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Development of an iron-fortified cake as an attempt to prevent anemia in adolescent girls

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Abstract
The unabated rise in the prevalence of anemia in adolescents especially females is one of the major public health concerns worldwide. Amongst other factors, faulty dietary practices, irregular meal patterns, frequent consumption of energy dense food, snacking in between main meals generally accounts for the above. To address this problem an iron-fortified cake was prepared. The variations of non-fortified and iron-fortified cakes were assessed by 50 panel members by the hedonic scale test method. Sensory evaluation assays portrayed the iron-fortified fruit cake to be the best variation in terms of appearance, colour, taste, texture, and odour. Moreover, biochemical estimations showed the iron-fortified cake to have increased amounts of protein, calcium, iron and vitamin C compared to the non-fortified cake. Therefore, the iron-fortified cake may help in preventing anemia in addition to other deficiency disorders among the teenage girls belonging to the middle income group.

Keywords: Fortification, anemia, iron, antioxidant activity, nutritional composition and sensory evaluation.

1. Introduction
Anemia is a disease condition where the hemoglobin level is less than 13 g/dL and 12 g/dL in men and women respectively. According to the World Health Organization (WHO), it is an epidemic afflicting one-quarter of the world's population. One of the major kinds of anemia is nutritional anemia that occurs due to a deficiency of one or more essential nutrients required for the synthesis of hemoglobin and the production of erythrocytes (McEvoy and A. 2013) [26]. According to the studies conducted by the WHO and the International Centre for Research on Women (ICRW), India is one of the countries in the world that has the highest prevalence of anemia accounting to approximately fifty five percent cases (Fund 2004) [36]. Conditions of anemia have majorly been found in the states of Karnataka, Haryana and Chennai and many more (Devi *et al.* 2015 [8]; Kappala *et al.* 2014 [21]; Premalatha *et al.* 2012) [29]. Furthermore, the 2005-2006 reports of the National Family Health Survey clearly documents 56% of adolescent girls to be anemic. The causes of anemia ranges from iron deficiency, inadequate dietary amounts of hematopoietic nutrients such as vitamin A, B2, B12, C and folic acid to severe pathological conditions including malaria, tuberculosis as well as Acquired Immune Deficiency Syndrome (AIDS) (Haidar 2010; Sayes *et al.* 2011) [32].
The word adolescence is derived from the Latin word, "adolescere". The WHO has defined adolescence as the age period between 10 to 19 years of age for both the sexes. Adolescence more broadly refers to the phase of human development which encompasses the transition from childhood to adulthood (health 1999). In females, adolescence marks the beginning of the menstrual cycle. Adolescents gain 30% of their adult weight and more than 20% of their adult height between 10-19 years, which is referred as the growth spurt. Due to this increased growth and physical activity, adolescence demands a rise in nutritional requirements. Therefore, adolescent girls are at a high risk for anaemia and malnutrition. Moreover, inadequate nutrition during adolescence can have serious consequences throughout the reproductive years of life and beyond including morbidity, cognitive impairment, endocrine disorders and decrease in work productivity (Bhaskaram and Reddy 1975; Haas and Brownlie 2001; Walter *et al.* 1983) [4].

Meals are everyday eating events structured not only by the frequency of the food consumption and the food items appropriate to a meal but also by the order of dishes in the menu. According to nutritional parameters, eating events can be categorized as either main meal or snacks. Whereas, the main meal comprises of structured meals, snacks include unstructured eating events between the meals (Zizza *et al.* 2007). Snacking can be an important part of eating and contribute to the overall health when done in the correct way. Snacks may provide long lasting hunger satisfaction and hence help a person to manage the calorie intake by avoiding overeating during the main meals (Kant and Graubard 2015) [20]. Therefore, choosing smart snacks can impact the nutrients received for the day, levels of hunger and energy and of course the risk of long term diseases.

Product and product development is a systematic as well as an oriented research to develop products and processes satisfying a known need. It is a combination and application of both natural food science as well as social sciences (Bhuiyan 2011) [5]. Cake generally serves as an energy dense food and is one of the preferred snack items of the adolescents (Giuliani *et al.* 2013) [11]. The current study therefore is concerned with the development of an iron-fortified cake for prevention of anemia in vegetarian adolescent girls. The physicochemical properties of cakes are largely dependent on the batter and the cake structure. Therefore, understanding the internal macro and microstructures of bakery products becomes essential. Cake ingredients normally include soft wheat flour and variable levels of fat, sugar, eggs, milk, baking powders, and emulsifiers. These agents have an important role to play in formation of the basic structure (Al-Dmoor 2013) [1]. Sugar and fat tenderize the cake, softening, giving the soft structure, and enhancing the flavor. The role of baking powder is to enlarge the bubbles causing the cake to rise to its potential. Flour works as a structure builder as it is involved in forming the crumb and crust structure of most types of cake and is hence considered as a toughener (Doke and Guha 2014) [10]. In addition to the above, the cake may occasionally also consist of ingredients such as cocoa powder, nuts, fruits, icings and certain flavoring agents.

A basic cake production process involves mixing, depositing, baking, cooling and packaging. Starch gelatinization and protein denaturation together with carbon dioxide formation gives cake its porous and soft structure. To produce a cake with an open structure and high volume, a procedure and recipe is needed in order to create a stable batter with many tiny air bubbles. These bubbles act as nuclei and grow in size when the carbon dioxide gas generated from baking powder leavens the product during baking (Kim and Walker 1992) [22].

The WHO and the United Nations Food and Agriculture Organization (FAO) have identified four main strategies for improving micronutrient malnutrition: food fortification, supplementation, nutrition education, and disease control measures. Of these, food fortification has proven to be one of the most cost-effective interventions to advance global welfare. In order to address the nutritional gap, this unique intervention involves adding critical vitamins and minerals to staple foods during the production process, which are commonly and consistently consumed by the target population (Horton 2006) [19]. Fortification provides nutritional benefits without requiring consumers to change their eating habits and often without requiring changes in purchase patterns since it targets foods the population is already consuming thereby overcoming the obstacles often experienced by other interventions such as supplementation and dietary diversity.

The cake prepared in this study contained wheat flour, oats and garden cress in addition to strawberries, rice bran, dates and raisins which were used as fortifying agents.

Wheat is considered good source of protein, minerals, B-group vitamins and dietary fiber therefore becoming the principal cereal for baking (Kumar *et al.* 2011) [11]. Oats scientifically known as *Avena sativa* is normally used as an animal feed (Decker *et al.* 2014) [7]. Nevertheless, the attractiveness of oat as a part of human diet is due to it containing significant quantities of dietary fibres, vitamins, minerals, antioxidants, sterols, proteins of high lysine content as well as oil having a favorable ratio of polyunsaturated to saturated lipid all of which are useful in prevention of significant disorders including cancer and heart disease (Chatuevedi *et al.* 2011) [6].

Garden cress or *Lepidium sativum* has been known to have health promoting properties and hence characterized as a functional food. Garden cress oil has a balanced amount of polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA) (Doke and Guha 2014) [14]. These seeds also possess several medicinal properties and can be used as galactogogue, diuretic, demulcent, aphrodisiac, carminative and antioxidant (Singh *et al.* 2015) [33]. They could also be utilized as a food supplement in human diet since it contains considerable amount of iron, calcium, vitamins A, B, C and E, dietary fibres, fatty acids and antioxidants viz., tocopherols and carotenoids (Divanji *et al.* 2012; Sarkar *et al.* 2014) [9].

Strawberries (*Fragaria sp)* are packed with vitamin-C and folate apart from flavonoids including anthocyanins and quercetin which impart antioxidant and anti-carcinogenic properties (Zheng *et al.* 2007) [38]. Rice bran on the other hand contains an array of neutraceuticals like oryzanols, tocopherols, tocotrienols, phytosterols, 20% oil and 15% protein, 50% carbohydrate (majorly starch,) dietary fibers like beta-glucan, pectin, and gum (Hernandez *et al.* 2000) [17]. They are also a rich source of antioxidants like polyphenols, carotenoids and vitamin E (Henk 2009) [16].

Raisins are dried grapes that are naturally endowed with both an array of valuable nutrients and a pleasant sweet flavour. They provide essential nutrients, soluble and insoluble fibres, health protective bioactive compounds or phytochemicals and flavonoids (Puglisi *et al.* 2008) [30]. It also functions as a source of carbohydrates, iron, vitamins and minerals. Raisins consist of approximately 60% sugars by weight and their sweetness is contributed mainly by glucose and fructose (Kostaropoulos and Saravacos 1995) [23]. Dates (*Phoenix dactylifera*) are one of the members of the palm family Palmae. There are more than two hundred varieties of dates available worldwide with each kind associated with a specific medicinal property (Al Jasser 2009) [3]. Date fruit play a significant role in diseases prevention through its anti-oxidant, anti-inflammatory and anti-bacterial activity (Hong *et al.* 2006 [18]; Mansouri *et al.* 2005) [25]. They are a potent source of essential fatty acids, flavonoids, sugars with low Glycemic Index (GI), dietary fibres, vitamins and essential minerals (Al-Farsi and Lee 2008) [8]. The cake involved in the present study utilized the above ingredients and their properties in order to address the problem of anemia in young girls.

2. Materials and Methods

2.1 Selection of place

The development of the cake was done in the Food and Nutrition Laboratory of J.D. Birla Institute, Kolkata. In order to conduct the survey, a random sampling was done which comprised of 50 vegetarian adolescent girls (15-20 yrs old) belonging to middle income group.

2.2 Selection of sample
While selecting the sample, the purpose was to know the best variation of the cake from 50 panel members comprising of students of the Department of Food Science and Nutrition Management of J. D. Birla Institute. All recipes were evaluated by the same panel members. Further biochemical estimations were carried out only with the best variant of the cake as assessed by the panel members compared to the basic cake. While conducting the survey, the purpose of investigation was to know the knowledge level of the adolescent girls regarding anaemia and the practices followed by the adolescent girls for the prevention of anaemia.

2.3 Construction of questionnaire
Keeping in mind the objective of the present study and to assess the awareness of the panel members about anemia, a questionnaire was prepared. The questionnaire was divided into two parts. The first part dealt with personal information of the panel members including name, age, sex, height, weight and Body Mass Index (BMI). The second part of the questionnaire dealt with basic questions with the purpose of finding out the knowledge level of the respondents about iron deficiency and anemia.

2.4 Sensory evaluation
Sensory evaluation of the final variation compared to the basic recipe was done by the well-known 9- point hedonic scale method developed by David Peryam and colleagues by all the 50 panel members (Peryam and Girardot 1952) [22]. The scale is represented in figure 1.

1-9 HEDONIC SCALE	
1	DISLIKE EXTREMELY
2	DISLIKE VERY MUCH
3	DISLIKE MODERATELY
4	DISLIKE SLIGHTLY
5	NEITHER LIKE NOR DISLIKE
6	LIKE SLIGHTLY
7	LIKE MODERATELY
8	LIKE VERY MUCH
9	LIKE EXTREMELY

Fig 1: 9-point Hedonic scale

2.5 Biochemical estimation of protein
The protein content of the samples was measured by the Biuret method. Bovine Serum Albumin (BSA) (Lobachemie, India) at a concentration of 1mg/ml was taken as the standard protein solution. Standard protein solution of 0.2ml, 0.4ml, 0.6ml, 0.8ml and 1ml was taken in a test tube and the volume was made up to 1ml with distilled water. For the test samples, 1ml of test sample was taken in separate test tubes. 4ml of Biuret reagent (Lobachemie, India) was added to all the test tubes and incubated at room temperature for 30mins. Thereafter, the optical density was recorded using spectrophotometer at 550nm. The concentration of the protein in the unknown samples was determined from the standard curve with BSA.

2.6 Determination of fat content
The fat content was determined by the Soxhlet method. The samples were extracted with petroleum ether in the Soxhlet continuous extractor. Samples were dried; powdered and 5gm of sample was weighed. They were placed in the middle part of Soxhlet apparatus. 150ml of petroleum ether was poured into the bottom flask and heat at 100 ±2 ^{0}C by the electrically controlled mantle heater. The volatile ether vapour ascended through the inlet tube and after condensation in the condenser fell drop by drop into the sample placed in the middle part. When the condensing liquid reached a certain height it went back into the flask through the outlet tube carrying the dissolving oil from the material. After extraction, the bottom flask with the residue was dried in an oven at 50-100°C. It was cooled and weighed to determine the fat content the following formula:
Fat content, percent = [100(B - C)]/A; where
A = Sample weight
B = Weight of flask after extraction
C = Weight of flask prior to extraction

2.7 Carbohydrate estimation
Determination of carbohydrate was done by the Anthrone method. 100mg of sample was taken in a boiling tube and hydrolysed by keeping it in a water bath for 3hrs with 5ml of 2.5N HCL. Thereafter, the sample was and cooled to room temperature and neutralized with sodium carbonate until the effervescence seized. The volume of the sample mixture was made up to 100ml and centrifuged at 3000-4000g. The supernatant was collected and 1ml of aliquot was taken for analysis. To it was added 4ml of Anthrone reagent (Lobachemie, India). It was heated for 8mins in a boiling water bath after which the absorbance was measured in at 630nm. D- Glucose at a concentration of 1mg/ml was used as the standard carbohydrate for the estimation. The concentration of the carbohydrate in the sample was calculated using the standard curve.

2.8 Preparation of ash solution
The ash solution was prepared in order to carry out the mineral estimations of calcium and iron. 20g of the sample was weighed, desiccated and placed in a muffle furnace at 600-800°C. The ash was weighed and divided equally for calcium and iron estimations. The measured ash was dissolved in 5ml of 6M HCL solution and warmed over a water bath and filtered trice through Whatman No 1 filter paper (Lobachemie, India). This ash solution was used for estimation of calcium and iron.

2.9 Determination of Calcium
Calcium content was estimated by the O- cresolphthalein Complexone (OCPC) Method, End point assay. OCPC combines with calcium at alkaline pH to form a purple colour complex, the absorbance of which is measured at 578nm. The recommended volumes of kit reagents (Coral Clinical System, India) were added to the test tubes labelled as Blank, Standard and Test. The reaction mixtures were incubated at room temperature for 5minutes followed by measurement of absorbance at 578nm. The concentration of calcium in mg/dl was determined by the following formula:
Calcium in mg/dl = [Absorbance of Test/ Absorbance of Standard] X 10

2.10 Determination of Iron
Determination of Iron was done by Ferrozine method where the Fe (II) ions react with Ferrozine to form a violet coloured complex. The recommended volumes of kit reagents (Coral Clinical System, India) buffer solution, colour reagent and standard solutions were added to the test tubes labelled as Blank, Standard and Test. The reaction mixtures were incubated at room temperature for 5minutes followed by

measurement of absorbance at 578nm. The concentration of iron in the sample was determined by the following formula:
Iron (µg/dL) = [Absorbance of Test/ Absorbance of Standard] X 200
Iron (µM) = [Absorbance of Test/ Absorbance of Standard] X 35.8

2.11 Vitamin C
Determination of Vitamin C was done by performing redox titration with the dye 2, 6- dichloroiodophenol (DCIP). In order to standardise the dye, 20ml of standard vitamin C solution (5mg/ml) was mixed with 20ml Meta phosphoric acid. 1ml of dye was titrated with this filtrate till the dye becomes colourless. For the unknown sample, 5gm of sample was taken and 50ml of water was added and filtered through filter paper. This solution was titrated against the dye solution until a persistent pink colour was observed. The concentration of vitamin C in the sample was determined by the following formula:
$V_1S_1 = V_2S_2$; where V_1, S_1 and V_2, S_2 represent the volumes and strength of the known and unknown solutions respectively.

2.12 Folic Acid and Energy content
Folic acid and energy content of the samples were calculated using the Indian Council of Medical Research (ICMR) table (Gopalan *et al.* 2012). All results were expressed per 100gms of the sample.

2.13 Microbiological Analysis
Shelf life study was performed. The final product (that is selected on the basis of sensory evaluation) was prepared once again and seal packed. The sealed products were kept under normal storage conditions at room temperatures in zipper pouches to estimate its shelf life and acceptability after storage. For shelf life study enumeration by plate count dilution agar plate technique was carried out. Serial dilutions of the sample were prepared starting from 10^{-1} to 10^{-5}. Each dilution was plated on a Nutrient Agar plate and incubated at 37 ^{0}C for 18-24 hrs. Thereafter the plates were analysed for growth of microbial colonies. The results were expressed as Colony Forming Units (CFU)/ml.

2.14 Analysis of data
The data was tabulated and organized according to the requirement of appropriate analysis. Percentages were calculated on the basis of the questionnaire and data. The results were measured as mean±s.e.m of ≥3 individual experiments. The results have been represented in the form of tables, pie-charts and bar graphs.

3. Results
3.1 Components of the iron-fortified cake vs the non-fortified cake: The basic recipe of the cake contained maida, sugar, butter, milk, baking powder, baking soda and essence of vanilla in the amounts mentioned in Table 1. In order to develop an iron-fortified product, the final variation was fortified with ingredients such as oats, whole wheat flower, roasted and powdered garden cress seeds, powdered rice bran, condensed milk, strawberries, dates, raisins and plain soda (Table 1). All of these ingredients not only enhanced the sensory parameters of the final product but also increased the content of protein, carbohydrate, energy, iron, calcium, vitamin C and folic acid (figure 4 and 5) thereby making it more suited as an energy dense snack for the prevention of anemia in adolescents.

3.2 Sensory evaluation
The basic recipe and the fortified product were both assessed by 50 panel members using the 9- point hedonic scale test method. After the assessment it was found that the iron-fortified cake scored higher in all sensory parameters including appearance, colour, taste, texture and odour compared to the non-fortified cake. The scores for the fortified cake were 8.3, 8.4, 8.25, 8.3 and 8.15 versus 7.55, 7.85, 7.4, 7.45 and 7.9 of the non-fortified cake for appearance, colour, taste, texture and odour respectively (figure 2). The cake was perfectly baked at appropriate baking conditions giving it an appealing appearance and a perfect light brown colour. The chopped raisins and dates decorated at the top of the cake made the cake attractive to the panel members. The garden cress seeds were roasted and then coarsely grinded which resulted in no after taste of the seeds. The fruits namely strawberries, dates and raisins added extra sweetness to the cake. Moreover, the amounts of the liquid ingredients used were brought down in this variation, which gave a good texture to cake without making the cake soggy. The odour of the garden cress seeds was subdued by the odour of vanilla essence and strawberries which resulted in pleasant odour. Hence, the fortified variant of the cake scored an overall rating of 8.37 compared to the basic cake that scored 7.9 (figure 2).

3.3 Consumer Survey Questions
In order to assess the awareness and knowledge level of the panel members about anemia, a questionnaire of twenty three questions was prepared. This study showed that the females exhibited higher levels of health consciousness compared to the males. The increase in health consciousness for females can be attributed to social and psychological factors that influence the way females think about their body shape and health in general. The results indicated that a large number of female respondents were concerned about their health and were interested in acquiring information about a health topic (figure 3A). It was found that adolescent girls were skipping meals more than adolescent boys especially the breakfast (figure 3B). The tendency of skipping meals was also more prominent in girls due to increasing trend to lose weight in addition to lack of appetite and time. It was also observed that most of the panel members had a fair knowledge of anemia as depicted in figure 3C, 3D, 3E and 3F. Seventy four percent of these members consumed three major meals and one snack in a day (figure 3G). Iron sources for eighty two percent of the respondents consisted of green leafy vegetables and pulses that constituted two major meals of the day for eighty percent of them (figure 3H, 3I and 3J). Figure 3K – 3R displays the knowledge level of the members about iron and anemia which included questions about haem iron, symptoms of anemia, food cravings, treatment, role of vitamin C in iron absorption and effect of exercise on anemia respectively. Moreover it was shown that these members were aware of at least one iron-fortified product (figure 3S). Figure 3T and 3U manifests the response of the panel members regarding the effect of beverages and family history respectively on anemia. However, none of them were aware of any anemia related government programme (figure 3V) but were of the opinion that counselling, health education programmes and training could help the adolescents in understanding and preventing anemia (figure 3W).

3.4 Estimation of food macromolecules and energy
After evaluation for the sensory parameters, both the variations of cake were biochemically assessed in order to study its

nutritional implications. The cakes were estimated for carbohydrate, protein, fat and the energy content. The fortified cake showed a carbohydrate content of 30mg versus 17mg for the non-fortified product (figure 4A). It was observed that the fortified cake displayed 13.63mg of protein compared to the basic cake that exhibited 7.43mg protein as estimated by the Biuret method (figure 4B). The incorporation of rice bran, oats and wheat might have contributed to the increased protein and carbohydrate content of the final variation. However, both the products showed similar fat percentages which were 12.3 % and 12.8% for the fortified and basic cake respectively as estimated by the Soxhlet method (figure 4C). On the whole the iron-fortified cake was shown to be more energy dense having 1202.05 kcal energy per 100gm compared to the non-fortified cake which possessed only 1137.3 kcal energy per 100gm as calculated by the ICMR table (Gopalan *et al.* 2012) [12] (figure 4D). Rice bran, whole wheat and oats being energy dense cereals contributed to enhancing the energy content of the iron-fortified cake. Moreover, addition of raisins, dates as well as condensed milk also played a significant role in increasing the energy content of the final variation of the cake. The experimental results are displayed as mean±s.e.m of atleast 3 independent experiments.

3.5 Determination of vitamins and minerals

The products were also assayed for minerals like iron and calcium. The non–fortified cake contained 80.06µg/dl of iron in contrast to the iron-fortified cake which contained 225.09µg/dl of iron as determined by the Ferrozine method which may be due to the incorporation of garden cress seeds, rice bran, dates and raisins in the final variation (figure 5A). Moreover, the non- fortified cake showed 2.142mg/dl of calcium as milk and refined flour was the only source of calcium in it compared to the iron-fortified cake which displayed 13.366mg/dl of calcium as the cake was fortified with oats, flour, garden cress seeds; all of which contributed to an increase in the calcium content of the cake (figure 5B). Since vitamin C plays a significant role in absorption of iron, attempt was made to increase the vitamin C content of the fortified cake and the same was also biochemically estimated. It was observed that the fortified cake showed 200mg/gm of vitamin C versus 120mg/gm for the basic recipe (figure 5C). The strawberries incorporated in the iron-fortified cake may be responsible for increased the Vitamin C. The folic acid content were 10.74µg and 3.4µg per 100gm for the non-fortified and iron-fortified cake respectively as estimated using the ICMR table (figure 5D). Therefore, the iron-fortified cake would serve as a better snack compared to the non-fortified cake for prevention of anemia in adolescents. The experimental results are displayed as mean±s.e.m of at least 3 independent experiments. *, **and ***denote P value <0.05, <0.005 and <0.001 respectively.

3.6 Estimation of microbiological shelf life

Both the iron-fortified and the non-fortified products were subjected to microbiological shelf life study. Serial dilutions of the sample were prepared starting from 10^{-1} to 10^{-5}. Each dilution was plated on a Nutrient Agar plate and incubated at 37 ^{0}C for 18-24 hrs. Thereafter the plates were analysed for growth of microbial colonies. The results were expressed as Colony Forming Units (CFU)/ml. Although initial dilutions displayed some microbial growth, further dilutions did not show any significant microbial colonies in both the samples thereby making it acceptable from the aspect of microbiological shelf life (Table 2). -, +, and ++ indicate 0, 1-10 and >10 CFU/ml.

4. Discussion

Iron is essential to all cells. Functions of iron include involvement in energy metabolism, gene regulation, cell growth and differentiation, oxygen binding and transport, muscle oxygen use and storage, enzyme reactions, neurotransmitter synthesis, and protein synthesis. Adolescents are vulnerable to iron deficiency because of increased iron requirements related to rapid growth. Iron needs are highest in females during peak pubertal development because of a greater increase in blood volume, muscle mass and myoglobin (Mesías *et al.* 2013) [27]. After menarche, iron needs continue to remain high in females because of menstrual blood loss.

In spite of increased iron needs, many adolescents, particularly females, may have iron intakes of only 10-11 mg/day of total iron, resulting in approximately 1 mg of absorbed iron. About three -fourths of adolescent females do not meet dietary iron requirements (U.S. Department of Agriculture 2012) [34]. Iron therapy, in combination with dietary strategies to increase iron and vitamin C intakes can effectively treat iron deficiency anemia by raising the haemoglobin level and replacing iron stores. Since snacks form an important part of the meal which is also desired especially by the adolescent age group, an iron-fortified cake was prepared to address the above issue. The products were assessed by 50 panel members by the 9 point hedonic scale test method. The structured questionnaire helped in attaining information regarding lifestyle and eating habits of the respondents. The iron-fortified variation of fruit cake was better accepted because all the sensory parameters like appearance, colour, taste, texture, odour was good compared to the standard recipe.

Moreover, biochemical estimations showed the final fruit cake fortified with garden cress seed and rice bran powder to have increased amounts of protein, calcium, iron, folic acid and vitamin C compared to the non-fortified cake. The fruits present in the cake like dates, raisins and strawberries contributed to the increase in vitamin C content which may henceforth help to absorb the iron obtained from garden cress and rice bran and thereby enable the adolescent girls to meet their dietary requirements (Hallberg *et al.* 1989) [15]. Moreover, one of the key ingredients in the fortified cake; garden cress seeds also functions as a galactogogue. Hence this cake can also be consumed by lactating women where it may help them in maintaining their iron requirements in addition to aiding in lactation. Therefore, this iron-fortified product may serve as a nutritional snack and along with Nutritional Education Programmes may help in prevention as well as treatment of anemia.

5. Conclusion

Prevalence of anemia in adolescent girls is alarmingly high. This study was conducted to assess the knowledge and practice regarding prevention of anemia among adolescent girls and to develop an iron fortified cake for the same. The study showed all the subjects to possess average knowledge and practice regarding diet and disease. The fortified cake was an energy dense snack with good amounts of iron, Vitamin C and folic acid apart from other macro and micro nutrients. Adolescents, especially vegetarians are usually susceptible to low iron intake as they are dependent on non-heme sources of iron, which are not readily absorbed and they do not usually contain the large amount of iron as is offered by heme sources. Food fortification and food supplementation are important alternatives that may help to satisfy the iron needs. Hence, this fortified cake may serve as a healthy snack option to prevent anemia among the adolescent girls in addition to meeting their dietary requirements.

Figures

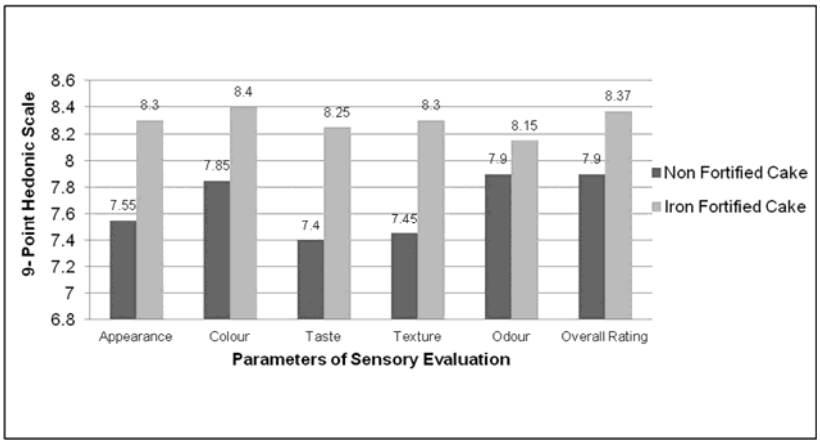

Fig 2: Sensory evaluation. The basic recipe and the fortified product were both assessed by 50 panel members using the 9- point hedonic scale test method. The iron-fortified cake scored higher in all sensory parameters compared to the non-fortified cake.

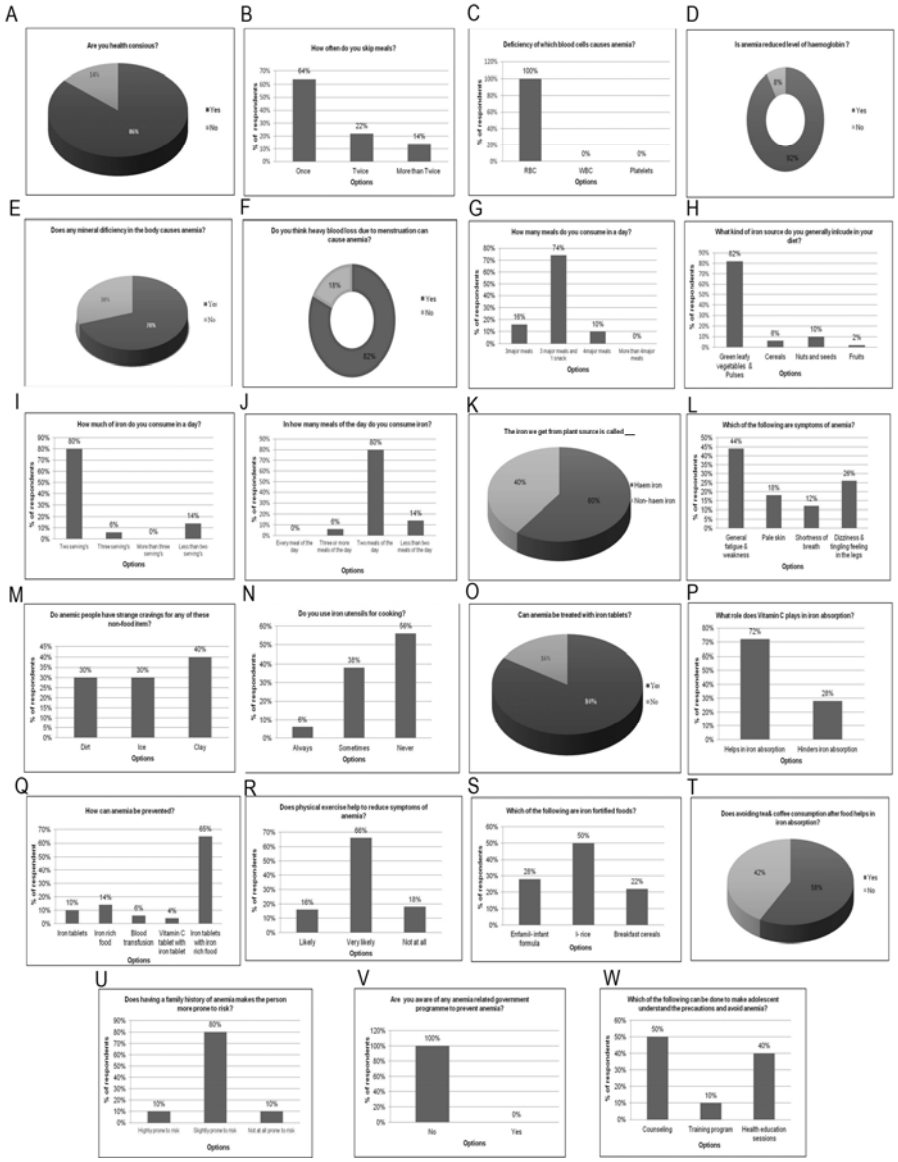

Fig 3: Consumer Survey Questions. A questionnaire of twenty three questions was prepared in order to assess the awareness and knowledge level of the panel members about anemia (A-W).

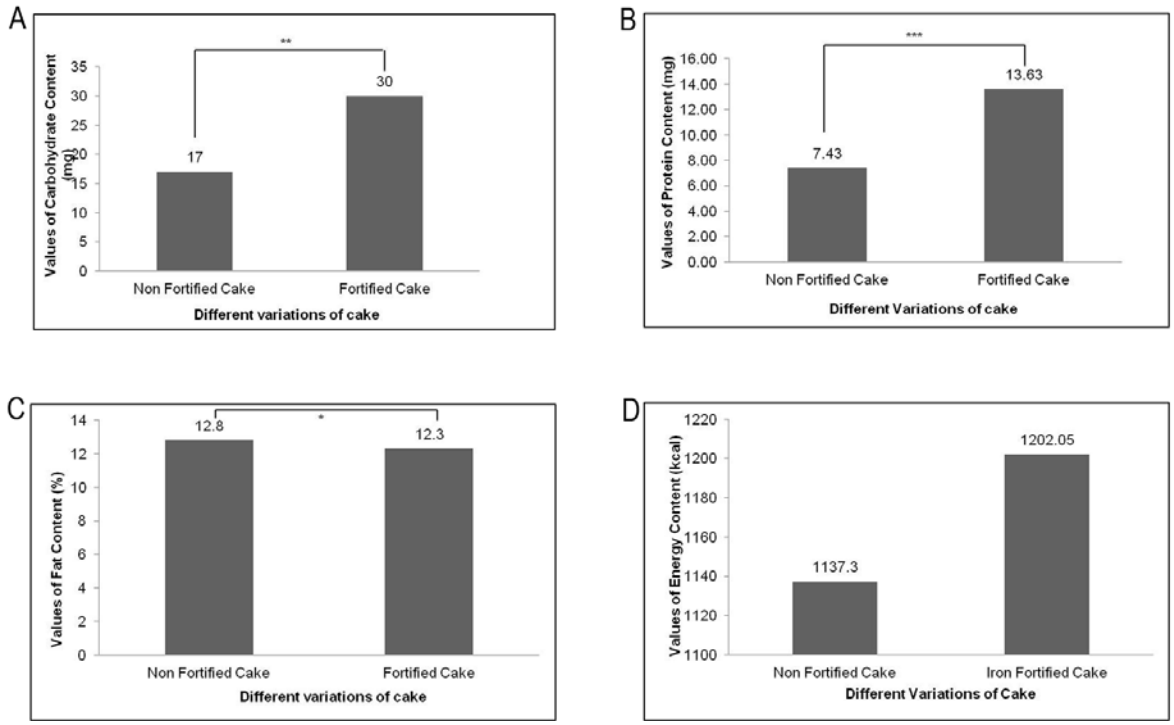

Fig 4: Estimation of food macromolecules and energy. Both the iron-fortified and the non-fortified cake were biochemical estimated for protein, carbohydrate, fat and energy content. The iron-fortified cake displayed increased carbohydrate, protein and energy content compared to the basic recipe (A, B and D). The fat percentages in both the products were similar (C). The experimental results are displayed as mean±s.e.m of atleast 3 independent experiments. *, **and ***denote P value <0.05, <0.005 and <0.001 respectively.

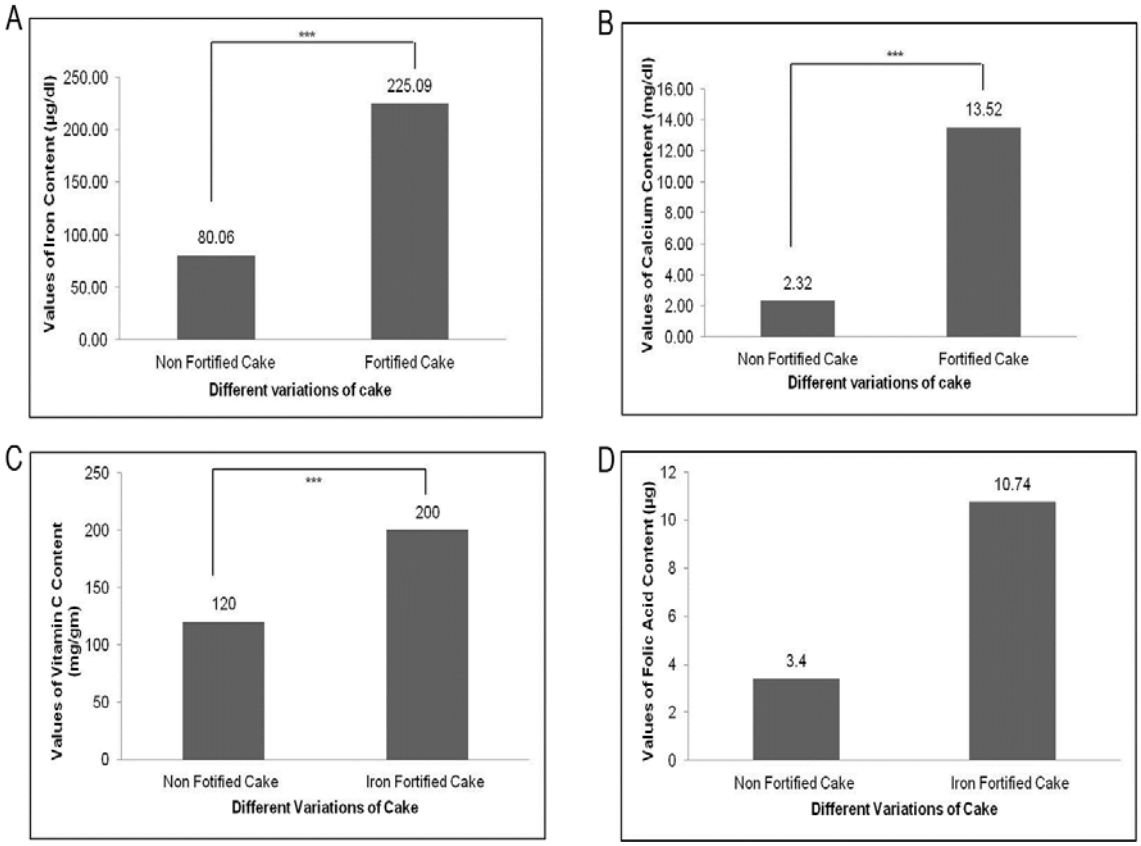

Fig 5: Determination of vitamins and minerals. Both the iron-fortified and the non-fortified cake were biochemical estimated for iron, calcium, vitamin C and folic acid content. The iron-fortified cake displayed increased iron, calcium, vitamin C and folic acid content compared to the basic recipe (A, B, C and D). The experimental results are displayed as mean±s.e.m of at least 3 independent experiments. *, **and ***denote P value <0.05, <0.005 and <0.001 respectively.

Table 1: Components of the iron-fortified cake vs the non-fortified cake

INGREDIENTS	BASIC RECIPE AMOUNT(gm)	FINAL RECIPE AMOUNT(gm)
MAIDA	100	40
SUGAR (POWDERED)	100	80
BUTTER	50	50
MILK	40	-
BAKING POWDER	5	5
BAKING SODA	2.5	2.5
VANILLA ESSENCE	2.5	2.5
OATS(POWDERED)	-	20
WHOLE WHEAT FLOUR	-	30
GARDEN CRESS SEEDS(ROASTED, POWDERED)	-	5
RICE BRAN (POWDERED)	-	10
CONDENSED MILK	-	15
STRAWBERRIES	-	60
DATES	-	10
RAISINS	-	10
SODA(PLAIN)	-	100
COST(Rs)	**32.87**	**55.865**

Table 2: Estimation of microbiological shelf life. Microbiological shelf life was estimated by serial dilution nutrient agar plating method. The results are expressed as CFU/ml. -, +, and ++ indicate 0, 1-10 and >10 CFU/ml.

	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}
Non -Fortified Cake	++	+	-	-	-
Iron -Fortified Cake	++	+	-	-	-

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