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Characterization of dehydrated functional fractional spinach powder

Ankita, K.Prasad*

Department of Food Engineering and Technology, Sant Longowal Institute of Engineering and Technology,
Longowal – 148106, Punjab, (INDIA)
E-mail: dr_k_prasad@rediffmail.com

ABSTRACT

Spinach (*Spinacia oleracea*) leaves respire and transpire even after harvesting. Moisture of spinach leaf during post harvest period loss rapidly due to transpiration, which reduces the cell turgor and results in the leaves of reduced acceptability and quality. The rate of faster deterioration of this important leafy winter vegetable thus categorized among highly perishable commodity. Processing of such commodities is therefore essential to utilize for their future use. Dehydration is an important unit operation applied mainly to remove the moisture in form of vapor from its surface into the surrounding to obtain relatively shelf stable dried form. For easier handling, storability and usability, the leaves are generally crushed into powdered form and often fractionated to incorporate the functionality in it. The efforts have been made under the present study to obtain the functional spinach powder. The prepared powder was characterized and suggested for the possible benefits and their uses during off season as the ready to use nutritional and functional material.

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KEYWORDS

Spinacia oleracea;
Spinach;
Dehydration;
Powder;
Characterization.

INTRODUCTION

Spinach (*Spinacia oleracea*) is a plant belongs to Amaranthaceae or Chenopodiaceae family and considered to be the native to central and south western Asia. It is cultivated throughout the world as cool season annual leafy vegetable^[1, 2]. It is preferably utilized in food as raw, boiled, canned, frozen, dehydrated, pureed form and cooked or baked into various dishes. Chlorophyll is the important constituent of leaf responsible for photosynthesis and green coloration. The color of the leaves changes from green to olive green or brown on thermal pro-

cessing mainly due to conversion of chlorophyll to pheophytin and pyropheophytin applying the non-enzymatic pathway^[3]. Formation of chlorophyllin and arresting further conversion is a major challenge and could be achieved through the controlled processing for obtaining the highly acceptable dehydrated leaf powder enriched with carotenoids, vitamin C and vitamin E content^[3-6].

Spinach is known for its antimicrobial, anti carcinogenic and antioxidant activity^[1]. Anti aging properties associated with spinach leaves with considerable amount of β -carotene (2-6mg/100g), folic acid (120 mg/100g) and riboflavin (0.25mg/100g) make

this leaf a unique food material^[5]. Apart from the benefits, the powder may be used as replacer of artificial color as demand of natural pigment is increasing in present era whether the carotenoids, porphyrin colors, flavonoids and betalains colours^[7,8].

Spinach contains high amount of moisture, which reduces rapidly due to transpiration resulting in the loss of cell turgor. Being enormous exposed surface area in respect to the occupied volume, the transpirational loss are more and resulted in the rapid quality deterioration thus spinach leaf is categorized under highly perishable category. Processing of such commodities is therefore essential to enhance the shelf life for its extended use. Dehydration is an important unit operation applied mainly to remove the moisture in form of vapour from the surface into the surrounding to obtain relatively shelf stable dried form^[9,10]. For easier handling, storability and usability the leaves are generally crushed into powder form and often fractionated to incorporate the functionality in it^[3,4].

However, major problems associated with the powders containing fine particles are the inherent difficulties in reconstitution, problems of dusting or lumping and improper flow properties during handling^[11]. But addition of small quantities of leaf powder not only improves the taste and color of food stuffs but also enhance the nutritional values. Considering the mentioned importance, the present study was conducted specifically in order to characterize the functional leaf powder of spinach so as to use as ready to use material in various food formulations.

EXPERIMENTAL

Sample Preparation

Spinacia oleracea var Palak RNG - All Green Akshit leaves were harvested and prepared for subjecting the process of blanching and dehydration^[3,4]. Steam blanching as pretreatment was provided to the prepared spinach leaves for a period of two minutes in a developed precision steam blancher consisted of two interconnected chambers, one as steam generation and another as blanching chamber for effective and reproducible blanching process^[12]. The untreated and blanched samples were spread as thin

layer uniformly on the aluminum tray hooked with the developed precision dehydration chamber to study the effect of isothermal dehydration temperatures (50, 60, 70, 80 and 90°C) having temperature fluctuation of $\pm 1^\circ\text{C}$ ^[12]. The dehydration kinetics and the resultant products were compared^[13,14] and time temperature combination was identified as initial dehydration temperature of 80°C for 1 hour and finishing temperature of 60°C for 2 hours (unpublished data).

The obtained dehydrated leaf samples under the identified time temperature of dehydration the dehydrated leaves were obtained and manual crushed before using mixer grinder to get spinach leaf powder. Further, 60 BSS screen was used to classify the leaf powder into fine and coarse fractions, which was characterized on the basis of physico-chemical properties^[3].

Physical characteristics

The physical properties were evaluated using standard method as described elsewhere^[15]. Dehydration characteristics as dehydration ratio with water absorption capacity (WAC) and water solubility index (WSI) were evaluated^[3].

Chemical characteristics

Chemical composition for moisture content, crude protein, crude fiber, crude fat, carbohydrate and ash was evaluated using standard method^[16]. The total antioxidant capacity (TAC) of spinach leaf powder was assessed using 1,1-diphenyl-2-picrylhydrazyl (DPPH) as free radical scavenging activity^[17].

Phytochemical identification

The presence of phytochemical in the extracts of spinach leaf powder was assessed using Shimadzu GC-2010. Nitrogen gas was used as carrier gas and maintained at 10 psi inlet pressure using FID and AB inno-wax column (60 m X 0.25 mm id, film thickness 0.25 μm). Injector and detector temperatures were maintained at 270 and 280°C, respectively. Column temperature programmed from 60 to 180°C at 3°C/min with hold time of 2 minutes and from 180 to 250°C at 5°C/min with hold time 20 min, respectively. The flow rate of carrier gas was 1.2

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ml/min and split ratio was 80:1. The data were processed on GC solutions software for leaf powder composition. GC-MS analysis was obtained on a Shimadzu Mass Spectrometer-2010 series system using same column and conditions as GC. Helium was used as carrier gas. EI source and mass range were 70 eV and 40-750 amu, respectively. To identify the presence of phytochemicals in the sample NIST and Wiley computer libraries have been used.

Optical characteristics

The optical characteristics of spinach powder samples were assessed using Hunter Colorimeter in terms of L (luminance or brightness), a [red (+) - green (-)] and b [yellow (+) - blue (-)] values^[18].

Thermal characteristics

Differential scanning calorimetric (DSC) characterization was carried out for the spinach powder using Mettler DSC- 30 (Mettler Toledo, Switzerland) instrument, equipped with STAR^e software. Heat flow calibration was carried out using indium standard at 10°C/min heat scan rate. Approximately, 20 mg sample was weighed, mixed with water (1:3 w/w) and were taken into standard aluminium pan. The pan was press sealed with a lid using sample press supplied with the instrument. Thermal scanning was carried out using sealed pan made in similar ways without any sample incorporated to consider as reference.

FTIR characteristics

Infrared spectra of the powdered sample was recorded at room temperature using FTIR spectrometer (SPECTRUM-2000, Perkin Elmer, USA) in the range 700–4000 cm⁻¹. The sample was placed directly in the sample holder. A background was collected before each sample was analyzed then subtracted from the spectra of sample prior to analysis. After every scan, a new reference background spectrum was taken. The ATR crystal cleaned with hot water and acetone was examined for spectral authenticity in order to avoid the chance of any contamination error.

Microstructure analysis

Scanning electron micrographs of functional

fractionated spinach fine powder was acquired in the range of 500 to 3500 magnifications using scanning electron microscope (Jeol JSM-6100, Jeol Ltd, Tokyo, Japan). The sample for acquiring the micrographs was prepared and mounted on double sided tape on the used aluminum stubs. Further, adhered sample was coated with gold - palladium (60:40) at an accelerated voltage of 15 kV under vacuum^[19] (Suksomboon and Naivikul, 2006). A digital camera head cooled with a Peltier mechanism was present for capturing the image by microscope.

Statistical analysis

The reported data are the average of triplicate observations. The mean \pm standard deviation (SD) was calculated for each treatment. In order to determine any statistically significant effects prevailed, Duncan multiple range test at Pd^{0.05} was applied using SPSS 16.0 and Microsoft excel software package (Microsoft Corporation, USA).

RESULTS AND DISCUSSION

Figure 1 represent the pictorial representations of dehydrated spinach leaf powder with the fractionated coarse and fine power fractions as obtained using 60 BSS (250 μ m) sieve. The particle size distribution characteristics of fine fraction with scanning electron micrograph are presented in Figure 2. Particle size distribution reflects the particles are narrowly distributed as measured through the use of laser diffraction technique. Rosin Rammler function also known as Weibull function was applied as it describes the particle distribution to fairly accurate level.

Physical characterization

The physical characterization was performed mainly on dimensional, gravimetric and frictional

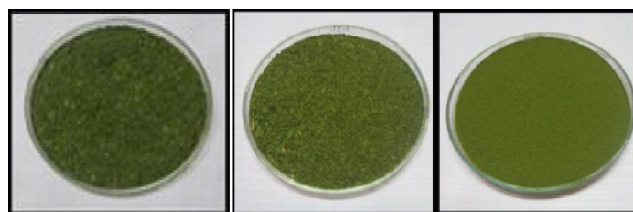


Figure 1 : Mixed, coarse and fine spinach powder fractions

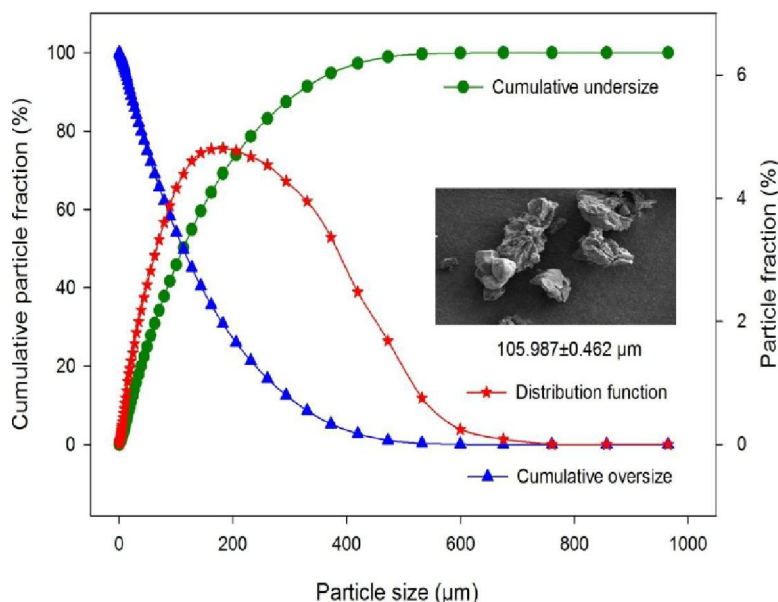


Figure 2 : Particle size distribution of fine spinach powder fraction

characteristics. The surface area to volume ratio for the spinach leaves were found to be enormous and in order of $37.80 \times 10^3 \text{ m}^{-1}$. This higher ratio of surface area to volume have favoured faster removal of water from the spinach during initial phase of dehydration and also helped to keep the material temperature around wet bulb temperature to convert chlorophyll into chlorophyllide through enzymatic pathway as judged the increased level of phytol presence in the dehydrating product^[3]. The average particle size of fine spinach powder fraction was found to be $105.987 \pm 0.462 \mu\text{m}$ Figure 2. Bulk density is the ratio of mass of sample to the occupied volume was found to be insignificant difference for the fine spinach powder fraction (422 kg/m^3) on comparison with the mix fractions (413 kg/m^3) from which it was separated TABLE 1.

The frictional properties were examined for the powders are angle of repose and coefficient of friction. Angle of repose for the fine powder (46.980°) was found to be significantly higher than the mix and coarse powder fractions (40.022 to 42.306°). The coefficient of friction for the spinach powders were determined on glass, plywood and galvanized steel sheet and found similar trend for fine powder fraction on comparing with coarse spinach powder fraction TABLE 1. These phenomenons are imperative in processing, particularly in designing the handling equipment.

Fine powder of leaves show higher degree of cohesiveness^[20] and thus exhibit higher frictional characteristics and so the coefficient of friction^[21,22] on different applied surfaces. Dehydration ratio was found to be 21.55. The rehydration studies were conducted in terms of water absorption capacity (WAC) and water solubility index (WSI). Fine powder fraction has shown significantly higher water binding characteristics with the 3.607 and 41.650 as the WAC and WSI, respectively^[23] TABLE 1.

CHEMICAL CHARACTERIZATION

Proximate composition

TABLE 2 represents the proximate composition with the chlorophyll content and antioxidant capacity as free radical quenching capacity of different powder fractions. The proximate analysis was concentrated on moisture, crude protein, crude fat, crude fiber, ash and carbohydrate content on difference method excluding the fiber content. Protein and crude fiber are found to be in good amount in leaf powder^[24]. The moisture content of spinach powder was found to be $4.64 \pm 0.28\%$. The crude protein content for the fine powder fraction was found to be 22.14%, which was significantly higher than mix or coarse spinach powder fractions TABLE 2. Proteins from leafy vegetables are utilized effectively and abundantly in different food applications. The mineral

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TABLE 1 : Physical properties of dehydrated spinach leaf as powder at combination temperature

Spinach leaf powder	Bulk density (kg/m ³)	Angle of repose (°)	Coefficient of friction			Water absorption capacity (g/g)	Water solubility index (%)
			Glass	Steel	Plywood		
Mix	413±4.041 ^a	42.306±2.459 ^b	0.279±0.002 ^b	0.341±0.007 ^a	0.359±0.007 ^a	2.746±0.019 ^b	40.200±0.707 ^b
Coarse	375±1.586 ^b	40.022±1.710 ^b	0.265±0.000 ^b	0.319±0.006 ^b	0.319±0.009 ^b	3.396±0.002 ^a	40.250±2.192 ^b
Fine	422±1.045 ^a	46.980±1.310 ^a	0.306±0.130 ^a	0.344±0.011 ^a	0.354±0.013 ^a	3.607±0.416 ^a	41.650±2.050 ^a

Means in the same column followed by the different letters are significantly different ($p \leq 0.05$)

TABLE 2 : Chemical characteristics of spinach leaf powder

Spinach leaf powder	Moisture (%)	Crude Protein (%)	Crude Fat (%)	Crude fiber (%)	Ash (%)	Carbohydrate (%)	Chlorophyll mg/100g	Antioxidant mg/g
Mix	4.64±0.28 ^a	20.84±0.51 ^b	3.63±0.11 ^b	25.39±0.69 ^{ab}	12.50±0.07 ^a	33.00±0.65 ^b	653.400±0.850 ^b	371.92±6.55 ^b
Coarse	4.43±0.06 ^b	18.36±0.60 ^c	3.54±0.45 ^b	27.80±0.37 ^a	12.33±0.62 ^a	33.55±1.24 ^b	615.700±6.650 ^c	354.94±6.53 ^c
Fine	4.48±0.11 ^b	22.14±0.27 ^a	3.87±0.01 ^a	20.10±0.93 ^b	11.37±0.04 ^b	38.05±0.50 ^a	673.400±1.980 ^a	390.72±2.80 ^a

Means in the same column followed by the different letters are significantly different ($p \leq 0.05$)

matter as expected was found to be lower in the fine powder fraction. Major health benefits coupled with functional properties such as fat replacement properties and water holding capacity have created the interest in the high fiber ingredient, particularly in pharmaceutical industry.

Studies have been shown the beneficial effects of fiber consumption in improving the gut health and protection against several ailments^[25].

Chlorophyll is predominant chemical responsible for imparting green colour to leaves. It masks carotene in plant leaves^[3] to exhibit yellow pigment. β -carotene content found in fresh spinach leaves was 4.498±0.084 mg/100g. Spinach contains 178.253±2.038 mg/100g of total chlorophyll. Higher levels of free radical quenching capacity (390.72mg/g) further makes fine spinach powder a nutritional fraction to be used for the therapeutic purposes and thus the powder may be considered to be food functional.

Phytochemical constituents

Blanched powder was found to be more compact with higher moisture content as compared to the powder obtained from untreated leaves at respective temperatures. The powder obtained from the spinach leaves dehydrated at 70°C was subjected for the phytochemical evaluation came in the hexane extracts through gas chromatography coupled with

mass spectroscopy. Interpretation of mass spectra was performed using National Institute of Standard and Technology (NIST) and Wiley database. Similarity index was used to select the identified probable phytochemical, which was then verified from the available literature. TABLE 3 depicts the possible compound of unblanched leaf powder whereas TABLE 4 represents the compound in blanched leaf powder. The role of different class of identified compounds is complied with their possible beneficial effects in TABLE 5. Thus the presence of various compounds in the GCMS analysis Figure 3 are hexadecanoic acid, phytol (7,11,15-Tetramethyl-2-hexadecen-1-ol), Campesterol (Ergost-5-en-3.beta.-ol), octadecanoic acid, n-eicosanol, alpha-tocopherol-beta-mannoside, stigmasterol, hexadecenal, vitamin E acetate, 1-pentadecyne, cis-9-hexadecenal, pentadecanoic acid, β -sitosterol, oleic acid, ethyl-4,5-dimethyl-phenol, linoleic acid (octadecatrienoic acid, methyl ester) and Beta-sitosterol (Stigmast-5-en-3.beta.-ol)^[26-31]. Presence of these compounds possibly contributes their role in exhibiting different properties such as antibacterial activity, antioxidant property, nutraceutical value with other important effects.

Optical characteristics

The L, a, and b values used to represent colour of the materials was found to be 30, -11 & 24; 37, -

TABLE 3 : Possible phyto-chemicals in the hexane extract of unblanched spinach leaf powder

Peak	RT(min)	Area (%)	Mol wt.	SI	Molecular formula	Name of the constituent
1	6.457	0.80	142	84	C ₆ H ₁₀ N ₂ O ₂	Pyrrrolidoneacetamide
2	6.565	0.83	155	90	C ₁₀ H ₂₁ N	3-Methyl-N-[(E)-3-methylbutylidene]-1-butanamine
3	8.094	1.08	116	79	C ₆ H ₁₂ O ₂	4-Vinyl-1-butanol
4	10.452	2.49	150	95	C ₉ H ₁₀ O ₂	2-Methoxy-4-vinylphenol
5	12.81	0.63	198	76	C ₁₁ H ₁₈ O ₃	3,3-Dimethyl-1,5-Dioxaspiro-Undecan-9-one
6	16.492	0.57	154	86	C ₁₁ H ₂₂	8-Methyl-1-decene
7	16.549	13.97	296	92	C ₂₀ H ₄₀ O	3,7,11,15-Tetramethylhexadecan-2-en-1-ol
8	16.611	2.69	280	90	C ₂₀ H ₄₀	(2E)-3,7,11,15-Tetramethyl-2-hexadecene
9	16.8	2.35	250	89	C ₁₈ H ₃₄	1-Octadecyne
10	16.996	3.85	278	88	C ₂₀ H ₃₈	1,4-Eicosadiene
11	17.408	0.61	270	92	C ₁₇ H ₃₄ O ₂	Hexadecanoic acid, methyl ester
12	19.084	0.65	294	89	C ₁₉ H ₃₄ O ₂	9,12-Octadecadienoic acid, methyl ester,,
13	19.16	2.27	292	95	C ₁₉ H ₃₂ O ₂	Linolenic acid, methyl ester
14	19.271	21.44	296	97	C ₂₀ H ₄₀ O	Phytol
15	20.567	0.54	258	81	C ₁₆ H ₂₂ N ₂ O	Cyclohex-2-enone, 3-(2-dimethylaminoethylamino)-5-phenyl
16	20.721	1.83	157	84	C ₈ H ₁₅ NO ₂	2-methyl-acrylic acid 2-dimethylamino-ethyl ester
17	20.855	0.70	198	77	C ₁₃ H ₂₆ O	11-Tridecen-1-ol
18	22.17	0.77	508	85	C ₃₄ H ₆₈ O ₂	Heptadecanoic acid, heptadecyl ester
19	22.248	2.89	155	87	C ₁₀ H ₂₁ N	Benzedrex
20	22.32	0.86	530	66	C ₂₉ H ₆₂ O ₄ Si ₂	Eicosanoic acid, 2,3-bis[(trimethylsilyl)oxy]propyl ester
21	22.417	5.68	294	93	C ₂₁ H ₄₂	10-Heneicosene
22	23.32	0.43	204	74	C ₁₁ H ₁₂ N ₂ O ₂	Tryptophan
23	23.846	0.56	500	68	C ₂₇ H ₅₆ O ₄ Si ₂	9-Octadecenoic acid
24	23.942	1.16	164	65	C ₂₀ H ₃₆ O ₂	9,12-Octadecadienoic acid (9z,12z)-, ethyl ester
25	24.165	11.89	298	95	C ₂₀ H ₄₂ O	1-Eicosanol
26	26.152	0.45	268	83	C ₁₉ H ₄₀	Nonadecane
27	26.255	7.70	452	94	C ₃₁ H ₆₄ O	N-hentriacontanol
28	29.016	0.63	270	80	C ₁₈ H ₃₈ O	1-Octadecanol
29	30.106	1.62	472	76	C ₃₁ H ₅₂ O ₃	Vitamin E acetate
30	34.712	8.07	414	82	C ₂₉ H ₅₀ O	Beta-sitosterol

12 & 32 and 40, -14 & 37, respectively for mix, coarse and fine fractions of spinach powders. Figure 1 clearly reflects brighter with intense green tinge reflected for fine fraction of spinach powder as compared to other powder.

Thermal characteristics

Differential scanning calorimetric (DSC) characterization describes thermal changes without altering the mass of the sample as processing time proceeded, Starch gelatinization, protein denaturation or melting are the important phenomenon occurring in various foods to provide unique texture and struc-

ture^[32]. The onset and peak temperatures were found 42.53 and 53.15 °C and 75.75 and 75.91 °C, respectively with differential enthalpy (ΔH) as 2.278 and 0.984 J/g Figure 4, which supports the associated changes happening during thermal treatments.

Fourier Transmission Infrared Spectroscopy (FTIR)

Infrared spectroscopy is the study of interactions between biomaterial and electromagnetic fields in the IR region of the electromagnetic spectrum. Molecules excite to the higher vibration state by absorbing infrared radiation and provide the fingerprint

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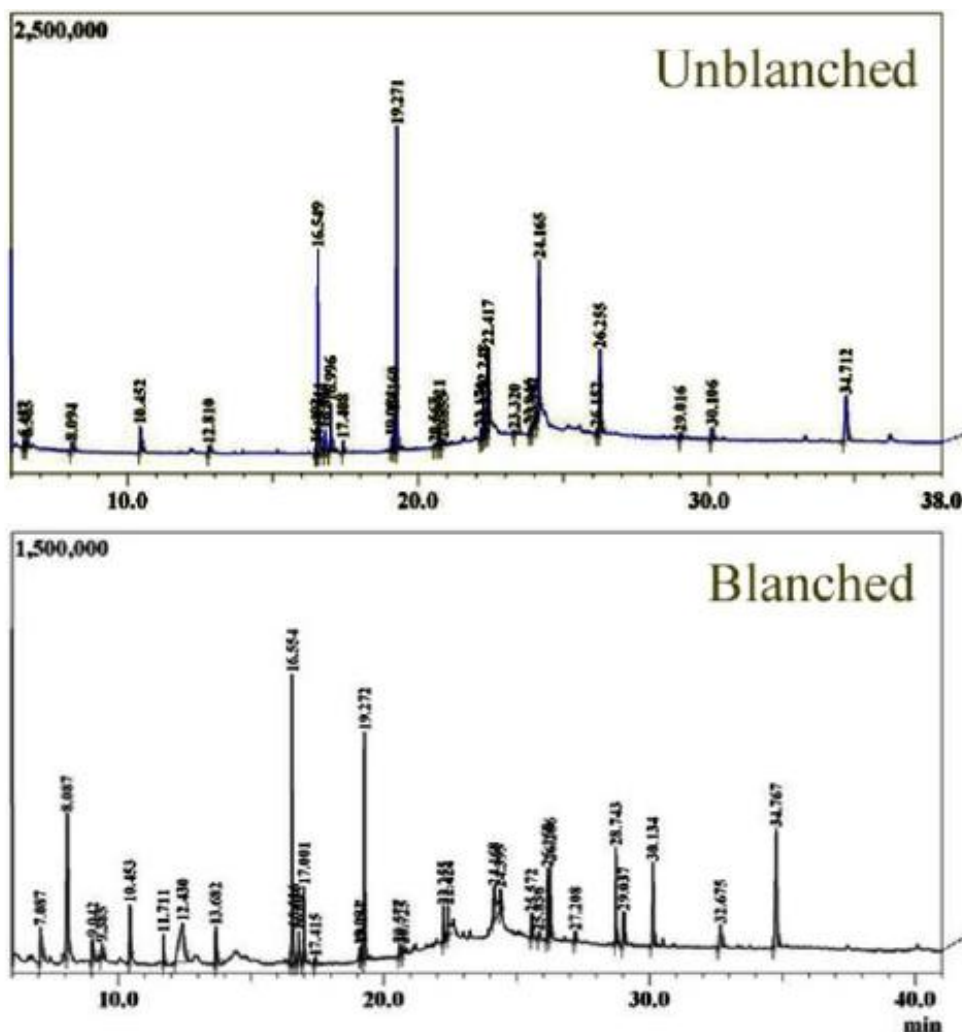


Figure 3 : Gas chromatograph of spinach powder

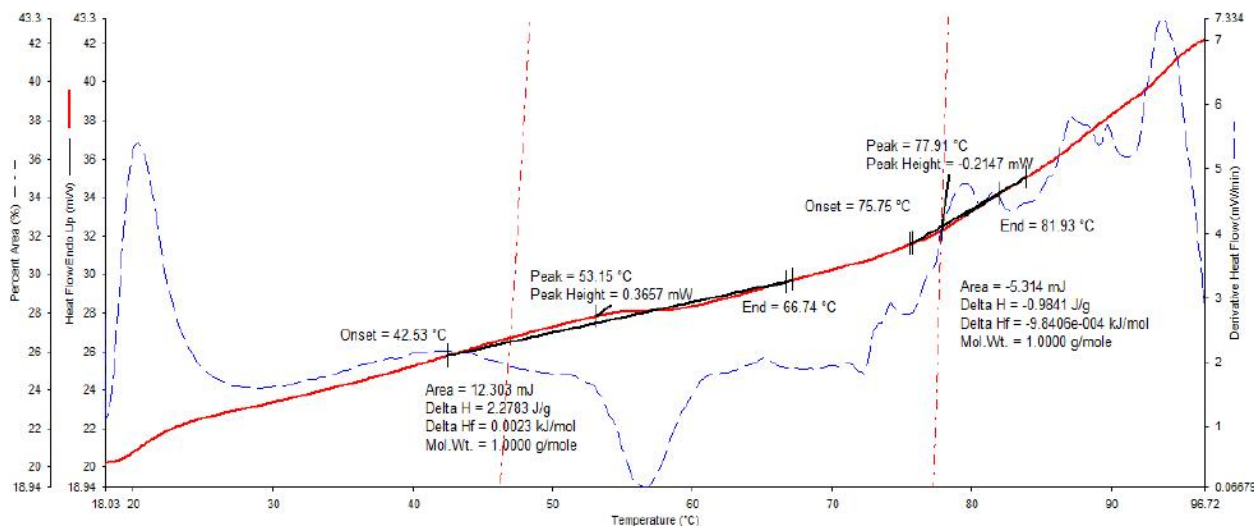


Figure 4 : DSC thermogram of fine spinach powder fraction

information for the chemical so as to get the information regarding the functional groups depending of the unique wave number, the reciprocal of wave

length.

Fourier Transmission Infrared Spectroscopy is the analytical technique, which monitors directly the

TABLE 4 : Possible phyto-chemicals in the hexane extract of blanched spinach leaf powder

Peak	RT(min)	Area (%)	Mol wt.	SI	Molecular formula	Name of the constituent
1	7.087	2.92	126	79	C ₅ H ₆ N ₂ O ₂	3-Diazo-2,4-Pentanedione
2	8.087	8.90	116	80	C ₆ H ₈ O ₄	3,5-Dihydroxy-6-methyl-2,3-dihydro-4H-pyran-4-one
3	9.042	1.60	120	74	C ₈ H ₈ O	2-Methylbenzaldehyde
4	9.385	1.09	126	7	C ₆ H ₁₀ N ₂ O	3,4,4-Trimethyl-5-pyrazolone
5	10.453	2.44	150	96	C ₉ H ₁₀ O ₂	2-Methoxy-4-vinylphenol
6	11.711	1.01	252	81	C ₁₂ H ₂₆ FO ₂ P	Phosphonofluoridic acid, propy-nonyl ester
7	12.43	9.98	243	82	C ₉ H ₁₃ N ₃ O ₅	Beta-ribofuranoside, cytosine
8	13.682	1.66	180	74	C ₁₁ H ₁₆ O ₂	3-Tert-Butyl-4-hydroxyanisole
9	16.554	11.61	362	89	C ₂₆ H ₅₀	2-Hexadecyl-bicyclopentyl
10	16.616	1.90	182	87	C ₁₃ H ₂₆	3,3,4-Trimethyl-1-decene
11	16.805	1.80	296	90	C ₂₀ H ₄₀ O	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
12	17.001	3.04	250	90	C ₁₈ H ₃₄	1-Octadecyne
13	17.415	0.18	256	87	C ₁₆ H ₃₂ O ₂	Pentadecanoic acid, methyl ester
14	19.091	0.32	194	84	C ₁₄ H ₂₆	7-Tetradecyne
15	19.167	0.65	292	88	C ₁₉ H ₃₂ O ₂	9,12,15-Octadecatrienoic acid, methyl ester
16	19.272	8.79	296	96	C ₂₀ H ₄₀ O	Phytol
17	20.577	0.44	155	86	C ₁₀ H ₂₁ N	Benzedrex
18	20.725	0.27	132	82	C ₅ H ₁₂ N ₂ O ₂	Carbamic acid, (2-dimethylamino_ethyl ester
19	22.255	1.33	292	81	C ₁₉ H ₃₂ O ₂	9,12,15 Octadecatrienoic acid
20	22.424	1.33	288	83	C ₁₈ H ₃₇ Cl	1-Chlorooctadecane
21	24.168	2.67	184	80	C ₁₃ H ₂₈	3,8-Dimethylundecane
22	24.395	1.45	264	85	C ₁₈ H ₃₂ O	9,12,15-Octadecatrien-1-ol, (Z,Z,Z)
23	25.572	1.08	410	90	C ₃₀ H ₅₀	Squalene
24	25.836	0.29	280	84	C ₂₀ H ₄₀	2-Methyl-7-nonadecene
25	26.168	3.21	478	95	C ₃₄ H ₇₀	Tetratriacontane
26	26.266	3.66	322	93	C ₂₃ H ₄₆	1-Tricosene
27	27.208	0.55	254	85	C ₁₁ H ₂₀ Cl ₂ O ₂	Dichloroacetic acid, nonyl ester
28	28.743	5.25	338	82	C ₂₃ H ₄₆ O	Diundecyl ketone
29	29.037	2.24	298	89	C ₂₀ H ₄₂ O	1-Eicosanol
30	30.134	5.50	472	89	C ₃₁ H ₅₂ O ₃	Vitamin E acetate
31	32.675	2.22	400	83	C ₂₈ H ₄₈ O	Campesterol
32	34.767	10.62	94	81	C ₂₉ H ₅₀ O	Beta.-sitosterol

functional groups for characterizing the molecular structure^[33]. Figure 5 shows the functional groups present in the spinach fine fraction and appearing in form of bands due to molecular vibrations. Regions of stretching and bending includes (-C-O-CH₂-) stretching, (C-H) bending, and C-O, C=C (ring) stretching, ring vibrations of carbohydrates in wave number range 1200-100 cm⁻¹, tocopherol (1089-1058 cm⁻¹), symmetric -C-H bending of methyl group (1400-1330 cm⁻¹), C-C (ring) stretching and C-H (ring) bending, stretching of -C-H of the CH₂ and

CH₃ aliphatic groups of fatty acids (1485-1425 cm⁻¹), C=O, C-N stretching of amide I, N-H, C-O bending and C-C, C-N stretching of amide II, C=C (ring) stretching, C-C, C-O bending (1690-1485 cm⁻¹), protein beta sheet (1639-1613 cm⁻¹), C=O stretching of carbonyl groups (ketone, esters and acids) (1800-1700 cm⁻¹). Presence of various groups at particular to the wave numbers is illustrated elsewhere^[34-36].

The spinach leaf powder in form of either mix, course or fine fractions being rich in nutrients with the available wide varieties of phytochemicals hav-

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TABLE 5 : Role of secondary metabolites in the body

Nature of compound	Activity/Function
Ester	Anti-inflammatory, antioxidant, hypo cholesterolemic, anti-inflammatory, cancer preventive, hepato-protective
Terpene	Antimicrobial, anticancer, anti-inflammatory, diuretic, antithrombotic
Steroid	Anti cancerous, hepato protective, antimicrobial, antiasthma, diuretic, antioxidant, hypo cholesterolemic, anti androgenic, hemolytic

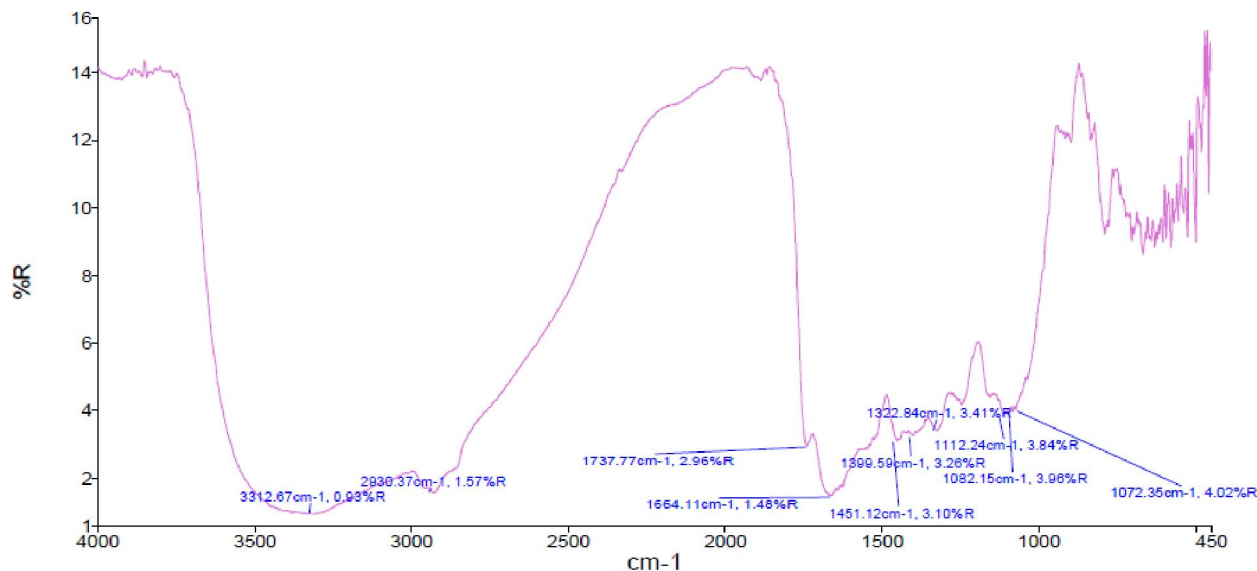


Figure 5 : FTIR spectra of fine spinach powder fraction

ing diverse functions make the developed biomaterial as unique one. This may find the application in fortification of various food products, use as natural nutritional food color, pharmaceutical application and may be used as ready to use ingredient in cousins.

CONCLUSION

Physical, chemical and optical characteristics of spinach leaf powder are particle size dependent. Blanching of spinach prior to dehydration, results in powder of more compact structures. Leaf powder as obtained under the combination dehydration temperature of 80°C as initial and 60°C as finishing temperature without blanching provide a quality powder, which has further improved on fractionation. The fine fraction could be considered as a functional material to be used to make the food nutritionally rich with addition of functionalities.

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