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Investigation of the antioxidant potential of black oat, rye and wheat cereals through multi-response extraction optimization with different solvents

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ABSTRACT. Cereals possess functional and nutritional properties already consolidated in the literature; they are also an excellent source of health-promoting bioactive compounds. This work aimed to determine the best solvents to extract phenolic compounds from black oat, rye, and wheat through a simplex centroid design, using pure solvents and binary and ternary mixtures. The response variables were the total phenolic compounds (TPC) and the antioxidants (DPPH, ABTS, FRAP) were quantified. For optimized extract, the phenolic compounds were identified by UHPLC. An optimization study for the recovery of antioxidant compounds from these cereals to obtain extracts with better antioxidant properties is reported. The water and acetone binary mixture extracted 18, 49, and 110% more than water and 3.2, 4.0, and 5.5 times more than TPC acetone for black oat, rye, and wheat, respectively. Chromatography identified that rye has the highest number of phenolic compounds, including vanillic acid (3008.64 μg g^{-1}), ellagic acid 352.05 μg g^{-1}, hesperetin 24.33 μg g^{-1}, and formononetin. To conclude, the binary mixture of water and acetone was the best condition to obtain a maximized extract for the analyses in the optimized proportions of solvents for the extract are as follows: 0.52/0.48 for oats, 0.46/0.54 rye and 0.33/0.67 for wheat, respectively. This study optimizes the time and improves quality in measuring antioxidants, both for cereals and derivatives, and for evaluating the potential of new products.

Keywords: Mixture design; phenolic profile; ABTS; DPPH; FRAP.

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Introduction

Cereals are the basis of human food, providing 50% of the calories and proteins of the diet to the world population. Besides the benefits mentioned above, these grains are an excellent source of health-promoting bioactive compounds (Wrigley, 2017; Zamaratskaia et al., 2021).

Among these compounds are those with antioxidant properties that aid in oxidative damage to cellular components such as membranes, proteins, and nucleic acids; therefore, they reduce the rate of cell death and, consequently, the effects of aging and related diseases (Leyane et al., 2022; Masisi et al., 2016).

Antioxidants in cereals are sensitive to several factors besides being specific to certain solvents (Kaur et al., 2019). Choosing an effective and appropriate method of extraction is essential to characterize and quantify active compounds. Many factors can influence the extraction process, including the properties of the matrix, solvent, temperature, pressure, applied time, and proportion of solvent/matrix (Hernandez et al., 2009; Ivanović et al., 2020).

Oats (*Avena sativa*), rye (*Secale cereale*), and wheat (*Triticum* spp) are among the most consumed cereals. They are the agricultural products widely used in food and feed, with potential applications in other sectors, such as the cosmetic and pharmaceutical industries, due to their functional, nutritional, and bioactive properties (Hadidi et al., 2023; Wrigley, 2017).

As mentioned, the dosage of antioxidant compounds in foods depends on the extraction solvents employed. Moreover, although there is a diversity of studies about oats, rye, and wheat (Masisi et al., 2016), the literature presents a gap concerning the identification of the best extraction condition to determine the bioactive compounds of these grains. Given the importance of the characterization of bioactive compounds

from cereals, this work aimed to evaluate the influence of the synergy between solvents (water, acetone, and methanol) on the extraction of phenolic compounds through a simplex-centroid design to understand which is the best extraction condition to obtain bioactive compounds from black oats, rye, and wheat.

Material and methods

For this study, three samples were analyzed: rye, cultivar IPR 89, category S2, donated by COPERCAMPOS, located in Santa Catarina State, Brazil (latitude -27.34237, longitude -51.21059); black oat, Embrapa cultivar; and wheat cultivar Supera, donated by the Alvorada Farm, located in Paraná State, Brazil (latitude -25.15767, longitude -50.53942). The initial cereals moisture (wet base) was: 14.33% for rye, 12.65% for black oat, and 14.45% for wheat.

Simplex-centroid design

This technique is well-used due to its efficient design. In this case, the goal was to understand from three solvents (water, acetone and methanol) which pure solvent or mixture would be more efficient for extracting antioxidant compounds. The design used pure solvents and binary or ternary mixtures (Table 1).

Table 1. Simplex-centroid planning for the extraction of antioxidant compounds from raw Black oat, Rye and Wheat.

Run	Water	Acetone	Methanol
1	1	0	0
2	0	1	0
3	0	0	1
4	1/2	1/2	0
5	1/2	0	1/2
6	0	1/2	1/2
7	2/3	1/6	1/6
8	1/6	2/3	1/6
9	1/6	1/6	2/3
10	1/3	1/3	1/3

Special cubic regression models were used since the coefficients of determination (R^2) were higher than 70% (Boscariol Rasera et al., 2019). The equation representing these models is described below (Equation 1).

$$Yi = \sum_{i=1}^{q} \beta i\, Xi + \sum \sum_{i<j}^{q} \beta ij\, XiXj + \sum \sum_{i<j>k}^{q} \sum \beta ijkXiXjXk \quad (1)$$

Where Y_i is the response predicted by the model; q is the number of independent variables in the assay; X *i, X j, X k* ' indicate the encoded independent variables; β_i, β_{ij} and β_{ijk} represent the regression coefficients for each linear effect term, the terms of a binary and ternary interaction effect, respectively. The software Statistica® 10 was used in the analysis.

Extraction of antioxidant compounds

The solvents and techniques followed similar studies with mustard seeds and lentils (Boscariol Rasera et al., 2019; Đorđević et al., 2021; Nguyen et al., 2023; Yeo & Shahidi, 2015; 2017). Water, acetone and methanol were used, and their mixtures, is shown in Table 1. The grains were ground (Marconi knife grinder, MA 630/01/Brazil) and mixed with solvent in a ratio of 1:4 (grain: solvent - w/v). Mixtures were kept under agitation (150 rpm) for 20 min. at 25º and centrifuged (17.000 rpm) for 15 min., and the supernatant was collected and stored in an amber vial at −18°C for further analysis.

Evaluation of antioxidant compounds

Total phenolic content (TPC)

The TPC of the raw materials was determined by the Folin-Ciocalteu method (Singleton & Rossi, 1965). In volumetric balloons of 10 mL, 0.2 mL of each extract was mixed with 0.5 mL of Folin-Ciocalteu reagent (2N) and 5 mL of distilled water. The solutions were stirred with the help of a vortex (Scientific Industries, G-560) for 1 min. Following, 2 mL of 15% Na_2CO_5 solution was added to the solutions and stirred again for 30 seconds. The flasks were filled with water and kept in the dark for 120 min. Subsequently, samples were analyzed in a UV-VIS spectrophotometer (Q898DPT, Quimis, São Paulo). The readings were performed at 760 nm, expressing the results in mgGAE 100 g^{-1} dry sample.

ABTS$^{\bullet+}$ radical cation scavenging activity

The ABTS$^{\bullet+}$ radical assay was performed according to the methodology adapted (Re et al., 1999). The ABTS$^{\bullet+}$ saline solution (7 mM) was prepared by diluting 0.0392 g of ABTS in 10 mL of sodium acetate buffer (20 mmol L^{1}; pH 4.5). Potassium persulfate was weighed (0.37845 g) and diluted in 10 mL of sodium acetate buffer (20 mmol L^{1}; pH 4.5). In the 10 mL of ABTS$^{\bullet+}$ solution, 176 µL of potassium persulfate was added and kept in the dark at room temperature for 16h to allow complete radical generation. After this period, sodium acetate buffer (80 mM) was used to adjust the absorbance of the ABTS$^{\bullet+}$ reagent. In 30 µL of the sample, 3 mL of ABTS$^{\bullet+}$ reagent was added, kept in the dark for 30 min., and absorbance was measured at 734 nm. The results were expressed in µM TE 100 g^{1} (µmol Trolox 100 g^{-1} dry sample).

DPPH radical scavenging activity

Antioxidant activity was determined by adapting the method 22-diphenyl-1-picrylhydrazyl (DPPH) proposed by Brand-Williams et al. (1995). 50 µL of the extract and 1.95 mL of DPPH solution were added to test tubes. The tubes were stirred in a vortex (NA 3600, Norte Científica, Brazil) and stored in the dark for 30 min. The measure was at 517 nm and the results were expressed in µM TE 100 g^{1} (µmol Trolox 100 g^{1} of dry sample).

Ferric Reducing Antioxidant Power (FRAP)

This technique determines the reduction of iron in biological fluids and aqueous solutions of pure compounds. The method consisted of adding 100 µL of the extract and 3400 µL of FRAP reagent in test tubes and 100 µL of the solvent used in the extract with 3400 µL of FRAP reagent. The samples were heated to 37°C for 30 min. in a water bath. Subsequently, samples were read in a UV-VIS spectrophotometer (Q898DPT, Quimis, São Paulo) at 593 nm. The results were expressed in µM TE 100 g^{-1} (µmol Trolox 100 g^{1} of dry sample) (Benzie & Strain, 1996).

Optimization of process conditions

The desirability methodology consists of transforming each response *Y*i into an individual desirability (*di*), which indicates the proximity of the adjusted value of the response in relation to the ideal configurations of the factors. The individual desirability functions of the significant responses are combined according to Equation (2) to obtain the global desirability (D) (Derringer & Suich, 1980):

$$D = \sqrt[k]{d_1 d_2 \dots d_k} \quad (2)$$

where *k* represents the number of responses considered. With $0 \leqslant D \leqslant 1$, a high value of D suggests that all *di* values are close to the target value, which is the optimal solution for the system.

Quantification of phenolic compounds by UHPLC-DAD

The phenolic compounds present in the optimized extracts were identified and quantified using an ultra-high pressure liquid chromatography (UHPLC) (Waters, Milford, MA, USA) equipped with a DAD diode array detector, a quaternary pump and BEH C18 column (2.1 mm × 50 mm x 17 µm) (Waters, Milford, MA, USA) and an automatic sampler. The mobile phase consisted of A (0.1% formic acid in ultrapure water) and B (0.1% formic acid in methanol), with a flow rate of 0.35 mL $min.^{-1}$. The elution gradient applied was 0 min. - 0% B, 8 min. – 20% B, 15 min. - 100% B, 18 min. - 0% B, and isocratic elution with 0% B at 20 min. (Melo et al., 2022; Turola Barbi et al., 2018). The standards used for the identification of phenolic compounds were as follows: gallic acid, 3,4- dihydroxybenzoic acid, theobromine, catechin, 2,4- dihydroxybenzoic acid, vanillic acid, theophylline, quinine, chlorogenic acid, salicylic acid, p-coumaric acid, m-coumaric acid, o-coumaric acid, ellagic acid, hesperidin, formononetin. The phenolic acid and flavonoid contents were quantified using external calibration curves, expressed in µg g^{-1}. All curves were constructed by varying the concentration from 1 to 50 µg g^{-1} and analyzed in triplicate.

Statistical analysis

The data were analyzed expressed as the mean ± standard deviation of the values, compared by Tukey's test ($p < 0.05$). Plotting and desirability plots were performed using Statistica® 10 software. The most appropriate condition determined in the mixture design was replicated experimentally to confirm the validity of the models.

Results and discussion

Optimization of the extraction of antioxidant compounds from cereals

Antioxidant compounds are substances of high interest for the beneficial health effects and minimization of chronic and degenerative diseases caused by oxidative stress (Boscariol Rasera et al., 2019). Bearing in mind that grains are one of the primary sources of these compounds, it is crucial to investigate the best extraction medium for further measurement since the extract yield can be affected by solvent chemistry, extraction time and temperature, among others.

For the study of the bioactive properties of a material, more than a single method is required, because of each technique has its specific actions and can present different results in the same sample (Melo et al., 2022). Therefore, this study chose to verify the antioxidant potential of cereals by three methods in addition to the solvent mixtures used.

Finding the most appropriate solvent is essential to obtain significant bioactive compounds since the complexity of the composition of plant matrices directly influences the result (Ivanović et al., 2020). Through simplex centroid design, it was possible to obtain the data of each run, its graphs of each analysis (Figure 1), the best extraction point evaluating all analyses (Figure 2), as well as, equations and adjustments for each analysis (Table 2).

As observed in Figure 1, each solvent and its mixtures directly influence the result of soluble antioxidants in the mix. The TPC (Figure 1) ranged from 35.50 to 126.12 mgEAG 100 g-1 for black oat, 50.63 to 200.95 mgEAG 100 g-1 for rye, and 31.93 to 176.53 mgEAG 100 g-1 for wheat. Rye presented the highest values among the highest concentrations, followed by wheat and black oat. The binary mixture of 1/2 water and 1/2 acetone had the best effect on extraction for rye and wheat. The ternary mixture of 2/3 water,1/6 acetone and 1/6 methanol had the best result for black oat.

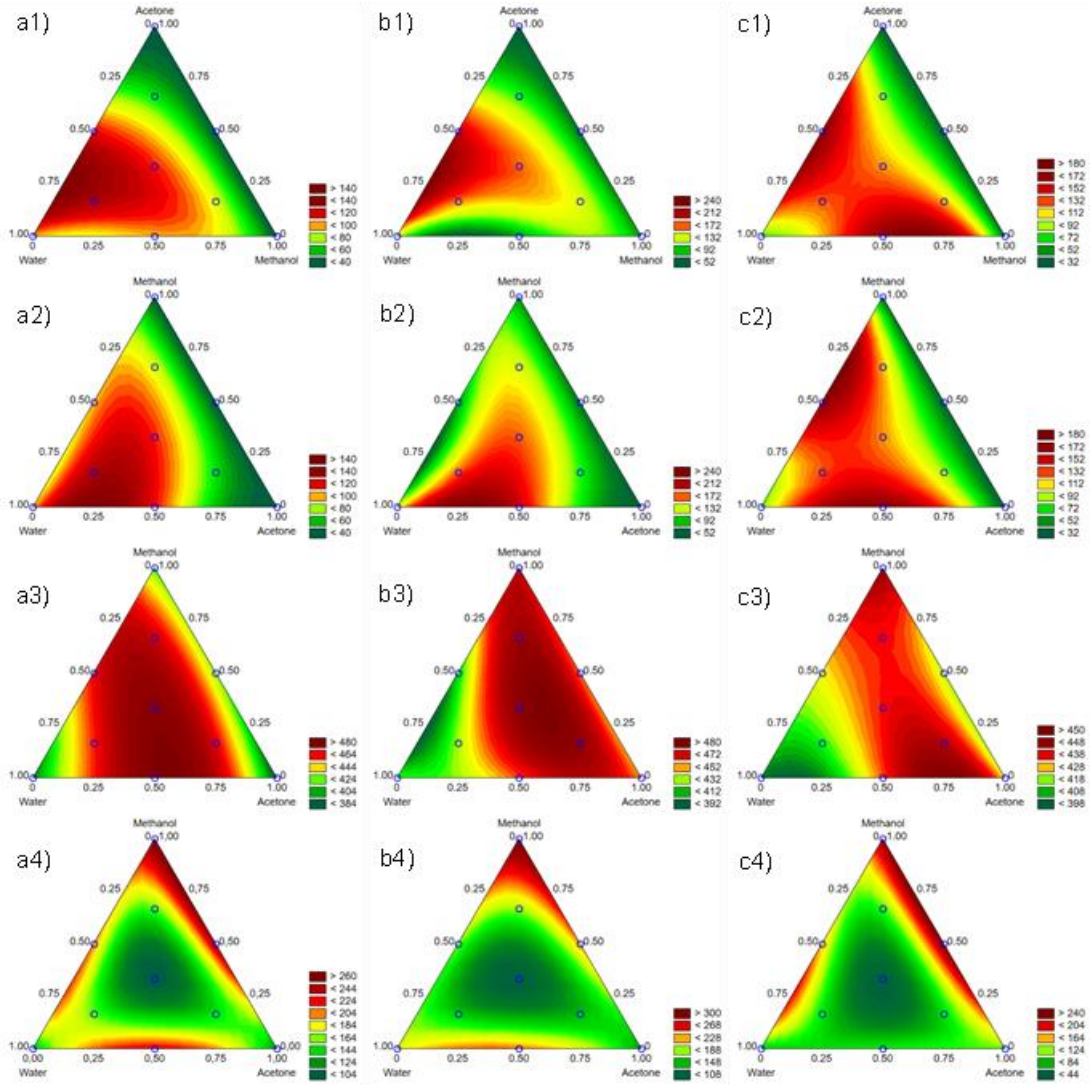

Figure 1. Contour charts for total phenolic compounds (TPC) (1), ABTS (2), DPPH (3) and FRAP (4) for raw Black oat (a), Rye (b) and Wheat (c).

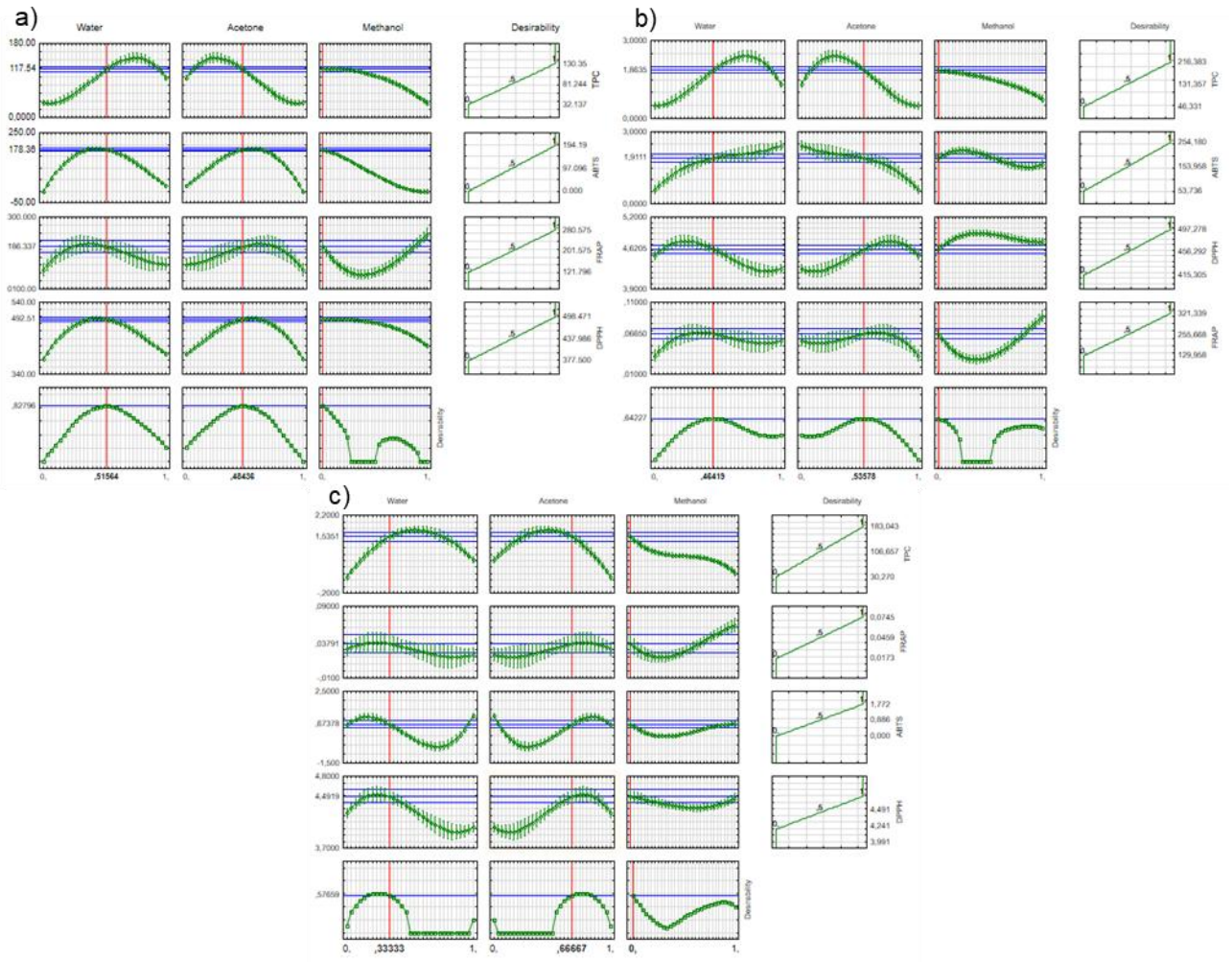

Figure 2. The desirability of the best extract for raw Black oat (a), Rye (b) and Wheat (c).

Table 2. Mathematical models and coefficient of determination of antioxidant analyses for raw Black Oat grains, Rye and Wheat.

Sample	Adjusted model equation	R2
	TPC	
Black oat	97.17x+37.28y+36.42z+188.18xy+117.72xz+548.84xyz+301.99(xy(x-y))-178.99(xz(x-z))	0.9808
Rye	133.93x+51.60y+78.04z+424.99xy-104.67xz+1266.36xyz+504.77(xy(x-y))-532.25xz(x-z)	0.9678
Wheat	82.60x+29.97y+41.82z+471.87xy+438.39xz-865.61xyz-427.12(xz(x-z))	0.9831
	FRAP	
Black oat	134.30x+123.39y+267.57z+390.33xy+187.98yz-3781.87xyz+460.39xz(x-z)	0.8773
Rye	201.43x+139.17y+317.18z+262.03xy-301.04xz-2939.14xyz	0.8747
Wheat	90.48x+116.33y+228.54z+283.60yz-3565.55xyz+737.83xz(x-z)	0.8617
	ABTS	
Black oat	22.95x-0.51y+4.51z+674.47xy+559.25xz-984.53xyz-231.91(xy(x-y))+675.77(xz(x-z))	0.9937
Rye	235.99x+62.17y+168.47z+165.35xy-430.52xz-118.92yz+2862.81xyz	0.9328
Wheat	113.51x+68.12y+72.62z-378.59xy+285.29xz-2474.00xyz-942.70(xy(x-y))+376.63(xz(x-z))	0.9643
	DPPH	
Black oat	395.68x+383.08y+418.92z+416.02xy+224.49xz+96.23yz+236.90xyz-123.77(xy(x-y))-174.17(xz(x-z))	0.9880
Rye	426.37x+450.85y+475.33z+77.30xy-139.02xz-35.42yz+1299.22xyz-186.72(xy(x-y))-244.78(xz(x-z))	0.9630
Wheat	402.37x+424.25y+445.96z+75.37xy-61.40yz+244.24xyz-193.10(xy(x-y))	0.8488

x = water, y = acetone and z = methanol

In the inhibition of ABTS and FRAP radicals, the maximum values were for rye, followed by black oat and DPPH. The black oat and rye obtained the same maximum value, and wheat showed a lower concentration. For the elimination of FRAP radical (Figure 1 - a4, b4, c4), the ternary mixtures presented the worst results; while pure methanol was better for black oat and rye, wheat had a better affinity with the binary mixture of acetone and methanol. The grains behaved differently by eliminating the ABTS radical (Figure 1- a2, b2, c2). However, the binary mixtures involving water were better for black oat and wheat, while rye had a better

extraction with pure water. The DPPH technique presented similar graphs for black oat and rye, although the results did not varied significantly between them.

The polarity of the extraction solvents influences the recovery of the content of antioxidant compounds. Phytochemical compounds with greater polar character are extracted with water (Uba et al., 2022). As is the case with acetone, less polar solvents have an affinity with low-density compounds (Palaiogiannis et al., 2023). The mixture of water with other solvents positively influenced the extraction of antioxidant compounds, this type of characterization in cereals is interesting and attractive.

Several processes, such as cooking, fermentation and germination, use the immersion of cereals in water, which can be discarded after processing. As water is a green solvent and can be reused in several processes, the values found for bioactive compounds in this solvent should be highlighted because, when benefiting a cereal with immersion, the residual water can be used to add to other products to enrich it.

With a significance level of 95%, the proposed models are reliable since all variance analysis (ANOVA) models presented *p-value* less than 0.05. The coefficient of determination (R^2) indicates that the models explain 84.2 -99.4% of the variability of the experimental data (Table 2).

Desirability is a response that depends on the control variables, that is, the optimization of the results to obtain a maximized response. When evaluating the responses of TPC and elimination of ABTS, DPPH and FRAP radicals (Figure 2), it can be concluded that the binary mixture of water: acetone is the most efficient for fresh grains. The ratio of solvents to the optimized extract is: 0.52 water:0.48 acetone, and 0.46 water:0.54 acetone, and 0.33 water:0.67 acetone for black oat, rye and wheat, respectively (Table 3).

Table 3. Predicted and experimental values of antioxidant analyses of raw Black oat grains, Rye and Wheat in optimized extract.

Sample	Predicted ^{Value}	Experimental value	Deviation (predicted/experimental)
		TPC (mgGAE 100 g^{-1})	
Black oat	095.61^{A}	128.42^{c} ± 13.34	0.74
Rye	225.37^{A}	197.45^{abc} ± 14.91	1.14
Wheat	076.62^{A}	162.39^{bc} ± 15.02	0.47
		ABTS (µmolET 100 $g^{-1)}$	
Black oat	177.72 ^{A}	197.17^{aA} ± 27.38	0.90
Rye	183.20^{A}	208.29$^{a\ A}$ ± 25.94	0.88
Wheat	069.04 ^{A}	155.91$^{a\ A}$ ± 0.32	4.32
		DPPH (µmolET 100 $g^{-1)}$)	
Black oat	492.23 ^{A}	472.96^{abA} ± 32.53	1.04
Rye	462.50 ^{A}	518.03^{abA} ± 42.00	0.89
Wheat	448.03 ^{A}	422.09^{abA} ± 10.77	1.06
		FRAP (µmolET 100 g^{-1})	
Black oat	226.49 ^{A}	237.22^{aA} ± 12.77	0.95
Rye	232.90 ^{A}	227.11^{aA} ± 10.99	1.28
Wheat	107.72 ^{A}	102.87^{bA} ± 10.83	1.05

Averages ± standard deviation. Averages followed by the same letters did not differ significantly by the Tukey's test (5%), being lowercase for rows and uppercase for columns.

The deviation between the predicted and experimental values of the optimized extracts was, on average, 1.23, with wheat presenting the most significant deviations (1.72), followed by rye (1.04) and black oat (0.91). Regarding the analyses, minor deviations were achieved in the ABTS, followed by FRAP, DPPH and the TPC. As the most significant deviation was under 4.32, the model prediction is satisfactory.

This study also allowed obtaining an optimized extract for the several characterizations of antioxidant compounds and the model (Table 3), where it can be verified that the experimental values do not differ statistically from those predicted. Another highlight is that rye presented the best results in all analyses with these extracts. At the same time, black oat does not differ from rye in the elimination of FRAP, ABTS and DPPH radicals, wheat does not differ from black oat in TPC and the elimination of DDPH radical in rye does not differ only in TPC, as well as other analyses, wheat is lower than other cereals.

The optimized black oat extract used the ratio of 0.52:0.48 (water: acetone), and the synergy of these solvents extracted amounts higher than tetrahydrofuran, with 95.61 mgGAE 100 g^{-1} for TPC and 69.04 µmolET 100 $g^{-1)}$ for ABTS analysis (Deng et al., 2012). It was also better than the mixture of acetone, water and acetic acid (0.700:0.295:0.005), obtained with mean values of 128.42 mgGAE 100 g^{-1} for TPC and 472.96 µmolET 100 g^{-1} for DDPH analysis (Rao et al., 2019).

The mixture of 0.46:0.54 (water: acetone) in the rye ensured a good yield of phenolic compounds, which extraction in pure water obtained values between 180.63 - 209.00 mgGAE 100 g^{-1} for TPC and methanol 80%, with

values of 103 mgGAE100 g^{-1} for TPC, but methanol 80% was more efficient when assessing the retention capacity of ABTS (13.00 µmolET 100 g^{-1}) and DPPH (12.17 µmolET 100 g^{-1}) (Mishra et al., 2017; Ragaee et al., 2006).

The best extract for wheat in this study was the ratio of 0.67:0.33 (water: acetone), the synergy of this mixture was better than 80% methanol (0. 50 mgGAE 100 g^{-1}), ethanol 80% (0. 52 - 1.03 mgGAE 100 g^{-1}), ethanol (0. 11-0.37 mg GAE 100 g^{-1}) and acetone 50% (0. 4 - 0. 8 mg GAE 100 g^{-1}) in TPC analysis. Methanol 80% (4. 33 µmolET 100 g^{-1}) was similar to the mixture of this study for the analysis of DPPH and superior for that of ABTS (8. 3 µmolET 100 g^{-1}) (Barros Santos et al., 2019; Luo et al., 2014; Moore et al., 2005; Ragaee et al., 2006; Yu et al., 2002).

In this case, the extraction of antioxidant compounds from black oat, rye and wheat showed that the combination of water and acetone was the most appropriate solution because of the synergy of this mixture which can maximize the extraction of the compounds for the analysis of TPC, ABTS, DPPH and FRAP.

Determination of phenolic compounds by UHPLC-DAD

The intake of whole grains and their flours increases the consumption of antioxidant compounds compared to refined flours. Among the patterns used, it was possible to identify 3 bioactive compounds in black oat, while in rye and wheat were found 4 and 2, respectfully (Table 4).

Table 4. Phenolic compound profile of optimized extracts of raw Black oat, Rye and Wheat.

Compounds	Detection limit (ng g^{-1})	Phenolic and flavonoid acids (µ g^{-1})		
		Black oat	Rye	Wheat
Vanillic acid	17.80	Nd	3008.64 ± 0.29	Nd
Chlorogenic acid	11.22	0057.92 ± 0.88	Nd	Nd
Ellagic acid	22.21	Nd	0352.05^{ab} ± 1.53	31.29^{ab} ± 0.13
Hesperidin	42.05	015.44^{ab} ± 0.23	024.33^{ab} ± 0.02	Nd
Formononetin	64.44	295.10^{ab} ± 1.46	0159.69^{ab} ± 0.56	89.26^{abc} ± 1.01

Average ± standard deviation. Means followed by the same letters in the lines did not differ significantly by the Tukey's test (5%). nd: not detected.

The compound most present in black oat and wheat, but also in the rye in a large amount was formononetin. This substance presents anti-cancer, antioxidant, anti-inflammatory and cardioprotective biological activities and inhibitory effects against enzymes (Machado Dutra et al., 2021). Rye has the highest concentration of vanillic acid, which has anti-cancer, antiobesity, antidiabetic, antibacterial, and anti-inflammatory, and is used as a flavoring agent in several food products (Kaur et al., 2022). Chlorogenic acid detected only in black oat is also found in green coffee and tea extracts; it is biologically active and has antioxidant, antibacterial, hepatoprotective, cardioprotective, anti-inflammatory, neuroprotective, antiobesity, antiviral, antimicrobial, antihypertensive extracts, central nervous system stimulator and can modulate lipid metabolism and glucose in disorders related to genetic and health metabolism (Naveed et al., 2018; Yanagimoto et al., 2022), strengthening the nickname of "healthy food by nature" given to this cereal.

The ellagic acid, also present in rye, presents similarities such as chlorogenic acid, is found in several plants and vegetables and has antioxidant activity, the ability to reduce the human lipid profile and metabolism, as well as alter pro-inflammatory mediators thus affecting metabolic syndrome and diabetes. It also includes antibacterial, antifungal, antiviral, anti-inflammatory, hepatic and cardioprotective, neuroprotective, antidiabetic, gastroprotective, anti-hyperlipidemic and antidepressant activities, among others (Evtyugin et al., 2020). On the other hand, hesperidin belongs to the class of flavonoids, having the common antioxidant and anti-inflammatory capacity of bioactive compounds, cardioprotective and anticancer effects, as well as indicating an improvement in aerobic performance, which is essential mainly for athletes (Martínez-Noguera et al., 2019).

Conclusion

The study to optimize the solvent for extracting antioxidant compounds was critical for properly combining each plant material. After evaluation, the binary mixture of water and acetone was chosen through simplex centroid design as the best solvent mixture for all three cereals. This study serves as a comparative basis for measuring how the processing of these cereals can influence the resultant amount of these important compounds, as well as understanding potential new products and utilization of the residues of these matrices. Other studies that measured the antioxidant compounds of cereals with other solvents showed lower or

similar values to this study. To this extent, it is understood that the diversity of composition of each cereal requires individually designed and optimized extraction studies of bioactive compounds by solvents, quantifying and valuing each matrix.

The bioactive compounds found in cereals have essential features that help combat comorbidity, improving the quality of life of those who consume them. These results can support future research on fresh and processed cereals since this is the first study that reports the optimization of the extraction of antioxidant compounds from black oat, rye, and wheat. In addition, it encourages the consumption of whole grains, as they have antioxidant activity and potential biological function.

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