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ARTICLE

Impact of Wheatgrass (*Triticum aestivum* L.) Supplementation on Atherogenic Lipoproteins and Menopausal Symptoms in Hyperlipidemic South Asian Women – A Randomized Controlled Study

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ABSTRACT

Objective: The purpose of this study was to evaluate the effect of wheatgrass (*Triticum aestivum* L.) on atherogenic lipoproteins, inflammation, and menopausal symptoms. *Methods*: Fifty-nine hyperlipidemic women were randomized into control ($n = 30$) and intervention groups ($n = 29$). Intervention group was administered 3.5 g of freeze-dried wheatgrass powder in encapsulated form daily for 10 weeks, while the control group received no intervention. *Results*: The intervention group experienced a reduction of 5.4% in total cholesterol (TC), 4.4% in low-density lipoprotein cholesterol, and 9.5% in triacylglycerols (TAG); however, high-density lipoprotein (HDL) also reduced by 6% following 10 weeks of intervention. Compared with the control group, the baseline-adjusted post-intervention levels of TC, TAG, and Apolipoprotein B (Apo B) were significantly lower in the experimental group compared with the control group ($p = 0.043$, 0.045, and 0.016, respectively). Prevalence of menopausal symptoms saw nonsignificant reductions: vasomotor, 42%; somatic, 33%; and psychological, 50%, while urogenital symptoms remained unaltered. *Conclusions*: Wheatgrass supplementation at a dose of 3.5 g per day for a period of 10 weeks results in significant reductions in Apo B fraction, TC, and TAG without significantly reducing the HDL cholesterol.

KEYWORDS
atherogenicity; hyperlipidemic; inflammation; menopausal symptoms; South Asian women; wheatgrass

Introduction

Physiology, etiology, and burden of cardiovascular disease (CVD) differ in women as compared with men. CVD has compound associations with the regulation of sex steroid metabolism and its endocrine effects on other organ systems in women. Therefore, one of the main attributable factors of CVD in women is estrogen withdrawal resulting during the onset of menopause. For the same reason; the manifestation, stage of diagnosis, and treatment of CVD in women tends to differ and has been purported to have underlying links with the menopausal stage in woman (Hart, Charkoudian, & Miller, 2011). For example, clinical presentation of CVD in women is not the same as in men: more women (16%) present with less common symptoms, such as back pain, dyspnea, and indigestion, as compared with men (6%), in whom chest pain is the most common symptom (Bajaj et al., 2016; Maas &

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Appleman, 2010). The body of evidence that exists for possible benefits of therapy for menopausal symptoms on CVD health is not conclusive; at the same time, the line of therapy for CVD is not of any assistance for menopausal symptoms (Dessapt & Gourdy, 2012; Guay et al., 2007).

On the other hand, nutraceutical compounds in many natural functional foods, such as red yeast rice, black cohosh, red ginseng and red clover (Dog, 2005; Geller & Studee, 2005; Leach & Moore, 2012; Lipovac et al., 2012; Kim et al., 2012), have been proposed to exert holistic health benefits, particularly with regard to cardiovascular, metabolic, and oncogenic events. At the same time, intervention with plant products, such as barley grass powder (*Horduem vulgare*) and Panchratna juice (containing *Ocimum sanctum, Emblica officinalis, Curcuma longa, Zingiber officinale*, and *Mentha spicata*), have shown hypolipidemic effects in diabetic subjects (Iyer et al., 2010; Venugopal & Iyer, 2010). One such plant substance that has been hypothesized to contain a plethora of nutraceutical polyphenols, mucopolysacccharides, vitamins, antioxidant enzymes, and generous amounts of antioxidant pigments, is wheatgrass or *Triticum aestivum* L. (Rana, Kamboj, & Gandhi, 2011).

Consumption of wheatgrass has been reported in Egyptian civilizations and as a part of Ayurveda, the ancient system of medicine in India (Shukla & Tripathi, 2005). Western interest in wheatgrass was piqued when Charles Schnabel, an agricultural chemist, patented wheatgrass for its beneficial chlorophyll content (Schnabel, 1934). Popularity of wheatgrass has increased immensely since then, with wheatgrass supplements taking up a sizeable share of the health supplements market globally (QY Research, 2016). However, still there are not enough reported large-scale clinical trials that have investigated the antioxidant benefits, hypoglycemic properties, or hypolipidemic properties of wheatgrass.

Laboratory-based molecular studies, in vitro studies, and animal model trials have reported that fresh wheatgrass has been observed to possess significant antioxidant properties (Kulkarni et al., 2006), decreases oxidative stress and atherogenic indices (Das, Hakim, & Mittal, 2012; Kothari et al., 2011; Sethi et al., 2010), and possesses anti-cancerous (Talas et al., 2008; Wheat & Currie, 2008) and immunosuppressive properties (Hemalatha et al., 2012). However, most of these beneficial effects of wheatgrass, especially on atherogenic indices and menopausal symptoms, have not been corroborated by conducting human trials.

Further, processing in the form of freeze-drying has been shown to preserve the antioxidant capacity of wheatgrass; freeze-dried wheatgrass samples have demonstrated higher Diphenyl Picrylhydrazyl (DPPH) free-radical scavenging activity and total antioxidant capacity compared with fresh and oven-dried wheatgrass, as assessed by Ferric Reducing Ability of Plasma (FRAP) assays (Das, Raychaudhuri, & Chakraborty, 2012). Yet, no in vivo human study has investigated the effects of freeze-dried wheatgrass to date.

Therefore, it is essential to investigate the protective effectiveness of wheatgrass in human model for advancement of the existing knowledge from animal model studies. Consequently, this randomized controlled study was planned with the objective of evaluating the impact of freeze-dried wheatgrass powder supplementation on atherogenic lipoproteins, inflammation, and menopausal symptoms in primary hyperlipidemic women.

Methods

Design

The study was conducted using an open label randomized controlled design, involving an experimental group, which was given wheatgrass powder capsules daily for 10 weeks, and a control group, which was maintained without administering any intervention. Although initially planned as a triple blind placebo-controlled trial, administering a placebo to the control group did not turn out to be feasible because a large proportion of control group subjects did not consent to taking a pill that "may or may not contain the active ingredient for cholesterol reduction," which had to be explained to the subjects while taking their informed consent. The data pertaining to background information, medical history, and dietary and lifestyle habits were collected prior to and after the intervention (pre- and post-intervention data). The study design was approved by the Institutional Ethics Review Committee (No. Fc Sc/FND/ME 158 dated September 30, 2010).

Preparation of treatments

Source of wheatgrass

Grass of the common wheat plant (*Triticum aestivum* L.), which taxonomically belongs to the family *Poaceae*, subfamily *Pooideae*, and tribe *Triticeae*, in powdered form using freeze-drying (subject to dehydration at 0–5°C and ground using cold water jacketing) was procured from AumAgri Freeze Foods, a local exporting firm based in Vadodara, Gujarat, India. The powdered wheatgrass was nitrogen-packed to ensure minimal moisture accumulation and contamination.

Encapsulation

The powdered wheatgrass was encapsulated into 350 mg gelatin capsules of size 0 (courtesy Centurion Laboratories, Vadodara, India). Prepared capsules were hermetically packed in sterile plastic jars, each jar having a capacity to contain 100 capsules.

Sample size estimation

Since no prior studies have been conducted on wheatgrass and its effect on atherogenic lipoproteins, our primary hypothesis (based on aforementioned properties of wheatgrass) was that intervention group would see a 15% reduction in the low-density lipoprotein (LDL) cholesterol levels. Therefore, sample size was calculated with Students t-test to detect one-tailed difference (reduction) in the post-intervention values of LDL in the intervention group compared with the control group, with an effect size of 0.7, 80% power, and 5% alpha error. The computed value came to be 23; to that we added a margin of 20% attrition rate and arrived at a final sample size of 28 per group. The calculations were performed using the software G*Power version 3.1.9.2.

Subjects and sample selection

The subjects were selected from a population of primary hyperlipidemic South Asian women from Gujarat in western India. The potential participants ($n = 78$) were explained the nature and purpose of the research in a language comprehensible to them and also the risks and benefits involved in the research, following which a written consent was obtained from those willing to participate ($n = 69$). The consenting women were subjected to scrutiny using the following inclusion criteria: subjects aged 30–60 years, had total cholesterol levels $>$200 mg/dL, and/or a triacylglycerol level $>$150 mg/dL. Since studying the impact of wheat-grass on menopausal symptoms was one of the secondary objectives, one of the inclusion criteria was the presence of menopausal symptoms. Although menopausal symptoms are related to the menopausal transition, they are not strictly specific to the stage of menopause.

Therefore, while enrolling participants, priority was given to the presence of symptoms themselves, which are indicative of changing endocrinology in the subjects, rather than the menopausal stage categorization, which does not always correspond very accurately to the clinical–biochemical changes in these women. The exclusion criteria were as follows: Subjects who suffered from diabetes, polycystic ovarian syndrome, hypothyroidism, or pituitary disorders; pregnant; or who had been initiated on statins for less than three months before the trial; or who were on hormone replacement therapy (HRT) (or had undergone HRT in the past one year) were not enrolled in the study ($n = 8$). After enrollment of the subjects ($n = 61$), they were randomized into two study groups – control and experimental groups – by following the chit method for equal random allocation into two groups as delineated by Giesbrecht and Gumpertz (2004). Only two subjects from the experimental group left the study, resulting in a total of 29 subjects in the experimental group and 30 subjects in the control group. The reason for leaving was the difficulty to comply with the intervention regimen of taking wheatgrass capsules every day.

Outcome parameters

The main outcome parameters that were studied to assess the impact of the supplementation were the serum lipoprotein fractions: LDL, total cholesterol (TC), triacylglycerols (TAG), very low-density lipoprotein (VLDL) cholesterol, high-density lipoprotein (HDL) cholesterol; apolipoproteins, Apo A and Apo B, and to study atherogenicity. For studying impact on inflammation, the high-sensitivity assay of C-reactive protein (hs-CRP) was studied. The fasting blood sugar (FBS) and hemoglobin (Hb) levels were also studied as secondary outcome parameters. The clinical data included height, which was measured using a non-stretch fiberglass tape to the nearest 0.1 cm, and weight, which was measured using a research grade Salter weighing scale to the nearest 0.1 kg. In this study, overweight was defined as a body mass index (BMI) >23.0 to 26.9 kg/m^2, and obesity was defined as a BMI of ≥ 27 kg/m^2 as per the Asia-Pacific high cut-off points delineated by the World Health Organization (WHO Expert Consultation, 2004). Waist circumference (WC) was also measured using a fiber-glass tape to the nearest 0.1 cm. In this study, increased WC was defined as >80 cm in women and >90 cm in men (WHO, 2000). The blood pressure was measured using a sphygmomanometer with standard procedure; high blood pressure was defined as a systolic blood pressure (SBP) value of 140 mm Hg or more and a diastolic blood pressure (DBP) of 90 mm Hg based on the Joint National Committee (JNC) VII guidelines (Chobhanian et al., 2003). Dietary intake was assessed using 24-hr diet recall method; wherein the subject was asked to remember the last typical day's meal. Recipes were noted down to ascertain information about individual food items. Standard measuring cups were used to enable the participants to indicate the quantities of cooked food consumed, which were extrapolated to raw food amount and subsequently the nutrient content using the standard food and nutrient composition database for foods indigenous to South-Asia region (Gopalan, Rama Shastri, & Balasubramanian, 2007). Information on menopausal symptoms (categorized as was assessed using the Greene Climacteric Scale) (Greene, 1998). Participants were also asked whether they smoked, consumed alcohol, and exercised regularly (defined as at least 30 min of brisk walking, jogging, gym workout, or other moderate-to-intense activity for five days a week).

The biochemical parameters studied included serum TC and TAG, both were estimated using enzymatic methods. Serum HDL was estimated using the cholesterol plus second generation assay for direct measurement of HDL. Serum LDL was estimated directly on

Table 1. Clinical profile of subjects (mean + SD).

Parameter	Control ($n = 30$)	Experimental ($n = 29$)	p value
Age (years)	50 ± 8.9	48 ± 7.6	0.47
Weight (kg)	63.4 ± 10.7	62.5 ± 11.5	0.75
BMI (kg/m^2)	27.2 ± 3.9	25.8 ± 4.0	0.18
WC (cm)	95.9 ± 7.3	93.7 ± 9.8	0.32
SBP (mmHg)	135 ± 15.6	133.4 ± 52.8	0.76
DBP (mmHg)	82 ± 7.4	81.8 ± 12.8	0.86

BMI = body mass index, WC = waist circumference, SBP = systolic blood pressure, DBP = diastolic blood pressure.

an Olympus AU2700 (Beckman Coulter, Tokyo, Japan) analyzer using Rapid Diagnostics' LDL direct measurement kit. Serum VLDL was derived by using the Freidewald formula (Freidewald, Levy, & Fredrickson, 1972). Plasma glucose was measured using the glucokinase method. Serum Apo A and Apo B and hs-CRP were measured using the nephelometric assays.

Statistical analyses

Difference between pre- and post-intervention values was tested using a paired t-test; difference between the values of control and experimental groups was tested using a two-sample t-test. Difference between post-intervention values in the study groups while adjusting for baseline values was computed by running an F-test for analysis of covariance, with pre-intervention values as covariates and post-intervention values as dependent variables. Effect size was ascertained by computing Cohen's d for continuous variables. All reported p values were two-tailed values and considered significant at <0.05. All analyses, except Cohen's d, were performed using SPSS version 23; Cohen's d was computed using G×Power version 3.1.9.2.

Results

The background characteristics revealed that the majority of the subjects were aged 40–60 years in both control and experimental groups (76.7% and 82.7%, respectively). The distribution of subjects based on their menopausal status revealed that women from all three stages of menopause, namely premenopause, peri-menopause, and post-menopause, were equally distributed in both the study groups: 30% premenopausal, 30% peri-menopausal, 27% post-menopausal, and 13% hysterectomized in the control group; and 24% premenopausal, 24% peri-menopausal, 38% post-menopausal, and 10% hysterectomized in the experimental group. The information on anthropometric variables and blood pressure (Table 1) revealed that clinically the subjects in both groups were comparable at the beginning of the study.

The nutrient intake of the subjects (Table 2) was similar across control and experimental groups except the iron intake, which was higher in the experimental group (11.2 g/dL) as compared with the control group (8.9 g/dl), with the difference being statistically significant ($p < 0.05$).

The unadjusted data on all outcome parameters in both control and intervention group are shown in Table 3. Here it can be seen that the pre-intervention (baseline) values between control and experimental groups were significantly different. Following the intervention, TC experienced a 5.4% reduction, LDL experienced a 4.4% reduction, and TAG experienced a 9.5% reduction in the experimental group. However, Apo A also saw 18.8% reduction and

HDL experienced 6% reduction in the experimental group at the end of the intervention period.

Owing to differences in the baseline values between the study groups, the post-intervention values were compared between the two after controlling for baseline values. These results have been illustrated in Table 4. After adjusting for baseline differences, the post-intervention

Table 2. Nutrient intake by subjects (mean + SD).

Nutrient	Control ($n = 30$)	Experimental ($n = 29$)	p value
Energy (kcal)	1523 ± 164	1592 ± 287	0.32
CHO (g)	186 ± 22.6	192 ± 36	0.44
Fat (g)	59 ± 12.6	64 ± 16.8	0.16
Protein (g)	43 ± 10.8	45 ± 13.2	0.34
Iron (mg)	8.9 ± 3.7	11.2 ± 4.6	0.03*
Vitamin C (mg)	70.4 ± 24.1	77.8 ± 18	0.18
β-carotene (µg)	1398 ± 754	1498 ± 833	0.63
TDF (g)	23.8 ± 3.5	23.8 ± 5.7	0.97

*$p < 0.05$. CHO = carbohydrates, TDF = total dietary fiber.

Table 3. Atherogenic and inflammatory parameters of subjects (mean + SD).

	Control ($n = 30$)		Experimental ($n = 29$)	
Parameters	Pre	Post	Pre	Post
TC (mg/dL)	215.6 ± 26.9	217.2 ± 34.2	223.4 ± 28.8	211.5 ± 39.5**
TAG (mg/dL)	138.0 ± 53.9	142.8 ± 54.7	146.4 ± 53.8	132.2 ± 44.7
LDL (mg/dL)	134.8 ± 27.9	136.0 ± 30.8	138.8 ± 22.9	132.6 ± 31.3
HDL (mg/dL)	52.9 ± 8.3	52.2 ± 8.8	56.6 ± 11.4	53.1 ± 10.4*
LDL/HDL	2.6 ± 0.6	2.6 ± 0.6	2.6 ± 0.7	2.5 ± 0.7
Apo A (mg/dL)	139.1 ± 19.8	126.1 ± 17.7	134.4 ± 21.1	109.0 ± 18.4***
Apo B (mg/dL)	111.9 ± 16.6	106.3 ± 17.1	111.1 ± 17.2	96.5 ± 20.5***,#
FBS (mg/dL)	83.4	80.4	85.3	80.9***
hs-CRP (mg/dL)	0.26	0.29	0.30	0.27

*Significantly different from baseline at $p < 0.05$, **$p < 0.01$, ***$p < 0.001$. #Significantly different from control at $p < 0.05$. TC = total cholesterol, TAG = triacylglycerols, LDL = low-density lipoprotein cholesterol, HDL = high-density lipoprotein cholesterol, Apo A = apolipoprotein A, Apo B = apolipoprotein B, FBS = fasting blood sugar, hs-CRP = high-sensitivity C reactive protein.

Table 4. Post-intervention atherogenic and inflammatory parameters after adjusting for baseline values (mean + SE).

Parameter	Control group (baseline adjusted post-test mean ± SE)#	Wheatgrass group (baseline adjusted post-test mean ± SE)#	p value	Effect size (Cohen's d)
TC	221 ± 4.5	207 ± 4.5	**0.043**	0.156
TAG	145 ± 5.7	129 ± 5.8	**0.045**	0.212
VLDL	28.5 ± 1.2	25.8 ± 1.2	0.095	0.270
LDL	137 ± 4.0	130 ± 4.1	0.240	0.109
HDL	53.4 ± 1.0	51.8 ± 1.05	0.303	−0.094
Apo A	128 ± 2.4	131 ± 2.4	0.341	**−0.423**
Apo B	106 ± 2.6	96 ± 2.6	**0.016**	**0.520**
LDL/HDL	2.6 ± 0.28	2.5 ± 0.28	0.633	0.04
hs-CRP	0.31 ± 0.03	0.26 ± 0.03	0.251	0.064
FBS	80.9 ± 1.1	80.4 ± 1.1	0.741	−0.054
Hb	12.2 ± 0.09	12.2 ± 0.09	0.766	−0.053
SBP	134 ± 0.36	132 ± 0.37	**0.012**	0.148
DBP	82.3 ± 0.21	81.2 ± 0.22	**0.001**	0.154

#Mean values depicted have been adjusted for the respective baseline values of each parameter.
TC = total cholesterol, TAG = triacylglycerols, LDL = low-density lipoprotein cholesterol, HDL = high-density lipoprotein cholesterol, Apo A = apolipoprotein A, Apo B = apolipoprotein B, FBS = fasting blood sugar, hs-CRP = high-sensitivity C reactive protein.

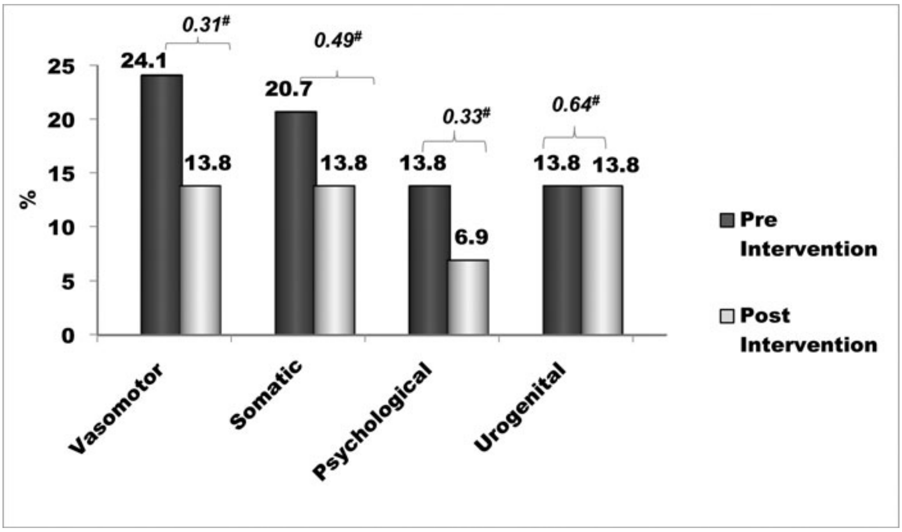

Figure 1. Impact of wheatgrass supplementation on menopausal symptoms. Menopausal symptom categories are not mutually exclusive. #*p* values for the McNemar test between baseline and post-intervention values.

values of TC, TAG, Apo B, SBP, and DBP were significantly lower in the wheatgrass group compared with the control group ($p = 0.043$, 0.045, 0.016, 0.012, and 0.001, respectively). HDL remained comparable between the two groups. However, the effect sizes indicated that reduction in Apo B had a medium size effect (Cohen's d = 0.520), whereas the rest of the differences in the intervention group had a small effect (Cohen's d for: TC = 0.156, TAG = 0.212, SBP = 0.148, and DBP = 0.154).

The impact of supplementation on menopausal symptoms revealed that in the experimental group the prevalence of vasomotor symptoms saw a decline of 42% from the initial prevalence of 24.1% (95% Confidence Interval [CI]: 8.6–39.6) to 13.8% (95% CI: 1.3–26.3) after the intervention period as shown in Figure 1. However, the Mantel–Haenszel Chi square *p* values did not indicate any statistical significance ($p = 0.31$). Similarly, the prevalence of somatic symptoms declined by 33% ($p = 0.49$). In case of psychological symptoms, the prevalence decreased by 50% ($p = 0.33$). Only in the case of urogenital symptoms, it was observed that the prevalence remained unchanged at 13.8% (95% CI: 1.3–26.3) from baseline till the end of supplementation.

Discussion

Supplementation with wheatgrass in the present study had a favorable impact on atherogenic indices as gauged by significant reductions in TC, TAG, and Apo B as well as blood pressure in the experimental group as compared with the control group. Similar results have been found in animal model studies, which are the only studies conducted to evaluate the effect of wheatgrass on atherogenicity. Results from a recent mouse model study (Kothari et al., 2011) on wheatgrass were similar to those found in our research; therein administration of wheatgrass juice at 5 mL/kg and 10 mL/kg in hypercholesterolemia-induced rats resulted in dose-dependent significant ($p < 0.05$) decline in TC, TAG, LDL, and VLDL levels. Another study in rabbit model (Das, Raychaudhuri, & Chakraborty, 2012) evaluated the effect of ethanol extract of wheatgrass fed to hyperlipidemic as well as normal animals. The authors found a significant ($p < 0.05$) decline in the serum TG, TAG, LDL, and malondialdehyde (MDA) levels of animals in both normal and hypercholesterolemic groups.

In our study, while TAG reduced, the Apo A fraction also saw significant decline after supplementation. One of the possible reasons could be the effect of lack of physical activity. The reported prevalence of regular physical activity (at least 30 min of brisk walking, jogging, gym workout, or other moderate-to-intense activity for five days a week) was very low to begin with (27%); one of the possible confounding factors could be drop in physical activity levels after the subjects started taking wheatgrass supplements. In this study, although information on prevalence of regular physical activity was obtained at the beginning of supplementation, the data on incidence of specific physical activities during the intervention period were not obtained. This is one of the limitations of the study.

Other possible reasons for decrease in Apo A could be alcohol intake and smoking. None of the subjects reported alcohol intake or smoking; however, before weighing the accuracy of this reported information, its cultural facet also needs to be considered. Since all the participants were middle aged women belonging to lower middle class to middle class, even if there was any alcohol intake or smoking, such information is highly unlikely to be reported because of the social stigma associated with these practices in Indian societies. Therefore, it is not possible to completely rule out the confounding effects of alcohol and smoking. Further investigations on impact of wheatgrass on lipid parameters need to monitor physical activity, alcohol intake, and smoking in more detail to rule out confounding factors.

Shakya et al. (2014) studied the effect of fresh wheatgrass on glucose metabolism in a rat model study. The results indicated a favorable effect, with significant reduction in plasma glucose upon supplementation with wheatgrass. However, our study did not find any impact on blood glucose.

The only human trial conducted with wheatgrass targeted at cardiovascular health is by Shyam et al. (2007), in which the authors evaluated the effect of wheatgrass supplementation on oxidative stress and antioxidant levels in healthy subjects. The study found that after supplementation with 500 mg wheatgrass for 30 days, there was significant ($p < 0.05$) reduction in MDA and significant enhancement in superoxide dismutase (SOD) and vitamin C levels in subjects. Experiments on lipemic response of wheatgrass contained in common Indian recipes (Iyer et al., 2010) have reported that incorporation of wheatgrass into recipes reduced glycemic index and the TAG level response of recipes as compared with recipes without addition of wheatgrass. However, in our study, the results from hs-CRP indicated only a marginal reduction in the experimental group compared with the control group.

In spite of iron intake being significantly higher in the experimental group, the Hb levels were not significantly different in both groups. Data on interaction of dietary inhibitors of iron absorption and wheatgrass need to be studied further in this regard before drawing sufficient conclusions.

Supplementation with wheatgrass saw nonsignificant reductions in menopausal symptoms in the intervention group at the end of 10 weeks. The most marked change was in the prevalence of vasomotor symptoms, which decreased by 42% after a period of 10 weeks, followed by somatic symptoms, which decreased by 33%, and psychological symptoms, which decreased by 50%. Only the prevalence of urogenital symptoms did not experience any decrease, which remained unchanged at 13.8% from the beginning until the end of supplementation period. The most effective line of therapy for treating menopausal symptoms is HRT. However, the number of health practitioners and patients who do not opt for HRT due to risk of breast cancer has increasingly gone up over the past decade (Guay et al., 2007; Kim et al., 2005; Lambe et al., 2010); and with studies reporting reduction in the incidence of breast cancer following decline in HRT use (Canfell et al., 2008; Katalinic & Rawal, 2008; Lambe et al., 2010; Verkooijen et al., 2009), most of the patients would prefer to resort to herbal supplements. Natural herbal products that have yielded positive results in relieving menopausal symptoms include red ginseng, which, in a study by Kim et al. (2012), reported significant decrease in aggregate menopausal symptom rating ($p < 0.05$) on menopause rating scale and the Kupperman index after a 12-week supplementation with 3 g red ginseng in a randomized double-blind placebo-controlled trial. However, a systematic review on the effectiveness of botanical herbal supplements in alleviating menopausal symptoms revealed that majority of the studies have indicated that extract of black cohosh (*Actaea racemosa* L.) improves menopause-related symptoms (Dog, 2005; Leach & Moore, 2012). The results of the clinical trials of soy isoflavones were mixed, and those of red clover extracts, dong quai (*Angelica sinensis* L.), ginseng (*Panax ginseng C.A. Mey*), or evening primrose seed oil were inconclusive and contradictory (Dog, 2005). In the present study, however, although the prevalence of various menopausal symptoms showed decline, it was not statistically significant for this duration of intervention; perhaps, long-term intervention trials are required to draw definite conclusions about the effect that wheatgrass has on menopausal symptoms.

Limitations: In our study the physical activity levels were not monitored during the entire duration of intervention. This information could have helped to explain, in part, the decrease in Apo A levels in the intervention group.

Conclusions

Supplementation with freeze-dried wheatgrass powder at a dosage of 3.5 g per day for 10 weeks appears to have a positive effect on atherogenic lipid fractions, especially on Apo B, TC, and TAG, in hypercholesterolemic women. Wheatgrass also seems to marginally improve vasomotor and somatic and psychological symptoms of menopause. However, without adequate physical activity, wheatgrass might not be able to retain the protective Apo A levels. Consequently, wheatgrass appears to be a promising holistic therapeutic supplement for menopausal women that can confer health benefits to maintain and improve their cardiovascular, metabolic, and reproductive systems. This is the first in vivo human study reporting the effects of freeze-dried wheatgrass on the atherogenic lipoproteins, inflammation, blood sugar, and menopausal symptoms.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the article.

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References

Bajaj S, Mahajan V, Grover S, Mahajan A, Mahajan N. Gender based differences in risk factor profile and coronary angiography of patients presenting with acute myocardial infarction in North Indian population. J Clin Diagn Res. 2016;10(5):OC05–OC07.
Canfell K, Banks E, Moa AM, Beral V. Decrease in breast cancer incidence following a rapid fall in use of hormone replacement therapy in Australia. Med J Australia. 2008;188(11):641.
Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL, et al. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. JAMA. 2003;289(19):2560–2571.
Das A, Raychaudhuri U, Chakraborty R. Effect of freeze drying and oven drying on antioxidant properties of fresh wheatgrass. Int J Food Sci Nutr. 2012;63(6):718–721.
Das S, Hakim A, Mittal A. Study of the antihyperlipidemic, antioxidative and antiatherogenic activity of aeglemarmeloslinn in rabbit receiving high fat diet. Asian J Pharm Clin Res. 2012;5(4):69–72.
Dessapt AL, Gourdy P. [Menopause and cardiovascular risk]. Journal de gynecologie, obstetrique et biologie de la reproduction. 2012;41(7 Suppl):F13–9.
Dog TL. Menopause: a review of botanical dietary supplements. Am J Med. 2005;118(12):98–108.
Freidewald W, Levy R, Fredrickson D. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem. 1972;18, 6:499–502.
Geller, S. E., Studee, L. Botanical and dietary supplements for menopausal symptoms: what works, what does not. J Women's Health. 2005;14(7):634–649.
Giesbrecht F, Gumpertz M. Planning, Construction, and Statistical Analysis of Comparative Experiments. 1st ed. New Jersey: John Wiley & Sons Inc.; 2004, 13–27.
Gopalan C, Rama Shastri BV, Balasubramanian SC. Nutritive value of Indian foods; revised and updated, 2007 ed. Hyderabad, India: Indian Council of Medical Research, 2007; 47–95.
Greene, J. G. Constructing a standard climacteric scale. Maturitas. 1998;29(1):25–31.
Guay MP, Dragomir A, Pilon D, Moride Y, Perreault S. Changes in pattern of use, clinical characteristics and persistence rate of hormone replacement therapy among postmenopausal women after the WHI publication. Pharmacoepidemiol Drug Saf. 2007;16(1):17–27.
Hart EC, Charkoudian N, Miller VM. Sex, hormones and neuroeffector mechanisms. Acta Physiol. 2011;203(1):155–165.
Hemalatha R, Karthik M, Babu KN, Kumar BD. Immunomodulatory activity of triticum aestivum and its effects on Th1/Th2 cytokines and NFκB P 65 response. Am J Biochem Mol Biol. 2012;2(1):19–25.
Iyer U, Sharma M, Dhruv S, Mani UV. Glycemic and lipemic response of wheat grass incorporated recipes. J Herbal Med Toxicol. 2010;4(1):161–164.
Katalinic A, Rawal R. Decline in breast cancer incidence after decrease in utilisation of hormone replacement therapy. Breast Cancer Res Treat. 2008;107(3):427–430.
Kim N, Gross C, Curtis J, Stettin G, Wogen S, Choe N, et al. The impact of clinical trials on the use of hormone replacement therapy. J Gen Intern Med. 2005;20(11):1026–1031.
Kim SY, Seo SK, Choi YM, Jeon YE, Lim KJ, Cho S, et al. Effects of red ginseng supplementation on menopausal symptoms and cardiovascular risk factors in postmenopausal women: a double-blind randomized controlled trial. Menopause. 2012;19(4):461–466.
Kothari S, Jain AK, Mehta SC, Tonpay S. D. Hypolipidemic effect of fresh Triticum aestivum (wheat) grass juice in hypercholesterolemic rats. Acta Pol Pharm. 2011;68(2):291–294.
Kulkarni SD, Tilak J, Acharya R, Rajurkar NS, Devasagayam TPA, Reddy AVR. Evaluation of the antioxidant activity of wheatgrass (Triticum aestivum L.) as a function of growth under different conditions. Phytotherapy Res. 2006;20(3):218–227.
Lambe M., Wigertz A, Holmqvist M, Adolfsson J, Bardage C, Fornander T. et al. Reductions in use of hormone replacement therapy: effects on Swedish breast cancer incidence trends only seen after several years. Breast Cancer Res Treat. 2010;121(3):679–683.

Leach MJ, Moore V. Black cohosh (Cimicifuga spp.) for menopausal symptoms. Cochrane Database Syst Rev. 2012;(9):CD007244. doi: 10.1002/14651858.CD007244.pub2.

Lipovac M, Chedraui P, Gruenhut C, Gocan A, Kurz C, Neuber B, et al. The effect of red clover isoflavone supplementation over vasomotor and menopausal symptoms in postmenopausal women. Gynecol Endocrinol. 2012;28(3):203–207.

Maas AH, Appleman YE. Gender differences in coronary heart disease. Neth Heart J. 2010;18(12):598–602.

QY Research. Global Wheat Grass Powder Market by Manufacturers, Regions, Type and Application, Forecast to 2021. Global Info Research Report ID 11293.

Rana S, Kamboj JK, Gandhi V. Living life the natural way—wheatgrass and health. Funct Foods Health Dis. 2011;1(11):444–456.

Rossi R, Grimaldi T, Origliani G, Fantini G, Coppi F, Modena MG. Menopause and cardiovascular risk. Pathophysiol Haemostasis Thromb. 2002;32(5–6):325–328.

Sethi J, Yadav M, Dahiya K, Sood S, Singh V, Bhattacharya SB. Antioxidant effect of Triticum aestivium (wheat grass) in high-fat diet-induced oxidative stress in rabbits. Exp Clin Pharmacol. 2010;32(4):233–235.

Schnabel CF. Feed. US patent No. 1942943; January 9, Washington, DC: United States Patent Office, 1934.

Shakya G, Randhi PK, Pajaniradje S, Mohankumar K, Rajagopalan R. Hypoglycaemic role of wheatgrass and its effect on carbohydrate metabolic enzymes in type II diabetic rats. Toxicology and industrial health. 2016;32(6):1026–32.

Shukla V, Tripathi R. Charaka samhita, Vol. 1, 1st ed. Varanasi, India: Chaukhamba Orientalia Press, 2005, 14.

Shyam R, Singh SN, Vats P, Singh VK, Bajaj R, Singh SB, et al. Wheat grass supplementation decreases oxidative stress in healthy subjects: a comparative study with spirulina. J Altern Compl Med. 2007;13(8):789–792.

Talas ZS, Ozdemir I, Yilmaz I, Gok Y, Orun I. The investigation of the antioxidative properties of the novel synthetic organoselenium compounds in some rat tissues. Exp Biol Med. 2008;233(5):575–579.

Venugopal S, Iyer UM. Management of diabetic dyslipidemia with sub-atmospheric dehydrated barley grass powder. Int J Green Pharm (IJGP). 2010;4(4):251–6.

Verkooijen HM, Bouchardy C, Vinh-Hung V, Rapiti E, Hartman M. The incidence of breast cancer and changes in the use of hormone replacement therapy: a review of the evidence. Maturitas. 2009;64(2):80–85.

Wheat J, Currie G. Herbal medicine for cancer patients: an evidence based review. J Altern Med. 2008;5(2):28–30.

WHO Expert Consultation. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. Lancet (London). 2004;363(9403):157.

WHO. Redefining obesity and its treatment. Western Pacific Region, WWP, IASO and IOTF: the Asia-Pacific Perspective. Sydney, Australia: Health Communications Australia, 2000; 15–8.