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Research Article

Formulation and preclinical evaluation of Anti-inflammatory activity of *Triticum aestivum*

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Abstract

A wide scope of medical advantages has been credited to wheatgrass, the young grass of the wheat plant *Triticum aestivum*. Wheat grass is a decent wellspring of mineral supplements. It contains critical measures of iron, phosphorous, magnesium, manganese, copper and zinc. Wheatgrass is a rich supplement of tocopherols with high vitamin E content. Wheatgrass is beneficial in restoring more infections due to its significant function that, it can arrest the development of antagonistic microbes which are responsible for spreading certain diseases. constituents of wheatgrass may be obtained from fresh juice, frozen juice, powder, tablets with compositions differing as per their production methods which otherwise depends on growing conditions of wheatgrass. Anti-inflammatory activity of wheatgrass tablets was assessed by using formalin induced rat paw edema model. The results obtained were compared with aceclofenac, standard drug.

Keywords: *Triticum aestivum*, Flaxseed gel, Anti-inflammatory, Aceclofenac, Formalin.

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INTRODUCTION:

The wheatgrass juice (WGJ) contains a high concentration of vitamin C. WGJ contains a lot of highly functional nutritive ingredients potent to unify the liver with the kidneys for detoxification of the organs and filtration of the blood to build a strong immune system. Wheatgrass extract has antimicrobial activities¹. Wheatgrass is known to help wellbeing and insistence both in humans and animals. *Triticum aestivum* has been utilized as natural medication in present and past and is highly esteemed for its therapeutic and nutritional properties². Wheatgrass is a food that is prepared from cotyledons of *Triticum aestivum*. It contains amino acids, chlorophyll, vitamins, minerals and enzymes. It is gluten free. Wheatgrass is a good source of potassium, likewise a generally excellent source of dietary fiber, vit A, vit E (alpha tocopherol), vit K etc. Wheatgrass is likewise very nutritious. Wheatgrass juice supports weight reduction since it is rich in fiber content. Leaves of wheatgrass rises the activities of liver elements, as well as lipid peroxidation³. Wheatgrass is effective in serious cases of stomach ache, gas, paralysis, contamination of stomach, heart attack, diabetes, asthma, constipation, leukemia and other malignancy⁴. Wheatgrass extract is also utilized as effective haemostatic agent, anti-inflammatory agent, stimulant of fibroblasts, with a wide scope of healing properties. Its significant levels of proteins and amino acids work like natural cleanser to detoxify the liver, eliminate poisonous heavy metals from the circulatory system, free the group of squander matter, and hinder the pre maturing

cycle⁵. The anti inflammatory properties of wheatgrass applies a constructive effect on diminishing pain and swelling⁶. The fermented wheatgrass extract improves high threat of survival of skin melanoma patients⁷.

MATERIALS AND METHODS

Extraction of wheatgrass powder

For extraction of wheatgrass powder for manufacturing of wheatgrass tablets, wheat grains should be washed thoroughly and then soaked in water for 6 hrs or over night. Later the water must be removed from grains and it should be kept in cotton cloth for 12 hrs until it gives sprouts, these sprouts should be spread in the tray containing soil. Use the spray bottle filled with water to lightly sprinkle on the soil before going to bed, so the seedlings will be moist over night. Grass is usually ready to harvest after 9 to 10 days of growth. It must be cut at the edges so that it can give the second crop. Rinse the grass lightly as it doesn't need heavy washing as it is grown from organic seeds from organic soil or compost. Juice it in a blender and strain it to remove the solids. This juice is filtered in a vacuum filtration to remove the particles. The wheat grass juice thus obtained is transferred to a spray drier to get powdered wheat grass.

Preparation of wheatgrass tablets

Flaxseed gel is added to the spray dried powder which acts as a binder in the preparation of tablets by wet granulation method. The tablets were compressed after addition of other excipients.

Table 1: Formulation table

Ingredients	F1	F2	F3	F4	F5	F6
Spray dried wheat grass powder(mg)	100	200	250	300	400	500
Flaxseed gel	q.s	q.s	q.s	q.s	q.s	q.s
Starch(gm)	0.025	0.025	0.025	0.025	0.025	0.025
Magnesium stearate (gm)	0.01	0.01	0.01	0.01	0.01	0.01
Talc(gm)	0.0075	0.0075	0.0075	0.0075	0.0075	0.0075

Evaluation of prepared formulations:**Hardness**

The hardness of a tablet was tested using Pfizer hardness tester⁸.

Friability

The friability of the tablets was calculated by using a Lab India tablet friability tester. Tablets were taken, weighed and initial weight was noted (W_0). The tablets were allowed to rotate in the drum at 25 rpm for 4 minutes and weighed (W) again. Percentage friability was calculated⁹.

Content uniformity

20 tablets were crushed in mortar and pestle, and weighed powder equivalent to 10mg of drug was taken and dissolved in 10ml of 0.1N HCL and the solution was then filtered through membrane filters of 0.45 μ m, after suitable dilution with 0.1N HCL, the drug content was spectrophotometrically analyzed at λ max 249 nm.

Invitro disintegration

The *invitro* disintegration was as carried out using a Lab India tablet disintegrating tester (Model No. 251 DT1000). Six tablets are placed in each tube of disintegrating test apparatus and the time for disintegration was noted at $37 \pm 0.5^\circ\text{C}$ using 900 ml of 0.1 N HCL.

Invitro drug release profile

The *in-vitro* dissolution was carried out using paddle apparatus, the dissolution medium was 0.1N HCL, the temperature was maintained at $37 \pm 0.5^\circ\text{C}$, and 50 rpm. Aliquots of 5ml samples were withdrawn at predetermined time intervals of 5, 10, 15, 20, 25, 30 minutes and maintained sink condition with the same dissolution medium. The withdrawn samples were filtered through a membrane filter of 0.45 μ m and analyzed by using a UV- spectrophotometer at 249 nm¹⁰.

RESULTS AND DISCUSSION**Table 2: Pre compression parameters for the formulation of F1 to F6**

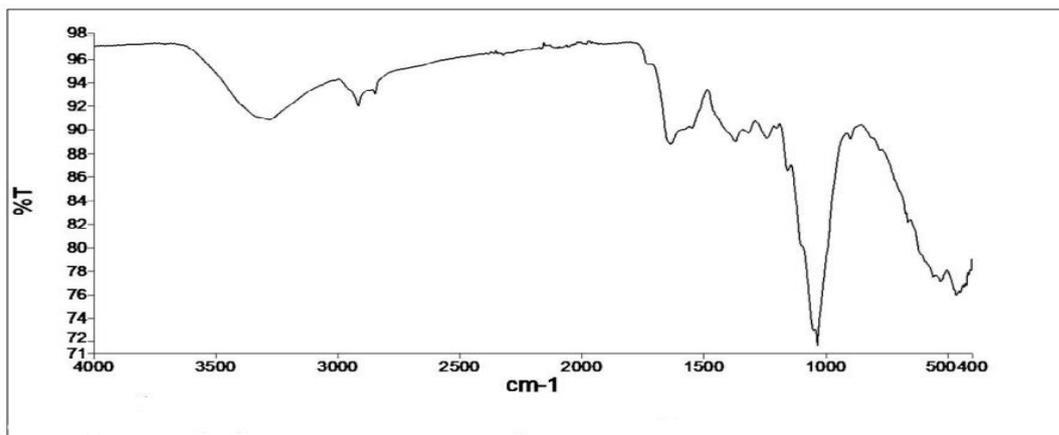
Formulation code	Angle of repose	Bulk density	Tapped density	Carr's compressibility	Hausner's ratio
F1	23.41 \pm 0.97	0.24 \pm 0.03	0.28 \pm 0.04	8.82 \pm 0.09	1.05 \pm 0.01
F2	24.58 \pm 1.22	0.27 \pm 0.03	0.27 \pm 0.04	6.01 \pm 1.26	1.09 \pm 0.01
F3	22.98 \pm 0.71	0.27 \pm 0.04	0.31 \pm 0.05	10.75 \pm 1.12	1.12 \pm 0.01
F4	23.81 \pm 1.89	0.27 \pm 0.03	0.29 \pm 0.05	11.40 \pm 2.32	1.12 \pm 0.02
F5	24.56 \pm 1.08	0.26 \pm 0.04	0.31 \pm 0.04	9.67 \pm 3.33	1.10 \pm 0.04
F6	24.24 \pm 0.87	0.25 \pm 0.03	0.30 \pm 0.04	11.71 \pm 0.16	1.13 \pm 0.02

Table 3: Post compression parameters for the formulation of F1 to F6

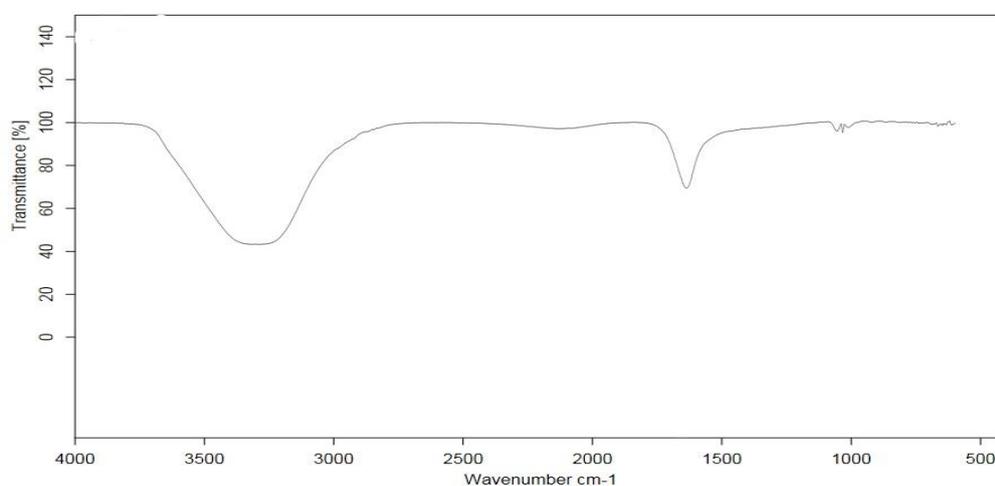
Formulation code	Hardness (kg/cm ²)	Friability (%)	Content uniformity	Disintegration time (sec)	Percent drug release
F1	4.3 \pm 0.11	0.71 \pm 0.04	98.1 \pm 1.14	7min 50 sec	96.62 \pm 0.75
F2	4.1 \pm 0.25	0.74 \pm 0.09	98.9 \pm 0.78	7min 10 sec	93.5 \pm 0.83
F3	4.6 \pm 0.54	0.72 \pm 0.02	98.6 \pm 0.58	6min 8 sec	97.6 \pm 0.51
F4	4.4 \pm 0.54	0.78 \pm 0.01	97.1 \pm 1.54	7min 6 sec	98.7 \pm 0.41
F5	4.3 \pm 0.51	0.75 \pm 0.01	98.2 \pm 1.15	8min 10 sec	97.6 \pm 0.42
F6	4.5 \pm 0.23	0.69 \pm 0.04	99.2 \pm 1.24	6min 4 sec	98.9 \pm 0.51

Fourier transform infrared (FT-IR) studies

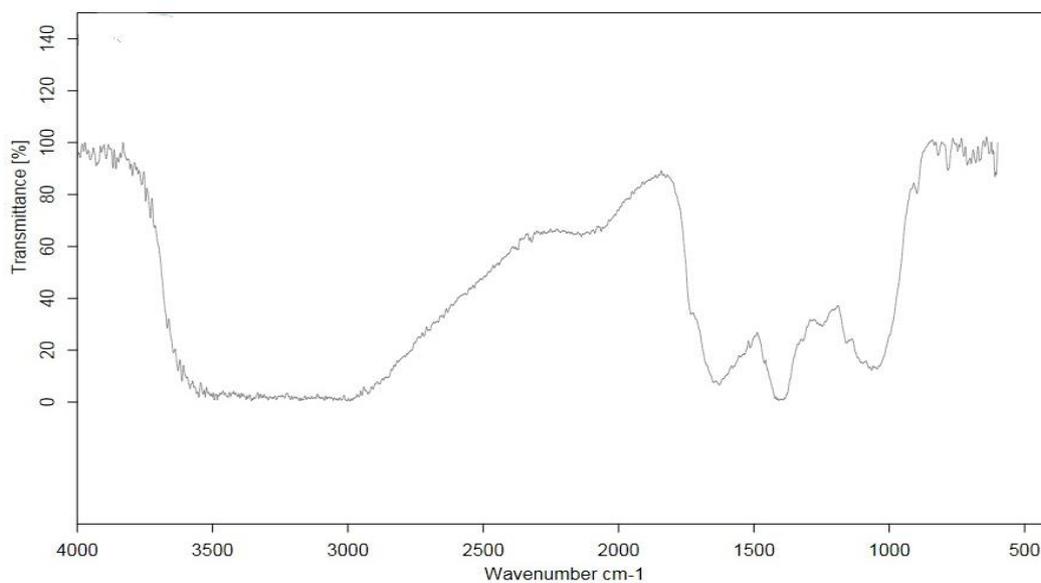
The FTIR spectrum of pure wheatgrass exhibited characteristic signals at 3282.98 cm^{-1} (O-H stretching vibrations), 2917.68 cm^{-1} (N-H stretching vibration), 1637.27 cm^{-1} (C=C stretching vibration), 1052.48 cm^{-1} (C-O stretching vibration).



The FTIR spectrum of flaxseed gel exhibited characteristic signals at 3384.28 cm^{-1} (N-H stretching vibration), 1636.34 cm^{-1} (C=C stretching vibration).



The FTIR spectrum of wheatgrass and flaxseed granules exhibited characteristic vibrations at 3356.04 cm^{-1} (N-H stretching vibration), 1408.16 cm^{-1} (O-H bending vibration), 1065.27 cm^{-1} (C-O stretching vibration).



Preclinical evaluation

Albino wistar rats weighing between 150-200 g kept under standard condition in the animal house of the college were

utilized for the investigation. Every animal was fasted with water over night. Edema was induced using formalin. Formulation F6 with high drug content was selected as the optimized formulation and was taken for preclinical study.

Table 4: Effect of vehicle on formalin induced paw edema in rats

Animal no.	Initial paw volume (ml)	Paw volume after 5 hours (ml)	Difference in paw volume (edema)
1	69	133	64
2	73	142	70
3	72	124	51
4	74	144	69
5	75	135	61
6	71	143	72
Mean \pm SEM	72.33 \pm 0.8819	136.83 \pm 3.156	64.5 \pm 3.17

Number of animals used are 6 in each group

Table 5: Effect of dispersed wheat grass tablet on formalin induced paw edema in rats

Animal no.	Initial paw volume (ml)	Paw volume after 5 hours (ml)	Difference in paw volume (edema)
1	68	81	13
2	74	95	21
3	65	86	21
4	55	74	19
5	69	84	15
6	64	78	14
Mean \pm SEM	65.83 \pm 2.60	83 \pm 2.996	17.166 \pm 1.47

Number of animals used are 6 in each group

Table 6: Effect of aceclofenac tablet on formalin induced paw edema in rats

Animal no.	Initial paw volume (ml)	Paw volume after 5 hours (ml)	Difference in paw volume (edema)
1	73	90	17
2	75	86	11
3	76	90	14
4	80	96	16
5	79	85	6
6	71	81	10
Mean \pm SEM	75.67 \pm 1.406	88 \pm 2.113	12.34 \pm 1.687

Number of animals used are 6 in each group

Table 7: Effect of various treatments on formalin induced paw edema in rats

Treatments	Initial paw volume (ml)	Paw volume after 5 hrs (ml)	Increase in paw volume (ml)	% Inhibition
Control	72.33 \pm 0.8819	136.863 \pm 3.156	64.5 \pm 3.17	-
Aceclofenac	75.67 \pm 1.406	88 \pm 2.113	12.34 \pm 1.687	80.87
Wheatgrass tablet	65.83 \pm 2.60	83 \pm 2.996	17.166 \pm 1.47	73.379

DISCUSSION

The beneficial effect of wheatgrass in skin diseases and anti-inflammatory activity was studied using formalin-induced rat paw edema as the experimental model in the present project. Results of the study have shown that the dispersed tablet of wheatgrass possess a significant anti-inflammatory activity against chronic paw edema induced by formalin. Maximum anti-inflammatory activity was observed after 5 hours of administration of the wheatgrass formulation.

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