Food Sciences & Applied Microbiology Reports
Vol. 1(1), 2022, 49-58
ISSN (Online) 2958-0684
https://finessepublishing.com/fsamr

Characterization of dietary fiber extracted from different oats (Avena Sativa. L) cultivars with respect to physicochemical, functional and phytochemicals

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Abstract: Five indigenous oat cultivars were grown under the standard agronomic practices. Dietary fiber from the selected oat cultivars was assessed for its various physicochemical, functional, and phytochemical potential. The results showed that selected cultivars varied significantly ($P \leqslant 0.05$) for dietary fiber contents. SGD81 has shown the highest values for TDF (16.153±0.5273), IDF (11.23±0.38), and SDF (4.95±0.18). The lowest fraction of TDF was shown by the Avon (11.727±0.77), IDF by PD2LV65 (7.42±0.38), and SDF by PD2LV65 (3.87±0.31), respectively. Oat cultivar SGD81 showed maximum WS, WHC, and OHC due to its higher dietary fiber content. Dietary fiber from selected oat cultivar SGD81 is also loaded with phytochemicals, including TPC, TFC, TFOC, and anthocyanin. The selected cultivars varied significantly ($P \leqslant 0.05$) with respect to functional ingredients. The highest fraction of Phenolic acid was shown by the SGD81, SGD2011, PD2LV65, S2000, and Avon, respectively. It is evident from the study that dietary fiber extracted from SGD81 is the best and most economical alternative to expensive commercial fibers.
Keywords: physicochemical characterization, mineral profile, functional properties, phytochemical quantification, color tonality

1. Introduction

Oat (Avena sativa L) is a rabi crop that is now broadly cultivated in all areas of Pakistan for livestock rearing. Punjab province is the 2nd largest contributor of agricultural production, having 16.68 Mha under agricultural production. Fodder crop occupies 2.05 Mha with a production of 45.97 metric tons. Oats are a major contributor among fodder crops and occupy about 35 percent (Ahmad et al., 2014).

In developing countries, value addition could be achieved through appropriate mechanization, post-harvest management, and value addition. Exploration of indigenous economic resources for various bioactive compounds is a matter of higher interest for scientists in the present scenario. Plant-based diets are now preferred by users for their potential health benefits (Alasmre & Alotaibi, 2020). Oat predominately cultivated as a forage crop. Its grains are still unexplored for its value added benefits. Dietary fiber is an important functional ingredient found in cereal grains. Oats have a maximum fraction of dietary fiber, so it has much room for their value addition. Cereals are a rich source of dietary fiber. Urbanization and economic growth have increased the use of energy-dense food, with more refined carbohydrates and a low dietary fiber fraction. The use of refined and milled products from grain is the major cause of this negative transition. This resulted in reduced intake of DF, photochemicals, and micronutrients, which impart various physiological functions in

[Received 19 Mar 2022; Accepted 21 June 2022; Published (online) 27 June 2022]

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DOI:10.61363/fsamr.v1i1.20

the body. The cell wall from the outer part of the grain is loaded with these functional moieties. The structure and composition of the cell walls of rye and wheat are more similar than in the case of barley, corn, and oats (Bach Knudsen et al., 2017).

Dietary fiber consists of NSP, RS, oligosaccharides, and lignin. The main NSPs in cereals are arabinoxylan (AX), SDF, and cellulose. These are major components of the cell wall, along with other polysaccharides, phenolic acid, and proteins (Izydorczyk & Dexter, 2008). The main NSP in cereals is AX, followed by cellulose and β-glucan. Corn grain contains SDF concentrations up to 0.1%, in wheat and rye, it ranged from 0.7-1.7%, and in oat, it ranged from 2.8-4.1% (Bach Knudsen et al., 2017). Dietary fiber is an NSP, not digestible by monogastric animals. Dietary fiber, when ingested, imparts various physiological and biological functions (Soliman, 2019). Dietary fiber has gained importance for its various potential health benefits. Dietary fiber was investigated for its quantification, physiological effect, and disease preventive and management perspectives, including hypotensive, anti-obesity, hypocholesterolemic, and anti-diabetic, having satiety function by delaying gastric emptying. A soluble fraction of dietary fiber produces a hypocholesterolemic effect by reducing total cholesterol and LDL (Dahl & Stewart, 2015). Dietary fiber plays a potential role in the management and prevention of various metabolic disorders, such as hypertension, diabetes, and cardiovascular diseases (Ötles & Ozgoz, 2014). Dietary fiber in ample quantity in food regulates gut microbiota (Carlson et al., 2018; Holscher, 2017) and attenuates the insulin sensitivity of the cell. A low-fiber diet promotes the activity and growth of colonic mucus-degrading bacteria and aggravates the risk of colitis by the enteric pathogen H. pylori (Desai et al., 2016).

"Inadequate" dietary fiber intake may lead to an increased incidence of metabolic disorders (Das et al., 2020). Dietary fiber has a wide range of applications in the food, feed, and nutraceutical industries. Different processing operations alter the physicochemical properties of the dietary fiber; hence, improving its biological activity (Dhingra et al., 2012). The physiological and biological potential of dietary fiber has gained the attention of researchers, processors, and consumers to be used as a food fortification ingredient. Dietary fiber fortification in food products lowers their energy value by substituting fat and sugar, making the final product healthier and more attractive (Limdi et al., 2016; Rodríguez-García et al., 2013).

This, when in a foodstuff, also alters the physical, functional, and structural properties of the final product, such as WHC, perishability, OHC, ES, FC, hydration, viscosity, texture, and sensory characteristics (Elleuch et al., 2011). Both fractions of dietary fiber, including soluble and insoluble fractions, have significant physiological functions. Physical properties, including surface area and pore size, define functional properties of the dietary fiber. The surface area enhances the absorption ability of dietary fiber in the gastrointestinal tract, hence producing health benefits. Phenolic contents are affected by the cultivars, biotic and abiotic factors, including UV radiation, herbicide application, fertilizer application, physiological stress, temperature fluctuation, and low iron contents, etc. (Kovacova & MaliNoVá, 2007).

Phenolic acid and its derivatives impart an anti-inflammatory role by inhibiting superoxide (Lee et al., 2008). Phenolic acids have anti-proliferative action even at low concentrations (Kampa et al., 2003). Anthocyanin is a water-soluble reddish to purple flavonoid primarily located in the aleuron layer of the grain and is equipped with potential health benefits like antioxidant activity, anti-inflammatory, anticancer, and hypoglycemic effects. These are widely used as colorants in various food products as functional ingredients. Potential health benefits associated with anthocyanins include anti-oxidative, anti-carcinogenic, hypoglycemic, anti-obesity, neurogenerative, retinal regulation, hypolipidemic, hepatoprotective, and anti-aging (Zhu, 2018). It is evident from the study that dietary fiber from oat is an economical alternative of commercial expensive fibers, ultimately has a wide range of applications in various industries, including baking, confectionery, beverages, and nutraceutical.

2. Experimentation

2.1. Oat cultivars

Dietary fiber from indigenous oat cultivars (SGD81, SGD2011, PD2LV65, Avon, and S2000) was analyzed for proximate, mineral profile, total phenol contents, total flavonoid contents, total flavones contents, radical scavenging activity, anthocyanin, total dietary fiber, soluble dietary fiber, insoluble dietary fiber, and color tonality.

2.2. Functional properties of dietary fiber from different cultivars

2.3. WHC-Water- and OHC- Oil-holding capacity

WHC of dietary fiber was determined as described by Raghavendra et al. (2004). OHC of dietary fiber was determined as per the method described by Zhang et al. (2009).

2.4. WS- Water solubility

WS was determined according to Zhang et al. (2009). One gram of fiber sample was mixed with 50 mL of distilled water. 1 g of fiber was mixed with 50 mL of distilled water and placed on a magnetic stirrer for thirty minutes. After stirring, the sample was centrifuged for fifteen min at 4500 rpm. The residue was weighed and dried to a constant weight. Water solubility was determined by following the equation WS (g/g) = Weight of dried sample / Weight of sample

2.5. Physico-chemical profiling of Oat Extract

Physico-chemical characterization of dietary fiber from five indigenous oat cultivars was done using the following protocols. All the results were recorded on a dry weight basis.

2.6. Proximate Composition

Moisture content of dietary fiber from selected oat cultivars was determined as narrated in method No. 44-15A AACC (2000). Protein contents were determined by Kjeldahl's method as indicated by the AACC (2000) method No. 46-30. Soxhelt extraction was done for fat determination of fiber as described in method No. 30-25 AACC (2000). Fiber contents were quantified by acid digestion and neutralization by using alkali according to method No. 32-10. Ash content was estimated as given in method No. 0801. Chemists (2000).

2.6.1. Mineral Profiling of Oat Flour Extract

DF was analyzed for micronutrients, including iron, zinc, copper, and manganese, using a spectrophotometer (U-2800, Hitachi, USA). All cultivar samples were ashed as per method No. 0801 AACC (2000).

2.6.2. Dietary fiber contents of fiber extracted from different oat cultivars

Dietary fiber from selected oat cultivars was characterized for its TDF, IDF, and SDF fractions by the gravimetric method; Megazyme Kit was used for the determination of three fiber fractions in fiber samples extracted from different cultivars. TDF was determined according to the Committee (2000) method No.32-05. SDF was determined according to the AACC (2000) method No. 32-07, and IDF was determined according to AACC (2000) Method No. 32-20.

2.6.3. Preparation of Extract

An extract of dietary fiber was prepared by dissolving the fiber in a 1:10 ratio of methanol, as narrated by Stankovic (2011).

2.7. TPC-Total Phenolic Contents

TPC of dietary fiber was determined as described by Stankovic (2011).

2.8. TFC-Total Flavonoid Contents

TFC of dietary fiber was determined by the method of Nongalleima et al. (2017).

2.9. TFoC-Total Flavonols Contents

Total Flavonols of dietary fiber from selected oat cultivars were determined by the procedure described by Miliauskas et al. (2004). 0.5 mL of AlCl3 (20%) and1.5mL of CH3COONa (10%) were added to 0.5 mL of the fiber. Samples were kept in the dark for 2.5 h, and then absorbance was measured at 440 nm using a spectrophotometer. TFoC were quantified by following the equation and were taken as mg of quercetin equivalent. TFoC (mg of Quercetin equivalent) = (Sample absorbance +0.006)/0.091

2.10. Total Anthocyanin

Total anthocyanin of dietary fiber from selected oat cultivars was determined as described by Lee et al. (2008). 0.4M solution of CH3COONa (pH 4) and 0.025M solution of KCl (pH 1) were prepared. 200 microliters of the extract was taken into the glass cuvette, and 2 mL of both reagents was added to a separate cuvette. Absorbance

of the sample was taken by the spectrophotometer at 510 and 700 nm, respectively. The result was expressed as mg of Cyanidine 3-glucoside kg-1 of fiber (mg C3G kg-1).

2.11. DPPH Radical Scavenging Activity

DPPH of the dietary fiber from selected oat cultivars was determined as described by the (Afify et al., 2012).

2.12. Determination of Phenolic Acids

HPLC for quantification of Phenolic acids was determined as described by Peanparkdee et al. (2018).

2.13. Color tonality of Oat Fiber

Color tonality of dietary fiber from selected oat cultivars was determined by the method of Rocha and Morais (2003) with the use of Konica MINOLTA Chroma meter-410 (Sensing Inc., Tokyo, Japan), with reference to illuminant D65 and a visual angle of 10°, using the CIE Lab system.

2.14. Statistical Analysis

The data was analyzed through one-way ANOVA using Statistix 8.1 software. Significantly different treatment means were separated through LSD ($P \leqslant 0.05$) (Steel & Torrie, 1981). The graphical presentation was done using Origin Prom 2016.

3. Results and Analysis

3.1. Functional properties of fiber extracted from different oat cultivars

Hydrocolloid water absorption, solubility, and oil holding capacity are potential features that define its ultimate industrial application. The results of WS, WHC, and OHC are shown in Table 2. All the cultivars vary significantly ($p \leqslant 0.05$) with respect to WS, WHC, and OHC. The highest values for WS (1.24±0.06), WHC (1.46±0.051), and OHC (1.97±0.10) were shown by the cultivar SGD81 due to the highest fraction of dietary fiber. The lowest values for WS (0.86±0.06bc), WHC (1.10±0.10), and OHC (0.96±0.12) were shown by the S2000. Water holding capacity of the dietary fiber attenuates the viscosity of the mix and ultimately prevents final products from shrinking (Elleuch et al., 2011). Dietary fiber fortification in food, especially high-fat products and emulsions, enhances the oil-holding capacity of the mix, hence stabilizes the end product (Lv et al., 2017). (Hydrophobic properties, surface area, and charge density of particles define their OHC (Chang et al., 2011).

3.2. Proximate of fiber extracted from different oat cultivars

The proximate composition of the fiber is shown in Figure 1. The highest fat, moisture, ash, fiber, and NFE were shown by PD2LV65 (1.34±0.21), Avon (5.31±0.25, SGD81 (83.20±0.446), PD2LV65 (35.04±0.32), and S2000 (2.93±.55), respectively. The lowest fraction of fat, moisture, ash, fiber, and NFE was shown by the SGD81 (1.03 ±0.03), Avon (3.42±0.40), SGD81 (11.85±0.17), PD2Lv65 (59.32±0.44), and SGD81 (0.78±0.72), respectively.The cultivars varied significantly ($\leqslant 0.05$) for moisture, ash, and fiber contents, while non-significant ($\leqslant 0.05$) differences existed among the cultivars for NFE and fat contents. Protein contents were not detected in all samples. In fiber, it is considered an impurity and removed by the manipulation of Sonication and pH. Proximate analysis showed that the explored cultivars are also a good source of minerals, along with fiber content.

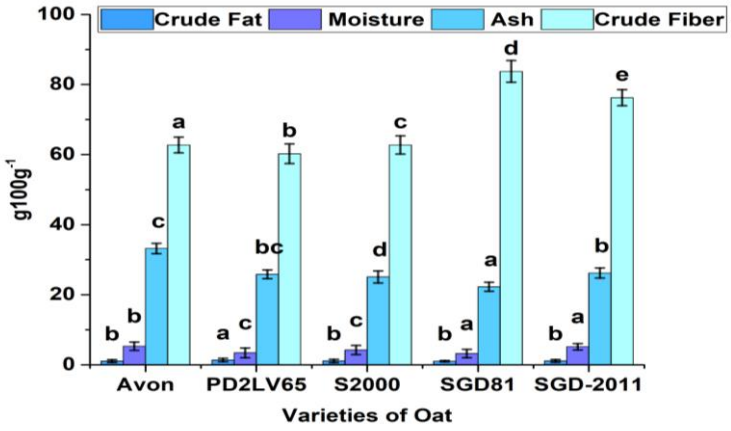

Figure 1. Proximate of the extracted fiber from different oat cultivars

Food Sciences & Applied Microbiology Reports
Vol. 1(1), 2022, 49-58
ISSN (Online) 2958-0684
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3.2.1. Mineral profile of fiber extracted from different oat cultivars

The results of the mineral profile of the fiber are shown in Figure 2. The highest fraction of Cu, Fe, Mn, and Zn was reported in SGD2011 (6.92±0.17), SGD2011 (218.84±0.20), PD2LV65 (39.66±0.51), and Avon (37.08±0.20), respectively. Avon was found to have the lowest in all micronutrient fractions, including Cu (0.99±0.21), Fe (8.13±0.23), Mn (1.29±0.35), and Zn (37.08± 0.20), respectively. All the cultivars are highly significant (⩽0.05) with respect to these micronutrients. Iron and zinc are among the most important micronutrients that play an important role in the regulation of various physiological and biochemical processes. Exploration of economical sources of these moieties will be a step forward in overcoming their deficiency.

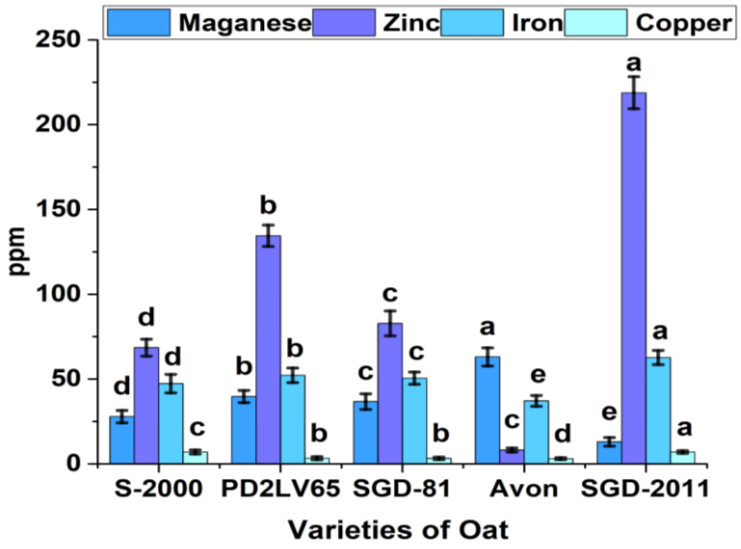

Figure 2. Mineral profile of the extracted fiber from different oat cultivars

Table 1. Phenolic acid contents of extracted fiber mg/100g on DW

Oat cultivars	Vanillic acid mg100g^{-1}	Gallic cid mg100g^{-1}	Caffeic acid mg100g^{-1}	4-Hydroxyphenylacetic acid mg100g-1	Protocatechuic acid mg100g^{-1}	Ferulic acid mg100g^{-1}	p-Coumaric cid mg100g^{-1}	Cinnamic cid mg100g^{-1}
Avon	2.99±0.11b	15.03±0.43e	13.18±0.16b	0.49±0.06d	42.26±0.16d	144.77±0.13c	2.24±0.15c	1.90±0.12c
PD2LV65	4.31±0.25a	16.32±0.28b	14.33±0.22a	0.60±0.02b	44.71±0.07c	145.31±0.25b	3.24±0.12ab	2.03±0.21c
S2000	3.78±1.14b	15.41±0.13d	15.54±0.35b	0.48±0.04c	43.56±0.08a	143.95±0.18a	4.26±0.09bc	2.40±0.17b
SGD81	5.19±0.26a	15.94±0.31c	16.13±0.13bc	0.36±0.03a	45.18±0.07b	146.75±0.19a	4.56±0.33a	3.38±0.27a
SGD2011	3.24±0.13a	16.33±0.07a	16.68±0.20c	0.41±0.03b	44.84±0.06e	147.13±0.15b	3.40±0.15c	1.35±0.13d
LSD(p≤0.05)	0.508	0.42	0.07	0.18	0.347	0.35	0.98	0.35

V1= Avon V2=S-2000 V3= PD_2LV_{65} V4=SGD-81 V5=SGD-2011.

Table 2. Functional properties of dietary fiber from selected oat cultivars

Varieties	WS g/g[1]	WHC g/g[1]	OHC g/g
V1	0.96±0.09bc	1.34±0.06ab	1.67±0.07b
V2	0.86±0.06bc	1.10±0.10ab	0.96±0.12b
V3	0.92±0.06c	1.33±0.05c	1.63±0.04c
V4	1±0.06a	1.23±0.08a	1.56±0.15a
V5	1.24±0.06b	1.46±0.051bc	1.97±0.10b
LSD (p≤0.05)	**0.19**	**0.13**	**0.12**

V1=Avon V2=S-2000 V3=PD2LV65 V4=SGD-81 V5=SGD-2011

3.2.2. Dietary fiber contents of fiber extracted from different oat cultivars

The results regarding total, insoluble, and soluble dietary fiber were shown in Figure 3. All the cultivars vary significantly (⩽0.05) with respect to all dietary fiber fractions, including TDF, IDF, and SDF. SGD81 has shown the highest values for TDF (16.153±0.5273), IDF (11.23±0.38), and SDF (4.95±0.18). The lowest fraction of TDF was shown by the Avon (11.727±0.77), IDF by PD2LV65 (7.42±0.38), and SDF by PD2LV65 (3.87±0.31), respectively. Chawla and Patil (2010) reported that TDF, IDF, and SDF range from 11.8 to 16.4%, 6.5% to 7.0%, and 5.3% to 8.7%, respectively. The difference in Insoluble dietary fiber fractions may be due to cultivars, climate, soil, and agronomic practices. Manthey et al. (1999) studied the oat genotype for its soluble and insoluble dietary fiber fractions and reported that 3.9–5.2% and 6.0–7.1%, respectively.

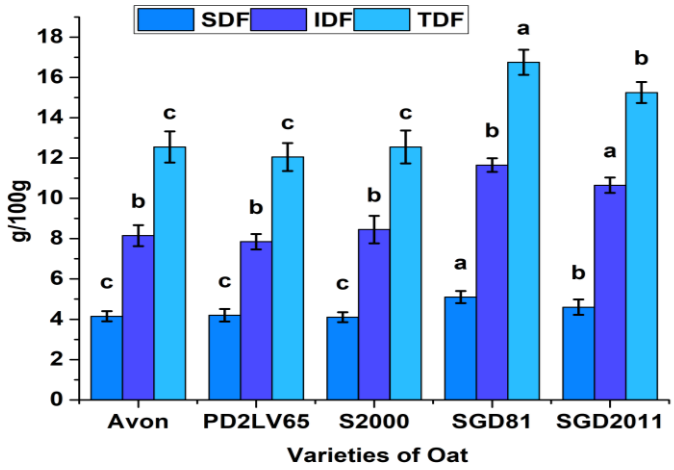

Figure 3. Total dietary fiber, Insoluble dietary fiber, and soluble dietary fiber contents of the extracted fiber from different oat cultivars

3.2.3. TPC, TFC, TFoC, DPPH, and Anthocyanin of fiber extracted from different oat cultivars

The result of TPC, TFC, TFoC, and anthocyanin contents of extracted fiber were shown in fig 4. The highest value of TPC, TFC, TFoC, DPPH, and anthocyanin was reported in SGD81 (125.78±0.2150, SGD81 (1270.2±0.2401), SGD2011 (714.21±0.3201), SGD81 (58.700±0.2000), and SGD2011 (2.9800±0.0800), respectively. The least value of these were reported in S2000 (43.253±0.1450), S2000 (900.16±0.3650), PD2LV65 (558.84±0.2103), SGD2011 (29.213±0.2750) and S2000 (0.6833±0.065). All the cultivars are highly significant (p⩽0.05) with respect to TPC, TFC, TFoC, DPPH, and anthocyanin contents. Ibrahim et al. (2020) reported the TPC (36.07 to 101.56 mg100 g-1), TFC (754.16 to 1147.08 mg100 g-1), TFoC (548.33 to 697.5 mg100 g-1), DPPH (24.33 to 55.88%), and anthocyanin contents (0.5 to 2.87 mg of C3G kg-1), respectively. The difference in the cited study and the reported study value is due to Sonication. This effectively released the bioactive from the flour. Oat flour was sonicated, which resulted in excessive release of these bioactive compounds by the acoustic effect. It is evident from the study that mechanical abrasion is an effective tool to maximize the bioactive recovery as compared to the simple extraction methodology.

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Vol. 1(1), 2022, 49-58
ISSN (Online) 2958-0684
https://finessepublishing.com/fsamr

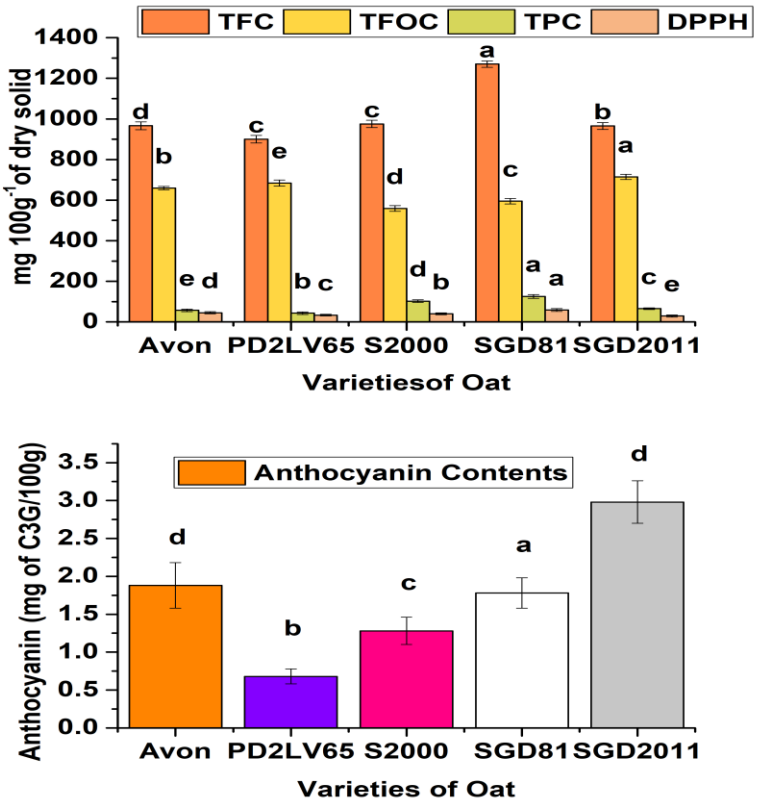

Figure 4. Phytochemical quantification of the extracted fiber from different oat cultivars

3.2.4. Phenolic acids of fiber extracted from different oat cultivars

Results regarding phenolic acid contents of different oat cultivars are shown in Table 1. The results indicated that there is a significant difference among the varieties ($p \leq 0.05$). The highest contents regarding vanillic, Gallic, cafeic, 4-hydroxyphenyl acetic acid, Protocatechuic acid, Ferulic acid, p-cumaric acid and cinnamic acid were shown by SGD81 (5.19±0.26), SGD2011(16.33±0.07), SGD2011(16.68±0.20), PD2LV65(0.60±0.02), SGD81(45.18±0.07), SGD2011(147.13±0.15), SGD81(4.56±0.33) and SGD81(3.38±0.27) respectively. The lowest fraction of these acids was shown by the Avon (2.99±0.11), Avon (15.03±0.43), Avon (13.18±0.16), SGD81 (0.36±0.03), Avon (42.26±0.16), PD2LV65 (143.95±0.18), Avon (2.24±0.15), and SGD2011 (1.35±0.13), respectively.

The results showed fiber from SGD 81and SGD 2011 has more phenolic acids as compared with all others, hence has potential to be used as functional ingredients. The results are consistent with the findings of Kilci and Gocmen (2014), Soycan et al. (2019), and Skrajda-Brdak et al. (2019). Twenty-one genotypes of oat were studied for ferulic and coumaric acids. The results showed that p-cumaric acid was higher than that of ferulic acid. Ferulic acid content varied in the range from 16.50 mg/100 g to 149.36 mg/100 g of grain. P-Coumaric acid ranged between 8.05 mg/100 g and 210.27 mg/100 g of grain.

3.2.5. Color tonality of Oat Fiber extracted from different oat cultivars

Color plays a potential role in the acceptability of foodstuffs by the processor, retailer, and consumer as well. The extracted fiber from all cultivars was checked for its color tonality to have better acceptability by all stakeholders. Fiber from all cultivars varied significantly ($p \leqslant 0.05$) for its L*, a*, b*, c*, and h* coordinates. The results of the color tonality are shown in Figure 5. The highest L* value was shown by the fiber from SGD2011 (80.4±0.23), and Avon showed the highest a* value (3.25±0.03).

The highest b* value was shown by the SGD81 (18.5±0.22). SGD 81 showed the highest c* value (18.653±0.2754) and the highest h* value, also accounted by the SGD2011 (85.137±0.2371). The lowest count for L*, a*, b*, c* and h*was shown by Avon (70.18±0.22), SGD2011 (1.36±0.05), SGD2011 (15.98±0.23), SGD2011 (16.07±0.23) and Avon (79.59±0.09). Hussein et al. (2011) reported that fiber addition imparts a dark color to the finished product.

Pasha et al. (2011) also reported that fiber fortification of food imparts a potential effect on the finished food product and determines its acceptability.

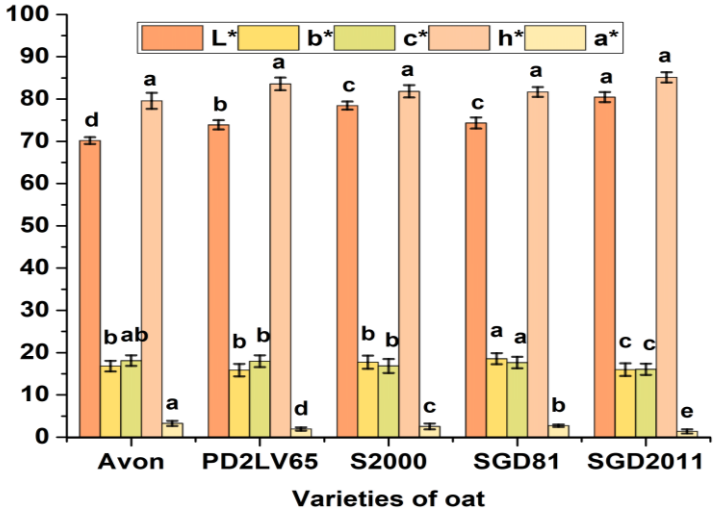

Figure 5. Color tonality of the extracted fiber from different oat cultivars

4. Conclusion

Fiber extracted by Sonication is characterized for its functional and nutraceutical potential. Among the five selected cultivars, SGD81 had the maximum fiber content and was also equipped with a plethora of potential bioactive. SGD81 has a lot of potential for value addition as a functional crop instead of a forage crop. This will also provide an economical fiber source for the various industrial applications. Bioactive compounds from other cultivars may be purified for use in nutraceutical products.

CRediT authorship contribution statement

Muhammad Suhail Ibrahim and Ishtiaq Hassan, Experiment and analyze data, Muhammad Javaid Asad, Maryum Arif, and Muhammad Nadeem, Data analysis, writing original draft, Amir Mumtaz, Ahmed Mujtaba, and Abdul Hafeez, editing and proofreading.

Funding

This work was not financially supported by any funding agency.

Declaration of competing interest

The authors declare no conflict of interest.

Acknowledgements

This manuscript is part of a Ph.D. study at the Institute of Food and Nutritional Sciences, PMAS Arid Agriculture University, Rawalpindi, Pakistan. The authors declare that there is no conflict of interest in the present work. I am thankful to Mr. Amir Mumtaz, Senior Scientific Officer, Food Science Research Institute, NARC, Islamabad, and Muhammad Nadeem, Assistant Professor, Institute of Food and Nutrition, University of Sargodha, Pakistan, for providing me with analytical facilities.

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