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WHEATGRASS JUICE AND ITS NUTRITIONAL VALUE AS AFFECTED BY SPROUTING CONDITION

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Dina A. Anwar²; A. Abou El-Yazied¹; Thanaa, F. Mohammadi² and M.M.F. Abdallah¹

Horticulture Department, Faculty of Agriculture, Ain Shams University, Cairo, Egypt
 Regional Center for Food and Feed, Agri. Res. Center, Giza, Egypt

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ABSTRACT

Wheatgrass juice is the young grass of the common wheat plant (Triticum aestivum) freshly juiced for human consumption. The objective of the investigation performed was to assess the nutritional value of wheatgrass juice under laboratory and open field conditions at two different cuts. Protein, chlorophyll contents, minerals content (Ca, Fe, Mg, Zn and Se) and amino acids content as well as phytochemical constituents were determined. Grown wheatgrass at laboratory caused an increase of the protein content of its juice over open field condition. High chlorophyll content was observed under open field especially at second cut. Most of minerals content underwent to increase under open field except Mg content. Aspartic acid was recorded the highest amino acid in both laboratory and open field. Total essential amino acids were increased under open field condition at both first and second cut followed by first cut at laboratory. No big changes of natural phytochemicals constituents can shown between laboratory and open field condition while it was more pronounced compare with wheat seeds. The study suggested that sprouting wheat seeds at laboratory and open field improve the nutritional value of grass juice with preferably to laboratory condition especially at first cut and for saving agricultural land.

INTRODUCTION

Wheatgrass juice (WGJ) is an extract squeezed from the mature sprouts of wheat seeds

(Received 22 January, 2015) (Accepted 29 January, 2015) (Triticum aestivum). Wheatgrass, young grass of common wheat plant, is freshly juiced or dried into powder for animal and human consumption - both the forms provide chlorophyll, minerals K, Ca, Fe, Mg, Na and S, vitamins such as A, B, C and E, enzymes and 17 forms of amino acids(Singh et al 2012). Germination is a natural process occurred during growth period of seeds in which they meet the minimum condition for growth and development (Sangronis et al 2006). During soaking and germination for producing sprouts, seeds loss dry matter. Chavan and Kadam (1989) stated that the original dry weight of the seeds decreased during soaking and subsequent sprouting process due to leaching of materials and oxidation of substances from the seeds. Chung et al (1989) measured 9.4% decreased in dry matter of sprouted barley seeds over 5 days, while Abdallah et al (2014) found 14.8% loss for 7 days. During this period the chemical composition changes drastically because biochemical activity produces essential compounds and energy (Moongngarm and Saetung (2010). Wheatgrass can be traced back in history over 5000 years, to ancient Egyptian and perhaps even early Mesopotamian civilizations. It is purported that ancient Egyptians found sacred the young leafy blades of wheat and prized them for their positive effect on their health and vitality. Wheatgrass can extracts for consume by two different types like squeezed or chewed then throw out. Wheatgrass has quickly become "the new age espresso" offered in smoothies and juices, salads, and even in powders and tablets and is one of the cereal grasses mostly used as a health drink (Ben-Arye et al 2002). Wheatgrass juice will provide you with more energy by fulfilling nutritional deficiencies and by removing wastes that clog your cells, blood, tissues and organs (Mujoriya and Ba-

buBodla, 2011). Wheatgrass is a complete food that contains bioflavonoids, proteins and other important nutrients and helps in maintaining body functions (Mogra and Rathi, 2013). A very few publications are reported in scientific literature on nutritive and antioxidant properties of wheat sprout extracts where it is reported that these extracts inhibit the DNA oxidative damage and effective in suppressing superoxide radical that can further lead to various diseases (Falcioni et al 2002). At present, the wheatgrass is available in the form of products such as healthy diets (powders, tablets) in USA, East Asian countries and Eastern Europe; however, it's not popular in Egypt and lack knowledge about growing conditions for good quality of wheatgrass.

To solve these problems and to make wheatgrass juice widely consumed, this research work has been carried out. Thus, the main objectives of the study were: (1) growing wheatgrass under different condition and their effect of its quality (2) evaluate the nutritional value of formed grass juice.

MATERIALS AND METHODS

Materials

Grains of wheat were obtained from Field Crop Research Institute, Agricultural Research Center. Rice and wheat straw have been collected from a private farm located in Sharkia Governorate. Wheat and rice straw were sun dried and chopped (2-4 cm) according to **Mohammadi and Abdallah** (2007).

Wheat sprout production methods

Production method for wheat grains sprout was tray method as described by Abdallah (2008) using about 250 gm of rice or wheat straw on the bottom as sprouting media. The plastic tray size was (40 x 24 x 11 cm). Grains of wheat were cleaned, washed and soaked in tap water placed in 0.7L. capacity glass jars for each replicate. The soaked period was 12 hr. (over night) to allow for initial germination before spread evenly on the wheat or rice straw medium as described by Mohammadi and Abdallah (2007). Wheat shoots were harvested after 8 days from grain sowing. The harvested shoot yield per unit area (m²) and per unit volume of seeds (kg) was calculated. Ten sprouts were randomizaly were chosen from each tray for sprout characters measurements.

Experimental trials

Many tray experiments were conducted in organic vegetable sprouts experimental laboratory in Horticulture Department, Faculty of Agriculture – Ain shams University during 2013-2014. From the growing media experiment (data not shown), rice straw media recorded the highest yield and wheat sprout characters. Therefore, it was selected as a medium from the following experiments. In seeding density experiments, four densities were studied (155.6, 116.7, 77.8, 38.9 g/tray). The 116.7 gm were selected for its best yield and shoot characters (data not shown).

1. Open field versus laboratory sprout production

Recommended rice straw medium with seeding density were used in laboratory to study number of shoot cuts and its juice production compared to open field wheat shoot production on one square meter plot area (clay soils) located in the vegetable experimental farm of the Faculty Agriculture, Ain shams university using seeding rates about 162 gm of dry grains per square meter in ten rows (one meter length). Shoots were harvested twice, the first cut when shoot length were more than 10 cm height (when shoots develop a split as another leaf emerges). The shoots of second cut were harvested when shoots reached about 10 cm height. Shoot characters in each cut and its juice volume and nutrition value were measured.

2. Nutrient composition of wheatgrass juice

Collected wheatgrass juice from first and second cuts after squeezed (either under laboratory or open field condition) were placed in a container for cold storage (in a common refrigerator or freezer) during analysis period. The major chemical constituents that make wheatgrass a valuable food such as protein according to AOAC (2012 a). Total chlorophyll according to Lichenthaler and Wellburn (1983), amino acids according to AOAC (2012b), minerals according to AOAC (2012c), phytochemicals constituent according to Santana et al (2013) were analyzed.

Statistical analysis

The production data were statistically analyzed by analysis of variance using completely randomized design and least significant difference (L.S.D)

Wheatgrass Juice and its Nutritional Value as Affected by Sprouting Condition 39

at 0.05 levels according to the method described by **Snedecor and Cochran (1980).**

RESULT AND DISCUSSION

1. Effect of open field versus laboratory production on wheat grass sprout characters

As a result of growing locations, it could be noticed that daily wheatgrass shoot height was significantly increased when produced grasses under laboratory condition in compare with open field condition (**Table, 1**) shoot weight (mg) per day was increased sharply under open field condition (11 mg) over laboratory (5.0 mg).

These result may be due to that low light (indirect sun light) under laboratory condition with resulted in etiolated growth and increased shoot height and may reduced seedling dry weight and that can be reflect to fresh seedling weight adverse to open field. The increase in shoot weight would be brought significant increase in fresh yield of shoots (kg/kg of seeds) or growing grass in open field not only increase shoot weight but also it caused an increase in fresh yield of wheatgrass shoots (kg/kg of seeds). Conversely, higher yield of fresh shoots (g/m2/day) was recorded at laboratory which reached (372.9 g) in compare with open field (24.53 g). This increment may be related to the differ of sowing method between laboratory and open field. While it was in rows at open field, it was sprinkled in trays at laboratory and this can decline the growing area per square meter at open field compare with laboratory (5 times increased in area) in addition to higher seeding density in laboratory (37.5 bold).

According to Schnabel's research on wheatgrass grown outdoors, the environment in which wheatgrass grows affects its vitality. The wheatgrass sown through the winter and harvested at the jointing stage has maximum concentration of active principles. At this stage the plant reaches its peak nutritional value; after jointing, concentrations of chlorophyll, protein, and vitamins decline sharply. Thus wheatgrass is harvested just prior to this jointing stage, when the tender shoots are at their peak of nutritional potency. Wheatgrass grown outdoors is harvested, dehydrated at a low temperature and sold in tablet and powdered concentrates. Growing wheatgrass indoors usually requires the grass to be grown in small trays with the wheat grains close together for a high yield (Singh et al 2012).

Quality and quantity of fresh wheatgrass juice and its residues per square meter also affected by growing locations (Table 2). At laboratory, the growth of wheatgrass in square meter by added five trays vertically, volume of juice and residues weight were built up under laboratory and the increment reached 1688 and 1855%, respectively. However, no significant effect in juice (ml) and residues weight g/kg of seeds/day between laboratory and open field was observed over open field. Concerning cutting effect, first wheatgrass cut was an efficient growth which was the highest in all shoot characters in addition to volume juice and their residue. This result can be expected because seeds were exhausted most of its storage materials at primary growth (first cut). For about interaction between growing locations and number of cutting (Tables 1 and 2) there is no significant variation in shoot height and juice's residues weight (per square meter and kg of seeds). However, higher shoot weight (mg) was recorded under field condition at first cut.

Fresh shoot yield (g/kg of seeds/day) and grass juice (ml/ kg of seeds/day) were increased at first cut under field condition but it is not significantly differ with first cut under laboratory condition. Nevertheless, fresh shoot yield per square meter and their juice were raised under laboratory over open field condition Tables (1 and 2). Variation in growth could be seen with changing conditions and environmental parameters were considered important for the growth of wheatgrass namely temperature, humidity, air circulation and time taken to reach a target height (Ashish et al 2012). Finally, wheatgrass was successfully grown under laboratory for its highly fresh wheatgrass yield in addition this production method can saving use of land and irrigation water. Concerning cutting effect, first grass cut was an efficient growth which was the highest in all shoots character in addition to volume of juice and their residues. This results can be expected because seeds were exhausted most of its storage materials at primary growth (first cut).

2. Effect of sprouting on wheatgrass seedlings weight and development

Soaking of seeds for 12 hr. enhanced fresh weight of dry seeds by 68% (Fig. 1). During growth of wheatgrass under non nutritional water culture, fresh seedling weight was increased in linear curve with growing days to be at the highest value after

Charae	Characters		Shoot weight	Fresh shoots	Fresh shoots weight g/kg of seeds/day	
Location (L)	Cutting (c)	Shoot height Shoot weig cm/ day mg/day		weight g/m2/day*		
Laboratory	First cut	1.38	6.0	565.7	93.22	
	Second cut	0.52	3.0	180.1	29.64	
	Mean	0.95	5.0	372.9	61.43	
Open field	First cut	0.77	14.0	33.23	102.5	
	Second cut	0.57	7.0	15.82	48.77	
	Mean	0.67	11.0	24.53	75.64	
Mean	First cut	1.08	10.0	299.5	97.86	
	Second cut	0.54	5.0	97.96	39.21	
	L	0.025	0.33	19.113	4.154	
L.S.D (0.05)	С	0.025	0.33	19.113	4.154	
	LxC	NS	3.74	NS	14.70	

Table 1. Effect of growing locations and number of cutting and their interaction on wheatgrass yield

*m²= 5 roof in lab exp. Only

Table 2. Effect of growing locations and number of cutting and their interaction on wheatgrass juice and residues

Characters		Juice	e(ml)/day**	Residues fresh weight (g)/day		
Location (I)	Cutting (c)	Cutting (c) m ^{2*} kg of s		m ² *	kg of seeds	
Laboratory	First cut	249.7	41.1	226.2	37.21	
	Second cut	79.2	13.1	71.47	11.76	
-	Mean	164.5	27.1	148.8	24.49	
On an field	First cut	15.1	46.6	8.451	26.05	
Open field	Second cut	3.4	10.3	6.774	20.89	
	Mean	9.3	28.45	7.613	23.47	
Mean	First cut	132.4	43.9	117.3	31.63	
	Second cut	41.3	11.7	39.12	16.33	
L.S.D (0.05)	L	8.48	NS	7.999	NS	
	С	8.48	1.996	7.999	1.612	
	L x C	237.86	12.416	NS	NS	

*m²= 5 roof in lab exp. Only

**Juice ml/kg shoots (F.W.)

Arab Univ. J. Agric. Sci., 23(1), 2015

40

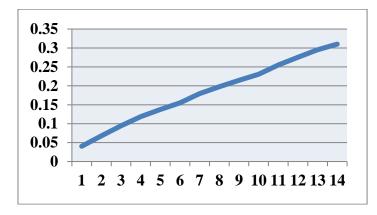


Fig. 1. Fresh seedling weight (gm)

twelve-sprouting days old (study period).From observed data, it was revealed that spread increment of fresh seedlings weight initiated with the first stages of seedling development followed by small increase until twelve days old. Increase in fresh weight denoted the growth (continuously increase of shoots and roots).

Depression in dry seedling weight was observed during growth of wheatgrass seedling started with first to twelfth days old of sprouting (Fig. 2). As growth of seedlings goes up, decrement in dry weight would be increase and this result would be expressed as the percentage of loss in dry weight (Fig. 3). After soaking seeds, 1% of dry seedling weight would be lost and these losses of dry weight continued during growing period to be 39% after twelfth growing day old.

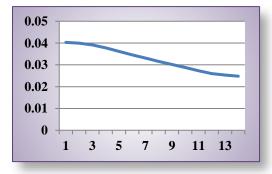


Fig. 2. Dry seedling weight (gm)

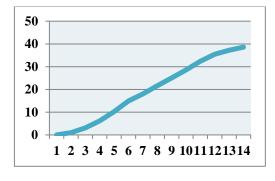


Fig. 3. % loss of dry matter

Decrement in dry weight may be due to that soaking and germination processes caused leakage of storage materials of seeds and no dry substance would be built. These results agreed with Morgan et al (1992) who showed that during soaking and germination, seeds lose dry matter as they use their own energy reserves for growth in the short growing cycle. There is most commonly a DM loss ranging from 7% to 47% and suggested that photosynthesis is not important for the metabolism of the seedlings until the end of day 5.When the chloroplasts are activated, light did not have a significant effect on DM content and losses continued to increase from a value of 5.2% after 3 days to 12.3% after 6 days, probably reflecting the losses due to respiration and negligible amount of photosynthesis by young seedlings at the low light intensity (800 lux).

3. Effect of sowing location, cutting and their interaction on protein content of wheatgrass juice residues

42

As affecting of growing location, higher content of protein of this factor was found in wheat grass's residues produced at open field condition (Table 3) however, low residues weight (4.3 kg) can provide one kilogram of protein content compared with laboratory condition (4.6 kg) on a dry weight basis. The influence of cutting on protein content was proved unequivocally. Dry residues collected from second cut were 259 gof protein while it was 204 g at first cut on a dry weight basis. Thus, big amount of residues were needed (4.9 kg) to obtained one kilo gram of protein at first cut compared with second cut (3.9kg). For about the interaction of growing location and number of cutting, data were in favor of residues collected from squeezed grass grown in open field at second cut which had the highest protein content over others and an opposite quantity of residues content.

4. Effect of growing locations and number of cutting on protein and chlorophyll contents of wheatgrass juice

Table (4) revealed the contents of protein and chlorophyll of wheatgrass juice grown under laboratory and open field conditions at first and second cut.

Grown wheatgrass in laboratory improve protein content of its juice either at first or second cut than open field condition. However, wheatgrass juice at second cut under laboratory condition contained higher protein content (3.39 g/100 ml) compared with first cut (3.03g / 100 ml). Conversely, on open field, grass juice at first cut contained high protein content (2.62g) over second cut (2.01g). These results encouraged the consumption of wheatgrass juice for its protein content under laboratory and open field preferably at laboratory for its high protein content and the raw materials are cheap and available throughout the year and saving the land for producing strategically crops.

Plant based food and food products are the major source of nutrients such as proteins and chlorophyll, one of such main source is wheatgrass and its juice which is consumed for healthy growth of human body (Ashish et al 2012). Concerning storage proteins in wheat grains, Glutens play significant role in technological and nutraceutical of cereal grains. However, the gluten fraction was reported to be responsible for celiac disease and later on food allergy (Larre et al 2011).on the other

hand, **Michalcova et al (2012)** showed that remarkable degradation of glutens started after three days and the lowest concentration was measured at the seventh day of germination of wheat grain. Therefore, wheatgrass juice can be recommended for celiac diseases patients to replace wheat bread nutrients and phytochemicals (need more research).

Influence of growing location and number of cutting were also affected the content of chlorophyll of wheatgrass juice (Table 4). Maximum chlorophyll content was determined from wheatgrass juice at second cut under open field condition (0.466 mg/ml). On the other hand, wheatgrass juice contained 0.424 mg of chlorophyll at first cut as grown on open field which it was the highest value in compare with laboratory condition either at first cut (0.223 ml) or second cut (0.138 ml). Wheatgrass juice is one of the best sources of chlorophyll and most of its health benefits are attributed to this chlorophyll, besides other vital nutrients (Mogra and Rathi, 2013). The high chlorophyll content under open field condition related to direct sunlight effect along growth period (10 days old) compare with laboratory (8 sprouting days old under indirect sunlight). On the other hand, grown in soils and consumed raw, contamination with bacteria or other substances may be a concern women who are pregnant or breast-feeding should not use wheatgrass (Mogra and Rathi, 2013). Therefore, growing in laboratory using sterilized rice straw medium can be recommended in this study.

Reference mean daily requirement of protein intake was 56 mg/day for male, 46 mg/day for female and 19 mg/day for children (DRI, 2002). Table (4) revealed that 100 ml of fresh wheatgrass juice under laboratory condition can supply about 5.4, 6.6 and 16% at first cut and 6.1, 7.4 and 17.8% at second cut for male, female and children, respectively, which be higher than the open field juice supply. Higher intake of protein was reported in children followed by females and males.

Finally, an idea could be implemented in future to introduce wheatgrass juice as a health drink.

5. Effect of growing locations and number of cutting on minerals content of wheatgrass juice

Table (5) represents the contents of Ca, Fe, Mg, Zn and Se of wheatgrass juice after first and second cut under laboratory and open field condition. The content of Ca of wheatgrass juice at second cut was found to be higher than that in the

Chara	cters	Residues dry	Protein (g)/kg of	
Location (I)	Cutting (c)	weight(g)/kg of protein	residues dry weight	% Protein
Laboratory	First cut	4975	201	20.1
Laboratory	Second cut	4132	242	24.2
	Mean	4554	222	22.2
Onen field	First cut	4866	206	20.6
Open field	Second cut	3623	276	27.6
	Mean	4245	241	24.1
Mean	First cut	4921	204	20.4
wean	Second cut	3878	259	25.9
	L	0.8593	0.384	
L.S.D (0.05)	С	0.8593	0.384	
	LxC	1.2153	0.543	

 Table 3. Effect of growing location, cutting and their interaction on protein content (kg) of wheatgrass juice residues

Table 4. Effect of growing locations and number of cutting on protein and chlorophyll contents of wheatgrass juice

			Protein				
Location	Cutting	Chlorophyll (mg/ml)	g/100 ml -	% DRI of 100 ml of grass juice			
				Male	Female	Children	
Laboratory	First cut	0.223	3.03	5.41	6.59	15.95	
	Second cut	0.138	3.39	6.05	7.37	17.84	
Open field	First cut	0.424	2.62	4.68	5.70	13.79	
	Second cut	0.466	2.01	3.60	4.38	10.60	

Table 5. Effect of growing locations and number of cutting on Minerals content (mg/l) of wheatgrass juice

		La	aboratory		Open field			
Minerals	mg/l	%DRI of 100 mI of juice		mg/l	%DRI of 100 mI of juice		nl of juice	
		Male	Female	Children		Male	Female	Children
				First cut				
Calcium	200	2	2	2	500	5	5	5
Ferric	63.6	80	35	64	44.96	56	25	45
Magnesium	2252	56	73	173	1089	27	35	84
Zinc	8.81	8	11	18	5.28	5	7	11
Selenium	0.0074	1.3	1.3	2.5	0.016	2.9	2.9	5.3
				Second cut				
Calcium	300	3	3	3	7500	75	75	75
Ferric	44.10	55	25	44	133.9	167	74	134
Magnesium	398	10	13	31	1186	30	38	91
Zinc	7.55	7	9	15	11.9	11	15	24
Selenium	ND	-	-	-	ND	-	-	-

first cut. On the other hand, grown wheatgrass on open field caused an increase on its content of Ca over laboratory either first or second cut. Highest Fe content was observed under field condition after second cut (133.9 mg/l) followed by first cut under laboratory (63.6 mg/l).Fe concentration of juice from first cut (field) and second cut (laboratory) were found to be almost constant around 44.0 mg/l. Lap tap water analysis for minerals content was 33.8 and 12.8 ppm for Ca and Mg, respectively while Fe was not detected (Anwar et al 2009). The wheatgrass juice contained 2252 mg per liter of Mg at first cut under laboratory condition which be higher than those contents at first and second cut under open field condition (1089 and 1186 mg, respectively) and second cut under laboratory (398 mg/l) (Table 5). Maximum and lower content of Zn was recorded at second and first cut under open field (11.9 and 5.28 mg/l, respectively) (Table 5). Wheatgrass juice contained 16 mg/l of Se if grass were produced under open field at first cut where as it was 7.4 mg under laboratory condition in the same first cut. Moreover, no Selenium content was detected in the juice obtained from second cut either under laboratory or open field condition (Table 5).

Previous data can revealed that growing wheatgrass under open field especially at second cut caused an increase in most of minerals juice content and these results can be explained to the higher chlorophyll juice content under open field condition at second cut (Table 4). Chlorophyll can be extracted from many plants, but wheatgrass is superior because it has been found to have over 100 elements needed by man. If grown in organic soil. It absorbs 92 of the known 102 minerals from the soil. Wheatgrass contains over 90 minerals, including high concentrations of the most alkaline minerals: potassium, calcium and magnesium. Consumption of 100 ml of fresh wheatgrass juice under laboratory at first cut supply about 2% of daily intake of calcium while it was 5% under open field condition at the same cut(Table 5) and this percentage rose at the second cut to be 75% of the daily intake. Dietary Recommended Intakes (DRI) of iron was 8.0 mg for male, 18 mg for female and 10 mg/day for children (DRI, 2004). As high recommended iron intake for female compare with male and children, grass juice can supply only about 35% and 25% of iron at first cut under laboratory and open field, respectively compared with 80%, 56% and 64%, 45% for male and children under laboratory and open field, respectively.

Same trend would be finding at the second cut to be 25% and 75% for female compared with 55%, 167% and 44%, 134% for male and children under laboratory and open field, respectively. Cereals exhibit inhibition of dietary iron absorption due to the endogenous phytate contained in their outer coats. Removal of the phytate in bran by different methods such as fermentation, germination and baking increases iron absorption by approximately 3.5 times (Hallberg et al 1986). At first cut, 100 ml of juice can supply 56, 73 and 173% of Mg; however, it was 27, 35 and 84% under open field for male children, respectively. Conversely, at second cut, grass juice supply about only 10, 13 and 31% under laboratory condition while it was 30, 38 and 91% of Mg under open field for male, female and children, respectively. Grass juice also can supply about 5 to 18% of Zn at first cut and 7 to 24% at second cut. Regarding selenium DRI data in Table (5) showed that grass juice under laboratory at first cut will covered 1.3%, 1.3% and 2.5% while it was 2.9%, 2.9% and 6.3% under open field at the same cut for male, female and children, respectively. Yoshida et al (2007) showed that the availability of selenium in the radish sprout ranged between 33 and 65 %. When the amount of selenium was increased to 2 µg/g both the selenite form and selenium taken in with the radish sprout prevented the tumour cells from growing. Hama et al (2008) examined that Japanese radish sprout on the oxidative stress of female rats, the selenium diet of 12.5 mg/kg reduced the increase of the body mass but increased the mass of the liver.30 ml of freshly squeezed wheatgrass juice is equivalent in nutritional value to 1kg of leafy green vegetables. Wheatgrass has quickly become "the new age espresso" offered in smoothies and juices, salads and even in tablets and powders and is one of the cereal grasses mostly used as a health drink (Ben-Arye et al 2002).

6. Effect of growing locations and number of cutting on the composition of amino acids of wheatgrass juice

The compositions of amino acids were expressed in terms of 100 g proteins (16 g N) (Table 6). In present work, the amino acid contents of wheat juice were differed slightly among laboratory and open field conditions. Aspartic acid recorded the highest concentration of amino acids of juice under laboratory condition either at first or second cut On the other hand, the first cut under open field

Sowing locations	Lab	oratory	Open field		
Cutting	First cut	Second cut	First cut	Second cut	
Non essential amino acids					
Aspartic acid	16.50	18.29	10.69	11.94	
Alanine	4.95	3.24	6.87	5.97	
Glutamic acid	8.91	5.60	12.21	10.45	
Serine	3.63	2.65	4.58	3.98	
Total	33.99	29.78	34.35	32.34	
Conditionally essential amino acids					
Argenine	5.28	3.24	6.87	5.97	
Proline	3.63	2.06	4.20	4.48	
Glycine	3.96	2.36	6.11	4.98	
Cysteine	0.99	1.18	1.15	1.49	
Tyrosine	0.99	1.77	1.91	2.49	
Total	14.85	10.61	20.24	19.41	
Essential amino acids					
Isoleucine	3.63	2.36	4.58	4.48	
Therionine	3.96	2.36	3.44	4.48	
Valine	4.62	3.24	6.11	5.47	
Phenylalanine	4.29	2.95	5.34	5.47	
Lysine	5.61	3.83	6.49	6.47	
Leucine	6.27	3.54	9.16	7.96	
Methionine	1.32	0.59	1.53	1.99	
Histidine	2.31	2.65	3.05	2.49	
Total	32.01	21.52	39.70	38.81	

Table 6. Effect of growing locations and number of cutting on the composition of amino acids of wheatgrass juice per 16 g of N $\,$

or laboratory condition increased the essential amino acids as compared with second cut and the data was more pronounced with lysine and leucine amino acids. However, Glutamic, Alanine and Serine were the highest non essential amino acids in first cut in both open field and laboratory conditions. Similar increment in first cut in both open field and laboratory were observed for two conditionally amino acids Arganine and Glycine (Table 6). Chavan and Kadam (1989) stated that an increase in proteolytic activity during sprouting is desirable for nutritional improvement of cereals because it leads to hydrolysis of prolamins and the liberated amino acids such as glutamic and proline are converted to limiting amino acids such as lysine. Lysine is an essential amino acid; diets deficient in lysine can impair growth in children and reduce immune function.

Usual doses of lysine supplementation range from 0.5 to 4.0 g/day. Lysine and arginine share a common transport system for intestinal absorption and uptake into cells of the body and brain. Because arginine competes for uptake with lysine, a high ratio of lysine/arginine in the diet enhance the effects of lysine supplementation (Zimmermann, 2001). However, differences in the amino acids content per 16 g N can revealed that (1) most of amino acids of juice were increased if grass are grown under open field over laboratory either at first or second cut. (2) Grass juice at first cut was succeeded high amino acids content in compare with second cut especially in essential amino acids.

7. Effect of growing locations on the percentage of natural phytochemicals of wheat seeds and wheatgrass juice

Wheat seeds showed a predominance of saturated aliphatic alkanes Nonacosane (21.1%), Triacontane (19.9%), Tetratriacontane (18.9%), Heneicosane (12.8%) and Docosane (12.3%). As it can be seen from **Table (7)**, it is clearly visible that sprouting had evidence influenced on wheat seeds

 Table 7. Effect of growing locations on the percentage of phytochemicals of wheat seeds and wheatgrass juice

Percentage of phytochemicals	Dry wheat seeds	Laboratory grass juice	Open field grass juice
Eicosane	0.76	0.66	0.70
Tetracosane	4.96	3.66	4.37
Heneicosane	12.78	11.06	11.68
Triacontane	19.90	18.63	19.52
1,3-Dioctadecyloxypropane	0.28	-	-
Nonacosane	21.09	20.22	21.50
2-Methyldocosane	0.63	0.40	0.40
Tetratriacontane	18.91	17.89	17.16
3-Ethyl-5-(2'-ethylbutyl)octadecane	0.59	0.58	0.38
1-[1-Methyl-2-(octadecyloxy)ethoxy]octadecane	0.39	0.23	0.37
Docosane	12.31	11.27	10.29
Hexadecanoic acid,2(octadecyloxy)ethyl ester	0.66	-	-
3-Methylheneicosane	0.41	-	-
Octacosane	6.33	5.06	4.51
Arachic acid	-	1.89	0.44
2,6-Di-tert-butylhydroquinone	-	0.84	0.01
Tritetracontane	-	3.15	-
3,7,11,15-Tetramethyl-2-hexadecen-1ol	-	2.07	0.41
Methyl 5,11,14-eicosatrienoate	-	0.18	0.33
Stearic acid, 2-(octadecyloxy) ethyl ester	-	2.21	2.97
Squalane	-	-	4.96

natural phytochemical constituents. 1, 3-Dioctadecyloxypropane, 3-Methylheneicosane and the fatty ester Hexadecanoic acid, 2(octadecyloxy) ethyl ester were recorded 0.28%, 0.66% and 0.41%, respectively in seeds, however, it is not detected or may be traces in wheatgrass juice.

Alkans play a role, if a minor role, in the biology of the three eukaryotic groups of organisms: fungi, plants and animals (Hendey, 1964), they protect the plant against water loss, prevent the leaching of important minerals by the rain, and protect against bacteria, fungi, and harmful insects (Baker, 1982). The higher melting point of these alkanes can cause problems at low temperature and in Polar Regions. In latter function they work at the same time as anti-corrosive agents, many solid alkanes find use as paraffine wax. Nonacosane is a straight-chain hydrocarbon with a molecular of C₂₉H₆₀, and evidence suggests it plays a role in the chemical communication of several insects (Beri et al 2004) Nonacosane has also been identified within several essential oils. It can also be prepared synthetically (Bentley et al 1955). Conversely, Arachic acid, Stearic acid. 2-(octadecyloxy) ethyl ester, Tritetracontane, 2,6-Ditert-butylhydroquinone, 3,7,11,15-Tetramethyl-2hexadecen-1ol and Methyl 5,11,14-eicosatrienoate were found at wheatgrass juice either under laboratory or open field conditions while it is not presence (or traces) in wheat seeds(Table 7). On the other hand, Squalane was absence in laboratory grass juice while it was 4.96% of total phytochemical constituents in open field grass juice.2,6-Di-tertbutylhydroquinone is a highly effective antioxidant (Richard, 2008). In foods, it is used as a preservative for unsaturated vegetable oils and many edible animal fats (Almeida et al 2011). It does not cause discoloration even in the presence of iron, and does not change flavor or odor of the material to which it is added (Richard, 2008). It can be combined with other preservatives such as butylatedhydroxyanisole (BHA). It is added to a wide range of foods, with the highest limit (1 g/kg) permitted for frozen fish and fish products. Its primary advantage is enhancing storage life. Squalane is a hydrocarbon and triterpene derived by hydrogenation of squalene, most notably in the livers of sharks (Allison, 1999). Squalane has low acute toxicity and is not an irritant at the concentrations used in cosmetics.

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