the participants with 100 grams of frozen bread for 3, 5, and 7 days to observe the effect of freezing and reheating white bread on blood sugar levels, as well as the impact of freezing duration.

Results: The glycemic response of the participants to frozen and reheated white bread was lower compared to fresh white bread at all time intervals. The maximum blood sugar level was reached after 30 minutes of consuming fresh bread, at 120 mg/dl. However, for reheated bread, it reached 132 mg/dl. The prolonged freezing time did not increase the amount of resistant starch, and similar results were observed for 3, 5, and 7 days of freezing in terms of decreasing blood sugar levels.

Conclusion: Frozen and reheated white bread leads to lower blood sugar levels compared to fresh white bread. This can be attributed to the formation of resistant starch during the freezing process.

Keywords: Resistant starch, Glycemic control, Frozen foods, Bread.

Introduction

Starch is composed of complex molecules and is found in a variety of shapes, structures, and qualities that impact digestion. Smaller grain sizes are more easily broken down by digestive enzymes due to increased surface area.[1] The characteristics of industrial foods depend on the presence of amylose and amylopectin, which make up the starch. Higher amylose-content starches typically offer more resistance to digestion.[2] Resistant starch (RS) is a type of starch which enters the small intestine undigested for at least 120 minutes.[3] Resistant starch can be found naturally in food or added to specific dishes in the form of isolated or synthetic natural RS.[4] It is categorized as both dietary fiber and functional fiber, depending on whether it is naturally in the foods or added to them.[5,6] Resistant starch is categorized into four types. RS1 is undigestible and naturally found in whole grains, legumes, and unprocessed seeds. RS2, found in fresh green banana starch and high-amylose maize starch, can resist enzymatic digestion due to its natural form, which counteracts the enzymes' effects and prevents it from passing undigested into the large intestine. RS3, or resistant starch 3, is created by boiling and chilling starchy foods like pasta. Reflux causes the dissolved starch to become less soluble after being heated, dissolved in water, and then cooled. This changes the structure of the amylose and amylopectin, making it more complex and harder for enzymes to digest. RS4 includes chemically modified starches that are manipulated to suit various industrial uses by altering the bonds between their molecules.[7,8] Food processing methods can have an impact on the composition of resistant starch. Generally, methods that remove obstacles to digestion can lead to a drop in resistant starch levels.[9] For instance, whole wheat has been found to contain up to 14% resistant starch, compared to only 2% in milled wheat flour.[10] Similarly, cooking or grinding rice can also result in a reduction of resistant starch content.[11] In contrast, certain processing techniques can increase the RS content of food. Excessive water during cooking of food can cause the starch to gel, making it more digestible. However, when these starch gels are cooled, grains and starch crystals form which are resistant to digestive enzymes, resulting in higher RS3 content.1 According to research, refrigerating boiled potatoes before consumption can also increase the amount of RS.[12] Additionally, Burton and Lightowler stated that freezing bread alters the composition and structure of starch molecules, leading to an increased resistance to digestion.[13] This results in a reduction of the glycemic response, as the breakdown of starch by digestive enzymes in the mouth and intestines becomes more challenging. Foods high in RS are known to escape small intestine digestion and instead move into the large intestine, where they are fermented by bacterial colonies. This process causes them to come into contact with more carbohydrates than any other meal component, including simple sugars crucial for colon health, non-starchy polysaccharide fiber, oligosaccharides, and RS. Fermentation of resistant starch not only increases the number of bacterial cells in the intestine and generates short-chain fatty acids, such as acetate, propionate, and butyrate, but also produces gases like carbon dioxide, methane, and hydrogen.[3,5,14,15] According to a study, the recommended daily intake of RS for adults could be as high as 45 grams, which is significantly more than the recommended intake of dietary fiber, ranging from 25-38 gm/day. Additionally, replacing flour with extracted RS in food can decrease its glycemic reaction.[4,16]

Objectives

This study aimed to investigate the impact of reheated white bread and frozen versions of bread on the glycemic responses of healthy individuals.

Methods

Study design

This study was designed as a randomized controlled trial to investigate the impact of freezing and reheating white bread on the glycemic response of healthy individuals.

Sample size

In this study, the sample size was estimated by considering the primary outcome measure, which is the effect of freezing and reheating white bread on the glycemic response of healthy individuals. To detect a significant difference in blood glucose levels between fresh bread and frozen or reheated bread, a power analysis was conducted.

Assuming a moderate effect size (Cohen's d=0.5), an alpha level of 0.05, and a power of 0.80 based on previous research in the field, the power analysis indicated that a minimum sample size of 28 participants would be required.

To account for possible dropouts or exclusions, a total of 32 healthy individuals were recruited for the study, comprising 14 males and 18 females. This distribution was based on the demographic characteristics of the target population and aimed to ensure a balanced representation of both genders.

Inclusion criteria

Participants included in the study were healthy individuals, both males and females, aged between 18 and 50 years, with a body mass index (BMI) ranging from 18.5 to 29 kg/m2. They also had to have a fasting blood sugar level between 75 and 100 mg/dL, be non-alcoholic and non-smokers, and have the habit of regularly consuming breakfast.

Exclusion criteria

The following individuals were excluded from the study: breastfeeding or pregnant women, individuals with Type 2 diabetes, a history of hypertension, taking medications that interfere with glucose metabolism, or a history of malabsorption. Additionally, those with a wheat allergy were excluded from the study.

Steps of the research

The study was conducted using the following steps: Initially, eligible participants were randomly chosen based on the criteria previously mentioned. The participants' weight and height were measured to determine their BMI. Fasting blood sugar levels were checked using a blood glucose meter, specifically the "On-Call Plus" model produced by ACON Laboratories, Inc. This portable and user-friendly device is commonly used in clinical and research settings to quickly and accurately measure fasting blood glucose levels. After cleaning participants' fingertips, a minimal amount of blood was collected via a small prick, and the meter used cutting-edge electrochemical technology to rapidly and accurately measure blood glucose levels. Results were reported in milligrams per deciliter (mg/dL).

Next, participants were given 100 grams of freshly baked white bread, purchased from a bakery in Mosul and made from Turkish flour (Al-Haramain Type), and their blood glucose levels were monitored at 30, 60, 90, and 120 minutes. Participants were then given 100 grams of frozen bread that was stored for seven days, reheated, and their blood glucose levels were measured at the same intervals to determine the effects of freezing on blood glucose levels in healthy individuals. To investigate the effect of the number of days for which the bread was frozen on blood glucose levels, the bread was frozen for 3, 5, and 7 days, reheated, and fed to the same participants. Blood glucose levels were measured for the same time intervals of 30, 60, 90, and 120 minutes to observe variations caused by the number of days of freezing.

Ethical considerations

The study was conducted in accordance with the Declaration of Helsinki. The study was conducted after obtaining ethical approval from the Scientific Research Ethics Committee of the Medical Group at the University of Mosul (No. 20, at 14/6/2023), then the participants were contacted individually by the researchers to obtain their written consent.

Statistical analysis

The continuous variables were expressed as the mean ± SD, and the categorical variables were presented as a percentage and frequency. All statistical analyses were performed with SPSS (version 16.0, SPSS Inc, Chicago, IL, USA). A “P-value” less than 0.05 was considered significant.

Results

Table 1 displays the sociodemographic data of the 32 healthy participants enrolled in the study. The majority of participants were married (62.5%), and a significant proportion were employed (40.62%), had tertiary education (46.88%), and lived in metropolitan areas (53.13%). The mean values and standard deviations for various numerical variables are also reported. The average age of participants was 34.5 years, their mean BMI was 23.6, and their mean fasting blood glucose level was 85 mg/dl with a standard deviation of 6.4. These statistics provide valuable information regarding the demographic and health characteristics of the participants.

Table 1. Sociodemographic characteristics of the study participants

Characteristics		**N (%)**
Gender	Males	14 (43.75)
	Females	18 (56.25)
Education	Primary	5 (15.62)
	Secondary	12 (37.5)
	Tertiary	15 (46.88)
Occupation	Student	9 (28.13)
	Employee	13 (40.62)
	Private Work	10 (31.25)
Marital Status	Single	12 (37.5)
	Married	20 (62.5)
Residence	Rural	15 (46.87)
	Urban	17 (53.13)
Characteristics	**Min-Max**	**Mean ± SD**
Age	18-50 Years	34.5 ± 15.3
BMI (Kg/m^2)	18.5-29	23.6 ±2.2
FBG (mg/dl)	75-100	85± 6.4

BMI: Body Mass Index, FBS: Fasting blood glucose

As illustrated in Figure 1, the blood glucose level in both frozen and reheated bread was lower than that of fresh bread at all observed time points, with the peak reaching

120 mg/dl and 132 mg/dl in fresh and frozen/reheated bread, respectively. This suggests that freezing and reheating white bread has a significant impact on lowering blood glucose levels.

Figure 2 indicates that freezing white bread for 3, 5, or 7 days had a similar effect on lowering blood sugar levels, but prolonging the freezing time did not increase the amount of resistant starch in the bread.

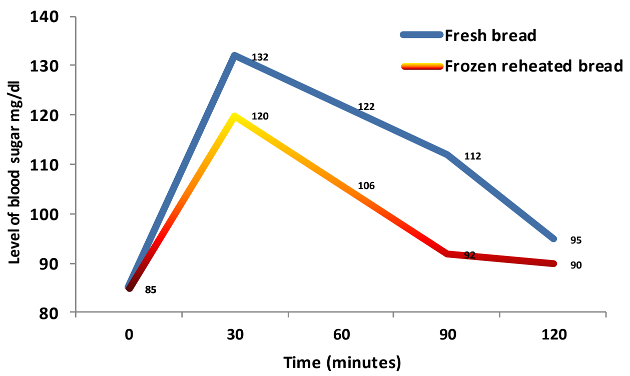

Figure 1. Difference in Blood Glucose Levels between Fresh and Frozen/Reheated Bread Over Time

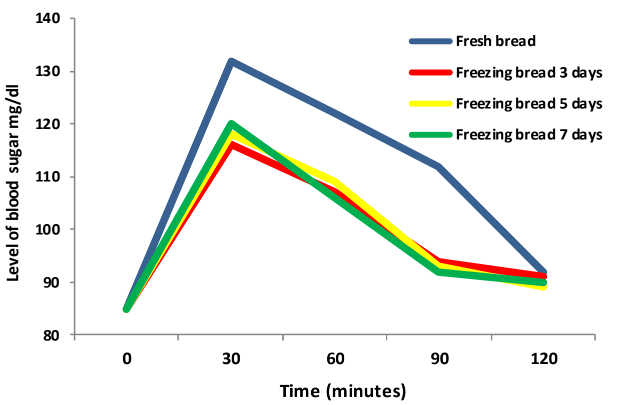

Figure 2. The effect of the duration of freezing white bread on the blood glucose level of the study sample compared with fresh bread

Discussion

In this study, out of 32 healthy individuals, 18 were female and 14 were male, ranging in age from 18 to 50 years, with a mean age of 34.5±15.3 years. The BMI of the participants ranged from 18.5 to 29 kg/m^2, with a mean of 23.6±2.2. In terms of fasting blood glucose levels, the participants ranged from 75 to 100 mg/dl, with a mean of 85±6.4. These criteria were used as key indicators in the analysis of the study participants.

Figure 1 displays the findings of the study, indicating a lower level of glucose in frozen and reheated bread compared to fresh bread across all timeframes. Notably, the glucose level peaked after 30 minutes of consuming the bread, reaching 120 mg/dl and 132 mg/dl in fresh and reheated bread, respectively. These differences in glucose levels can be attributed to the structural changes of starch molecules during the freezing process, rendering them more resistant to digestion. Similar observations were made by Burton and Lightowler in their previous study, demonstrating that frozen bread's starch structure disintegrates, thus making it challenging for digestive enzymes in our mouth and intestines to digest it.[13] Furthermore, any attempt to return these starch granules to their original state after thawing does not restore their former composition. Interestingly, freshly toasted bread resulted in 25% lower glycemic responses as compared to freshly baked bread. However, toasting frozen bread increased the distortion of starch molecules, reducing the glycemic response by approximately 40%. These results indicate that frozen and reheated bread has a slower digestion rate than fresh bread, leading to a lower rise in blood sugar levels and reducing the risk of obesity and type 2 diabetes.

Long-term ingestion of resistant starch has been demonstrated to increase insulin secretion and metabolism by stimulating colonic enteroendocrine cells and minimizing abrupt fluctuations in adipose tissue lipolysis, according to research conducted by Johnston et al.[17] and Bodinham et al.[18] These findings are consistent with other studies[8,19,20] that have shown that serving white rice to healthy individuals at a temperature of 4 C and reheating it later can result in lower blood glucose levels than freshly cooked rice. These studies suggest that resistant starch is formed during the cooling process.

The research findings indicate that cooling, reheating, and freezing food items like rice and bread may encourage the development of resistant starch, which can decrease glycemic reactions and potentially minimize the risk of type 2 diabetes and obesity. These outcomes highlight food processing and preparation techniques' significance in considering the nutritional qualities and health impacts of high-carbohydrate foods.

Figure 2 indicates that freezing white bread for varying durations of 3, 5, and 7 days all resulted in a similar decrease in blood sugar levels. Prolonged freezing periods did not increase the amount of resistant starch in the bread, suggesting that an extended freezing period does not lead to an increased development of resistant starch.

These findings could have important implications for dietary recommendations for individuals at risk of type 2 diabetes and obesity. Incorporating frozen and reheated rice and bread into meal planning may help to regulate blood sugar levels, and food producers could work to improve resistant starch development in their products to meet dietary requirements. However, it is important to note that our study has limitations.

For healthy individuals, it may be beneficial to include frozen and reheated white bread, rice, potatoes, and pasta in daily meals to take advantage of their resistant starch content as a preventive measure against high blood sugar and weight gain. Additionally, for individuals with type 2 diabetes, consuming reheated frozen bread or taking resistant starch in the form of daily nutritional supplements could lead to an improvement in glucose response and help to avoid high blood sugar levels after eating processed white bread.

Conclusions

When consumed by healthy people, freezing and reheating white bread can improve its glycemic response compared to freshly baked bread due to the creation of resistant starch, which improves insulin sensitivity and lowers blood sugar levels. Moreover, our findings indicate that prolonged freezing of the bread does not lead to an increased production of resistant starch.

Acknowledgment

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Competing interests

The authors declare that they have no competing interests.

Abbreviations

Resistant starch: RS;

Body Mass Index: BMI;

Fasting blood glucose: FBS.

Authors' contributions

Study concept; first draft writing; data collection; data analysis; and final draft review by all authors. All authors read and approved the final manuscript. All authors take responsibility for the integrity of the data and the accuracy of the data analysis.

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Role of the funding source

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Availability of data and materials

The data used in this study are available from the corresponding author on request.

Ethics approval and consent to participate

The study was conducted in accordance with the Declaration of Helsinki. The study was conducted after obtaining ethical approval from the Scientific Research Ethics Committee of the Medical Group at the University of Mosul (No. 20, at 14/6/2023), then the participants were contacted individually by the researchers to obtain their written consent.

Consent for publication

By submitting this document, the authors declare their consent for the final accepted version of the manuscript to be considered for publication.

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