

Original Research Article

Nutritional evaluation of cereal-pulse based extruded snacks supplemented with dehydrated herbs.

ABSTRACT

AIM: To Do Nutritional Evaluation Of Cereal-Pulse Based Extruded Snacks Supplemented With Dehydrated Herbs.

STUDY DESIGN: experimental

place and duration of study: sample: department of food and nutrition and department of food science and technology, punjab agricultural university, ludhiana, punjab, india, between jan 2015 to dec, 2017.

METHODOLOGY: IN THE PRESENT INVESTIGATION, A COMBINATION OF WHEAT AND CHICKPEA (80:20) WAS USED FOR FORMULATION OF EXTRUDED SNACKS WHICH WERE SUPPLEMENTED WITH VARYING LEVELS OF 1-5 PERCENT DRIED HERBS NAMELY BASIL (BL), MINT (ML), DRUMSTICK LEAVES (DL) AND A MIXTURE OF ALL THESE HERBS (MXL) HAVING ONE PERCENT OF EACH HERB. THE ACCEPTABLE EXTRUDATES WERE ANALYSED FOR DIFFERENT NUTRITIONAL PARAMETERS NAMELY: PROXIMATE, VITAMINS MINERALS, BIOACTIVE COMPONENTS AND *IN VITRO* NUTRIENT DIGESTIBILITY.

RESULTS: ORGANOLEPTICALLY EXTRUDATES WERE FOUND TO BE MOST ACCEPTABLE AT THREE PERCENT LEVEL OF SUPPLEMENTATION FOR ALL THE HERBS. OVERALL ACCEPTABILITY OF VARIOUS EXTRUDATES WAS IN ORDER OF MXL>DL>ML>BL. THE MOISTURE, ASH, CRUDE PROTEIN, FIBRE AND FAT CONTENT OF SUPPLEMENTED EXTRUDED SNACKS RANGED FROM 3.40 TO 3.88, 2.32 TO 2.90, 11.35 TO 12.20, 3.0 TO 3.36 AND 1.67 TO 1.75 G/100G, RESPECTIVELY. ASCORBIC ACID AND B-CAROTENE CONTENT OF SUPPLEMENTED EXTRUDED SNACKS RANGED FROM 9.72 TO 12.98 MG/100G AND 54.71-98.10 µG/100G, RESPECTIVELY. TOTAL IRON, CALCIUM AND ZINC CONTENT IN SUPPLEMENTED EXTRUDED SNACKS WERE FOUND TO INCREASE IN THE RANGE OF 19-29, 38-77 AND 52-63%, RESPECTIVELY IN COMPARISON TO CONTROL. THE TOTAL PHENOL AND FLAVONOID CONTENT OF SUPPLEMENTED EXTRUDED SNACKS VARIED FROM 153.45 TO 184.76 MG GAE/100G AND 222.38 TO 384.40 MG RE/100G, RESPECTIVELY. THE PERCENT INCREASE IN TOTAL ANTIOXIDANT CAPACITY (TAC) IN SUPPLEMENTED SNACKS RANGED FROM 97-124, 93-125, 96-154 AND 25-46%, RESPECTIVELY BY DPPH, ABTS, FRAP AND RPA, RESPECTIVELY AS COMPARED TO CONTROL.

CONCLUSION: NON IT CAN BE CONCLUDED THAT SUPPLEMENTATION OF SOME OF THE COMMONLY USED DEHYDRATED HERBS LEAVES IN THE POWDER FORM TO THE CEREAL-PULSE BASED EXTRUDED SNACKS CAN HELP TO INTRODUCE A NEW TYPE OF VALUE ADDED SNACKS WHICH WILL NOT ONLY SATISFY CONSUMERS SHORT TIME HUNGER BUT ALSO PROVIDE NUMEROUS HEALTH BENEFITS ESPECIALLY IN TERMS OF BIOACTIVE COMPONENTS.

Keywords: extrusion, snacks, herbs, supplementation, bioactive components

1. INTRODUCTION

CEREALS HAVE IMPORTANT POSITION IN INTERNATIONAL NUTRITION OF ALL OVER THE WORLD DUE TO THEIR UBIQUITOUS CONSUMPTION. CEREAL GRAINS ARE OPTIMAL SOURCE OF ENERGY, CARBOHYDRATES, PROTEINS, FIBRE AND MICRONUTRIENTS. AMONG THE DIVERSE CEREALS CONSUMED ALL OVER THE WORLD, WHEAT IS EXTREMELY IMPORTANT BEING EXTENSIVELY GROWN AS A STAPLE FOOD. IT IS A GRASS BELONGING TO FAMILY GRAMINEAE AND GENUS (*TRITICUM AESTIVUM* L.) WHICH IS GROWN IN EACH AND EVERY PART OF WORLD. IN MAJORITY OF INDIAN POPULATION, 70 TO 80 PERCENT OF DAILY ENERGY INTAKE IS CONTRIBUTED BY WHEAT. WHEAT CAN BE USED AS FLOUR, SEMOLINA AND MANY OTHER FORMS, WHICH FORM THE BASIC INGREDIENTS FOR BREAD, BAKERY PRODUCTS AND PASTAS ETC. THEREFORE, WHEAT IS THE MAIN SOURCE OF NUTRIENTS TO THE MAJOR PART OF THE WORLD POPULATION.

THE QUALITY OF CHICKPEA PROTEIN HAS BEEN CONSIDERED TO BE BETTER THAN THAT OF MANY OTHER PULSES. IT HAS CONSIDERABLE AMOUNTS OF ALL THE ESSENTIAL AMINO ACIDS EXCEPT FOR SULPHUR CONTAINING AMINO ACIDS, WHICH CAN BE COMPLEMENTED BY ADDING CEREALS TO DAILY DIET ALONG WITH CHICKPEA. CHICKPEA CONTAINS GOOD AMOUNTS OF MINERALS INCLUDING CALCIUM, MAGNESIUM, PHOSPHORUS AND POTASSIUM AND ALSO A GOOD SOURCE OF VITAMINS LIKE RIBOFLAVIN, NIACIN, THIAMIN, FOLATE AND VITAMIN A PRECURSOR I.E. B-CAROTENE. VARIOUS COOKING METHODS HAVE BEEN FOUND TO REDUCE ANTI-NUTRITIONAL FACTORS PRESENT IN CHICKPEA. DUE TO ITS SEVERAL HEALTH BENEFITS, CHICKPEA WHEN USED IN COMBINATION WITH OTHER PULSES AND CEREALS CAN HAVE BENEFICIAL EFFECTS ON SOME OF THE LIFESTYLE DISEASES RELATED TO DIGESTIVE SYSTEM, CARDIOVASCULAR, TYPE II DIABETES, AND SOME CANCERS [1].

PROCESSED FOODS INCLUDE CONVENIENT FOODS LIKE INSTANT MIXES, EXTRUDED SNACKS, CANNED AND DEHYDRATED FOODS. THE EXTRUDED PRODUCTS HAVE BETTER SHELF LIFE AND ARE EASILY ACCEPTABLE BY ALL THE AGE GROUPS AS REPORTED BY KOWSALYA & INDRA [2]. THE EXTRUDED SNACKS THAT ARE PREPARED FROM CEREAL GRAINS ARE USUALLY LOW IN NUTRIENT DENSITY ESPECIALLY IN PROTEIN CONTENT AND ESSENTIAL AMINO ACIDS WHICH CAN BE INCREASED BY ADDING PULSES HAVING HIGH QUALITY PROTEIN. EXTRUSION COOKING IS ONE OF THE VERY POPULAR TECHNIQUES IN FOOD AND FEED INDUSTRIES. IT INVOLVES HIGH TEMPERATURE SHORT TIME (HTST) COOKING WITH A NUMBER OF ADVANTAGES SUCH AS SHORTER PROCESSING TIME AND ENERGY SAVING, LEADING TO PRODUCTION OF COST-EFFECTIVE AND OIL FREE PUFFED SNACKS. THE EXTRUSION TECHNOLOGY HAS BEEN USED FOR MAKING A WIDE RANGE OF SNACKS FROM CEREAL FLOURS, STARCH GRANULES, TUBERS AND LEGUMES ETC INTO SEMI-COOKED OR COMPLETELY COOKED ACCEPTABLE FOOD PRODUCTS LIKE BREAKFAST CEREALS, PASTA PRODUCTS, FLAKES, BREAKFAST GRUEL AND TEXTURISED VEGETABLE PROTEIN IN DEVELOPING COUNTRIES [3].

HOWEVER, EFFECTS OF EXTRUSION COOKING ON NUTRITIONAL QUALITY OF PRODUCTS ARE STILL UNCERTAIN. DUE TO THE EXPOSURE OF FOOD TO A VERY HIGH TEMPERATURE, SOME OF THE HEAT- LABILE VITAMINS MAY BE LOST TO VARYING EXTENTS. NUTRITIONAL QUALITY OF THE FOODS CAN BE AFFECTED IN A FAVOURABLE OR ADVERSE WAY DUE TO CHANGES IN PROTEINS AND AMINO ACID PROFILE, CARBOHYDRATES, DIETARY FIBRE, MINERAL CONTENT AND SOME OF THE

BIO-ACTIVE COMPONENTS. ON ONE SIDE, DESTRUCTION OF ANTINUTRITIONAL FACTORS, INCREASED SOLUBLE DIETARY FIBRE, GELATINISATION OF STARCH AND REDUCTION OF LIPID OXIDATION ARE SOME OF THE BENEFICIAL EFFECTS, WHILE ON OTHER SIDE MAILLARD REACTION BETWEEN PROTEIN AND SUGARS DURING EXTRUSION CAN RESULT IN REDUCTION OF PROTEIN QUALITY [4].

HERBS NOT ONLY ENHANCE THE TASTE AND FLAVOUR OF FOODS BUT THEIR ANTIMICROBIAL AND ANTIFUNGAL PROPERTIES ALSO HELP TO INCREASE THE SHELF LIFE. MANY LOW COST BUT VALUABLE MEDICINAL HERBS ARE EASILY AVAILABLE AND ARE VERY USEFUL DUE TO THEIR NEUTRACEUTICAL PROPERTIES. HOWEVER, THEIR USE IN CULINARY IS LIMITED ONLY AS A FLAVOUR ENHANCER IN FRESHLY COOKED FOODS. TULSI HAS BEEN BEING USED AS A COMMON HERB IN MANY OF THE INDIAN HOUSEHOLDS FOR TREATMENT OF MINOR AILMENTS LIKE FROM REGULAR FEVER TO SOME OF THE MOST FATAL BACTERIAL AND VIRAL INFECTIONS SINCE ANCIENT TIMES. IT IS ALTERNATIVELY KNOWN AS HOLY BASIL (*OCIMUM SANCTUM*) AND CONATINS A SIGNIFICANT AMOUNT OF PHYTONUTRIENTS, ESSENTIAL OILS, VITAMIN A AND C [5].THE DRUMSTICK LEAVES HAVE BEEN REPORTED TO PREVENT AND TREAT PROTEIN-ENERGY MALNUTRITION AND OTHER NUTRITION RELATED DISEASES [6]. HOWEVER, ITS SEEDS AND FLOWERS ALSO HAVE GOOD NUTRITIONAL AND THERAPEUTIC VALUES. LEAVES OF DRUMSTICK ARE LOW IN FAT AND CARBOHYDRATE BUT ARE GOOD SOURCE OF AMINO ACIDS MAINLY SULPHUR CONTAINING AMINO-ACIDS SUCH AS METHIONINE AND CYSTINE [7]. MINT BELONGS TO THE LAMIACEAE FAMILY WHICH HAS 25–30 SPECIES. THE MOST POPULAR SPECIE IS COMMON MINT OR SPEARMINT. PEPPERMINT (*MENTHA PIPERITA*) HAS BEEN USED AS A COMMON REMEDY FOR THE TREATMENT OF SOME OF COMMON DIGESTIVE TRACT PROBLEMS LIKE NAUSEA, INDIGESTION, FLATULENCE AND EVEN FOR HICCUPS.

AS EVIDENT FROM THE ABOVE DISCUSSION, HERBS HAVE BEEN USED AS AN ALTERNATIVE MEDICINE FOR PREVENTION AND CURE OF MANY HEALTH PROBLEMS. HOWEVER, VERY SCANTY INFORMATION IS AVAILABLE ON THEIR NUTRITIVE VALUE AND EFFECT OF PROCESSING ON VARIOUS SENSORY AND NUTRITIONAL PARAMETERS. THEREFORE, SUPPLEMENTATION OF SOME OF THE COMMONLY USED HERBS NAMELY BASIL, DRUMSTICK AND MINT LEAVES IN THE POWDER FORM TO THE CEREAL-PULSE BASED EXTRUDED SNACKS WILL HELP TO INTRODUCE A NEW TYPE OF VALUE ADDED SNACKS WHICH WILL NOT ONLY SATISFY CONSUMERS SHORT TIME HUNGER BUT ALSO PROVIDE NUMEROUS HEALTH BENEFITS.

2. MATERIAL AND METHODS / EXPERIMENTAL DETAILS / METHODOLOGY

2.1 Formulation of extruded snacks

The standardized recipe for control samples was prepared from wheat flour (WF) and Chickpea flour (CF). The chickpea flour (CF) was added to all the extruded snacks at the 20 % level. Dehydrated herbs Basil leaves (*Ocimum tenuiflorum*), Mint leaves (*Mentha*) and Drumstick leaves (*Moringa Oleifera*) were added at 1,2,3,4 and 5% level respectively with the replacement of wheat flour. One combination was prepared by mixing the powder of all three herbs in equal proportion. A standardized amount of salt i.e. 2g was added in all types of extrudates. The extruded snacks were made by using a co- rotating intermeshing twin screw extruder (Cletral, Firminy, France). Twenty one types of extruded products were developed and standardized using the following combinations.

2.2 Organoleptic evaluation of extruded snacks

The sensory evaluation of the formulated products was carried out by a panel of the semi-trained judges. The judges were served with one control and four preparations with different levels of incorporation. The panel was provided 9 point hedonic scale for attributes like appearance, colour, texture, aroma, taste and overall acceptability [8]. Different sample codes were given to different levels of products and exact composition of the levels were not revealed to the panelists to get their exact judgment of the samples. The mean scores for each product were then calculated.

2.3 Nutritional analysis

2.3.1 Proximate analysis: All the samples were analyzed in duplicates. Moisture, crude ash, crude protein, crude fat and crude fibre content was evaluated on dry matter basis by [9] Carbohydrate and energy was calculated on the basis of above parameters.

2.3.2 Vitamins and minerals: Estimation of Ascorbic Acid was done by [9] and β -Carotene by [10]. Elements namely iron, zinc and calcium were estimated using atomic absorption spectrophotometer (AAS, Varian model) [11]. Sample was digested with diacid mixture (nitric acid: perchloric acid, 5:1 v/v) and then analyzed by AAS.

2.3.3 Bioactive components: Extraction of Bioactive components was done with known quantity of weighed sample was taken in 100 ml conical flask. Added 15 ml of 80% methanol acidified to pH 2.0 with 6N Hydrochloric acid by shaking at room temperature for 30 minutes. Supernatants were decanted and re-extracted the residue for complete removal of phenolic and antioxidant compounds. This procedure was repeated for two times. The three supernatants were pooled, centrifuge at 6000 rpm for 15 min and filter through Whatman No.1 filter paper. The volume was made up to 50 ml with the solvent. The sample was transferred to micro centrifuge tubes and stored at -20°C for total phenolic content (TPC) and total antioxidant capacity (TAC) determination. Total phenols was estimated by [12] and Flavonoid content by [13] method. Determination of Total antioxidant capacity by Ferric Reducing Antioxidant Power (FRAP) assay [14] and modified by [15], Reducing Power Assay (RPA) [16], ABTS Radical Scavenging Activity [17] and DPPH (2,2-Diphenyl-1-picrylhydrazyl) Radical Scavenging Activity [18] as modified by [15].

2.3.4 Estimation of *In vitro* nutrient digestibility: *In vitro* iron bioavailability was analysed by using method of [19] and *in vitro* protein digestibility was determined by method given by [20] as Modified [21]. *In vitro* digestibility of carbohydrates was determined by estimating the quantity of maltose formed by the dinitrisalicylic acid method [22].

2.3.5 Statistical Analysis: Data obtained from the mean values and standard deviation for each sample were computed using MS Excel. Non parametric test such as Kruskal-Wallis were used for selecting the best formulations (extruded snack) through sensory evaluation. Analysis of variance (ANOVA) (Tukey's test) was employed to assess the significant difference between the treatments in terms of their nutrient content. The statistical procedures were performed using SPSS (version 16.0) SPSS Inc (Chicago, USA).

3. RESULTS AND DISCUSSION

3.1 Organoleptic evaluation of extruded snacks

One control and four types of extruded snacks supplemented with different level of herbs ranging from 1-5% were prepared and subjected to organoleptic evaluation by a panel of semi-trained judges using a point hedonic scale. Whenever, the extrudates supplemented with any herb were prepared, a control was also prepared every time. The results of the organoleptic evaluation have been presented in Table 1.

Among extrudates supplemented with mint leaves (7.88) the highest score for overall acceptability was found in case of the ones supplemented at 3% level of mint leaves. When various parameters of sensory evaluation of supplemented extrudates were compared with those of control sample, it was found that scores for appearance and texture were slightly lower as compared to those for control. The values being 7.85 and 7.85 for mint extrudates and 8.08 and 7.92 for control, respectively. This might be due to the reason that incorporation of herbs led to slight decrease in the fluffiness of extrudates. However, the scores for other parameters viz. Colour, taste and flavour were at par or even higher than those for control ones.

The results of organoleptic evaluation of extrudates supplemented with basil leaves also showed a similar trend as in case of mint leaves i.e. the highest overall acceptability score (7.80) was found in the ones supplemented at 3% of basil powder. However, in this case the scores regarding appearance, colour and texture were lower as compared to those of control, but for taste and flavour, the values were higher when compared to the control. Statistical analysis revealed non-significant difference in appearance, colour and texture while it was significant ($p<0.05$) in taste, flavor and overall acceptability. Pasta containing refined wheat flour (97%) and tulsi powder (3%) resulted in better quality having more nutritional elements and highest overall acceptability [5].

Drumstick leaves have been reported to contain significant amounts of iron, calcium and other bioactive compounds when extrudates incorporated with varying levels of DL were prepared, the highest overall acceptability was found to be 7.93 which was slightly less than those of control (8.01). The scores obtained for colour and texture were at par with those obtained for the control. However, the score for taste was even higher than that of control, the values being 8.06 and 7.92, respectively. The appearance and flavour parameters scored less than those for the control. Cream crackers were developed from cassava and sweet potato flour incorporated with drumstick leaves using wheat flour as controls. Sensory evaluation of the cream crackers developed show that they have good sensory properties and since the cassava and sweet potato flour crackers do not contain gluten, they can be consumed by those who are gluten intolerant [23].

A mixture of all the above mentioned three herbs was prepared by using equal proportion of each herb. This mixture of herbs was also incorporated into extrudates at level ranging from 1-5% and compared to the control. In this case also, the highest overall acceptability was found at 3% level (8.06) and the lowest at 5% level (6.94). Scores for all parameters including appearance, colour, texture and taste were even higher when compared to control. However, the score for flavour was lower (7.89) as compared to that for control (8.08). Statistical analysis revealed non significant difference in appearance and taste in different levels but it was significant ($p<0.05$) in colour, texture and flavor and overall acceptability. Low calorie ready-to-eat snack (rice flakes mix) by incorporating edible dehydrated herbs and found Treatment T4 was highly acceptable with overall acceptability score of 8.77 followed by T2 (8.31) and T1 (8.20). In T4 3% of herbs (basil, mint and drumstick leaves, one percent each) was incorporated [24].

Overall acceptability of various extrudates was in order of MXL>DL>ML>BL.

3.2 Nutritional evaluation

Various nutritional parameters including proximate composition, vitamin and mineral content, bioactive components and *in vitro* digestibility of nutrients were analysed in the extrudates supplemented

with levels of various herbs. For all the herbs the most acceptance was found to be 3%. The results of the nutritional evaluation of extrudates are being discussed here:

3.2.1 Proximate composition

The proximate composition of the most acceptable extruded snacks has been presented in the Table 2. The moisture content of the supplemented extruded snacks ranged from 3.80 to 3.88 g/100g being maximum in BL followed by MXL, ML and DL. Results showed that extruded snacks supplemented with herbs had more moisture content as compared to control. The percent increase in moisture content in supplemented snacks ranged from 8-11% as compared to control. The extruded snacks contained moisture within the range of 4-8% [25]. Moisture content of BL was significantly ($p<0.05$) higher than ML and control extrudates. The ash content of food products is an index of mineral content. The ash content of supplemented extruded snacks ranged from 2.32 to 2.90 g/100g, being maximum in ML 2.90 g/100g, followed by 2.84, 2.80 and 2.75 g/100g in DL, BL and ML, respectively. MXL extrudates had significantly ($p<0.05$) higher ash content than other extrudates.

MXL supplemented snacks had highest protein content and it was minimum in BL supplemented extruded snacks. The percent increase in crude protein content in supplemented snacks ranged from 4-10% compared to control. MXL extrudates had significantly ($p<0.05$) higher crude protein content than other extrudates. Extruded snack products are predominantly made from cereal flour or starches and tend to be low in protein and have low biological value [26]. In extrudates, the addition of legume flour to cereal-based formulations has proven to positively impact their essential amino acid balance [27]. Incorporation of dehulled legumes (black gram, green gram, lentil and peas) at the level of 5, 9, and 15% with corn was done to make extruded snacks. Protein content of extrudates showed a marginal increase, found to be maximum for lentil from 8.7 % to 10.19 %, 10.8 % and 11.8 % at 5 %, 10 % and 15 % incorporation levels, respectively. This increased protein content of extrudates may be attributed to their inherent higher protein content of legumes [28].

Crude fibre content of the supplemented extruded snacks was highest in MXL (3.25) and lowest in ML (3.00). Herb supplemented extruded snacks had more fibre content than control. The percent increase in fibre content in supplemented snacks ranged from 8-16% as compared to control. Supplemented snacks with MXL and ML showed significantly ($p<0.05$) higher crude fibre content as compared to other extrudates. Crude fat content was lowest in ML and maximum in BL supplemented extruded snacks. Crude fat was significantly ($p<0.05$) lower in control than BL, DL and MXL supplemented snacks. The percent increase in fat content in supplemented snacks ranged from 2-5% compared to control. ML extrudates had significantly ($p<0.05$) higher crude fat content than control and MXL extrudates. 0.5% fat content in extrudates made from mixture of soy and sweet potato flour [29]. fat content was found to decrease from 3.45% - 3.02% indicating that extrusion process plays a role in fat reduction [28].

More carbohydrate content was found in control as compared to supplemented extruded snacks. The percent decrease in carbohydrate content in supplemented snacks ranged from 1-3% compared to control. ML extrudates had significantly ($p<0.05$) higher carbohydrate content than other extrudates. The carbohydrate content of soybean extruded snacks was 64 g/100g [30]. 76.4g/ 100g of carbohydrate content was reported in chickpea extruded snacks [31]. The similar trend was observed [29] in extruded snacks developed from soyabean and sweet potato flour and found that with increase in sweet potato content the carbohydrate values of extruded products increased. The carbohydrate content of extruded snacks of sweet potato and soy flour in the ratio of (1:4) and (4:1) was 52.84 and 73.58g/100g whereas, the protein content was 38.15g and 16.32 g/100g respectively.

ML supplemented extruded snacks was found to have highest energy content i.e. 371.75 kcal/100g. Control had more energy content than supplemented extruded snacks. Carbohydrate content in extruded snacks did not differ significantly. However, Energy content was significantly ($p<0.05$) more in control than supplemented snacks. The percent decrease in energy content in supplemented snacks ranged from 0.7-1% as compared to control. The nutritive value for the extruded snacks as protein (3.76%), fibre (6.41%), carbohydrates (79.50%) and energy (343.03 kj/100g) was reported in a study [32]. Energy content of 382 kcal/100g in soybean extruded snacks was reported [30]. On the basis of physicochemical & nutritional

properties, cooking time and sensory qualities, pasta containing refined wheat flour (97%) and tulsi powder (3%) resulted in better quality having more nutritional elements and highest overall acceptability [5].

3.2.2 Vitamins

The data regarding vitamin C and β -carotene content of the developed extruded snacks along with that of control has been presented in Table 3. Ascorbic acid content was significantly ($p<0.05$) higher in DL (17.46 mg/100g), followed by MXL (12.98 mg/100g) and ML (10.12 mg/100g). It was significantly ($p<0.05$) lower in BL (9.72 mg/100g) and was negligible in control. The percent increase in vitamin C content in supplemented snacks was found to be more than 100% as compared to control. DL extrudates had significantly ($p<0.05$) higher vitamin C content than MXL, ML and BL extrudates. Reported vitamin C content in extruded snacks was 1.03mg/100g [32].

The β -carotene content was significantly ($p<0.05$) higher in DL (98.10 μ g/100g) followed by BL, MXL and ML i.e. 86.30, 77.40 and 54.71 μ g/100g, respectively. Content of β -carotene was significantly ($p<0.05$) lower in control i.e 32.60 μ g/100g. The percent increase in β -carotene content in supplemented snacks ranged from 137-200% as compared to control (Fig.1). A significant higher ($p<0.05$) content of vitamin C and β carotene in the supplemented extruded snacks might be due to their higher content in the herbs.

3.2.3 Mineral content

As discussed previously, the herbs have been reported to be a good source of minerals including iron, calcium and zinc etc. So in the present study, the effect of supplementation of herbs on the mineral content of extrudates was determined.

3.2.3.1 Iron

The data regarding total iron of extruded snacks have been presented in Table 4. The analyzed total iron content of extruded was significantly ($p<0.05$) higher in ML extrudates and lowest in control. Total iron content in supplemented extruded snacks was found to increase in the range of 19-29 % in comparison to control (Fig. 2). On the basis of intake data and isotope studies, iron bioavailability has been estimated to be in the range of 5–12% in vegetarian diet [33]. *in vitro* availability of iron from different green leafy vegetables was neither a function of their total iron content nor ascorbic acid but was slightly affected by oxalic acid content [34].

3.2.3.2 Calcium

The total calcium of extruded snacks has been presented in Table 4.

Total calcium content of supplemented extrudates was found to be in the range of 69.00-88.50 mg/100g with significantly ($p<0.05$) highest content in DL (88.50 mg/100g) and lowest in ML (69.00 mg/100g) supplemented extruded snacks. Percent increase in calcium content of supplemented snacks was highest in DL i.e. 77% followed by MXL, BL and ML i.e. 64, 56 and 38 %, respectively (Fig.2). The nutritive value of the extruded snacks for Ca was 238.50 mg/100g [32]. Total calcium content of DL supplemented snacks were significantly ($p<0.05$) higher than other extrudates.

3.2.3.3 Zinc

Zinc is an important element needed in body as it is involved in normal function of immune system. The percent increase in zinc content was 63, 54, 52 and 56 % in BL, ML, DL and MXL, respectively (Fig. 2). zinc content was 1.23 mg/ 100g in extruded snacks made from a composite blend of rice, soybean and sorghum [35]. Total zinc content of supplemented snacks were significantly ($p<0.05$) higher in BL than MXL and control.

291 3.2.4 Bioactive compounds

292 The content of bioactive compounds (total phenol, flavonoids) and antioxidant activity (DPPH, ABTS.
293 FRAP and RPA) has been presented in table 5 and percent increase in supplemented snacks as
294 compared to control has been depicted in Fig 3.

295 Until recently, phenolic compounds were regarded as non-nutritive compounds and it was reported that
296 excessive content of polyphenol inhibits the bioavailability of iron, protein etc and blocks digestive
297 enzymes in the gastrointestinal tract. Later on, the significance of phenolic compounds was gradually
298 recognized and several researches have now reported that phenolics offer many health benefits and are
299 vital in human nutrition [36]. Studies have also reported that polyphenols are involved in defense against
300 ultraviolet radiation or aggression by pathogens [37]. The total phenol content was significantly ($p<0.05$)
301 maximum in BL extrudates followed by ML, MXL and DL i.e. 178.72, 153.45 and 162.65 mg/100g. Control
302 had significantly ($p<0.05$) less content of phenol than all supplemented extruded snacks i.e. 124.73
303 mg/100g. The percent increase in total phenol content in supplemented snacks ranged from 23-48%
304 compared to control. In the present study the polyphenol contents in extrudates was high and this is
305 attributed to the effect of extrusion. The results were in accordance with previous works on polyphenols of
306 raw and extruded common beans and it was seen that the effect of extrusion on the phenolic content of
307 beans increased upto 14% in extrudates compared to raw beans [38].

308 Significantly ($p<0.05$) highest content of flavonoid was found to be in the MXL extrudates (384.40
309 mg/100g) followed by DL, ML and BL i.e. 322.69, 309.87 and 222.38 mg/100g, respectively. Control had
310 significantly ($p<0.05$) lower content of flavonoid than all supplemented extruded snacks i.e. 124.73
311 mg/100g. The percent increase in flavonoids content of supplemented snacks ranged from 78-207%
312 compared to control.

313 The antioxidant content of supplemented snacks (Table 4) was found to be significantly ($p<0.05$) higher in
314 ML by DPPH and FRAP and in DL by ABTS and in MXL by RPA and minimum in BL by DPPH and ABTS,
315 RPA and in DL by FRAP. Antioxidant activity in control was significantly ($p<0.05$) lower by all methods
316 than supplemented extruded snacks. The percent increase in TAC content in supplemented snacks
317 ranged from 97-124, 93-125, 96-154 and 25-46 % by DPPH, ABTS, FRAP and RPA, respectively as
318 compared to control. The high antioxidant content in all extrudates was may be due to the formation of
319 higher Maillard products during the high temperature short time processing (HTST) used in extrusion as
320 reported [39]. The high antioxidant can also be due to the presence of high TPC in extruded products
321 since the percentage DPPH inhibition is directly correlated with TP.

322 3.2.5 In vitro nutrient digestibility

323 The *In vitro* nutrient digestibility has been presented in Table 5.

324 MXL supplement extruded snacks had significantly ($p<0.05$) higher value for *in vitro* protein
325 digestibility, and however, *in vitro* starch digestibility was highest in ML extrudates. Control had
326 significantly ($p<0.05$) lesser *in vitro* protein digestibility (69.12%). Similar results were reported [40] for
327 protein digestibility of 68.2 and 79.9% in sorghum extruded snacks. Extrusion cooking as the best method
328 in comparison to other processing methods as it significantly decrease the antinutrients like trypsin,
329 chymotrypsin and alpha amylase activity in extruded snacks made from faba and kidney beans and thus
330 improving the protein digestibility [38]. The thermal treatment during the extrusion might have caused a
331 partial inactivation of the trypsin inhibitors, which improved the protein digestibility. In addition, the
332 denaturation of proteins in the extrusion process can expose sites that are susceptible to the enzymatic
333 hydrolysis [40]. In the present study, the *in vitro* protein digestibility values of extruded snacks were high
334 and this may be attributed to the extrusion process which significantly reduced the antinutrients and thus
335 increasing digestibility. Low calorie ready-to-eat snack was developed (rice flakes mix) by incorporating
336 edible dehydrated herbs and reported that except carbohydrate and energy remarkable increase was
337 observed in the nutrients such as protein, fat, fiber, calcium and iron in the developed rice flakes mix
338 compared to control [24].

339 In the present study, the carbohydrate digestibilities of the snacks were high and this may be attributed to
340 the effect of extrusion. Extrusion process favours starch digestibility properties while maintain availability

of other nutrients [41]. Extrusion cooking significantly increased the *in vitro* digestibility of barley starch [42]. whereas, extrusion treatment significantly increased the *in vitro* digestibility of pea starch [38]. This may be explained in such a way that the increased shearing action develops heat through dissipation of mechanical energy and causes loss of structural integrity and increases enzyme susceptibility. Study also reported that extrusion cooking was the best method to abolish antinutrients in faba and kidney beans. Furthermore, this thermal treatment was most effective in improving starch digestibility when compared with dehulling, soaking and germination.

The *in vitro* iron bioavailability of the snacks was lowest in ML extrudates and highest in BL extrudates. The iron bioavailability of BL supplemented extruded snacks was significantly ($p<0.05$) higher than all other extrudates, while control had significantly ($p<0.05$) lower *in vitro* iron bioavailability. Total and soluble iron content of supplemented snacks were significantly ($p<0.05$) higher than control and so iron bioavailability was also significantly higher ($p<0.05$).

4. CONCLUSION

It can be concluded that supplementation of some of the commonly used dehydrated herbs leaves in the powder form to the cereal-pulse based extruded snacks can help to introduce a new type of value added snacks which will not only satisfy consumers short time hunger but also provide numerous health benefits especially in terms of bioactive components.

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Table 1 Sensory evaluation of extruded snacks

Treatments	Appearance	Colour	Texture	Taste	Flavour	Overall Acceptability
Mint extrudates						
Control WF+CF (20%)	8.08±0.76	7.84±0.69	7.92±0.64	7.69±0.75	7.62±0.87	7.83±0.64
WF+CF (20%)+ML (1%)	7.69±0.95	7.77±0.72	7.54±0.88	7.38±0.96	7.38±0.87	7.55±0.80
WF+CF (20%)+ML (2%)	7.76±0.60	7.77±0.72	7.77±0.72	7.46±0.66	7.61±0.77	7.68±0.59
WF+CF (20%)+ML (3%)	7.85±0.69	7.84±0.69	7.85±0.69	7.74±0.90	7.81±0.96	7.88±0.74
WF+CF (20%)+ML (4%)	7.54±0.78	7.38±0.77	7.31±0.76	6.46±0.66	6.38±0.51	7.02±0.56
WF+CF (20%)+ML (5%)	7.23±0.76	7.07±0.64	7.15±0.81	6.08±0.64	6.23±0.83	6.75±0.49
χ^2 -value	NS	p<0.05	NS	p<0.01	p<0.01	p<0.01
Basil extrudates						
Control	7.90±0.81	8.00±0.82	7.90±0.74	7.75±0.98	7.70±0.94	7.85±0.75
WF+CF (20%)+BL (1%)	7.41±0.52	7.40±0.69	7.60±0.82	7.30±0.68	7.30±0.67	7.40±0.57
WF+CF (20%)+BL (2%)	7.70±0.48	7.70±0.82	7.70±0.82	7.60±0.70	7.60±0.70	7.66±0.62
WF+CF (20%)+BL (3%)	7.81±0.82	7.90±0.74	7.80±0.63	7.80±0.82	7.80±0.82	7.80±0.71
WF+CF (20%)+BL (4%)	7.20±0.79	7.20±0.79	7.50±0.71	7.40±0.84	7.30±0.82	7.32±0.73
WF+CF (20%)+BL (5%)	6.90±0.99	7.10±0.88	6.90±0.71	6.50±0.85	6.50±0.85	6.78±0.78
χ^2 -value	NS	NS	NS	p<0.05	p<0.05	p<0.05
Drumstick extrudates						
Control	8.07±0.76	8.0±0.76	8.0±0.90	7.92±1.38	8.0±0.98	8.01±0.84
WF+CF (20%)+DL (1%)	7.84±0.91	7.85±0.80	7.86±0.78	7.86±0.77	7.61±1.16	7.67±1.16
WF+CF (20%)+DL (2%)	7.87±0.88	7.89±0.86	7.89±0.86	7.92±0.78	7.64±1.02	7.75±0.98
WF+CF (20%)+DL (3%)	7.89±0.56	7.99±0.49	8.0±0.64	8.06±0.55	7.84±0.98	7.93±0.56
WF+CF (20%)+DL (4%)	7.27±0.85	7.43±0.96	7.32±0.85	7.35±0.95	7.10±0.97	7.14±0.95
WF+CF (20%)+DL (5%)	6.85±0.76	6.82±7.76	6.92±0.76	6.71±0.86	6.78±0.94	6.82±0.68
χ^2 -value	p<0.05	p<0.05	p<0.05	p<0.01	p<0.01	p<0.01
Mix extrudates (ML+BL+DL)						
Control	7.92±1.38	8.076±1.187	7.92±1.38	7.38±0.87	8.08±1.04	7.98±1.22
WF+CF (20%)+MXL (1%)	8.076±1.03	8.16±1.03	8.01±1.03	7.54±0.52	7.38±0.86	7.80±0.94
WF+CF (20%)+ MXL (2%)	7.90±0.87	7.92±1.031	7.99±0.85	7.77±0.83	7.54±0.52	7.52±0.69
WF+CF (20%)+ MXL(3%)	8.23±0.599	8.28±0.493	8.11±0.71	7.95±0.75	7.89±0.94	8.06±0.49
WF+CF (20%)+ MXL (4%)	7.69±0.85	7.54±0.66	7.61±0.65	7.72±1.25	7.31±0.75	7.49±0.45
WF+CF (20%)+ MXL (5%)	7.153±0.95	7.153±1.068	6.92±0.86	6.76±1.09	6.69±0.94	6.94±0.91
χ^2 -value	NS	p<0.05	p<0.05	10.82 ^{NS}	p<0.05	p<0.05

(WF: Wheat flour; CF: Chickpea flour; ML: Mint leaves, BL: Basil leaves, DL: Drumstick leaves MXL: Mix (Basil +Mint +Drumstick) leaves

Scores (Nine point hedonic scale, 9- Excellent, 8- Extremely good, 7- Very good, 6- Moderately good, 5- Good, 4- Fair, 3-Very fair, 2- Poor, 1- Very poor).

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463 **Table 2: Proximate composition of extruded snacks (per 100g)***

Treatments	Moisture	Ash	Crude protein	Crude fibre	Crude fat	CHO	Energy (Kcal)
C (WF+CF(20%))	3.50 ^c ±0.21	2.32 ^e ±0.02	11.07 ^c ±0.5	2.78 ^c ±0.05	1.66 ^c ±0.1	78.75 ^{ab} ±10.10	374.22 ^a ±12.30
C+BL(3%)	3.88 ^a ±0.12	2.80 ^c ±0.07	11.35 ^d ±0.2	3.20 ^{ab} ±0.01	1.75 ^a ±0.1	77.00 ^a ±9.32	369.15 ^d ±9.56
C+ML(3%)	3.80 ^{bc} ±0.14	2.75 ^d ±0.12	11.52 ^c ±0.08	3.00 ^b ±0.12	1.76 ^a ±0.09	77.66 ^a ±12.36	371.75 ^b ±8.52
C+DL(3%)	3.82 ^{ab} ±0.19	2.84 ^{bc} ±0.22	12.02 ^b ±0.05	3.20 ^{ab} ±0.05	1.71 ^{ba} ±0.14	77.01 ^a ±5.60	371.51 ^{cb} ±9.54
C+MXL(3%)	3.85 ^{ab} ±0.29	2.90 ^a ±0.10	12.20 ^a ±0.9	3.25 ^a ±0.09	1.69 ^{bc} ±0.13	76.11 ^a ±4.56	368.45 ^c ±8.45

464 Values are expressed as mean ± SD
465 (WF: Wheat flour; CF: Chickpea; C: Control, ML: Mint leaves, BL: Basil leaves, DL: Drumstick leaves MXL: Mix (Basil +Mint +Drumstick) leaves
466 *Results are on dry weight basis.
467 Values followed with different superscripts are significantly different (p < 0.05) using Tukey’s test.

468 **Table 3: Vitamin content of extruded snacks per 100g***

Treatments	Vitamin C (mg/100g)	β - carotene ($\mu\text{g}/100\text{g}$)
Control :WF+CF(20%)	0.05 ^c ±0.01	32.60 ^c ±0.96
C+BL(3%)	9.72 ^d ±0.21	86.30 ^b ±8.2
C+ML(3%)	10.12 ^c ±0.62	74.71 ^d ±7.2
C+DL(3%)	17.46 ^a ±0.12	98.10 ^a ±5.2
C+MXL(3%)	12.98 ^b ±0.54	77.40 ^c ±9.2

469 **Values are expressed as mean $\pm$ SD**

470 **(WF: Wheat flour; CF: Chickpea flour; C: Control, ML: Mint leaves, BL: Basil leaves, DL: Drumstick**
 471 **leaves MXL: Mix (Basil +Mint +Drumstick) leaves**

472 **Values followed with different superscripts are significantly different ($p < 0.05$) using Tukey's test.**

473 ***Results are on dry weight basis**

474 Table 4: Total and soluble mineral content in extruded snacks per 100g*

Treatments	Total iron (mg/100g)	Soluble iron (mg/100g)	% Solubility	Total calcium	Soluble calcium	% Solubility	Total Zinc	Soluble zinc	% Solubility
Control:WF+CF(20%)	3.76 ^c ±0.50	1.25 ^d ±0.21	33	49.84 ^c ±0.01	19.12 ^c ±0.25	38	1.86 ^d ±0.02	0.72 ^d ±0.20	39
C+BL(3%)	4.48 ^d ±0.12	2.56 ^b ±0.23	57	77.94 ^c ±0.03	60.23 ^b ±0.12	76	2.96 ^a ±0.03	1.56 ^a ±0.19	53
C+ML(3%)	4.96 ^a ±0.26	2.82 ^a ±0.12	57	69.00 ^d ±0.02	45.27 ^d ±0.30	69	2.81 ^{ba} ±0.12	1.42 ^a ±0.21	50
C+DL(3%)	4.68 ^c ±0.70	2.12 ^c ±0.09	52	88.50 ^a ±0.05	63.56 ^a ±0.25	76	2.78 ^{cba} ±0.20	1.23 ^b ±0.14	44
C+MXL(3%)	4.86 ^b ±0.60	2.15 ^c ±0.31	44	81.78 ^b ±0.12	52.95 ^c ±0.09	72	2.85 ^a ±0.12	1.12 ^{cb} ±0.22	39

475 Values are expressed as mean ± SD
476 (WF: Wheat flour; CF: Chickpea flour; C: Control, ML: Mint leaves, BL: Basil leaves, DL: Drumstick leaves MXL: Mix (Basil +Mint +Drumstick) leaves
477 Values followed with different superscripts are significantly different (p < 0.05) using Tukey’s test.
478 *Results are on dry weight basis

479 **Table 5: Bioactive components in extruded snacks***

Treatments	Total phenols	Flavonoids	DPPH	ABTS	FRAP	RPA
	(mg/100g)	(mg RE/ 100g)	(mg GAE/100g)	(mg TE/ 100g)	(mg TE/ 100g)	(mg TE/ 100g)
Control:WF+CP(20%)	124.73 ^c ±0.23	124.95 ^c ±3.6	150.61 ^c ±5.23	28.07 ^d ±1.2	98.89 ^c ±1.23	470.25 ^c ±1.23
C+BL(3%)	184.76 ^a ±0.56	222.37 ^d ±2.5	296.81 ^d ±6.30	54.27 ^c ±2.36	199.99 ^c ±6.78	585.92 ^d ±5.48
C+ML(3%)	178.72 ^b ±1.2	309.87 ^c ±2.6	331.81 ^a ±3.03	58.39 ^b ±3.56	250.85 ^a ±7.48	675.06 ^b ±8.32
C+DL(3%)	162.65 ^c ±0.96	322.69 ^b ±1.2	315.75 ^c ±5.23	63.09 ^a ±2.36	194.00 ^d ±4.89	655.71 ^c ±4.47
C+MXL(3%)	153.45 ^d ±0.21	384.40 ^a ±1.6	327.85 ^b ±6.32	58.43 ^b ±2.14	220.98 ^b ±7.02	687.93 ^a ±5.82

480 Values are expressed as mean ± SD
481 (WF: Wheat flour; CF: Chickpea flour; C: Control, ML: Mint leaves, BL: Basil leaves, DL: Drumstick leaves MXL: Mix (Basil +Mint +Drumstick) leaves
482 Values followed with different superscripts are significantly different (p < 0.05) using Tukey’s test.
483 *Results are on dry weight basis

484 **Table 6: *In vitro* nutrient digestibility of extruded snacks***

Treatments	Protein digestibility (%)	Carbohydrate digestibility (%)	Iron digestibility (%)
Control:WF+CF(20%)	69.12 ^c ±1.02	76.02 ^a ±7.23	12.96 ^d ±2.31
C+BL(3%)	72.22 ^c ±0.96	75.03 ^{ab} ±5.20	16.52 ^a ±2.00
C+ML(3%)	70.96 ^d ±2.30	76.12 ^a ±4.02	15.92 ^b ±1.20
C+DL(3%)	74.56 ^b ±2.36	74.30 ^b ±6.90	15.90 ^b ±1.20
C+MXL(3%)	76.15 ^a ±2.02	73.00 ^c ±5.23	15.00 ^c ±1.96

485 Values are expressed as mean ± SD

486 (WF: Wheat flour; CF: Chickpea flour; C: Control, ML: Mint leaves, BL: Basil leaves, DL: Drumstick

487 leaves MXL: Mix (Basil +Mint +Drumstick) leaves

488 Values followed with different superscripts are significantly different (p < 0.05) using Tukey's test.

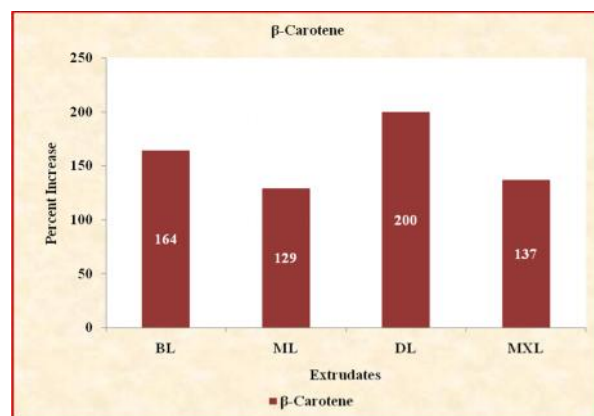
489 *Results are on dry weight basis

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496 **Fig. 1 Percent increase in β-Carotene content of supplemented snacks**

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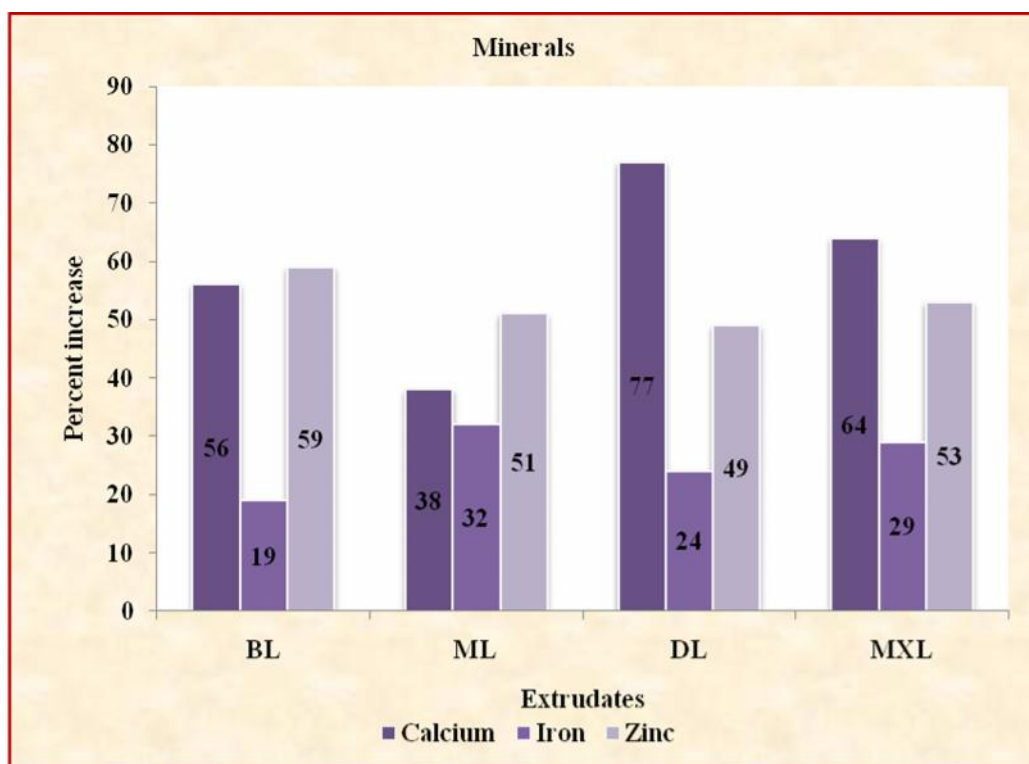


Fig. 2 Percent increase in minerals content of supplemented snacks

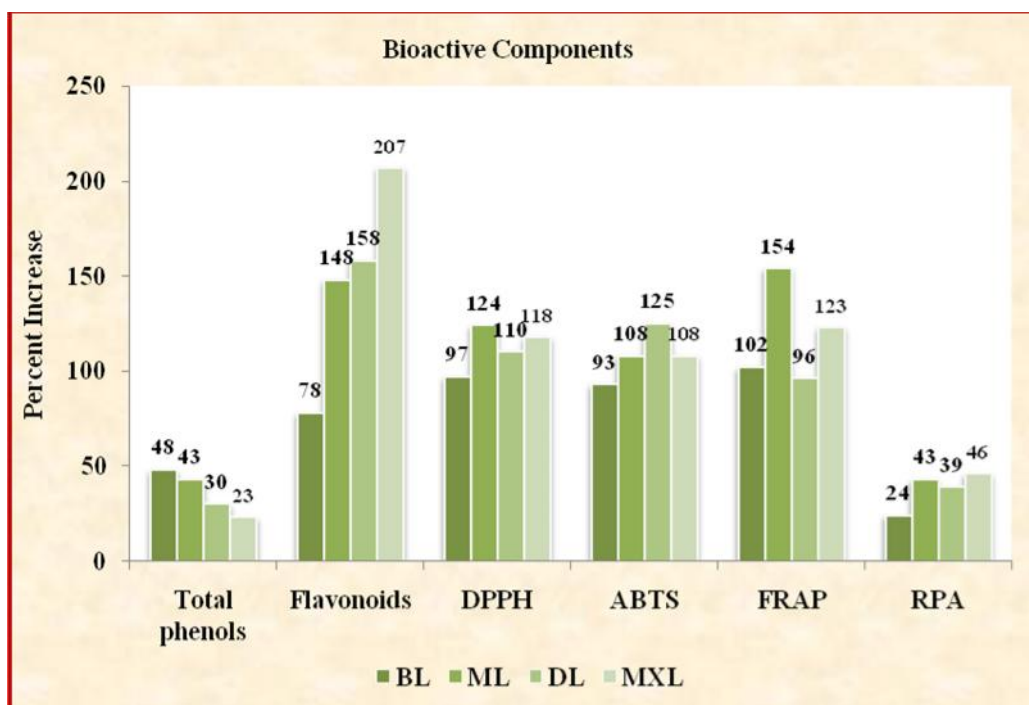


Fig. 3 Percent increase in bioactive components content of supplemented snacks