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Dietary intake of polyphenolic compounds from major food groups: results of analytical determinations

JOANNA TOKARCZYK^{1*}, MARCIN CZOP², WOJCIECH KOCH¹

¹ Department of Food and Nutrition, Medical University of Lublin, Poland

² Department of Clinical Genetics, Medical University of Lublin, Poland

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ABSTRACT

Polyphenols are widely distributed in plants and plant-based foods. Although they are not considered essential nutrients, they exhibit a broad spectrum of biological properties, particularly antioxidant activity. The aim of this study was to evaluate dietary intake of polyphenolic compounds derived from major food groups among young Polish adults. Dietary intake was assessed using a 24-hour dietary recall method supported by dedicated computer software. Liquid chromatography-mass spectrometry (LC-MS) was applied to identify major polyphenolic compounds, while the Folin-Ciocalteu assay and the DPPH radical scavenging test were used to evaluate the antioxidant activity of basic food groups. The primary sources of polyphenolic compounds were fruits, vegetables, and, most notably, beverages – mainly tea – which accounted for over 65% of beverage consumption in both interviewed groups. The predominant phenolic compounds identified were gallic acid, epigallocatechin gallate (EGCG), quercetin, and rutin. Gallic acid was the main polyphenolic component in daily food rations, with beverages constituting its primary dietary source. Only trace amounts of polyphenols were detected in eggs, fish, oils, milk, and meat products. The antioxidant activity of female diets (682.6 mg GAEq) was slightly higher compared with male food rations (680.04 mg GAEq).

INTRODUCTION

Food intake is one of the essential physiological requirements, and appropriate nutrition guarantees normal growth and development of the human body. Food products contain major and minor nutrients, non-nutritive components, and other compounds that may be either beneficial or disadvantageous to health [1]. Phenolic compounds are secondary plant metabolites responsible for the organoleptic quality and biological activity of fruits, vegetables, and other plant-derived foods; therefore, these products and beverages are particularly rich sources of this class of compounds [2]. Although phenolic compounds are not essential nutrients, they may exert beneficial effects on human health and are thus classified as biologically active non-nutrients [3].

Dietary polyphenols exhibit a wide spectrum of biological properties, especially antioxidant activity, which is associated with the presence of hydroxyl groups in their chemical structure [4]. Oxidative stress is considered a key trigger in the development of numerous diseases, including cardiovascular, renal, hepatic, and neurodegenerative disorders

[5-7]. Moreover, reactive oxygen species play a significant role in aging processes and carcinogenesis [5,8]. It has been demonstrated that the consumption of leafy vegetables rich in flavonoids and other phenolic compounds with antioxidant properties significantly reduces the risk of diabetes, cancer, cardiovascular diseases, and neurodegenerative disorders [9]. However, excessive intake of certain polyphenols may also lead to adverse effects, as some of these compounds exhibit anti-nutritional properties, including the chelation of iron and other trace elements, thereby reducing their bioavailability [10,11].

Polyphenolic compounds occur in plant and plant-derived foods in various forms and concentrations. Isoflavones are mainly present in legumes, flavanols (primarily catechins) are abundant in tea, whereas flavanones are characteristic of citrus fruits. Other compounds, such as quercetin, are ubiquitously distributed among plant-derived foods [12,13]. Among phenolic acids, caffeic acid and its esters (e.g., chlorogenic acid) are the most abundant and are commonly found in coffee, fruits, and vegetables – products that are frequently consumed and in relatively large quantities.

* Corresponding author

e-mail: joanna.tokarczyk@umlub.edu.pl

Nevertheless, flavonoids constitute the largest and most widespread group of polyphenols in plants and foods [14].

Due to their chemical instability, polyphenols undergo various transformations during food storage and processing, which may affect their concentration, intestinal absorption, and bioavailability [15]. Food processing generally leads to polyphenol degradation; however, in some cases, it may improve their bioaccessibility [16,17]. Arts *et al.* demonstrated that catechin content in fruits and vegetables is significantly reduced during thermal processing and that their concentration in processed products is usually lower than in fresh materials [18]. In turn, Miglio *et al.* showed that steaming is more effective than boiling in preserving phenolic compounds in vegetables [19]. Overall, the type of processing, its duration, intensity, and the heating medium applied have a significant impact on the polyphenol content of final food products [15].

The aim of this study was to identify the major dietary sources of selected polyphenolic compounds based on nutritional interviews combined with analytical determinations using liquid chromatography-mass spectrometry (LC-MS). An additional objective was to evaluate the total intake of polyphenols using the Folin-Ciocalteu method and to assess the antioxidant activity of reconstructed diets, with consideration of their division into major food product groups.

MATERIALS AND METHODS

1. Chemicals (Reagents)

Standards of gallic acid (GA), quercetin, epigallocatechin gallate (EGCG), and rutin of chromatographic purity (>98%), as well as 2,2-diphenyl-1-picrylhydrazyl (DPPH), Trolox, and citric acid, were purchased from Sigma-Aldrich (St. Louis, MO, USA). The Folin-Ciocalteu reagent was obtained from Stanlab (Lublin, Poland). Methanol (HPLC grade) was purchased from Avantor Performance Materials (POCH, Gliwice, Poland). All reagents used in the study were of analytical grade.

2. Sampling

The study material consisted of food samples prepared on the basis of previously conducted nutritional surveys. The study group included young adults (274 women and 305 men) aged 20–30 years who completed validated questionnaires concerning their dietary habits. Participants were asked to provide a detailed description of their daily food intake, including the type and mass (in grams) of consumed meals and food products.

Data were collected between October 2016 and March 2018 in the Lublin region (Poland). All participants were informed about the purpose of the study and assured of anonymity and voluntariness. After data verification, 41 incorrectly completed questionnaires were excluded from further analysis.

Combined qualitative and quantitative dietary data were processed using two nutritional software programs: Dietetyk 2006 (Jumar, Poznań, Poland) and Dieta 5.0 (National Institute of Public Health, Warsaw, Poland). These programs enabled the calculation of mean dietary intake values separately for women and men and their division into 12

major food groups according to the standards of the Polish National Institute of Public Health. Detailed information on the composition of the food groups is provided in the Supplementary Material (Table S1) and has been published previously [20].

Briefly, the mean daily mass of food intake was higher in men (2310 ± 351 g) than in women (2139 ± 264 g), although the difference was not statistically significant. In both groups, the diet consisted mainly of vegetables and beverages. Other major food groups included fruits, cereals, and meat and meat products. A substantial proportion of consumed beverages was black tea. Among vegetables and fruits, tomatoes, carrots, apples, and bananas were consumed in the highest quantities. The intake of vegetables and fruits was higher in women (384.1 ± 45.2 g and 227.2 ± 35.6 g, respectively) than in men (343.8 ± 38.4 g and 192.2 ± 26.2 g, respectively).

Based on the obtained data, representative daily diets were reconstructed separately for women and men, considering the division into 12 major food groups. Food products were purchased from local markets in the Lublin region, and various commercial brands were included to reflect typical consumer choices.

Each food group was reconstructed in three independent replicates. Considering two gender-based diet groups, 12 food groups, three replicates, and three analytical repetitions, a total of 216 samples were subjected to analytical determinations. None of the food groups was excluded from any analysis.

3. Extraction and Preparation of Analytical Samples

Reconstructed food groups were weighed and homogenized. Samples were extracted with methanol at a ratio of 1:1 (w/v). The resulting extracts were filtered through tissue paper filters (type 388; Munktell, Falun, Sweden), and the residues were re-extracted under identical conditions. This extraction procedure was repeated three times, and all obtained extracts were combined.

Water and beverage samples constituted an exception. These were measured as 100 mL aliquots and evaporated under reduced pressure at low temperature ($T < 37^\circ\text{C}$) using a vacuum evaporator (Ingos RVO 400, Prague, Czech Republic). The residues were subsequently dissolved in a water-methanol mixture (1:1, v/v) to a final volume of 50 mL.

4. Determination of Antioxidant Activity

4.1. Determination of Total Phenolic Content (TPC)

Antioxidant activity of the obtained extracts was assessed using two *in vitro* methods. The total phenolic content (TPC) was determined using the Folin-Ciocalteu assay, which is commonly applied to estimate the total reducing capacity of samples [21].

Briefly, 0.5 mL of each extract was mixed with 30 mL of ultrapure water and 2.5 mL of Folin-Ciocalteu reagent. After 1–8 minutes, 7.5 mL of a 20% (w/v) sodium carbonate solution was added. The mixture was adjusted to a final volume of 50 mL with ultrapure water and incubated for 2 h at room temperature in the dark. Absorbance was measured at 760 nm using a UV-VIS spectrophotometer (Evolution

300, Thermo Fisher Scientific, Waltham, MA, USA). Ultra-pure water served as a blank.

Calibration curves were prepared using gallic acid solutions in the concentration range of 50-500 mg/L. Results were expressed as gallic acid equivalents (mg GAE/L).

4.2. DPPH Radical Scavenging Assay

Antioxidant activity was also determined using the DPPH radical scavenging assay. A freshly prepared DPPH solution (60 µmol/L) was used. In brief, 3.9 mL of DPPH solution was mixed with 0.1 mL of the extract. The mixture was incubated for 30 minutes at room temperature in the dark, and absorbance was measured at 515 nm. Methanol was used as a blank.

The percentage of DPPH radical scavenging was calculated using the following equation:

$$\text{DPPH scavenging (\%)} = [(A_0 - A_1)/A_0] \times 100$$

where A_0 is the absorbance of the control solution and A_1 is the absorbance of the sample.

Additionally, Trolox Equivalent Antioxidant Capacity (TEAC) was determined using Trolox calibration curves (0.1-1.0 mmol/L). Results were expressed as mmol Trolox equivalents per liter.

5. LC-MS Analysis of Polyphenolic Compounds

Prior to chromatographic analysis, sample pH was adjusted to 3.2 using citric acid. The samples were then filtered through 0.45 µm syringe filters (Cronus, Gloucester, UK).

LC-MS analyses were performed using a time-of-flight mass spectrometer (Agilent G3250AA LC-MSD TOF, Agilent Technologies, Santa Clara, CA, USA) coupled with an Agilent 1200 Series liquid chromatograph equipped with an autosampler, degasser, diode array detector (DAD), and binary pump. Separation was achieved on a Zorbax RP-18 column (150 × 2.1 mm, 3.5 µm; Agilent Technologies).

The mobile phase consisted of water with 0.1% formic acid (solvent A) acetonitrile containing 0.1% formic acid (solvent B). Detailed chromatographic and mass spectrometric conditions are presented in Table 1.

Table 1. LC-MS operating conditions

Parameter	Setting
Injection volume	10 µL
Flow rate	0.2 mL/min
Post-run time	4 min
Ionization mode	Negative
Ion source	Electrospray ionization (ESI)
Capillary voltage	4000 V
Fragmentor (Slicer) voltage	150-225 V
Skimmer voltage	65 V
Nebulizer pressure	130 psi
Drying gas flow	10 L/min
Drying gas temperature	350°C
Column thermostat temperature	25°C
Mass range	m/z 100-1000

Identification of gallic acid, quercetin, EGCG, and rutin was based on retention times and mass spectra compared with authentic standards. Quantification was performed using calibration curves derived from standard solutions, based on peak area measurements. Dietary intake of each compound was calculated by combining analytical concentrations with food consumption data obtained from dietary questionnaires.

6. Statistical Analysis

Quantitative data are presented as mean values with standard deviation (SD). Differences between groups were analyzed using two-way analysis of variance (ANOVA) followed by Tukey's post hoc test. A p-value < 0.05 was considered statistically significant. Statistical analyses were performed using Statistica software (version 13.3, StatSoft, Poland).

RESULTS AND DISCUSSION

1. Total Phenolic Content (TPC)

The Folin-Ciocalteu method enabled the determination of total phenolic content in all analyzed food groups, separately for women and men. Although this assay is widely used to estimate the total phenolic content expressed as gallic acid equivalents (GAE), it should be noted that the obtained values more accurately reflect the overall reducing capacity of the sample rather than the absolute concentration of phenolic compounds [21].

Figure 1 presents the daily intake of polyphenolic compounds from major food groups, determined using the Folin-Ciocalteu method and expressed as gallic acid equivalents.

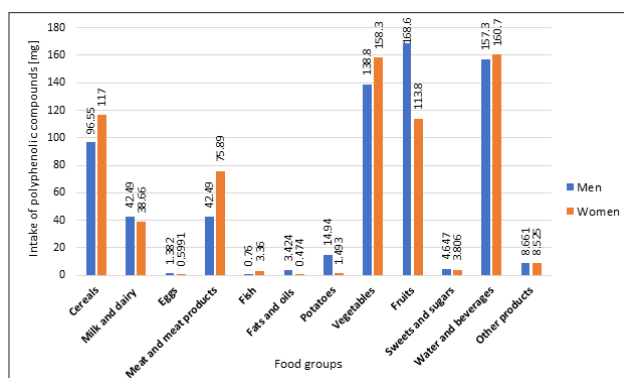


Figure 1. Daily average intake of polyphenolic compounds expressed as gallic acid equivalents – distribution across major food groups

The average daily intake of polyphenols in the women's diet was 682.6 mg and was slightly higher than that observed in men (680.04 mg). When calculated per 100 g of diet, the polyphenol content also differed between genders, amounting to 31.91 mg for women and 29.44 mg for men.

Previous analyses performed by Koch *et al.* reported higher total polyphenol intake from daily food rations, with greater intake observed in men compared to women; however, 1 g of the female diet contained higher concentrations of these compounds, indicating a greater density of phytochemicals in women's diets [22]. Similar observations were reported by Grosso *et al.* [23]. Importantly, in the study

by Grosso *et al.*, polyphenol consumption was estimated by matching questionnaire data with the Phenol-Explorer database. In contrast, the present study and the earlier work by Koch *et al.* relied on direct analytical measurements, providing more realistic estimates of dietary polyphenol intake.

The polyphenol content of food products depends on numerous factors, including climate, light exposure, maturity stage, as well as food processing and preparation. Culinary practices such as abrasion, peeling, and leaf removal may lead to substantial losses of flavonoids, particularly in products where these compounds are concentrated in peels or leaves [23]. Analytical methods account for these variables, unlike database-based estimations, which may oversimplify intake calculations.

Most published studies focus on selected polyphenolic compounds and limited food categories, often relying on U.S.-based databases to estimate flavonoid intake [24,25]. In contrast, the present study considered all major food categories, including those with low polyphenol content. The obtained results illustrate the contribution of individual food groups to total dietary polyphenol intake and antioxidant activity. These data are summarized in Table 2.

Among women, beverages constituted the richest source of dietary polyphenols (160.7 mg/day), primarily due to frequent consumption of tea, particularly black tea, which is abundant in simple and condensed catechins [26,27]. Significant amounts of polyphenols were also supplied by vegetables, fruits, and cereals. Vegetables ranked second among women, contributing 158.3 mg/day.

In men, fruits were the leading source of polyphenols (168.6 mg/day), followed by beverages (157.3 mg/day). Cereals, known to be rich in phenolic acids, also represented an important source, providing 117.0 mg/day in women and 96.55 mg/day in men.

These findings are consistent with previous reports identifying beverages, cereals, fruits, and vegetables as major contributors to dietary polyphenol intake. A study conducted among Finnish adults indicated coffee as the primary source, followed by cereals, tea, and fruits [28]. Similarly, Grosso *et*

al. reported the highest polyphenol intake from beverages, especially tea and coffee [23]. In contrast, Wilczyńska and Retel observed vegetables and fruits as the main contributors, accounting for over 50% of total intake, while coffee and tea contributed only 9.5% [29].

Although cereals are generally considered a minor source of polyphenols, their consumption – particularly of grain-based products – contributes mainly to gallic acid intake. Despite the high consumption of potatoes in Poland [30,31], this food group did not emerge as a significant source of polyphenols in the present study.

2. Antiradical Scavenging Activity (DPPH Assay)

The DPPH radical scavenging assay was used to evaluate the antioxidant activity of individual food groups. As expected, antioxidant activity was strongly correlated with polyphenol concentrations determined using the Folin-Ciocalteu method. Water and beverages (dominated by tea, accounting for over 60% of consumed liquids), vegetables, and fruits exhibited the highest antioxidant activity.

Due to similar activity levels observed among these food groups, samples were diluted to enhance differentiation. This approach revealed that fruits in men and water and beverages in women exhibited the highest antioxidant potential. Interestingly, the food group classified as “other products,” which included items consumed in very small amounts (e.g., nuts, almonds, tomato concentrate), also demonstrated high antioxidant activity. However, owing to their low consumption, these products contributed minimally to overall dietary polyphenol intake.

Animal-derived products, including eggs, meat and meat products, fats and oils, fish, milk, and dairy products, exhibited negligible antioxidant activity, reflecting their minimal polyphenol content. Trace amounts of polyphenols detected in these products are typically associated with plant-derived additives used during industrial processing [32].

In addition to percentage inhibition, antioxidant activity was expressed as Trolox Equivalent Antioxidant Capacity (TEAC) [33]. Food groups devoid of polyphenols or

Table 2. Total phenolic content (TPC) and antioxidant activity of major food groups consumed by women (W) and men (M)

Study group	Cereals	Eggs	Fats and oils	Fish	Fruits	Meat and meat products	Milk and dairy	Other products	Potatoes	Sweets and sugars	Vegetables	Water and beverages
Total Phenolic Content (TPC) [mg/L]												
M	339.0 ±11.16 ^a	30.37 ±4.076 ^a	136.4 ±11.54 ^{bc}	38.00 ±6.666 ^a	877.4 ±91.07 ^g	399.0 ±16.24 ^{ef}	182.91 ±9.031 ^{cd}	470.7 ±6.263 ^f	181.11 ±9.465 ^{cd}	67.25 ±5.139 ^{ab}	403.7 ±10.71 ^{ef}	220.6 ±11.31 ^d
W	522.033 ±30.656 ^d	32.92 ±3.13 ^a	33.86 ±8.083 ^a	38.79 ±4.686 ^a	500.8 ±46.35 ^d	395.8 ±10.22 ^c	139.6 ±4.67 ^e	247.1 ±6.016 ^b	29.80 ±4.423 ^a	61.00 ±2.62 ^a	412.02 ±20.92 ^c	251.6 ±7.74 ^b
Trolox Equivalent Antioxidant Capacity (TEAC) [mmol/L]												
M	0.343 ±0.02 ^c				0.591 ±0.008 ^d	0.276 ±0.001 ^b		0.672 ±0.028 ^e	0.154 ±0.004 ^a	0.135 ±0.001 ^a	0.592 ±0.008 ^d	0.596 ±0.007 ^d
W	0.31 ±0.02 ^b				0.622 ±0.047 ^c	0.187 ±0.034 ^a		0.559 ±0.01 ^c	0.172 ±0.041 ^a	0.596 ±0.009 ^c	0.609 ±0.002 ^c	
TEAC after dilution [mmol/L]												
M					0.523 ±0.028 ^c			0.347 ±0.01 ^b			0.244 ±0.013 ^a	0.324 ±0.009 ^b
W					0.349 ±0.03 ^b			0.28 ±0.009 ^a			0.261 ±0.015 ^a	0.36 ±0.006 ^b
DPPH radical scavenging activity [% inhibition]												
M	49.30 ±3.06 ^f	2.17 ±0.435 ^a	11.47 ±0.07 ^b	6.84 ±1.021 ^{ab}	81.7 ±0.104 ^g	40.05 ±0.076 ^e	6.88 ±0.01 ^{ab}	80.893 ±4.433 ^g	22.6 ±0.675 ^d	16.353 ±0.52 ^c	81.757 ±0.2 ^g	82.09 ±0.25 ^g
W	44.78 ±2.961 ^f	1.447 ±0.145 ^a	3.18 ±0.145 ^{ab}	6.32 ±0.315 ^{bc}	82.36 ±0.964 ^g	26.41 ±3.601 ^e	8.927 ±0.144 ^c	78.81 ±0.936 ^g	6.91 ±0.397 ^{bc}	17.52 ±0.433 ^d	81.88 ±0.458 ^g	82.88 ±0.19 ^g

DPPH – 2,2-diphenyl-1-picrylhydrazyl

Data are presented as mean ± standard deviation (SD)

Values within the same row followed by different superscript letters (a-g) are significantly different ($p < 0.05$)

Underlined values within the same variable indicate significant differences between men and women

containing them only in trace amounts (e.g., eggs, fats and oils, fish, milk and dairy products) were excluded from TEAC analysis. The highest TEAC values were observed in fruits among men (0.523 ± 0.028 mmol/L) and in water and beverages among women (0.360 ± 0.006 mmol/L).

Notably, the “other food products” group also exhibited substantial antioxidant capacity (0.347 ± 0.010 mmol/L in men and 0.280 ± 0.009 mmol/L in women), exceeding that of vegetables (0.244 ± 0.013 mmol/L in men and 0.261 ± 0.015 mmol/L in women). Overall, TEAC values correlated well with TPC results in both groups. The only exception was cereals: despite lower TPC in men (339.0 ± 11.16 mg/L) than in women (522.03 ± 30.66 mg/L), higher antioxidant activity was observed in men (TEAC 0.343 ± 0.02 mmol/L vs. 0.310 ± 0.02 mmol/L in women). These differences likely reflect variations in the qualitative composition of cereal products rather than total consumption.

3. LC-MS Analysis of Selected Polyphenols

High-performance liquid chromatography coupled with mass spectrometry (LC-MS) enabled the separation, identification, and quantitative determination of four major phenolic compounds present in all analyzed food groups: epigallocatechin gallate (EGCG), gallic acid (GA), quercetin, and rutin.

Gallic acid was the predominant phenolic compound, with total daily intakes of 14.87 ± 1.30 mg/day in men and 10.51 ± 1.16 mg/day in women. Water and beverages – primarily black tea infusions – were the main contributors, accounting for approximately 90% of total GA intake. These findings are consistent with the study by Ovaskainen *et al.*, which identified phenolic acids as the dominant polyphenol group in Finnish adults [28].

Water and beverages were also the primary source of EGCG, reflecting the high content of this compound in tea. However, EGCG intake was considerably lower than GA, amounting to 1.007 ± 0.070 mg/day in men and 1.181 ± 0.018 mg/day in women. Overall, the present results align with previous studies demonstrating that beverages

contribute the largest share of polyphenols to the daily diet [28,34,35].

Quercetin was the second most abundant polyphenol after gallic acid. Daily intake was estimated at 12.76 ± 0.77 mg/day in men and 6.64 ± 1.08 mg/day in women. Fruits constituted the primary source, accounting for 77% of total intake in men and 65% in women. Although vegetables were consumed in larger quantities than fruits, fruits exhibited higher antioxidant activity, likely due to their elevated quercetin content.

Vegetables also contributed significantly to quercetin intake, albeit to a lesser extent than fruits. Notably, many vegetables were consumed in processed forms (e.g., soups and salads), which may reduce polyphenol content. In contrast, fruits are typically consumed fresh and often with peels, where polyphenols are concentrated. Thermal processing may further contribute to flavonoid degradation and reduced antioxidant potential.

Quercetin was previously identified as a major dietary flavonoid in our earlier study [22], although reported intake levels were higher (>30 mg/person/day) than those observed in the present research. De Vries *et al.* similarly reported quercetin as a dominant dietary phenolic, accounting for 68-73% of total polyphenol intake [36].

Mean dietary intake of rutin was higher in women (5.85 ± 0.47 mg/person/day) than in men (4.41 ± 0.54 mg/person/day). Water and beverages were the principal source in both genders, contributing 70-80% of total rutin intake. As rutin is a glycoside of quercetin (quercetin-3-O-rhamnoglucoside), it may undergo hydrolysis during food storage or extraction, which could explain the relatively high quercetin intake from fruits despite lower total flavonoid content.

Dietary intake of rutin from fruits and vegetables was also notable but substantially lower than that from beverages. In previous work by Koch *et al.* [22], rutin intake was not quantified. Data on dietary rutin intake remain limited; however, animal studies have demonstrated that high-dose rutin supplementation significantly enhances antioxidant enzyme activity and reduces oxidative stress markers

Table 3. Dietary intake of selected polyphenols from major food product groups in women (W) and men (M)

Study group	Cereals	Eggs	Fats and oils	Fish	Fruits	Meat and meat products	Milk and dairy	Other products	Potatoes	Sweets and sugars	Vegetables	Water and beverages	Total intake [mg/person/day]
Polyphenols: Epigallocatechin gallate EGCG [mg]													
M					<u>0.19</u> $\pm 0.02^c$			<u>0.017</u> $\pm 0.001^a$		<u>0.06</u> $\pm 0.007^{ab}$	<u>0.1</u> $\pm 0.01^b$	<u>0.64</u> $\pm 0.05^d$	<u>1.007</u> ± 0.07
W					<u>0.24</u> $\pm 0.03^c$			<u>0.011</u> $\pm 0.001^a$		<u>0.08</u> $\pm 0.005^b$	<u>0.09</u> $\pm 0.009^b$	<u>0.76</u> $\pm 0.035^d$	<u>1.181</u> ± 0.018
Polyphenols: Gallic acid (GA)													
M	<u>0.18</u> $\pm 0.01^a$				<u>0.44</u> $\pm 0.04^a$			<u>0.12</u> $\pm 0.01^a$	<u>0.14</u> $\pm 0.02^a$		<u>0.59</u> $\pm 0.03^a$	<u>13.4</u> $\pm 1.28^b$	<u>14.87</u> ± 1.3
W	<u>0.12</u> $\pm 0.01^a$				<u>0.42</u> $\pm 0.03^a$	<u>0.11</u> $\pm 0.01^a$		<u>0.064</u> $\pm 0.002^a$	<u>0.19</u> $\pm 0.02^a$	<u>0.04</u> $\pm 0.003^a$	<u>0.41</u> $\pm 0.03^a$	<u>9.16</u> $\pm 1.12^b$	<u>10.51</u> ± 1.161
Polyphenols: Quercetin [mg]													
M	<u>0.004</u> $\pm 0.001^a$				<u>9.84</u> $\pm 1.12^c$			<u>0.03</u> $\pm 0.001^a$	<u>0.005</u> $\pm 0.001^a$		<u>2.85</u> $\pm 0.35^b$	<u>0.03</u> $\pm 0.002^a$	<u>12.76</u> ± 0.77
W	<u>0.008</u> $\pm 0.001^a$				<u>4.32</u> $\pm 0.62^c$			<u>0.014</u> $\pm 0.002^a$	<u>0.04</u> $\pm 0.008^a$		<u>2.227</u> $\pm 0.446^b$	<u>0.03</u> $\pm 0.003^a$	<u>6.639</u> ± 1.075
Polyphenols: Rutin [mg]													
M	<u>0.11</u> $\pm 0.01^{ab}$				<u>0.53</u> $\pm 0.06^b$	<u>0.009</u> $\pm 0.001^a$		<u>0.04</u> $\pm 0.003^a$	<u>0.01</u> $\pm 0.001^a$		<u>0.52</u> $\pm 0.07^b$	<u>3.19</u> $\pm 0.42^c$	<u>4.409</u> ± 0.536
W	<u>0.1</u> $\pm 0.01^a$				<u>0.68</u> $\pm 0.07^b$	<u>0.036</u> $\pm 0.046^a$	<u>0.01</u> $\pm 0.001^a$	<u>0.023</u> $\pm 0.002^a$	<u>0.05</u> $\pm 0.006^a$		<u>0.2</u> $\pm 0.02^{ab}$	<u>4.75</u> $\pm 0.52^c$	<u>5.849</u> ± 0.474

Data are presented as mean \pm standard deviation (SD)

Values within the same row followed by different superscript letters (a-c) are significantly different ($p < 0.05$)

Underlined values indicate significant differences between men and women within the same polyphenol and food group

[37,38]. Despite these promising effects, such supplementation levels greatly exceed those achievable through a typical mixed diet.

A human intervention study involving supplementation with 500 mg of rutin in healthy women demonstrated increased plasma concentrations of quercetin, kaempferol, and isorhamnetin, without significant changes in oxidative stress biomarkers, suggesting extensive metabolic transformation of rutin during absorption [39]. This observation underscores the broader conclusion that, although flavonoids exhibit strong antioxidant activity *in vitro*, their *in vivo* effects are often less pronounced and more difficult to demonstrate.

CONCLUSIONS

The conducted experiments revealed that beverages, fruits, and vegetables were the food groups contributing most substantially to the total daily intake of polyphenols. Although the individual structure of consumption determines the primary dietary sources of polyphenols, it was clearly demonstrated that only foods of plant origin constitute significant contributors to polyphenol intake. Total daily polyphenol intake, polyphenol concentration per 100 g of diet, and antiradical activity measured using the DPPH assay were slightly higher in women than in men; however, these differences were not statistically significant.

LC-MS analysis demonstrated that gallic acid was the predominant polyphenol in the diets of both women and men, followed by quercetin, rutin, and epigallocatechin gallate. The study also confirmed that total phenolic content values determined using the Folin-Ciocalteu method are not fully consistent with results obtained from chromatographic analysis. This observation supports previous reports indicating that the Folin-Ciocalteu assay primarily reflects the overall reducing capacity of the sample and is therefore more suitable for estimating antioxidant potential rather than precise phenolic content. Consequently, only a weak correlation between spectrophotometric and chromatographic results was observed.

The present study has certain limitations. The most important is that only four polyphenolic compounds were identified and quantified using the applied chromatographic method. Undoubtedly, crude extracts obtained from major food groups contain a much broader spectrum of polyphenolic compounds. Therefore, further studies are required to develop and optimize analytical methods enabling comprehensive determination of a wider range of polyphenols in daily food rations.

Despite these limitations, the conducted research allowed for a preliminary assessment of dietary polyphenol intake from major food groups using spectrophotometric techniques and for the development of a chromatographic approach that enables the identification and quantification of selected polyphenols consumed in the highest amounts with the diet.

ORCID iDs

Joanna Tokarczyk <https://orcid.org/0009-0002-3330-1224>

Marcin Czop <https://orcid.org/0000-0002-8113-5841> Wojciech

Koch <https://orcid.org/0000-0001-8749-9657>

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