IJFANS

Volume 01, Issue 01, Oct-Dec 2012, www.ijfans.com ISSN: 2319-1775

International Journal of Food And Nutritional Sciences

Official Journal of IIFANS

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IJFANS International Journal of Food And Nutritional Sciences

INTERNATIONAL JOURNAL OF FOOD AND NUTRITIONAL SCIENCES

ISSN 2319 – 1775 www.ijfans.com
Volume 01, Issue 01, Oct-Dec 2012

RESEARCH PAPER OPEN ACCESS

IMPACT OF SUPPLEMENTATION OF HEALTH DRINK ON HAEMATOLOGICAL PROFILE AND SERUM RETINOL LEVEL OF ADOLESCENT GIRLS

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ABSTRACT

Sixty adolescent girls in the age group of 16 to 18 years were selected from Punjab Agricultural University, Ludhiana to determine the impact of supplementation of health drink using under utilized foods on the nutritional status of adolescent girls. Health drink was prepared by using whey water, pearl millet, broccoli leaf powder, banana and jaggery at three different levels i.e. S 1, S 2, and S 3.The developed health drink was organoleptically evaluated by using nine-point hedonic scale and the highest overall acceptability scores was observed for S1 level with 5 g Pearl millet, 2.5 g Broccoli leaf powder, 50 g Banana, 10 g Jaggery, and 82.5 ml whey water. The most acceptable level was chemically analyzed for proximate composition, available carbohydrates, vitamins and minerals and anti-nutritional factors by using standard methods. The subjects were divided into two groups i.e. control group who were observed without the supplementation of health drink and experimental group who were supplemented with 150 ml of developed health drink for a period of three month.. The percent increase in serum retinol level, blood heamoglobin, mean corpuscular volume(MCV), packed cell volume (PCV), mean corpuscular haemoglobin concentration (MCHC) and total iron binding Capacity (TIBC) was 4.65, 7.13, 4.88, 5.56, 2.35 and 4.88 percent, respectively was observed in the subjects after the supplementation of health drink. Hence, it can be inferred from the results that supplementation of health drink improved the nutritional status of the adolescent girls.

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KEY WORDS:
Adolescent girls, banana, broccoli leaves, health drink, jaggery, pearl millet, whey water, serum retinol level, blood heamoglobin, mean corpuscular volume(MCV), packed cell volume (PCV), mean corpuscular haemoglobin concentration (MCHC) and total iron binding Capacity (TIBC).

INTRODUCTION

Adolescence being a transition period between childhood and adulthood makes particularly girls vulnerable to malnutrition because of body's increased nutritional needs imposed by physical changes taking place during this period. With the onset of adolescence, the need for iron increases as direct consequence of rapid growth and the expansion of blood volume and muscle mass. Compared to adolescent boys, girls are at high risk of developing iron deficiency anaemia as the onset of menstruation imposes additional iron needs for girls. Compared to adolescent boys, girls are at high risk of developing iron deficiency anaemia as the onset of menstruation imposes additional iron needs for girls. Thus increasing the iron status in girls through supplementation and maintaining it through continued emphasis and attention towards developing adequate diet and good nutrition habit through dietary counseling can be the effective and important measure of iron deficiency anemia. The young girls who are at the brink of womanhood constitute the most crucial segment of our population from the point of view of the quality of our future generation (Kapil et al 1993). Any nutritional deficiency experienced during this critical period of life can have an effect on the future health of the individual and their offspring as adolescent girls are the "future mothers". Thus increasing the iron status in girls through supplementation and maintaining it through continued emphasis and attention towards developing adequate diet and good nutrition habit through dietary counseling can be the effective and important measure of iron deficiency anemia. Any nutritional deficiency experienced during this critical period of life can have an effect on the future health of the individuals. Food based strategy is used as a tool for combating micronutrient deficiencies. It is also referred as dietary modification, which encompasses a wide variety of intervention that aim at increasing the production, availability and consumption of food products, which are rich in micronutrients (Joshi and Mehta 2010). Many underutilized foods like whey water, pearl millet and broccoli leaves which are available at low cost can be used to improve the nutritional status of the adolescent girls. Whey usually contains 6.0 percent of milk solids, made up largely of lactose, together with small quantities of protein and fat. It also contains most of the water soluble B complex vitamins of milk. Pearl millet is particularly rich in iron and zinc and has high levels of fat and considered to have one of the best protein quality or amino acid score as compared to other cereals. Broccoli leaves (Brassica oleracea var. italica) are available at no cost and is very rich in all the micronutrients like β carotene (16000 IU), vitamin C (93.2 mg), calcium (48mg). It is an exceptionally nutritious vegetable with a variety of potential uses.

MATERIALS AND METHODS

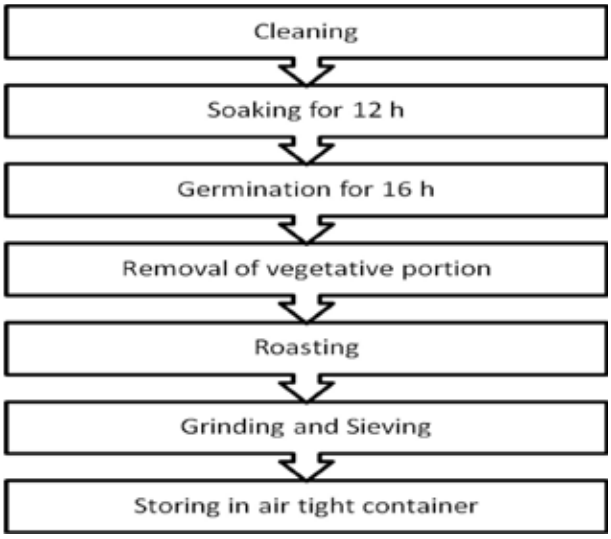
PROCUREMENT AND PROCESSING OF THE RAW MATERIAL

Broccoli leaves were procured by contacting the local vegetable grower. Whey water was procured on daily basis from the local dairy shop. Pearl millet, Bananas and jaggery were obtained from the local market of Ludhiana.

Preparation of broccoli leaf powder: The leaves were washed, blanched in boiling distilled water for 10-15 sec and dried at room temperature for 1-2 h by spreading on filter paper followed by drying in hot air oven at 50±5 °C for 48+6 hours. The dried leaves were grounded to fine powder.

MALTING OF PEARL MILLET

Pearl millet grains were cleaned, soaked in water for 12 hours, germinated for 36 hours at room temperature (27°C) and dried in hot air oven at 60° C for 8 hours and then seedlings were removed by hand rubbing to prepare malt (Malleshi and Desikachar 1981). The malt was roasted in skillet at slow heat till the aroma developed (5 min for 100 g of grain), ground in electric mixer to obtain fine flour. This whole flour was used for the development of health drink.



DEVELOPMENT AND STANDARDIZATION OF HEALTH DRINK

The health drink was prepared in the Food Laboratory of Department of Food and Nutrition, College of Home Science by using whey water, pearl millet, broccoli leaf powder, banana and jaggery at three different levels i.e. S1, S2, and S3.

S 1: 5 g Pearl millet, 2.5 g Broccoli leaf powder, 50 g Banana, 10 g Jaggery, and 82.5 ml whey water was added to make it 150 ml.

S 2: 5 g Pearl millet, 3 g Broccoli leaf powder, 50 g Banana, 10 g Jaggery, and 82.5 ml whey water was added to make it 150 ml.

S 3: 5 g Pearl millet, 3.5 g Broccoli leaf powder, 50 g Banana, 10 g Jaggery, and 82.5 ml whey water was added to make it 150 ml.

ORGANOLEPTIC EVALUATION OF DEVELOPED HEALTH DRINK

The organoleptic evaluation was done to select the most acceptable level of ingredients used in the development of health drink. The panel of judges including faculty of Department of Food and Nutrition and students were provided with score card of Hedonic Rating Scale to score the test samples for their color, flavor, texture, taste and overall acceptability. The drink was prepared and for sensory parameters twice. Mean scores and standard error were calculated after the second trial.

CHEMICAL ANALYSIS OF DEVELOPED FUNCTIONAL BEVERAGE

The most acceptable level was analyzed chemically for the following:

- Proximate composition (AOAC 1990).
- Total soluble sugars (Dubois et al 1956).
- Reducing sugars (Nelson 1944) and Non-reducing sugars (by calculation).
- Mineral content: Iron (AOAC 2000) and calcium (AOAC 1980).
- Vitamin content: β-carotene (Rao 1967) and vitamin – C (AOVC 1996)
- Anti-nutritional factors: Phytin Phosphorous (Haung and Lantzsch 1983), Total Phenols (AOAC 2000) and Oxalate (Abeza et al 1968)

SELECTION OF SUBJECTS

A group 0f 60 Adolescent girls ageing between 16 to 18 years and having heamoglobin level less than or equal to 10g/dl were selected from Punjab Agricultural University, Ludhiana. Selected 60 adolescent girls were divided randomly in two groups of 30 subjects each i.e. one control and the other as the experimental group and all the observations were recorded before the study and after three months study period. The subjects in control group were observed without the supplementation and in the experimental group the subjects were supplemented with 150 ml of developed health drink for a period of three month following five days a week pattern.

OBSERVATIONS RECORDED

Blood samples of the subjects were collected to analysis the following parameters:

- Haemoglobin (Dacie and Lewis 1975)
- PCV (Raghuramula et al 2003)
- MCV (by calculations , PCV) RBC count * 10
- MCHC (Raghuramula et al 2003)]
- TIBC (Teitz 1976)
- Serum retinol level (Raghuramula et al 2003)

RESULTS AND DISCUSSION

The result of the undertaken study has been discussed under the following headings:

- Organoleptic evaluation of the developed health drink
- Nutritional evaluation of the developed health drink
- Effect of health drink supplementation on haematological profile of the subjects
- Effect of health drink supplementation on serum retinol level of the subjects

ORGANOLEPTIC EVALUATION OF THE DEVELOPED HEALTH DRINK

The data of organoleptic evaluation has been presented in table 1.Three different levels of health drink were prepared using broccoli leaf powder, pearl millet, jaggery, banana and whey water. The developed health drink was organoleptically evaluated by a panel of judges from the department of food and nutrition and students by using nine-point hedonic scale to judge the acceptability of the product.

The scores given by the trained panel for colour, flavor, texture, taste as well as overall acceptability of the developed health drink varied from 7 to 7.4, 6.5 to 7.6, 6.6 to 7.4, 6.3 to 7.5 and 6.5 to 7.7, respectively. According to the trained panel the most acceptable level of drink was having 2.5 g broccoli leaf

powder, 5 g pearl millet, 10 g jaggery, 50 g banana and 82.5 ml whey. It had the overall acceptability score of 7.7+0.18.

According to the scores given by the student's panel, the scores for colour, flavor, texture, taste as well as overall acceptability of the developed health drink varied from 5.9 to 7.2, 6 to 7.8, 5.9 to 7.4, 5.8 to 7.9 and 5.8 to 7.8, respectively. The overall acceptability was highest for the same recipe i.e. S1 level with acceptability score as 7.8±0.18. Both the panels gave the highest overall acceptability scores to the drink with 2.5 g broccoli leaf powder, 5 g pearl millet, 10 g jaggery, 50 g banana and 82.5 ml whey water.

According to trained panel a non-significant difference was observed in colour and texture of the three levels.

NUTRITIONAL EVALUATION OF THE DEVELOPED HEALTH DRINK

The developed health drink was analyzed chemically for proximate composition, available carbohydrates, mineral content, vitamin content and anti-nutritional factors by using standard methods. The values have been calculated for 100 ml of health drink.

Proximate composition of developed health drink

Developed health drink had 61.43 g of moisture, 2.9 g of crude protein, 0.4 g crude fat, 1.2 g crude fibre, 2.2 g ash, 31.87 g carbohydrates and provided 142.68 Kcal of energy (Table. 2).

Available carbohydrates

Developed health drink had 21.7 g of total sugars, 7.9 g of reducing sugars, 13.8g of non-reducing sugars and 9.8 g starch (Table. 3).

Mineral and vitamin content

The concentration of iron and calcium in developed health drink was 2.96 mg and 167 mg respectively. The concentration of vitamins in developed health drink was 5.6 mg of vitamin C and 1568 µg of β- carotene (Table. 4).

Anti-nutritional factors

Developed health drink had 32.68 mg of phytin phosphorus and 18.2 mg of total phenols and 22.5 mg of oxalates (Table. 5).

HEMATOLOGICAL PROFILE OF THE SUBJECTS BEFORE AND AFTER SUPPLEMENTATION OF HEALTH DRINK

The data pertaining to hematological profile of the subjects is given in Table 6.

Heamoglobin (Hb)

The mean heamoglobin of the subjects in control and experimental group before the study was 9.89±0.12 and 9.81±0.12 g/dl and after the three months of study period the corresponding value came out to be 9.99±0.12 and 10.51±0.15 g/dl in control and experimental group respectively. The Hb of the subjects was less than the reference Hb value of 12 g/dl (WHO 2001). However, the heamoglobin level of the subjects revealed a significant ($p<0.01$) rise after the supplementation of developed health drink. Harsh (1992) and Singla (2011) also reported a haemoglobin level of 10.97 and 9.14 g/dl respectively, in adolescent girls. Batra (2009) also reported that the mean heamoglobin level was 9.9±1.0 and

10.7±0.09 g/dl in vegetarian and non-vegetarian girls.

Packed Cell Volume (PCV)

The value of mean PCV of the subjects in control and experimental group before the study was 33.70±0.55 and 32.74±0.40 percent. The corresponding values after the study period were 32.99±0.52 and 34.34±0.39 percent. The reference PCV range is 37-44 percent (Dacie and Lewis 1974). The corresponding value of PCV obtained was below the normal range indicating anaemic condition of the subjects. However, the statistical analysis revealed a significant ($p<0.01$) increase in PCV after the supplementation to the experimental group. Malkit (1987) reported a range of 33.9 to 35.0 percent of PCV of the girls. Kaur (2009) also reported that the mean PCV values in adolescent girls were 32.5±0.55 which was comparable to the present study.

Mean Corpuscular Volume (MCV)

The initial mean MCV value of the subjects in control and experimental group was 80.11±1.53 and 77.86±1.67 fl. The corresponding values after the three month study period were 78.25±1.72 and 82.19±1.73 fl, in control and experimental group respectively. The normal MCV range is 77-95 fl (Dacie and Lewis 1974). The corresponding value of MCV before and after the study was in-between the normal range. There was a significant ($p<0.01$) increase in the MCV value of the subjects after the supplementation of health drink to the experimental group. Kaur (2009) conducted a study in Ludhiana and reported the MCV values of 85.52 fl and 86.90 fl. Al –Sayes et al (2011) reported that the mean MCV values in adolescents was 76.24±7.95 fl in South Arabia.

Mean Corpuscular Haemoglobin Concentration (MCHC)

The initial mean MCHC value of the subjects in control and experimental group was 29.5±0.37 and 28.86±0.27 percent and the corresponding values after the study period were 29.9±0.31 and 29.54±0.22 percent, in control and experimental group respectively. The normal MCHC range is 30-33 percent (Dacie and Lewis 1974). The corresponding value of MCHC after the supplementation was slightly lower than the normal range but there was a significant ($p<0.01$) increase seen in the MCHC value of the subjects after the study period. Singla (2011) reported the MCHC levls among adolescent girls as 30.97±2.63 %.

Total Iron Binding Capacity (TIBC)

The initial mean TIBC value of the subjects in control and experimental group was 262.23±9.86 and 266.2±10.67 µg/dl. The TIBC of the subjects after the study period was 263.13±8.52 and 279.2±10.37 µg/dl, in control and experimental group respectively. There was a significant ($p<0.05$) change in the TIBC of the subjects after the supplementation programme. It was observed that the TIBC value was in the normal range. The normal TIBC range is 250-416 µg/dl (Goodhart and Shills 1990). Bains (1995) reported that the mean value of TIBC for young girls was 429µg/dl.

SERUM RETINOL LEVEL OF THE SUBJECTS BEFORE AND AFTER THE SUPPLEMENTATION OF HEALTH DRINK

The data regarding glucose and serum retinol level of the subjects is shown in Table 7.

Serum retinol level

The mean serum retinol level of the subjects in control and experimental group before the study period was 23.87±0.32 and 24.07±0.41 µg/dl, respectively. After the three months study period the corresponding values were 24.20±0.37 and 25.19±0.47 µg/dl in control and experimental group respectively. The normal value for serum retinol level is 25µg/dl. There was a significant ($p<0.01$) increase in the serum retinol level after the supplementation programm. The increase could be due to the consumption of broccoli leaf powder in the form of developed health drink.

CONCLUSION

The present study was conducted to see the impact of supplementation of health drink using underutilized foods on the nutritional status of adolescent girls. A group of sixty adolescent girls in the age group of 16-18 years were selected from Punjab Agricultural University, Ludhiana.

The subjects were divided into two groups i.e. control group and experimental group. All the observations were recorded before the study and after three months study period. The subjects in control group were observed without the supplementation and in the experimental group the subjects were supplemented with 150 ml of developed health drink for a period of three month following five days a week pattern.

Blood samples of the subjects were collected for the blood analysis of the hematological profile, and serum retinol level, before the study, and after the three months experimental period.

Health drink was prepared by using whey, pearl millet, broccoli leaf powder, banana and jaggery at three different levels i.e. S1, S2 and S3. The S1 level was prepared by using 5 g pearl qq31aqqmillet, 2.5 g broccoli leaf powder, 50 g banana, 10 g jaggery, and 82.5 ml whey was added to make it 150 ml whereas in S2 and S3 level, the amount of broccoli leaf powder was increased to 3 ad 3.5 g, respectively.

The average Heamoglobin value of the selected subjects were 9.89 and 9.81 in control and experimental group which was 17.58 and 18.25 per cent respectively below the reference value i.e. 12 g/dl.After supplementation to the experimental group, the percent increase in average heamoglobin and Serum retinol level was found to be 7.13 and 4.65 per cent, respectively. The percent increase in mean corpuscular volume, packed cell volume and mean corpuscular haemoglobin concentration and TIBC was 4.88, 5.56, 2.35 and 4.88 percent, respectively.

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Table 1: Organoleptic evaluation of developed health drink

S. No	Color	Flavor	Texture	Taste	Overall Acceptability
Trained Panel					
S 1	7.4±0.22	7.6±0.21	7.4±0.18	7.5±0.1	7.7±0.18
S 2	7.2±0.29	7.0±0.31	6.8±0.36	6.7±0.33	6.9±0.29
S 3	7.0±0.24	6.5±0.26	6.6±0.26	6.3±0.26	6.5±0.26
F Ratio	0.56NS	3.64*	2.08NS	4.73*	5.30*
CD	_	0.77	_	0.79	0.71
Students Panel					
S 1	7.2±0.2	7.8±0.18	7.4±0.2	7.9±0.19	7.8±0.17
S 2	6.2±0.36	6.8±0.27	6.6±0.34	6.6±0.28	6.6±0.26
S 3	5.9±0.35	6.0±0.27	5.9±0.34	5.8±0.35	5.8±0.31
F Ratio	4.61*	13.92*	5.75*	13.54*	16.67*
CD	0.89	0.70	0.87	0.82	0.73

S 1 = Health drink with 2.5 g broccoli leaf powder, 5 g pearl millet, 10 g jaggery and 50 g banana and 82.5 ml whey water to make the volume 150 ml.
S 2 = Health drink with 3 g broccoli leaf powder, 5 g pearl millet, 10 g jaggery and 50 g banana and 82.5 ml whey water to make the volume 150 ml.
S 3 = Health drink with 3.5 g broccoli leaf powder, 5 g pearl millet, 10 g jaggery and 50 g banana and 82.5 ml whey water to make the volume 150 ml.

Values are mean± S.E. *significant at 5% level NS- Non-Significant

Table 2: Proximate composition of developed health drink

Proximate Composition	FW (g/100ml)
Moisture	61.43
Crude protein	2.9
Crude fat	0.4
Crude fibre	1.2
Ash	2.2
NFE	31.87
Energy (Kcal)	142.68

Table 3: Available Carbohydrates in developed health drink

Available Carbohydrates	FW (g/100ml)
Total soluble sugars	21.7
Reducing sugars	7.9
Non reducing sugar	13.8
Starch	9.8

Table 4: Mineral and Vitamin content of developed health drink

Minerals and Vitamins	FW (g/100ml)
Iron	2.96
Calcium	167
Vitamin C	5.6
β Carotene(µg/100ml)	1568

Table 5: Anti-nutritional factors of developed health drink

Anti-Nutritional Factors	FW (g/100ml)
Phytin Phosphorus	32.68
Total phenols	18.2
Oxalates	22.5

Table 6: Haemotological profile of the subjects before and after the supplementation of developed health drink

	Before	After	% Change	t-Value	Normal Values
Control Group					
HB, g/dl	9.89±0.12	9.99±0.12	1.01	1.39NS	>12 a
PCV, %	33.70±0.55	32.99±0.52	2.1	1.28 NS	37-44 b
MCV, fl	80.11±1.53	78.25±1.72	2.32	0.86 NS	77-95 b
MCHC, %	29.5±0.37	29.9±0.31	1.35	1.08 NS	30-33 b
TIBC, ug/dl	262.23±9.86	263.13±8.52	0.34	0.16 NS	250-416 c
Experimental Group					
HB, g/dl	9.81±0.12	10.51±0.15	7.13	4.34**	>12 a
PCV, %	32.74±0.40	34.34±0.39	4.88	2.78**	37-44 b
MCV, fl	77.86±1.67	82.19±1.73	5.56	1.90*	77-95 b
MCHC, %	28.86±0.27	29.54±0.22	2.35	1.71*	30-33 b
TIBC, ug/dl	266.2±10.67	279.2±10.37	4.88	1.80*	250-416 c

a WHO (2001) b Dacie and Lewice (1974) c Goodhart and Shills (1990)
*Significant at 5 % **Significant at 1 %
NS Non significant

Table 7: Serum retinol level of the subjects before and after the supplementation of developed health drink

	Before	After	% Change	t-Value	Normal Values
Control Group					
Serum Retinol, ug/dl	23.87±0.32	24.20±0.37	1.38	0.98 NS	25 ug/dl ^
Experimental Group					
Serum Retinol, ug/dl	24.07±0.41	25.19±0.47	4.65	2.88 **	25 ug/dl ^

^ WHO (1998)
*Significant at 5 %
**significant at 1 %
NS Non significant