



Bioactive and nutritional properties of kilned whole-grain snacks: focus on buckwheat, oat, and rye

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Abstract

This study evaluated the effects of controlled heat treatment on the glycemic index (eGI), bile acid-binding capacity (BABC), total phenolics, antioxidant activity, and in vitro bioaccessibility of whole grain buckwheat (*Fagopyrum esculentum*), rye (*Secale cereale* L.), and oats (*Avena sativa*). Cereals were subjected to thermal processing at three temperatures (120, 150, and 180 °C) for 30 min. The eGI values increased in rye and oat following heat treatment but decreased in buckwheat at 150 and 180 °C. Thermal processing significantly affected BABC in all cereals ($p \leq 0.05$), with the highest values observed at 180 °C for oats (6.56 $\mu\text{mol/g}$), buckwheat (6.65 $\mu\text{mol/g}$), and rye (5.77 $\mu\text{mol/g}$). The total phenolic content (TPC) increased with heat treatment. The highest TPC values were recorded at 180 °C for rye (9.33 mg GAE/g dw), 120 °C for buckwheat (15.39 mg GAE/g dw), and 150 °C for oats (10.04 mg GAE/g dw). Antioxidant capacities, measured using DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) assays, were enhanced in buckwheat and oats after thermal processing, while a significant decrease was observed in rye. The in vitro bioaccessibility of TPC significantly improved during the gastric phase for all samples except oat, with buckwheat showing the highest increase (86.56% at 180 °C). Oats maintained the highest bioaccessibility index (BI) for total antioxidant capacity (TAC) across all treatments, while rye peaked at 444.09% TAC during the intestinal phase at 120 °C. These findings underscore the influence of thermal processing on the functional and nutritional properties of these cereals, with notable variations based on their unique compositions. Overall, these findings demonstrate that controlled heat treatment can enhance the nutraceutical value and bioaccessibility of phenolics in whole grains, providing a promising strategy for developing minimally processed, functional cereal-based products with cardiometabolic and digestive health benefits.

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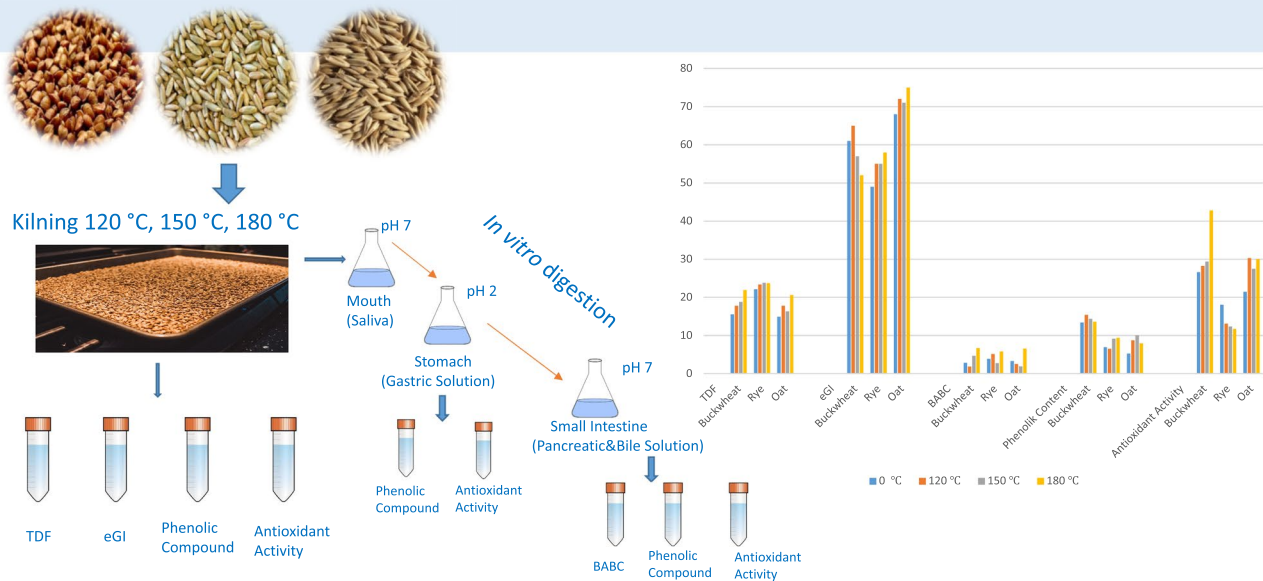
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Graphical Abstract

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Highlights

- Heat treatment enhances dietary fiber content and bile acid binding capacity.
- Phenolic compounds and antioxidant activity increase with kilning.
- Buckwheat showed the highest improvement in TPC bioaccessibility.
- Thermal processing significantly enhanced rye's TAC bioaccessibility.

Keywords Functional snack · Whole grain · Bile-acid binding capacity · Phenolic compounds

Introduction

Unhealthy dietary patterns have contributed to the global rise in obesity, diabetes, and cardiovascular diseases and colon cancer [1]. Increasing the intake of whole grains and dietary fiber is widely recognized as an effective strategy to prevent these conditions [2]. Numerous international health authorities emphasize the consumption of nutrient-dense foods, particularly whole grains, as a key source of dietary fiber, phenolics, essential nutrients, vitamins, minerals, and bioactive phytochemicals [3]. Whole grains and their bran fractions are rich in dietary fiber and bioactive compounds concentrated in the outer layers, making them attractive ingredients for functional food formulations [4]. However, whole-grain consumption remains limited due to sensory and technological challenges, including coarse texture, bitter taste and reduced consumer acceptance [5]. Moreover, in many cases, these grains are added in minimal levels to

facilitate technological processing and often undergo extensive refinement, resulting in the loss of valuable nutrients. Therefore, developing palatable whole grain-based snacks represents an effective approach to promote whole-grain intake while preserving nutritional quality.

Buckwheat, oats, and rye are particularly relevant in this context due to their distinct nutritional and functional profiles and health promoting components. Buckwheat is a highly valuable food source because of its high biological value, well-balanced amino acid composition, high fiber content, and substantial levels of essential minerals such as zinc (Zn), copper (Cu), manganese (Mn), and selenium (Se). From a nutritional perspective, buckwheat is also a rich source of antioxidants including flavonoids such as rutin and tocopherols, quercetin, and phenolic acids [6]. The absence of phytic acid, an anti-nutritional factor commonly found in wheat, further enhances the nutritional value of buckwheat fiber [7].

Rye contains more dietary fiber (22%), particularly arabinoxylans, than other cereals [4]. Rye dietary fiber and proteins (albumins and globulins) are essential for human nutrition. Additionally, rye fructans and fructooligosaccharides function as prebiotics in the large intestine, where they undergo complete fermentation into short-chain fatty acids [8]. Rye also contains five primary phenolic acids: ferulic, sinapic, caffeic, *p*-coumaric, and vanillic acids [9].

The superior nutritional profile of oats compared to other cereals has long been recognized. Oats contain high amounts of soluble fibers, proteins, unsaturated fatty acids, vitamins, minerals, and antioxidants. Oats are particularly rich in dietary fiber (14%–17%). The predominant fiber fraction consists of β -glucans, with concentrations varying from 4 to 7% in dehulled oats and 6% to 9% in oat bran [4].

These cereals were selected due to their distinct nutritional profiles and relevance in functional food development. These grains are particularly rich in bioactive compounds such as dietary fiber, polyphenols, and resistant starch, which have been associated with various health benefits [10]. Moreover, they exhibit differing responses to thermal processing, making them suitable candidates for evaluating heat-induced changes in functional properties. Their potential to bind bile acids—a mechanism linked to cholesterol-lowering effects—further supports their relevance in functional food research [11]. Additionally, the gluten-free or low-gluten nature of these grains enhances their applicability in the development of health-oriented products, particularly for individuals with gluten-related disorders [12]. Therefore, these cereals were selected to explore the impact of thermal treatment on bile acid-binding capacity and to compare their functional potential under different thermal conditions. Their differing compositions also allow for comparative evaluation of thermal effects across grain types. The findings aim to support the development of minimally processed, functional whole-grain snacks that promote digestive and cardiovascular health.

Mild thermal processing techniques such as kilning can improve sensory properties while preserving nutritional value [13]. While previous studies have examined individual grains or roasting effects, few have systematically compared the effects of kilning across multiple cereals using harmonized conditions. Additionally, limited research has explored how kilning influences glycemic index, bile acid-binding capacity, total phenolic content, and in vitro bioaccessibility. Therefore, this study addresses that gap by evaluating heat treatment effects on key nutritional and bioactive parameters. This study aims to enhance the nutritional utilization of these cereals in their most natural state by applying a single, dry-heat treatment. This approach supports the development of palatable whole-grain snacks and encourages their integration into health-promoting food formulations.

Materials and methods

Materials

Commercially dehulled common buckwheat (*Fagopyrum esculentum*) groats, whole oats (*Avena sativa*), and whole rye (*Secale cereale* L.) were procured from a local market. All samples were harvested during the 2022 crop year and cleaned manually to remove impurities prior to analysis. Enzymes used in the in vitro glycemic index analyses, including pepsin (P7000) and pancreatin (P7545), as well as bile salts, protease (P3910), α -amylase (A3306, from *Bacillus licheniformis*, 20000–60000 U/mL), Folin–Ciocalteu reagent, DPPH (2,2-diphenyl-1-picrylhydrazyl), and ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) and Total Dietary Fiber Assay Kit (TDF100A-1KT) were purchased from Sigma–Aldrich (St. Louis, MO, USA). Amyloglucosidase (3300 U/mL), the Glucose Oxidase–Peroxidase (GOPOD) Kit and Total Starch Kit (AA/AMG) were purchased from Megazyme International Ireland Ltd. (Bray, Co. Wicklow, Ireland). All other solvents and chemicals used in the analyses were of analytical grade. All solutions were prepared using deionized water until analysis.

Methods

Sample preparation

Buckwheat, oat, and rye samples were soaked in a 15% NaCl solution, thermally processed, and analyzed to investigate their quality and nutritional properties (dietary fiber, BABC, phenolic and antioxidant contents, and eGI).

The soaking time was chosen following Liu et al.'s [14] and Mohammadi Moghaddam et al. [15] method. In preliminary trials, seeds (50 g) were washed and soaked at room temperature in 350 mL of saline solutions at 5%, 10%, and 15%. In the preliminary trials, the weight gain of the samples and the perception of saltiness were evaluated at 30 min intervals. Based on sensory and hydration responses, a 15% NaCl solution and 30-min soaking period were selected. After soaking, the samples were drained to remove excess solution and then subjected to heat treatment at 120, 150, and 180 °C for 30 min, ground, and stored at 4 °C until analysis.

Sensory evaluation was conducted to determine optimal soaking conditions, and detailed procedures are provided in Supplementary Materials S1.

Following this optimization, the cereals were kilned in an oven (model KWS1528LQ-F2 (SS); 1500 W, 38 L; Galanz, Zhongshan, China). The oven was preheated for 10 min before kilning to reach the selected internal temperature.

Subsequently, pre-soaked buckwheat, oats, and rye (100 g dry weight) were uniformly kilned at 120, 150, and 180 °C, respectively, for 30 min. The kilning duration was determined based on preliminary experiments, ensuring that the seeds achieved a crispy texture. The kilned samples were ground into powder and stored at 4 °C until analysis.

Proximate analyses

The moisture content was determined using the AACC Method 44–15 [16]. The protein content was assessed using Kjeldahl's method, with a nitrogen-to-protein conversion factor of 6.25 [16]. The total lipid content was determined gravimetrically by extracting samples with hexane in a Soxhlet system [16]. The ash content was determined gravimetrically by incinerating a sample with a known weight at 550 °C for 8 h in an ash furnace [16]. The results of the analyses were expressed based on dry matter.

Total dietary fiber analysis

Prosky et al.'s [17] methods were used to determine the total dietary fiber (TDF) content using Sigma-Aldrich kits. Duplicate samples underwent sequential enzymatic digestion, involving heat-stable α -amylase (30 min at 98–100 °C), protease (30 min at 60 °C), and amyloglucosidase (30 min at 60 °C, pH=4.5).

1 g of the sample was weighed and transferred into a vessel. Subsequently, 40 mL of MES-TRIS buffer solution was added, followed by 50 μ L of α -amylase enzyme. The mixture was incubated in a shaking water bath at 95 °C for 30 min with gentle agitation to facilitate starch hydrolysis. After cooling, 100 μ L of protease enzyme was added, and the sample was further incubated at 60 °C for 30 min under mild shaking to promote protein degradation. Following this step, 5 mL of 0.561 N HCl was added to adjust the pH to the range of 4.1–4.8. Then, 200 μ L of amyloglucosidase enzyme was introduced, and the sample was incubated once again at 60 °C for 30 min with gentle agitation.

After enzymatic digestion, the mixture was filtered to separate the undigested residue. The residue was then dried at 105 °C for 16 h in an oven. Finally, the dried residue was incinerated in a ash furnace at 525 °C for 5 h for ash content determination.

Estimation of in vitro glycemic index value

The estimated glycemic index (eGI) values of the samples were determined under in vitro conditions according to the methods described by Englyst et al. [18]. As a pre-treatment, the ground samples were passed through a 212 μ m sieve.

A 100 mg portion of the kilned sample was weighed into centrifuge tubes, and ten glass beads (5 mm diameter) were added to each tube. Subsequently, 2 mL of hydrochloric acid solution (0.05 M) containing pepsin (5 mg/mL) was introduced. The tubes were incubated horizontally in a shaking water bath at 37 °C for 30 min to simulate gastric digestion. After incubation, 4 mL of sodium acetate buffer (0.5 M, pH 5.2) and 1 mL of an enzyme solution—comprising 14.45 U of amyloglucosidase and 0.104 g of pancreatin—were added to each tube. The intestinal digestion continued at 37 °C in the shaking water bath.

Aliquots (100 μ L) were collected at 0 and 90 min and mixed with 1 mL of absolute ethanol to terminate the reaction. Subsequently, the solutions were centrifuged at 6500 rpm for 10 min, and the glucose content was determined using glucose oxidase–peroxidase (GOPD) reagent and a spectrophotometer (Shimadzu 150 UV-1800, Tokyo, Japan) operated at a 510 nm wavelength.

The hydrolysis index (HI) was calculated based on the starch hydrolysis curves plotted over time. The HI served as an indicator of the starch digestion rate. The estimated glycemic index (eGI) provided insights into the digestibility of the samples relative to white bread (reference). The hydrolysis index (HI) was calculated as the ratio of the hydrolyzed (digested) starch content of the sample to that of white bread.

The eGI was then calculated using Eq. 1 [19].

$$eGI = 0.549HI + 39.71 \quad (1)$$

Extraction of kilned buckwheat, oat, and rye samples

The raw and kilned cereal samples were finely ground into powder. To each 1.0 g of powder, 50 mL of 50% aqueous methanol (v/v) was added. The mixtures were placed on a magnetic stirrer and stirred overnight at 25 °C. Next, the mixtures were centrifuged at 6000 rpm and 4 °C, and the supernatants were carefully collected and stored at 4 °C for further analysis.

The obtained extracts were used for the determination of total phenolic content (TPC) and antioxidant activity. TPC was measured using the Folin–Ciocalteu method and results were expressed as mg gallic acid equivalents (GAE) per gram of dry weight (dw). Antioxidant capacity was evaluated by the DPPH radical scavenging activity and the ABTS assay, and results were reported as Trolox equivalents (TE) per gram dw.

Total phenolic content (TPC) assay

TPC was determined using a modified version of the method described by Singleton and Rossi [20] 0.5 mL of kilned

cereal extracts, 2.5 mL of Folin-Ciocalteu reagent (0.2 N), and 2 mL of a 2% (w/v) Na_2CO_3 solution were combined. Next, the samples were incubated in the dark at room temperature for 30 min, and the absorbance was recorded using a spectrophotometer (Shimadzu 150 UV-1800, Tokyo, Japan) at 760 nm. The results are expressed in milligrams of gallic acid equivalent (GAE) per gram of dry, kilned sample.

Antioxidant capacity

Antioxidant capacity was evaluated using the DPPH and ABTS radical scavenging assays. The DPPH assay was performed according to the method described by Singh et al. [21]. After 60 min of incubation at 30 °C, the absorbance was measured at 515 nm using the same spectrophotometer. The ABTS radical cation scavenging activity was assessed following a modified version of the procedure developed by Re et al. [22], with absorbance measured at 734 nm. Results from both assays were reported as milligrams of Trolox equivalents (TE) per 100 g dry weight (dw).

In-vitro bioaccessibility of phenolic compounds

The digestion protocol outlined by Ozkan et al. [23] was applied. Briefly, 2.5 mL of kilned cereal extract was mixed with 17.5 mL of distilled water and 1.5 mL of a freshly prepared pepsin solution (60 mg pepsin in 1.5 mL of HCl). The pH was adjusted to 1.7–2.0 using 5 M HCl, and the samples were incubated in a shaking water bath at 37 °C for 2.5 h at 100 rpm to simulate gastric digestion. After this step, 5 mL aliquots were collected as post-gastric (PG) samples.

For the intestinal phase, 4.5 mL of a pancreatin–bile salt solution (18 mg pancreatin and 112.5 mg bile salts dissolved in 4.5 mL distilled water) was added to the remaining mixture. Dialysis bags with a molecular weight cut-off of 12,000 Da were filled with 0.5 M NaHCO_3 to neutralize the acidic environment and adjust the pH to approximately 7.0. The digestion continued by incubating the samples at 37 °C for an additional 2.5 h in a shaking water bath at 100 rpm.

Following the intestinal phase, samples were collected and classified into three fractions: post-gastric (PG), “serum-available” and “bioavailable” content (IN, inside the dialysis bags), and “colon-available” content (OUT, outside the dialysis bags). A blank control, containing all reagents but no sample, was subjected to the same digestion conditions. All collected fractions were centrifuged at 6500 rpm for 10 min and stored at –20 °C until analysis. Total phenolic contents (TPC) and antioxidant capacities were assessed for PG, “serum-available,” “bioavailable” (IN), and (OUT) contents, using the previously described methods.

In vitro digestion and bile acid-binding capacity analyses

In vitro digestion analyses were conducted following Minekus et al.’s [24] procedure, with modifications. Saliva amylase hydrolysis was not performed. Additionally, pancreatin was substituted for individual enzymes such as trypsin and chymotrypsin for intestinal digestion. Each minced sample (5 g, dry basis) was dissolved in 3.5 mL of the salivary solution (SSF, pH 7.0) to simulate oral conditions and incubated at 37 °C for 2 min with continuous shaking. Subsequently, the pH of the mixture was adjusted to 3.0 (using 1 M HCl), and 7.5 mL of gastric solution (SGF) and 1.6 mL of pepsin solution (2000 U/mL) were added to simulate gastric conditions. After 2 h of incubation at 37 °C, the pH of the mixture was readjusted to 7.0 (using 1 M NaOH). Subsequently, 11 mL of intestinal solution (SIF), 5 mL of pancreatin solution (adjusted to 100 U/mL trypsin activity), and 2.5 mL of bile acid solution (1.6 mM) were incorporated to simulate intestinal conditions. The mixture was further incubated at 37 °C for 2 h with continuous agitation. The digested samples were centrifuged at 5000 rpm for 5 min, and the supernatant was used for BABC analyses.

The quantity of the bound bile acid was estimated by subtracting the total amount of bile acids introduced during the in vitro digestion from the amount measured in the supernatant. BABC assessments were conducted with three sets of parallels and three replicates per sample. Bile acid-binding in the samples was evaluated following Sayar et al.’s [25] method. BABC was quantified as $\mu\text{mol/g}$ dry sample, using a commercial kit from Sigma-Aldrich. In this assay, 3-hydroxysteroid dehydrogenase interacts with all 12 bile acids, converting NAD into NADH. This reduction reaction transforms a probe into a highly fluorescent product. The resultant fluorescence intensity (excitation=530 nm/emission=585 nm) is proportional to the bile acid concentration in the sample. Cholestyramine was incorporated as a positive control in every test.

Statistical analysis

Statistical analysis was conducted using SPSS version 16 (SPSS Inc., Chicago, IL). A two-way ANOVA analysis was performed to assess potential differences (the effects of cereal type and temperature) between means when comparing more than two groups regarding a specific variable. Duncan’s test was used to evaluate these differences with a confidence interval of 95%. At least two repetitions were conducted, based on the sensitivity of the specific analysis.

Results and discussion

Proximate composition

Table 1 presents the proximate composition of the samples. The moisture content decreased as the heat treatment temperature increased. This reduction in moisture content is an inherent outcome of the kilning process. Thermal processing not only affects moisture content. It also enhances flavor and aroma, while contributing to desirable textural properties. Therefore, the thermal processing temperature that results in the lowest moisture content is considered optimal for these products [26].

According to the literature, buckwheat contains 12%–18.9% protein, with a lipid content ranging from 1.5% to 4%. The TDF content of whole buckwheat is approximately 7%, while its bran fraction contains up to 40% TDF [27, 28]. The TDF content in rye varies between 14% and 20.9%, and its protein content ranges from 8 to 15%, comparable to wheat [29, 30]. Whole oats have 6.7%–11.1% lipid, 18.1% protein, and 10.8% TDF [31–34].

Generally, the values obtained here are consistent with those reported in the literature. However, according to the literature, the TDF content of buckwheat and oats is comparatively higher. This difference can be explained by the fact that the TDF content of buckwheat can reach up to 15%. Hulled or unprocessed buckwheat tends to have higher fiber content, which may account for the relatively elevated TDF levels reported for buckwheat in the literature [35]. Similarly, oats can exhibit elevated TDF contents. This can be attributed to several factors, including high bran proportion, minimal processing, beta-glucan concentrations, and cultivar-specific characteristics. Collectively, these factors contribute to the higher dietary fiber values reported for oats [36]. Higher thermal processing temperature significantly increased the TDF content in buckwheat and oats. By contrast, although an increase in TDF was also seen in rye, no significant correlation was found between the degree

of thermal processing and the TDF content ($p>0.05$). The increase in TDF depends on both the processing method and the structural composition of the food matrix [37]. Heat treatment increases the proportion of soluble fiber in certain foods. For instance, studies have demonstrated that the soluble fiber content in some heat-treated vegetables and grains increases following thermal processing [38–40]. Similarly, Dong et al. [41] reported that thermal processing improved the dietary fiber content in whole oats, attributing this effect to the breakdown of glycosidic bonds in polysaccharides, which facilitates the release of oligosaccharides, increasing the proportion of soluble dietary fiber (SDF).

Diets rich in fiber and polyphenols are both recognized for their health benefits, and their interactions may modulate the bioavailability and functionality of phenolic compounds [42].

Overall, heat processing enhances the dietary fiber content of oats, buckwheat, and rye while influencing their structural properties. Another possible explanation of this increment is to decrease the moisture content of the samples due to thermal treatment, indicating an increase in dry matter.

Bile acid-binding capacity (BABC)

Table 2 presents the BABC of the samples. Cholestyramine was used as a positive control because of its extensively characterized bile acid-binding capacity. BABC results are reported in two formats: the amount of bile acid bound ($\mu\text{mol/g}$) and the percentage of bile acid bound relative to cholestyramine (%). Here, to facilitate a meaningful comparison with previous studies, the BABC was calculated relative to cholestyramine, with its binding capacity set at 100% to ensure that the results could be evaluated independently of any methodological variations [43].

Rye samples demonstrated a non-linear response to thermal processing. The sample treated at 180 °C exhibited a significantly elevated BABC ($5.77\pm0.15\ \mu\text{mol/g}$; 83.87%),

Table 1 Moisture, protein, lipid, and total dietary fiber (TDF) content of the evaluated cereals

Samples	Temperature	Moisture (%)	Protein (%)	Lipid (%)	TDF (%)
Rye	Raw	13.67 $\pm$ 0.5 ^a	12.80 $\pm$ 0.3 ^c	1.00 $\pm$ 0.0 ^d	22.16 $\pm$ 0.0 ^b
	120 °C	3.26 $\pm$ 0.0 ^c	12.60 $\pm$ 0.2 ^{c,d}	1.04 $\pm$ 0.0 ^d	23.44 $\pm$ 0.6 ^a
	150 °C	3.23 $\pm$ 0.1 ^c	12.60 $\pm$ 0.4 ^{c,d}	1.43 $\pm$ 0.1 ^c	23.80 $\pm$ 0.1 ^a
	180 °C	2.09 $\pm$ 0.0 ^f	13.80 $\pm$ 0.3 ^{a,b}	0.93 $\pm$ 0.0 ^d	23.68 $\pm$ 0.2 ^a
Buckwheat	Raw	11.89 $\pm$ 0.4 ^b	12.25 $\pm$ 0.2 ^d	2.68 $\pm$ 0.2 ^b	15.46 $\pm$ 0.3 ^{f,g}
	120 °C	2.87 $\pm$ 0.0 ^{c,d}	11.70 $\pm$ 0.2 ^e	2.71 $\pm$ 0.1 ^b	17.84 $\pm$ 0.9 ^e
	150 °C	2.70 $\pm$ 0.0 ^{d,e}	12.40 $\pm$ 0.3 ^{c,d}	2.84 $\pm$ 0.1 ^b	18.79 $\pm$ 0.5 ^d
	180 °C	2.38 $\pm$ 0.0 ^{e,f}	11.80 $\pm$ 0.2 ^e	2.74 $\pm$ 0.1 ^b	21.96 $\pm$ 1.0 ^b
Oats	Raw	13.84 $\pm$ 0.9 ^a	13.90 $\pm$ 0.3 ^a	5.56 $\pm$ 0.3 ^a	14.91 $\pm$ 0.4 ^g
	120 °C	3.25 $\pm$ 0.0 ^c	13.40 $\pm$ 0.3 ^b	5.58 $\pm$ 0.3 ^a	17.77 $\pm$ 0.9 ^e
	150 °C	3.17 $\pm$ 0.0 ^{c,d}	13.40 $\pm$ 0.3 ^b	5.54 $\pm$ 0.3 ^a	16.32 $\pm$ 0.8 ^f
	180 °C	2.92 $\pm$ 0.1 ^{c,d}	14.10 $\pm$ 0.3 ^a	5.36 $\pm$ 0.2 ^a	20.59 $\pm$ 0.0 ^c

Values are expressed as the mean $\pm$ standard deviation of triplicate analyses. Values within a column followed by the same letter are not significantly different ($p>0.05$). Protein, lipid, and total dietary fiber (TDF) contents were expressed on a dry matter basis

Table 2 Bile acid-binding capacity (BABC) of the evaluated cereals

Samples	Temperature	BABC ($\mu\text{mol/g}$)	BABC, % (relative to cholestyramine)
Cholestyramine	-	6.88 ± 0.06^a	100
Rye	Raw	$3.86 \pm 0.10^{d,e,f}$	56.10
	120 °C	$5.14 \pm 1.44^{a,b,c}$	74.71
	150 °C	$2.74 \pm 0.09^{f,g}$	39.82
	180 °C	$5.77 \pm 0.15^{a,b}$	83.87
Buckwheat	Raw	$2.84 \pm 0.18^{e,f,g}$	41.28
	120 °C	1.86 ± 0.07^g	27.09
	150 °C	$4.66 \pm 0.14^{b,c,d}$	67.72
	180 °C	6.65 ± 0.09^a	96.56
Oats	Raw	$3.29 \pm 0.11^{e,f,g}$	47.75
	120 °C	$2.52 \pm 0.07^{f,g}$	36.54
	150 °C	1.88 ± 0.03^g	27.37
	180 °C	6.56 ± 0.09^a	95.22

Values are expressed as the mean $\pm$ standard deviation of triplicate analyses. Values within a column followed by the same letter are not significantly different ($p > 0.05$)

whereas the sample processed at 150 °C showed the lowest capacity ($2.74 \pm 0.09 \mu\text{mol/g}$; 39.82%). Interestingly, moderate heating at 120 °C also enhanced the binding capacity ($5.14 \pm 1.44 \mu\text{mol/g}$; 74.71%) compared to the raw sample ($3.86 \pm 0.10 \mu\text{mol/g}$; 56.10%). Buckwheat samples displayed the most pronounced increase in BABC following high-temperature treatment. The sample processed at 180 °C reached $6.65 \pm 0.09 \mu\text{mol/g}$, corresponding to 96.56% of the cholestyramine reference. In contrast, the lowest value was observed at 120 °C ($1.86 \pm 0.07 \mu\text{mol/g}$; 27.09%), indicating a substantial thermal sensitivity. Oat samples followed a similar trend, with the highest BABC observed at 180 °C ($6.56 \pm 0.09 \mu\text{mol/g}$; 95.22%). The raw sample exhibited moderate binding ($3.29 \pm 0.11 \mu\text{mol/g}$; 47.75%), while the lowest capacity was recorded at 150 °C ($1.88 \pm 0.03 \mu\text{mol/g}$; 27.37%). High-temperature processing, especially at 180 °C, significantly enhances the bile acid-binding capacity of some cereal grains. However, the effect appears to be both grain-specific and temperature-dependent.

The raw samples exhibited similar BABC values ($p > 0.05$). Dongowski [44] reported BABC values of 4.29 for rye flour and 3.11 for oat flour, which is close to our findings. Here, the BABC values of the samples increased following heat processing, although this increase occurred at different temperatures.

Increased thermal processing of rye led to fluctuations in its BABC. The differential effects of temperature on the samples may be attributed to structural variations. Notably, all samples reached their highest BABC values at 180 °C. At 180 °C, the samples showed BABC values comparable to cholestyramine ($p \leq 0.05$). This indicates a high binding capacity.

Rye, buckwheat, and oats are important cereal crops that serve as rich sources of dietary fiber. Dietary fiber-rich foods or components bind to bile acids [43]. Dziedzic et al. [45] investigated the effects of technological processing on the dietary fiber content of buckwheat groats and related products and reported an increase in TDF from 28.09% to 34.02% after roasting. Furthermore, the buckwheat product with the highest TDF content also exhibited the highest BABC. They also found that roasted buckwheat groats bound approximately 10% more deoxycholic acid than other buckwheat samples [45]. Our results are also in line with the study of Dziedzic et al. [45] since regular increase in TDF content of buckwheat (Table 1) was caused by kilning, leading to higher BABC at 180 °C kilning temperature (Table 2).

Bile acids are synthesized in the liver from cholesterol and secreted into the small intestine, where they facilitate the digestion of dietary fats and oils. However, dietary fiber binds bile acids in the small intestine, hindering their reabsorption. The primary mechanism underlying the cholesterol-lowering effect of dietary fiber is the formation of a dietary fiber-bile acid complex, which prevents bile acid reabsorption from the small intestine [46].

These findings further support the hypothesis that thermal processing enhances BABC, likely due to an increase in the soluble fiber content of the samples. In addition to fiber solubilization, the observed improvement in BABC at higher kilning temperatures—particularly at 180 °C—may also be attributed to the formation of Maillard reaction products. These compounds can interact with bile acids through hydrophobic and electrostatic interactions, potentially increasing their binding affinity. Furthermore, thermal degradation of dietary fibers and proteins may expose or modify functional groups (e.g., hydroxyl, carboxyl, amino), enhancing their capacity to bind bile acids. Similar observations were reported for oxidized or degraded -glucans, which exhibited enhanced bile acid-binding due to the exposure of additional reactive sites [47, 48]. Therefore, both Maillard reaction products and heat-induced structural modifications are likely to act synergistically, contributing to the higher BABC observed at elevated temperatures. Hence, incorporating high-fiber foods into the diet is essential for maintaining healthy cholesterol levels [49].

Estimated glycemic index (eGI) and hydrolysis index (HI) of kilned samples

The HI represents the ratio of the starch digestion rate in a food sample to that in a reference food, typically white bread. As anticipated, white bread (control) exhibited the highest HI. Among the grains, raw rye demonstrated the lowest HI.

The relationship between thermal processing and glyce-mic response varied among the cereal samples. The HI and eGI values of the samples are presented in Table 3. Kilning significantly affected both eGI and HI. The eGI values of rye and oats increased with rising temperature ($p \leq 0.05$), likely due to heat-induced modifications in starch structure that enhance water absorption and enzymatic susceptibility. Additionally, the amylose-to-amylopectin ratio in rye and oats facilitates rapid hydrolysis under heat, contributing to higher eGI values [50, 51]. A similar trend in HI supports this mechanism, reflecting greater starch digestibility under intensified thermal treatment.

In contrast, buckwheat exhibited a decrease in eGI at higher temperatures. This result may be linked to the formation of Maillard reaction products that inhibit starch digestion and stronger protein–starch interactions that limit enzymatic access. Specifically, the eGI values for samples kilned at 120 °C were significantly higher than those kilned at 150 and 180 °C. Previous research suggests that Maillard reactions during thermal processing can reduce the amount of digestible carbohydrates, thus lowering the glyce-mic response [52, 53]. These grain-specific responses suggest that kilning alters the physicochemical properties of starch differently depending on cereal type. Bhinder et al. [54] similarly reported a marked increase in intermediate Maillard reaction products during roasting, exhibiting a strong positive correlation with roasting temperature (the highest value was observed at 170 °C). In our study, the lowest eGI values were obtained at 180 °C, suggesting that the formation of Maillard reaction products may have contributed to this reduction in eGI. This aligns with the notion that Maillard reaction products reduce the availability of digestible carbohydrates, lowering the eGI values. Further

investigation is warranted to elucidate the exact mechanisms underlying this relationship. Similarly, Małgorzata et al. [55] demonstrated that Maillard reactions in thermally treated buckwheat are associated with changes in its chemical composition.

Zamaratskaia et al. [56] reported that buckwheat-based products generally have a lower predicted GI compared to wheat-based products. This can be due to the flavonoids profile of buckwheat. Flavonoids, particularly quercetin, have been associated with a reduced risk of type 2 diabetes mellitus (T2DM). These compounds inhibit α -glucosidase and α -amylase, key enzymes involved in starch digestion, thereby reducing starch digestibility. The fiber content in buckwheat also plays a crucial role in improving metabolic conditions related to T2DM. Studies have shown that incorporating buckwheat bran at 0.5%, 1%, and 2% into the diet reduces blood glucose levels [56].

Oats showed higher eGI values than rye and buckwheat (Table 3), suggesting that its lower effectiveness in moderating postprandial glyce-mic responses. Also, kilning caused significant increase in eGI value of whole oat ($p < 0.05$), especially at 180 °C (Table 3). These findings align with previous studies which reported an increment in the eGI of oat starch because of conventional heat treatment methods [34]. This was explained by the reduction of large starch granule content and the molecular weight of amylose that cause a higher GI value [57].

Both raw rye and kilned rye kernels in this study exhibited a low eGI to some extent. Medium eGI value (56 to 69) was obtained for kilning at 180 °C (Table 3). Rye has a low eGI because of its high soluble dietary fiber content [58]. Rye is commonly processed to flour and used in bread making [59]. Previous studies have demonstrated that whole-meal rye bread and rye bread baked with whole kernels resulted in a lower insulin response than refined wheat bread. Juntunen et al. [60] conducted a study comparing the eGI of rye bread and white bread and concluded that less insulin is required to regulate postprandial glucose levels following rye consumption. This effect is primarily attributed to the fiber content and unique structural characteristics of rye. The findings also indicate that the starch granules in rye bread undergo swelling, accompanied by partial amylose leaching. Moreover, the starch granules are densely packed, forming a continuous matrix in the rye bread structure. The firmer structure of rye bread, in contrast to refined wheat bread, can be linked to these structural differences [60]. The differences in eGI responses to temperature variations among the three evaluated grain products can also be explained by their structural characteristics. Numerous studies have demonstrated that plant-derived polysaccharides play a key role in glucose and insulin metabolism regulation. These bioactive compounds contribute to blood sugar

Table 3 Hydrolysis index (HI) and estimated glyce-mic index (eGI) of the evaluated cereals

Samples	Temperature	HI	eGI
White bread	-	74.00 ± 0.0 ^a	80.33 ± 0.0 ^a
Rye	Raw	17.60 ± 3.3 ^k	49.40 ± 1.8 ^k
	120 °C	28.24 ± 7.0 ^{h,i}	55.21 ± 3.8 ^{h,i}
	150 °C	27.63 ± 1.6 ⁱ	54.88 ± 0.8 ⁱ
	180 °C	34.33 ± 0.8 ^g	58.56 ± 0.4 ^g
Buckwheat	Raw	38.75 ± 1.5 ^f	61.00 ± 0.8 ^f
	120 °C	46.15 ± 1.4 ^e	65.05 ± 0.7 ^e
	150 °C	32.20 ± 2.5 ^{g,h}	57.40 ± 1.4 ^{g,h}
	180 °C	22.80 ± 2.7 ^j	52.20 ± 1.5 ^j
Oats	Raw	52.27 ± 2.6 ^d	68.40 ± 1.5 ^d
	120 °C	58.41 ± 0.2 ^c	71.78 ± 0.1 ^c
	150 °C	57.90 ± 3.9 ^c	71.49 ± 2.1 ^c
	180 °C	63.80 ± 2.1 ^b	74.74 ± 1.1 ^b

Values are expressed as the mean ± standard deviation of triplicate analysis. Values within a column followed by the same letter are not significantly different ($p > 0.05$). HI: Hydrolysis index, eGI: Estimated glyce-mic index

regulation, improved insulin sensitivity, and overall metabolic health [34]. Thus, low-GI functional food products are of interest for addressing metabolic health concerns.

Total phenolic content (TPC) and total antioxidant capacity (TAC) of undigested and in vitro-digested samples

The total phenolic content (TPC), total antioxidant capacity (TAC), and bioaccessibility index (BI) of kilned rye, buckwheat, and oat were given in Table 4. The findings indicate that the TPC increased in all three cereals following thermal processing, although the extent of the increase varied with kilning temperature and cereal type.

In rye, a significant increase in TPC was observed after kilning, except at 120 °C. Samples kilned at 150 and 180 °C exhibited higher phenolic levels than their raw counterparts, with the highest value recorded at 180 °C (Table 4). The TPC of raw rye was 6.91 mg GAE/g dw, increasing by 24.56% and 25.94% at 150 °C and 180 °C, respectively. This rise is likely due to the release of bound phenolic compounds at elevated temperatures.

In contrast, buckwheat reached peak at 120 °C, followed by a decline at 150 °C and 180 °C. Notably, the TPC at 180 °C was comparable to that of the untreated buckwheat sample. These findings align with the TPC values reported for common buckwheat varieties, which noted TPC increases between 30.98%–49.55% after roasting of buckwheat varieties [61]. By comparison, the TPC values in unroasted buckwheat flours typically range from 8.90 to 14.72 mg GAE/g dw [54].

In oats, TPC gradually increased at 120 °C and 150 °C, reaching the highest value at 150 °C, but declined at 180 °C. Nevertheless, kilned oat samples exhibited higher TPC levels than raw oats at all tested temperatures, with increases of 66.28%, 91.24%, and 51.24% observed at 120 °C, 150 °C, and 180 °C, respectively. In the present study, TPC values in methanolic extracts of oats ranged from 5.25 to 10.04 mg GAE/g dw, although no consistent trend was observed across temperatures.

These results demonstrate that thermal processing enhances TPC in selected cereal grains, although with variations depending on the cereal type and temperature. The differences in phenolic responses to heat can be attributed to the distinct phenolic composition of each cereal. The content and activity of polyphenols are influenced by multiple factors, including plant species, cultivar, maturity state, the nature, and location of phenolic compounds, and the duration and intensity of the thermal treatment [9, 62]. Previous studies have shown that thermal processing facilitates the release of bound phenolic acids by disrupting cellular structures and breaking down cell walls [63]. For instance,

Liukkonen et al. [64] reported that the content of bioactive compounds in baked rye bread was comparable to or slightly higher than those in whole rye flour, with the exception of tocopherols and tocotrienols. This suggests that baking can preserve or even enhance bioactive compounds in cereals, thus maintaining its nutritional quality. The literature consistently indicates that free phenolics represent only a small fraction of total phenolic content in cereals, as the majority exist in insoluble, bound forms. Iftikhar et al. [65] have reported that hydrothermal treatment increases phenolic content by promoting the release of bound phenolics [65]. Similarly, in the present study, the use of whole cereal grain, kilned without milling or baking, likely minimized structural degradation and contributed to the retention of phenolic compounds.

On the other hand, thermal processing may also stimulate chemical reactions that produce new antioxidant compounds. Browning reactions, including Maillard reaction, caramelization, and chemical oxidation of phenols, are known to enhance both TPC and free radical-scavenging capacity. This phenomenon may be attributed to the dissociation of conjugated phenolic moieties, followed by polymerization and/or oxidation reactions [63]. While some studies have reported TPC losses due to thermal degradation [55], others noted that antioxidant compounds formed during the Maillard reaction may compensate for such losses and even contribute to an increase in TPC [54]. Ma et al. [61] reported that TPC in flour does not substantially decrease after roasting. Therefore, the increase in TPC observed in the present study may result from both the release of bound phenolic compounds and the formation of Maillard-derived antioxidants, reducing the effects of heat induced phenolic degradation. The positive influence of thermal processing on TPC has also been confirmed in other cereal studies. Lee et al. [66] evaluated the effects of roasting oats at different temperatures (150, 200, and 250 °C) and durations (15 and 30 min), finding that methanolic extracts produced the highest TPC, reaching 135 mg GAE/g after 30 min at 250 °C. Similarly, Sandhu et al. [67] reported that toasting oat flour (115±2 °C for 3 h) increased the TPC by 11.5% to 27.1% ($p \leq 0.05$). These combined effects suggest that kilning offers a promising strategy for enhancing the nutritional and functional value of cereal grains through both the release and transformation of phenolic compounds.

In addition, the current study showed significant ($p \leq 0.05$) variations in antioxidant capacity among different kilning treatments of selected cereals. Previous literature also reports a positive correlation between total phenolic content and antioxidant activity [68]. Given the differing mechanisms of antioxidant activity, the antioxidant capacities of buckwheat, oats, and rye were evaluated before and after in vitro simulated digestion using DPPH and ABTS assays.

Table 4 Phenolic and antioxidant bioaccessibility of the evaluated cereals

TPC (mg GAE/g)													
Sample Name	Rye	Buckwheat						Oats					
		Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C
Undigested	6.91±0.2 ^b	6.49±0.1 ⁱ	9.16±0.1 ^e	9.33±0.2 ^e	13.38±0.1 ^c	15.39±0.2 ^a	14.35±0.1 ^b	13.60±0.0±0.1 ^c	5.25±0.1 ^j	8.73±0.0±0.1 ^f	10.04±0.1 ^d	7.94±0.0±0.0 ^g	
Gastric Phase	26.90±1.0 ^b	30.10±0.3 ^g	33.41±0.7 ^e	29.11±0.1 ^g	76.98±0.6 ^d	92.91±0.9 ^e	99.32±0.7 ^b	101.22±0.8 ^a	31.91±1.0 ^f	29.40±1.6 ^g	24.00±1.6 ⁱ	25.60±0.2 ^h	
Intestinal Phase	28.80±0.2 ^{d,e}	28.80±1.2 ^{d,e}	30.60±1.8 ^d	30.61±0.7 ^d	40.42±0.6 ^a	37.42±1.4 ^b	38.42±2.3 ^b	33.31±0.3 ^e	23.30±1.0 ^f	27.40±1.2 ^e	23.10±1.0 ^f	30.61±0.7 ^d	
BI%	416.92±1.9 ^b	444.09±22.5 ^a	334.27±22.3 ^e	328.10±10.7 ^{c,d}	302.19±7.8 ^e	243.23±11.4 ^g	268.05±14.2 ^f	244.08±4.0 ^g	443.60±11.0 ^a	313.85±10.8 ^{d,e}	229.99±8.0 ^g	399.64±10.8 ^b	
DPPH (mg TE/g)													
Sample Name	Rye	Buckwheat						Oats					
Undigested	Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C	
		12.71±0.0 ^b	11.93±0.1 ^{c,d}	8.19±0.4 ^g	8.06±0.0 ^g	11.20±0.2 ^e	11.28±0.2 ^e	11.73±0.6 ^d	13.85±0.1 ^a	9.77±0.1 ^f	11.77±0.0 ^{c,d}	11.94±0.1 ^{c,d}	
	Gastric Phase	3.12±0.1 ^g	4.55±0.2 ^f	5.55±0.1 ^e	5.80±0.4 ^e	11.74±0.3 ^a	10.36±0.2 ^e	11.55±0.1 ^a	9.30±0.3 ^d	9.26±0.0 ^d	10.05±0.1 ^c	10.89±0.1 ^b	
Intestinal Phase	Raw	2.72±0.1 ⁱ	4.20±0.0 ^b	4.36±0.1 ^b	5.05±0.1 ^g	2.31±0.2 ^j	5.68±0.3 ^f	6.12±0.3 ^e	6.68±0.2 ^d	11.77±0.0 ^e	12.83±0.3 ^a	13.51±0.3 ^a	
	BI%	21.44±1.0 ⁱ	35.20±0.1 ⁱ	54.53±2.5 ^f	62.75±0.8 ^e	20.83±2.1 ^j	50.43±4.2 ^{a,b}	52.17±0.2 ^{a,g}	48.22±2.0 ^b	120.46±1.4 ^a	101.85±0.1 ^d	111.56±2.7 ^b	
	ABTS (mg TE/g)												
Sample Name	Rye	Buckwheat						Oats					
Undigested	Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C	Raw	120 °C	150 °C	180 °C	
		18.04±0.2 ^b	13.12±0.2 ^l	12.33±0.1 ^j	11.68±0.3 ^k	26.60±0.8 ^f	28.21±0.5 ^d	29.36±0.5 ^c	42.79±0.4 ^a	21.42±0.5 ^g	30.32±0.1 ^b	27.48±0.2 ^e	
	Gastric Phase	12.79±0.3 ^b	14.27±0.1 ^g	14.98±0.3 ^g	16.52±0.1 ^e	28.04±0.9 ^d	14.84±0.6 ^g	15.68±0.9 ^f	12.87±0.3 ^h	36.55±0.6 ^e	36.49±0.4 ^e	37.99±0.1 ^b	
Intestinal Phase	Raw	9.08±0.1 ^d	7.47±1.1 ^c	5.76±0.2 ^f	4.89±0.5 ^f	10.37±0.2 ^d	5.76±0.2 ^f	6.29±0.1 ^{e,f}	5.51±0.3 ^f	43.60±1.2 ^e	52.60±1.1 ^b	53.75±1.6 ^b	
	BI%	155.47±4.0 ^d	113.07±3.3 ^f	127.13±7.2 ^e	110.25±5.5 ^f	39.01±1.7 ^g	20.42±0.2 ^h	21.43±0.3 ^h	12.87±0.9	203.73±13.3 ^a	173.50±3.2 ^e	195.65±7.9 ^a	
	ABTS (mg TE/g)												

Values are expressed as the mean±standard deviation of triplicate analysis. Values within a column followed by the same letter are not significantly different (*p*>0.05). TPC: Total phenolic content, DPPH: 2,2-diphenyl-1-picrylhydrazyl, ABTS: 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)

Values are expressed as the mean±standard deviation of triplicate analysis. Values within a column followed by the same letter are not significantly different ($p>0.05$). TPC: Total phenolic content, DPPH: 2,2-diphenyl-1-picrylhydrazyl, ABTS: 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)

In rye, a 36% reduction in DPPH values was observed following kilning (Table 4). A similar decreasing trend was observed for ABTS values. This decline in antioxidant capacity aligns with findings by Cammerata et al. [69], who reported reduced antioxidant activity in boiled wheat-based pasta compared to raw samples. This reduction may be attributed to the structural degradation or polymerization of phenolics due to thermal processing, or the interference of Maillard reaction products with TPC measurements. The inverse relationship observed between TPC and antioxidant capacity in thermally processed rye can be attributed to two concurrent mechanisms. At lower temperatures and higher moisture conditions phenolic degradation predominates, resulting in the loss of phenolic acids such as ferulic and p-coumaric acids leading to decreased antioxidant capacity despite measurable increases in total phenolics [70]. Conversely, high-temperature or low-moisture treatments promote the formation of Maillard reaction products which can interact with phenolics or proteins, leading to the generation of polymerized or bound complexes [71]. This interpretation is consistent with prior findings showing that the formation of Maillard reaction products under certain thermal conditions may contribute to antioxidant capacity. Recent studies [71, 72] confirm that these Maillard-derived structures can contribute to measured TPC values via Folin–Ciocalteu reactivity while possessing lower radical-scavenging efficiency than native phenolics. Consequently, the apparent increase in TPC accompanied by a reduction in antioxidant capacity in rye strongly suggests a shift from bioactive free phenolics to less active Maillard-derived derivatives under high-temperature conditions, as supported by previous findings in thermally processed cereals.

Conversely, in buckwheat and oats, antioxidant capacity increased after kilning. Untreated buckwheat and oat samples exhibited lower antioxidant capacities than thermally processed samples, indicating an enhancement in antioxidant potential upon heating. Bhinder et al. [54] reported a decrease in antioxidant activity in roasted buckwheat flour (6.84–11.53 $\mu\text{mol TE/g}$) compared to unroasted samples (11.37–12.74 $\mu\text{mol TE/g}$), while Ma et al. [61] observed an increase in TPC across all buckwheat varieties after thermal processing. These inconsistent results suggest that the effect of roasting on the antioxidant activity of different cereals may vary according to the specific phenolic profiles of the different cereals. Inglett et al. [73] similarly demonstrated that the extraction temperature significantly influenced both the phenolic content and the antioxidant capacity of buckwheat extracts, with the phenolic content ranging from 1.43 to 18.45 mg/g. The results of this study are consistent with these trends.

In oats, antioxidant activity consistently increased with thermal treatment. Calinoiu and Vodnar [62] reported that

the thermal treatment of oat bran enhanced its phenolic content, further supporting the present results. These correlations reinforce the conclusion that thermal processing can modulate antioxidant potential.

Effects of in vitro gastrointestinal digestion on the antioxidant activity and phenolic compounds of kilned rye, buckwheat, and oat

Table 4 presents the variations in TPC and TAC across the phases of in vitro gastrointestinal digestion, along with the bioaccessibility index (BI), which quantifies the fraction of phenolic compounds capable of crossing the simulated intestinal barrier (cellulose dialysis membrane) and thus represents the serum-available portion. Raw buckwheat, rye, and oats, and those kilned at different temperatures (120, 150, and 180 °C), were subjected to consecutive in vitro digestion simulating oral, gastric, and intestinal conditions. The TPC and antioxidant values at the oral phase were lower than those of the undigested sample due to the poor solubility of phenolics in the salivary fluid and the short duration of this phase. Therefore, the data for this phase are not included. Compared to the oral phase, the TPC of all three kilned samples significantly ($p \leq 0.05$) increased during the gastric phase. In plants, phenolic compounds exist in both soluble and insoluble-bound forms. Soluble phenolics are typically extractable using aqueous–organic solvent mixtures, while insoluble-bound phenolics require enzymatic treatment or extraction under variable acidic or alkaline conditions [74]. Therefore, the increase in phenolic content observed under gastric conditions in this study suggests that acidic pH and digestive enzymes may facilitate the release of phenolics bound to the cereal matrix. The most substantial relative rise was observed in buckwheat samples, with TPC increasing from 82.61% in the untreated sample to 83.43%, 85.55%, and 86.56% at 120, 150, and 180 °C, respectively. Similarly, Li et al. [75] demonstrated that extracts obtained from buckwheat hulls exhibited higher TPC and TAC compared to those derived from buckwheat flour. Jan et al. [76] also reported that baking reduced TPC in wheat cookies enriched with buckwheat flour; however, the TPC of buckwheat cookies remained higher than that of wheat cookies. As observed in the case of wheat cookies, in the present study, the oat sample exhibited lower TPC in the gastric phase compared to the undigested sample, despite an increase from the oral phase. This reduction may be attributed to the thermal degradation of phenolic compounds under gastric conditions.

A significant increase in bioaccessible TPC of rye samples was also detected in all soluble fractions produced following in vitro digestion compared to the samples prior to digestion. In vitro gastric digestion resulted in a progressive

increase in the TPC of rye samples compared to the untreated control, with increments of 11.89%, 24.20%, and 8.22% at 120 °C, 150 °C, and 180 °C, respectively. These results indicate that the optimum values were obtained from rye samples kilned at 150 °C. A similar effect has also been reported in previous studies [77, 78]. This finding is in agreement with the results of Chandrasekara and Shahidi [79], who reported increased phenolic content in millet extracts obtained after enzymatic digestion in the gastric phase. This may be attributed to the enhanced breakdown and release of phenolic compounds during the extended incubation period of the gastric phase, which increases their accessibility for absorption and contributes to the observed increase in TPC.

During intestinal digestion, the material was divided into the IN fraction (free soluble dialyzable fraction) and the OUT fraction (intestinal digest). In the dialyzed fraction following intestinal digestion (IN), the TPC was significantly lower than its initial value. After the intestinal phase, the TPC of the IN samples consistently remained lower than that of the OUT samples. Similar results were reported by Ozay Arancioglu et al. [80], who investigated the effects of different drying methods on the *in vitro* bioaccessibility of bioactive compounds in pomegranate arils. The TPC values of fresh pomegranate arils were 0.22 mg GAE/g for IN and 3.94 mg GAE/g for OUT. For freeze-dried pomegranate arils, the TPC values were 0.23 mg GAE/g for IN and 5.32 mg GAE/g for OUT, while ultrasound-assisted vacuum-dried samples indicated 0.66 mg GAE/g for IN and 6.03 mg GAE/g for OUT. These results demonstrate that the food matrix impacts bioaccessibility.

In the present study, a significant decrease was observed in the intestinal phase compared to the gastric phase, which may be related to the instability of phenolic compounds under the alkaline conditions of the small intestine. This instability may result from oxidation and polymerization reactions, the formation of high-molecular-weight phenolic derivatives with low solubility, and alterations in their chemical properties [81]. The limited release of phenolics is mainly attributed to strong physical interactions between free phenolic compounds and cell wall components, as well as the covalent bonding of phenolic acids, which are resistant to cleavage by human pancreatic enzymes [82]. The limited bioavailability facilitates the transport of phenolics to the large intestine, where they undergo microbial metabolism, potentially yielding beneficial health effects. This explanation provides meaningful insights into the higher TPC values in the OUT phase, highlighting the role of the food matrix in modulating phenolic availability.

Oxidative damage to biomolecules caused by excess free radicals has been associated with numerous chronic diseases, including cardiovascular disease, type 2 diabetes, obesity, and cancer. One possible mechanism by which

whole grains may help prevent these conditions is through the free radical scavenging activity of their phytochemicals [83]. The TAC of kilned whole grains was evaluated by ABTS and DPPH assays in the present study. The TAC of the rye samples was lower in the methanolic extracts of kilned samples at different temperatures compared to the initial levels in the control samples. In the gastric phase, both TPC and total antioxidant capacity increased, suggesting that the amount of biologically available polyphenols may have risen during gastric digestion. Previous studies have similarly reported that the majority of polyphenols released during simulated gastrointestinal digestion are obtained in the gastric phase. Most phenolic compounds in rye are known to exist in bound forms within the aleurone and bran layers [84]. Therefore, prolonged incubation during the gastric phase, combined with the acidic environment and enzymatic activity, may have facilitated the release of these bound phenolics. After intestinal digestion, the DPPH and ABTS analyses of the rye samples exhibited inconsistent patterns. While DPPH results showed a significant increase, antioxidant activity measured by ABTS decreased. This discrepancy is likely due to the two assays operating under different solubility environments and targeting radicals with different chemical structures [81]. ABTS assays interact with a broader range of compounds and are more sensitive to pH changes and structural transformations, which could explain the observed decrease. Moreover, transformation products generated during digestion may exhibit antioxidant activity that remains detectable by DPPH but is diminished in ABTS analysis. Similarly, in blackberry fruit, ABTS antioxidant activity was reported to be higher after gastric digestion compared to the intestinal phase [85].

Although oat samples had lower TPC values compared to buckwheat and rye, their TAC values, for the *in vitro* simulated digestion, were significantly higher. This is mainly due to the higher concentration of phenolic acids present in the digested oat fractions. This was in agreement with a previous study by Gong et al. [83], which found that the TAC of whole oat samples increased significantly during the digestion process. Bączek et al. [86] similarly reported that the bread-making process led to an increase in antioxidant compounds of oat and buckwheat flours. Despite the observed increase in TPC in buckwheat and rye samples, no corresponding enhancement in TAC was detected in the gastric and intestinal phases. This suggests that Maillard reaction products, which may be quantified as phenolics in TPC assays, contributed to the elevated TPC without necessarily enhancing TAC.

In addition to antioxidant capacity, Table 4 presents the bioaccessibility of phenolic compounds in kilned samples of buckwheat, oat, and rye. The bioaccessibility of a phytochemical refers to the proportion released from the food

matrix during digestion, rendering it available for absorption in the small intestine or biotransformation by the gut microbiota. This process is influenced by various factors, including the chemical nature of the phytochemical, the initial state of the food, and the dynamic conditions within the gastrointestinal tract.

The phenolic content of buckwheat was higher than that of kilned oat and rye samples. However, the bioaccessibility index (BI) of buckwheat was significantly lower, ranging between 243.23% and 302.19%. By contrast, oats exhibited higher bioaccessible TPC values (229.99% for the sample kilned at 150 °C and 443.60% for raw sample), whereas rye indicated 328.10% at 180 °C and 444.09% at 120 °C. These results imply that oats exhibited higher bioaccessibility than rye under similar conditions. These varying outcomes may be due to differences in cereal type, cultivation conditions, geographical origin, and postharvest processes, all of which help shape the phenolic composition and fate of these compounds during digestion [87].

The BI of TAC values revealed distinct patterns among the kilned grains. For oats, TAC remained consistently higher across all thermal treatments, ranging from 120.46% for the untreated oat sample to 111.56% at 180 °C, indicating high antioxidant bioaccessibility compared to other grains. By contrast, rye demonstrated notable improvements in TAC post-kilning, increasing from 21.44% to 62.75% at 180 °C (Table 4). Buckwheat, while exhibiting moderate TAC values, showed variability with kilning conditions. Buckwheat's TAC peaked at 52.17% at 150 °C but declined at higher temperatures (48.22% at 180 °C), suggesting the potential thermal sensitivity of its antioxidants at elevated temperatures. These findings illustrate the varying impacts of thermal treatment on the bioaccessibility of antioxidants in different grains, with oats maintaining consistently high bioaccessibility, rye benefiting from kilning, and buckwheat displaying potential thermal sensitivity at elevated temperatures.

These diverse outcomes can be attributed to variations in the chemical composition of oats, rye, and buckwheat. Rye and buckwheat have a higher fiber content than oats, and thermal treatment can modify the structure of fibers, thereby influencing digestibility. Dietary fibers, particularly insoluble fibers in cereal grains, reduce the bioaccessibility of phenolic compounds by entrapping these compounds and increasing gut fluid viscosity [88]. This increased viscosity can hinder enzymatic activity by limiting effective peristaltic movement, hindering enzyme diffusion and substrate interaction. In turn, this may lead to the accumulation of phenolic compounds in the large intestine and reduce bioaccessibility in the small intestine [87]. This may explain the enhanced TPC digestibility of rye and buckwheat following heat treatment, and the reduced

digestibility of oats. Furthermore, the higher fat content in oats, compared to the other cereals, may also play a role, potentially affecting the bioaccessibility of its antioxidants after thermal processing. The fat content plays a crucial role in digestibility because it influences enzymatic activity and emulsification. Fat digestion requires lipase, an enzyme responsible for breaking down lipids. A high fat content can overwhelm lipase activity, reducing its efficacy and making digestion more challenging. Moreover, fats require emulsification by bile salts for proper digestion [89]. The increase observed in the intestinal phase of oat sample, and consequently in bioavailability, can be attributed to the effect of lipase enzymes and bile salts, which facilitate this condition.

Conclusion

Heat treatment significantly influenced the functional and nutritional properties of buckwheat, rye, and oats. This study represents one of the comparative evaluations of three whole grains under harmonized kilning conditions and therefore provides novel insights into grain-specific thermal responses. The processing temperature differentially affected glycemic index, bile acid-binding capacity, antioxidant activity, and phenolic bioaccessibility, demonstrating that controlled dry-heat treatment can optimize health-promoting properties while preserving whole grain integrity.

From an application standpoint, these findings suggest that controlled heat treatment can be used to enhance the health-promoting properties of whole grains, supporting their use in functional food formulations. The findings have practical implications for the development of minimally processed, fiber-rich snacks that contribute to digestive and cardiovascular health through improved phenolic accessibility and bile acid-binding capacity. Future research should further investigate sensory acceptance, shelf-life stability, industrial scalability, as well as explore advanced processing techniques to validate their commercial potential.

Authors' contributions Betül Bay Yılmaz: Conceptualization, Writing – original draft, Data curation, Validation, Investigation, Formal analysis, Methodology, Funding acquisition, Project administration. Seher Serin: Conceptualization, Writing – original draft, Data curation, Formal analysis, Validation, Visualization, Software. Hatice Bekiroğlu: Writing – review & editing, Methodology, Formal analysis, Data curation, Investigation. Görkem Özülkü: Writing – review & editing, Methodology, Investigation. Osman Sağdıç: Writing – review & editing, Supervision, Resources.

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Data availability Data will be made available on request.

Declarations

Ethics approval Ethical approval for the involvement of human participants in the present study was obtained from the Research Ethics Committee of Yildiz Technical University, Faculty of Chemical and Metallurgical Engineering, Department of Food Engineering, in February 2025.

Consent to participate All participants provided written informed consent prior to their involvement in the study. They were fully informed about the purpose of the research, their right to withdraw at any time, and the confidentiality of their data.

Conflict of interest The authors declare no competing interests.

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References

- Ergun R (2014) Türkiye'ye Özgü Bazı Ekmek Türlerinin Glisemik İndeks Değerlerinin Saptanması. MSc Thesis, Hacettepe University, Department of Nutrition Sciences, Ankara
- K. Srikaeo, S. Mingyai, P.A. Sopade, Physicochemical properties, resistant starch content and enzymatic digestibility of unripe banana, edible canna, taro flours and their rice noodle products. *Int. J. Food Sci. Technol.* (2011). <https://doi.org/10.1111/j.1365-2621.2011.02724.x>
- P.G. Williams, The benefits of breakfast cereal consumption: a systematic review of the evidence base. *Adv. Nutr.* **5**(5), 636S–673S (2014). <https://doi.org/10.3945/an.114.006247>
- A. Makowska, A. Polcyn, S. Chudy, J. Michniewicz, Application of oat, wheat and rye bran to modify nutritional properties, physical and sensory characteristics of extruded corn snacks. *Acta Sci. Pol. Technol. Aliment.* **14**(4), 375–386 (2015). <https://doi.org/10.17306/J.AFS.2015.4.37>
- J.M. Jones, J. Adams, C. Harriman, C. Miller, J.W. Van der Kamp, Nutritional impacts of different whole grain milling techniques: a review of milling practices and existing data. *CFW* **60**(3), 130–139 (2015). <https://doi.org/10.1094/CFW-60-3-0130>
- S. Kowalski, A. Mikulec, B. Mickowska, K. Buksa, Nutritional properties and amino acid profile of buckwheat bread. *J. Food Sci. Technol.* **59**(8), 3020–3030 (2022). <https://doi.org/10.1007/s13197-022-05518-w>
- A. Ahmed, N. Khalid, A. Ahmad, N.A. Abbasi, M.S.Z. Latif, M.A. Randhawa, Phytochemicals and biofunctional properties of buckwheat: a review. *J. Agric. Sci.* **152**(3), 349–369 (2014). <https://doi.org/10.1017/S0021859613000166>
- M. Sluková, L. Jurkaninová, I. Švec, P. Skřivan, Rye—the nutritional and technological evaluation in Czech cereal technology—a review: grain and flours. *Czech J. Food Sci.* (2021). <https://doi.org/10.17221/203/2020-CJFS>
- D. Gumul, J. Korus, K. Czechowska, H. Bartoń, M. Folta, The impact of extrusion on the content of polyphenols and antioxidant activity of rye grains (*Secale cereale* L.). *Acta Sci. Pol. Technol.* **9**(3), 319–330 (2010)
- D. Žadeikė, C. Copperstone, O. Aleksandrova, D.Ö. Ünal, K. Šavikin, J. Živković, M. Knez, Rye (*Secale cereale* L.) revisited—nutritional composition, functional benefits, and role in sustainable diets. *Front. Nutr.* **12**, 1–18 (2025). <https://doi.org/10.3389/fnut.2025.1666455>
- S.A. Joyce, A. Kamil, L. Fleige, C.G. Gahan, The cholesterol-lowering effect of oats and oat beta glucan: modes of action and potential role of bile acids and the microbiome. *Front. Nutr.* **6**, 171 (2019). <https://doi.org/10.3389/fnut.2019.00171>
- S.T. Koç, F. Coşkun, Buckwheat: nutritional value, health effects and applications in foods. *TURJAF* **13**(6), 1665–1674 (2025). <https://doi.org/10.24925/turjaf.v13i6.1665-1674.7565>
- H. Zielińska, H. Kozłowska, Antioxidant activity and total phenolics in selected cereal grains and their different morphological fractions. *JAFAC* **48**(6), 2008–2016 (2000). <https://doi.org/10.1021/jf990619o>
- Y. Liu, C. Cai, Y. Yao, B. Xu, Alteration of phenolic profiles and antioxidant capacities of common buckwheat and tartary buckwheat produced in China upon thermal processing. *J. Sci. Food Agric.* **99**(12), 5565–5576 (2019). <https://doi.org/10.1002/jsfa.9825>
- T. Mohammadi Moghaddam, S.M. Razavi, M. Taghizadeh, A. Sazgarnia, Sensory and instrumental texture assessment of roasted pistachio nut/kernel by partial least square (PLS) regression analysis: effect of roasting conditions. *J. Food Sci. Technol.* **53**, 370–380 (2016). <https://doi.org/10.1007/s13197-015-2054-2>
- AACC (1995) Approved methods of the AACC, 9th ed. Method 30–20 A., Method 44–15 A, Method 46–13 A, Method 08–01 A, American Association of Cereal Chemists, St. Paul, MN
- L. Prosky, N.G. Asp, T.F. Schweizer, J.W. DeVries, I. Furda, Determination of insoluble, soluble and total dietary fiber in foods and food products: interlaboratory study. *J. AOAC Int.* **71**, 1017–1023 (1988). <https://doi.org/10.1093/jaoac/71.5.1017>
- H.N. Englyst, J. Veenstra, G.J. Hudson, Measurement of rapidly available glucose (RAG) in plant foods: a potential *in vitro* predictor of the glycaemic response. *Br. J. Nutr.* **75**(3), 327–337 (1996). <https://doi.org/10.1079/bjn19960137>
- I. Goni, A. Garcia-Alonso, F. Saura-Calixto, A starch hydrolysis procedure to estimate glycemic index. *Nutr. Rev.* **17**(3), 427–437 (1997). [https://doi.org/10.1016/S0271-5317\(97\)00010-9](https://doi.org/10.1016/S0271-5317(97)00010-9)
- V.L. Singleton, J.A. Rossi, Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *AJEV* **16**(3), 144–158 (1965). <https://doi.org/10.5344/ajev.1965.16.3.144>
- R.P. Singh, K.N. Chidambara Murthy, G.K. Jayaprakasha, Studies on the antioxidant activity of pomegranate (*Punica granatum*) peel and seed extracts using *in vitro* models. *J. Agric. Food Chem.* **50**(1), 81–86 (2002). <https://doi.org/10.1021/jf010865b>
- R. Re, N. Pellegrini, A. Proteggente, A. Pannala, M. Yang, C. Rice-Evans, Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radic. Biol. Med.* **26**(9–10), 1231–1237 (1999). [https://doi.org/10.1016/S0891-5849\(98\)00315-3](https://doi.org/10.1016/S0891-5849(98)00315-3)
- K. Ozkan, H. Bekiroglu, Y. Bayram, O. Sagdic, S. Erbas, *In vitro* bioaccessibility, antioxidant and antibacterial activities of three different safflower (*Carthamus tinctorius* L.) genotypes. *Food Sci Technol* **42**, e08921 (2021). <https://doi.org/10.1590/fst.08921>
- M. Minekus, M. Alming, P. Alvito, S. Ballece, T. Bohn, C. Bourlieu et al., A standardised static *in vitro* digestion method

- suitable for food—an international consensus. *Food Funct.* **5**, 1113–1124 (2014). <https://doi.org/10.1039/C3FO60702J>
25. S. Sayar, J.L. Jannink, P.J. White, *In vitro* bile acid binding of flours from oat lines varying in percentage and molecular weight distribution on β -glucan. *J. Agric. Food Chem.* **53**, 8797–8803 (2005). <https://doi.org/10.1021/jf051380g>
 26. M. He, T. Guo, D. Li, C. Xie, P. Wang, R. Yang, Effects of roasting on physicochemical characteristics and flavor substances of germinated brown rice. *Food Sci. Biotechnol.* **34**(1), 125–135 (2025). <https://doi.org/10.1007/s10068-024-01655-4>
 27. K. Ikeda, Buckwheat composition, chemistry, and processing. *Adv. Food Nutr. Res.* **44**, 395–434 (2002). [https://doi.org/10.1016/S1043-4526\(02\)44008-9](https://doi.org/10.1016/S1043-4526(02)44008-9)
 28. K. Christa, M. Soral-Smietana, Buckwheat grains and buckwheat products—nutritional and prophylactic value of their components—a review. *Czech J Food Sci* **26**(3), 153–162 (2008)
 29. H.B. Hansen, B. Møller, S.B. Andersen, J.R. Jørgensen, Å. Hansen, Grain characteristics, chemical composition, and functional properties of rye (*Secale cereale* L.) as influenced by genotype and harvest year. *J. Agric. Food Chem.* **52**(8), 2282–2291 (2004). <https://doi.org/10.1021/jf0307191>
 30. Wrigley C, Bushuk W (2017) Rye: grain-quality characteristics and management of quality requirements. In *Cereal Grains*. 153–178. Woodhead Publishing. <https://doi.org/10.1016/B978-0-08-100719-8.00007-3>
 31. D.M. Peterson, Composition and nutritional characteristics of oat grain and products. *Oat Sci Technol* **33**, 265–292 (1992). <https://doi.org/10.2134/agronmonogr33.c10>
 32. F.A. Mantey, G.A. Hareland, D.J. Huseby, Soluble and insoluble dietary fiber content and composition in oat. *Cereal Chem.* **76**(3), 417–420 (1999). <https://doi.org/10.1094/CCHEM.1999.76.3.417>
 33. D.C. Doehlert, S. Simsek, D. Thavarajah, P. Thavarajah, J.B. Ohm, Detailed composition analyses of diverse oat genotype kernels grown in different environments in North Dakota. *Cereal Chem.* **90**(6), 572–578 (2013). <https://doi.org/10.1094/CCHEM-09-12-0111-R>
 34. K. Zhang, R. Dong, X. Hu, C. Ren, Y. Li, Oat-based foods: chemical constituents, glycemic index, and the effect of processing. *Foods* **10**(6), 1304 (2021). <https://doi.org/10.3390/foods10061304>
 35. K.J. Steadman, M.S. Burgoon, B.A. Lewis, S.E. Edwardson, R.L. Obendorf, Buckwheat seed milling fractions: description, macronutrient composition and dietary fibre. *J. Cereal Sci.* **33**(3), 271–278 (2001). <https://doi.org/10.1006/jcrs.2001.0366>
 36. E.A. Decker, D.J. Rose, D. Stewart, Processing of oats and the impact of processing operations on nutrition and health benefits. *Br. J. Nutr.* **112**(S2), S58–S64 (2014). <https://doi.org/10.1017/S000711451400227X>
 37. V.M. Mathobo, H. Silungwe, S.E. Ramashia, T.A. Anyasi, Effects of heat-moisture treatment on the thermal, functional properties and composition of cereal, legume and tuber starches—a review. *J. Food Sci. Technol.* **58**(2), 412–426 (2021). <https://doi.org/10.1007/s13197-020-04520-4>
 38. M.C. Chang, W.C. Morris, Effect of heat treatments on chemical analysis of dietary fiber. *J. Food Sci.* **55**(6), 1647–1650 (1990). <https://doi.org/10.1111/j.1365-2621.1990.tb03591.x>
 39. E. Takeyama, M. Fukushima, A. Tanimura, Effect of heat-treatment on the content and polysaccharide composition of dietary fiber. *Food Sci. Technol. Res.* **8**(2), 125–130 (2002). <https://doi.org/10.3136/fstr.8.125>
 40. A. Căpriță, R. Căpriță, V.O. Simulescu, R.M. Drehe, The effect of temperature on soluble dietary fiber fraction in cereals. *J. Agroaliment. Process. Technol.* **17**(3), 214–217 (2011)
 41. J.L. Dong, M. Yang, R.L. Shen, Y.F. Zhai, X. Yu, Z. Wang, Effects of thermal processing on the structural and functional properties of soluble dietary fiber from whole grain oats. *FSTI* **25**(4), 282–294 (2019). <https://doi.org/10.1177/1082013218817705>
 42. M. Tomas, J. Beekwilder, R.D. Hall, C.D. Simon, O. Sagdic, E. Capanoglu, Effect of dietary fiber (inulin) addition on phenolics and *in vitro* bioaccessibility of tomato sauce. *Food Res. Int.* **106**, 129–135 (2018). <https://doi.org/10.1016/j.foodres.2017.12.050>
 43. S. Serin, S. Sayar, *In vitro* digestibility, glycaemic index and bile acid-binding capacity of foods containing different types of resistant starch in comparison with the commercial resistant starches. *Qual. Assur. Saf. Crops Foods* **15**(3), 68–76 (2023). <https://doi.org/10.15586/qas.v15i3.1215>
 44. G. Dongowski, Interactions between dietary fibre-rich preparations and glycoconjugated bile acids *in vitro*. *Food Chem.* **104**(1), 390–397 (2007). <https://doi.org/10.1016/j.foodchem.2006.11.053>
 45. K. Dziedzic, D. Górecka, M. Kucharska, B. Przybylska, Influence of technological process during buckwheat groats production on dietary fibre content and sorption of bile acids. *Food Res. Int.* **47**(2), 279–283 (2012). <https://doi.org/10.1016/j.foodres.2011.07.020>
 46. Chirirang P, Oonsivilai R (2018) Physicochemical properties, *in-vitro* binding capacities for lard, cholesterol, bile acids and assessment of prebiotic potential of dietary fiber from cassava pulp. *Int. Food Res J* **25**
 47. de Moura FA, Pereira, JM, da Silva, DO, Zavareze, E, da, R, da Silveira Moreira, A (2011) Effects of oxidative treatment on the physicochemical, rheological and functional properties of oat β -glucan. *Food Chem.* **128**(4): 982–987. <https://doi.org/10.1016/j.foodchem.2011.04.003>
 48. H.J. Kim, P.J. White, *In vitro* bile-acid binding and fermentation of high, medium, and low molecular weight β -glucan. *J. Agric. Food Chem.* **58**(1), 628–634 (2010). <https://doi.org/10.1021/jf902508t>
 49. B. Cetiner, S. Tömösközi, E. Schall, A. Salantur, H. Koksels, Bile acid binding capacity, dietary fibre and phenolic contents of modern and old bread wheat varieties and landraces: a comparison over the course of around one century. *Eur. Food Res. Technol.* **248**(2), 589–598 (2022). <https://doi.org/10.1007/s00217-021-03906-8>
 50. P.A. Sopade, Cereal processing and glycaemic response. *Int. J. Food Sci. Technol.* **52**(1), 22–37 (2017). <https://doi.org/10.1111/ijfs.13237>
 51. H. Kaur, B.S. Gill, B.L. Karwasra, *In vitro* digestibility, pasting, and structural properties of starches from different cereals. *Int. J. Food Prop.* **21**(1), 70–85 (2018). <https://doi.org/10.1080/10942912.2018.1439955>
 52. S.Y. Chung, S.H. Han, S.W. Lee, C. Rhee, Effect of Maillard reaction products prepared from glucose–glycine model systems on starch digestibility. *Starch-Stärke* **64**(8), 657–664 (2012). <https://doi.org/10.1002/star.201100176>
 53. E.M. Ucar, M. Kizil, A review on healthier bread making: focusing on the underlying mechanisms for decreasing glycemic index. *Food Rev. Int.* (2025). <https://doi.org/10.1080/87559129.2025.2450046>
 54. S. Bhinder, B. Singh, A. Kaur, N. Singh, M. Kaur, S. Kumari, M.P. Yadav, Effect of infrared roasting on antioxidant activity, phenolic composition and Maillard reaction products of Tartary buckwheat varieties. *Food Chem.* **285**, 240–251 (2019). <https://doi.org/10.1016/j.foodchem.2019.01.141>
 55. W. Małgorzata, P.M. Konrad, H. Zieliński, Effect of roasting time of buckwheat groats on the formation of Maillard reaction products and antioxidant capacity. *Food Chem.* **196**, 355–358 (2016). <https://doi.org/10.1016/j.foodchem.2015.09.064>
 56. Zamaratskaia G, Gerhardt K, Knicky M, Wendin K (2023) Buckwheat: An underutilized crop with attractive sensory qualities and health benefits. *Crit Rev Food Sci Nutr* **1–16**. <https://doi.org/10.1080/10408398.2023.2249112>

57. M. Ovando-Martínez, K. Whitney, B.L. Reuhs, D.C. Doehlert, S. Simsek, Effect of hydrothermal treatment on physicochemical and digestibility properties of oat starch. *Food Res. Int.* **52**(1), 17–25 (2013). <https://doi.org/10.1016/j.foodres.2013.02.035>
58. B. Borczak, M. Sikora, E. Sikora, A. Dobosz, J. Kapusta-Duch, Glycaemic index of wheat bread. *Starch-Stärke* **70**(1–2), 1700022 (2018). <https://doi.org/10.1002/star.201700022>
59. A. Ikram, F. Saeed, R.A. Noor, A. Imran, M. Afzaal, A. Rasheed, F. Islam, A. Iqbal, T. Zahoor, S. Naz, W. Waheed, M.Z. Shahid, A.W. Khan, A.B. Kinki, A comprehensive review on biochemical and technological properties of rye (*Secale cereale* L.). *Int. J. Food Prop.* **26**(1), 2212–2228 (2023). <https://doi.org/10.1080/10942912.2023.2244697>
60. K.S. Juntunen, D.E. Laaksonen, K. Autio, L.K. Niskanen, J.J. Holst, K.E. Savolainen, H.M. Mykkänen, Structural differences between rye and wheat breads but not total fiber content may explain the lower postprandial insulin response to rye bread. *AJCN* **78**(5), 957–964 (2003). <https://doi.org/10.1093/ajcn/78.5.957>
61. Q. Ma, Y. Zhao, H.L. Wang, J. Li, Q.H. Yang, L.C. Gao, B.L. Feng, Comparative study on the effects of buckwheat by roasting: Antioxidant properties, nutrients, pasting, and thermal properties. *J. Cereal Sci.* **95**, 103041 (2020). <https://doi.org/10.1016/j.jcs.2020.103041>
62. L.F. Călinoiu, D.C. Vodnar, Thermal processing for the release of phenolic compounds from wheat and oat bran. *Biomolecules* **10**(1), 21 (2019). <https://doi.org/10.3390/biom10010021>
63. S. Ragaei, K. Seetharaman, E.S.M. Abdel-Aal, The impact of milling and thermal processing on phenolic compounds in cereal grains. *Crit. Rev. Food Sci. Nutr.* **54**(7), 837–849 (2014). <https://doi.org/10.1080/10408398.2011.610906>
64. K.H. Liukkonen, K. Katina, A. Wilhelmsson, O. Myllymaki, A.M. Lampi, S. Kariluoto, V. Piironen, S.-M. Heinonen, T. Nurmi, H. Adlercreutz, A. Peltoketo, J.-M. Pihlavan, V. Hietaniemi, K. Poutanen, Process-induced changes on bioactive compounds in whole grain rye. *Proc. Nutr. Soc.* **62**(1), 117–122 (2003). <https://doi.org/10.1079/PNS2002218>
65. M. Iftikhar, H. Zhang, A. Iftikhar, A. Raza, M. Khan, M. Sui, J. Wang, Comparative assessment of functional properties, free and bound phenolic profile, antioxidant activity, and in vitro bioaccessibility of rye bran and its insoluble dietary fiber. *J. Food Biochem.* **44**(10), e13388 (2020). <https://doi.org/10.1111/jfbc.13388>
66. J.H. Lee, B.K. Lee, B.W. Lee, H.J. Kim, J.Y. Park, S. Han, Y.Y. Lee, Evaluation of bioactive compounds and antioxidant activity of roasted oats in different extraction solvents. *Korean J Food Sci Technol* **50**(1), 111–116 (2018). <https://doi.org/10.9721/KJFST.2018.50.1.111>
67. K.S. Sandhu, P. Godara, M. Kaur, S. Punia, Effect of toasting on physical, functional and antioxidant properties of flour from oat (*Avena sativa* L.) cultivars. *J. Saudi Soc. Agric. Sci.* **16**(2), 197–203 (2017). <https://doi.org/10.1016/j.jssas.2015.06.004>
68. V. Verardo, V. Glicerina, E. Cocci, A.G. Frenich, S. Romani, M.F. Caboni, Determination of free and bound phenolic compounds and their antioxidant activity in buckwheat bread loaf, crust, and crumb. *LWT-Food Sci Technol* **87**, 217–224 (2018). <https://doi.org/10.1016/j.lwt.2017.08.063>
69. A. Cammerata, B. Laddomada, F. Milano, F. Camerlengo, M. Bonarrigo, S. Masci, F. Sestili, Qualitative characterization of unrefined Durum wheat air-classified fractions. *Foods* **10**, 2817 (2021). <https://doi.org/10.3390/foods10112817>
70. D. Gumul, J. Korus, B. Achremowicz, The influence of extrusion on the content of polyphenols and antioxidant/antiradical activity of rye grains (*secale cereale* L.). *Acta Sci. Pol. Technol.* **6**(4), 103–111 (2007)
71. B. Solmaz, H. Levent, N. Şahin, The influence of various cereal brans, stabilized through hot air, microwave, and autoclave methods, on the physicochemical properties of cookies. *Food Sci. Nutr.* **13**(6), e70328 (2025). <https://doi.org/10.1002/fsn3.70328>
72. M. Schutte, S. Hayward, Manley, M (2024) Nonenzymatic browning and antioxidant properties of thermally treated cereal grains and end products. *J. Food Biochem.* **1**, 3865849 (2024). <https://doi.org/10.1155/2024/3865849>
73. G.E. Inglett, D.J. Rose, D. Chen, D.G. Stevenso, A. Biswas, Phenolic content and antioxidant activity of extracts from whole buckwheat (*Fagopyrum esculentum* Möench) with or without microwave irradiation. *Food Chem.* **119**(3), 1216–1219 (2010). <https://doi.org/10.1016/j.foodchem.2009.07.041>
74. T.M. Ucar, A. Karadag, The effects of vacuum and freeze-drying on the physicochemical properties and *in vitro* digestibility of phenolics in oyster mushroom (*Pleurotus ostreatus*). *J. Food Meas. Charact.* **13**(3), 2298–2309 (2019). <https://doi.org/10.1007/s11694-019-00149-w>
75. F. Li, Y. Yuan, X.I. Yang, S. Tao, J. Ming, Phenolic profiles and antioxidant activity of buckwheat (*Fagopyrum esculentum* Moench and *Fagopyrum tartaricum* L. Gaerth) hulls, brans and fours. *J. Integr. Agric.* **12**, 1684–1693 (2013). [https://doi.org/10.1016/S2095-3119\(13\)60371-8](https://doi.org/10.1016/S2095-3119(13)60371-8)
76. U. Jan, A. Gani, M. Ahmad, U. Shah, W.N. Baba, F.A. Masoodi, S. Maqsood, A. Gani, I.A. Wani, S.M. Wani, Characterization of cookies made from wheat four blended with buckwheat four and effect on antioxidant properties. *J. Food Sci. Technol.* **52**, 6334–6344 (2015). <https://doi.org/10.1007/s13197-015-1773-8>
77. W. Tian, G. Chen, M. Tilley, Y. Li, Changes in phenolic profiles and antioxidant activities during the whole wheat bread-making process. *Food Chem.* **345**, 128851 (2021). <https://doi.org/10.1016/j.foodchem.2020.128851>
78. M.B. Durairajan, V.V. Sundararajan, G. Kannan, B.M. Paul, K. Muniyandi, P. Thangaraj, Elicitation of nutritional, antioxidant, and antidiabetic potential of barnyard millet (*Echinochloa esculenta* (A. Braun) H. Scholz) sprouts and microgreens through *in vitro* bio-accessibility assessment. *Food Chem.* **441**, 138282 (2024). <https://doi.org/10.1016/j.foodchem.2023.138282>
79. A. Chandrasekara, F. Shahidi, Bioaccessibility and antioxidant potential of millet grain phenolics as affected by simulated *in vitro* digestion and microbial fermentation. *J. Funct. Foods* **4**, 226–237 (2012). <https://doi.org/10.1016/j.jff.2011.11.001>
80. Ozay-Arancioglu I, Bekiroglu H, Karadag A, Saroglu O, Tekin-Çakmak Z H, Karasu S (2021) Effect of different drying methods on the bioactive, microstructural, and in-vitro bioaccessibility of bioactive compounds of the pomegranate arils. *Food Sci Technol* e06221. <https://doi.org/10.1590/ft.06221>
81. E. Yener, O. Saroglu, O. Sagdic, A. Karadag, The effects of different drying methods on the *in vitro* bioaccessibility of phenolics, antioxidant capacity, and morphology of European plums (*Prunes domestica* L.). *ACS Omega* **9**(11), 12711–12724 (2024). <https://doi.org/10.1021/acsomega.3c08383>
82. F. Giusti, E. Capuano, G. Sagratini, N. Pellegrini, A comprehensive investigation of the behaviour of phenolic compounds in legumes during domestic cooking and *in vitro* digestion. *Food Chem.* **285**, 458–467 (2019). <https://doi.org/10.1016/j.foodchem.2019.01.148>
83. E.S. Gong, N. Gao, T. Li, H. Chen, Y. Wang, X. Si, J. Tian, C. Shu, S. Luo, J. Zhang, Z. Zeng, W. Xia, B. Li, C. Liu, R.H. Liu, Effect of *in vitro* digestion on phytochemical profiles and cellular antioxidant activity of whole grains. *J. Agric. Food Chem.* **67**(25), 7016–7024 (2019). <https://doi.org/10.1021/acs.jafc.9b02245>
84. D. Dziki, Rye flour and rye bran: New perspectives for use. *Processes* **10**(2), 293 (2022). <https://doi.org/10.3390/pr10020293>
85. L. Liang, X. Wu, T. Zhao, J. Zhao, F. Li, Y. Zou, G. Mao, L. Yang, *In vitro* Bioaccessibility and Antioxidant Activity of Anthocyanins from Mulberry (*Morus Atropurpurea* Roxb.) Following

- Simulated Gastro-Intestinal Digestion. *Food Res. Int.* **46**(1), 76–82 (2012). <https://doi.org/10.1016/j.foodres.2011.11.024>
86. N. Bączek, A. Jarmułowicz, M. Wronkowska, C.M. Haros, Assessment of the glycaemic index, content of bioactive compounds, and their *in vitro* bioaccessibility in oat-buckwheat breads. *Food Chem.* **330**, 127199 (2020). <https://doi.org/10.1016/j.foodchem.2020.127199>
87. E.F. Aydar, Z. Mertdinç, E. Demircan, S.K. Çetinkaya, B. Özçelik, Kidney bean (*Phaseolus vulgaris* L.) milk substitute as a novel plant-based drink: fatty acid profile, antioxidant activity, *in-vitro* phenolic bio-accessibility and sensory characteristics. *Innov. Food Sci. Emerg. Technol.* **83**, 103254 (2023). <https://doi.org/10.1016/j.ifset.2022.103254>
88. C.C. Ng, S.P. Loh, Effect of selected cereal grains on *in vitro* bioaccessibility of isoflavones in soymilk. *Int. Food Res. J.* **25**(1), 303–309 (2018)
89. Cheng HM, Mah KK, Seluakumaran K (2020) Fat digestion: Bile salt, emulsification, micelles, lipases, chylomicrons. *Defining Physiology: Principles, Themes, Concepts. Neurophysiology and Gastrointestinal Systems* 63–65. https://doi.org/10.1007/978-3-030-62285-5_18

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