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Assessment of biochemical yield variation in small millets germplasm lines

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Abstract
Small millets are the group of six crops comprising of finger millet, kodo millet, little millet, foxtail millet, barnyard millet and proso millet. These are considered as nutri-cereals and are source of food, feed and fodder. The crops were grown in a variety of agro-ecological situations including plains, coast and hills as well as in diverse soils and varying rainfall. Three general types of methods have been developed for isolating and analyzing dietary content of fibers, carbohydrates, phenol and protein in small millets *i.e:* enzymatic-gravimetric, DNS acid, Folin-Ciocalteu's assay method and Lowrys method respectively. Protein was estimate according to the procedure of Lowry *et al.,* (1951). The protein in the crude extracts was estimated after precipitation with an equal volume of 10% (w/v) trichloro acetic acid. The mixture was incubated for 30 min at -20 degree celsius and then centrifuged at 10,000g for 10 min. The precipitate was dissolved in 0.5 ml of 1 N NaOH. A standard curve for the protein estimation was prepared by using bovine serum albumin fraction V. The absorbance of the protein sample was measured at 660 nm after 30 min incubation with the reagent mixture.3,5-Dinitrosalicylic acid can be prepared by the nitration of salicyclic acid they reacts with reducing sugars and other reducing molecules to form 3-amino-5-nitrosalicylic acid, which strongly absorbs light at 540 nm. The original AOAC enzymatic-gravimetric method for the determination of total dietary fibre (TDF) was developed on basis of the joint experience.

Keywords: small millets, lowrys methods, dns methods, folin-ciocalteu's assay method

Introduction
Small millets are warm-season cereals largely grown in the semi-arid tropical regions of Asia and Africa, under rainfed farming systems. Small millets includes finger millet (*Eleusine coracana*), kodo millet (*Paspalum scrobiculatum*), proso millet (*Panicum miliaceum*), foxtail millet (*Setaria italica*), little millet (*Panicum sumatrense*) and barnyard millet (*Echinochloa frumentacea*).

Small millets grains are rich in dietary energy, vitamins, several minerals (especially micronutrients such as iron, calcium and zinc), insoluble dietary fiber and phyto-chemicals with antioxidant properties and are considered as "Nutri-cereals". They are rich in compounds that help against several chronic diseases like is themic strokes, cardiovascular diseases, cancers, obesity and Type II diabetes. Small millets are also used as substrate for bio fuels or bio ethanol, biopolymers, distilleries and syrups. Nutritional composition of some of the major cereals including coarse cereals and millets (per 100g) is presented as below. Millets are nutritionally comparable or even superior to major cereals such as wheat and rice, owing to their higher levels of protein with more balanced amino acid profile (good source of methionine, cystine and lysine). The main constituents of millet kernel are seed coat (testa), embryo and endosperm. Among the several varieties of finger millet such as yellow, white, tan, red, brown, only the red colored are cultivated extensively throughout the world. The presence of five layered testa in finger millet makes it unique compared to other millets such as foxtail millet, kodo millet and proso millet. This could be one of the possible reasons for the higher dietary fiber content in the finger millet Gopalan C. *et.al.* (2004)

Millets are important crops of Asia and Africa (especially in India, Nigeria and Niger), with 97% of millet production in developing countries. Millets, considered as important food staples in human history. They have been in cultivation in East Asia for the last 10,000 years. India is the world's largest producer of millet.

The total area of small millets in India around as 1.92 m ha, of and finger millet alone occupies 1.19 m ha. In five yearly analyses of data indicate a steady decline in the area of small millets other than finger millet from 5.29 to 0.93 m ha. The production of finger millet fluctuated between 1.61 m t in 1951-55 and 1.94 m t in 2011-15 with high of 2.65 m t during 1976-80 despite huger education in area. They was achieved because of doubling of productivity of finger millet from 704 kg/ha to 1631 kg/ha due to wide spread cultivation of high yielding blast tolerant varieties (www.aicrpsm.res.in).

Millets contain 60-70% carbohydrates, 7-11% proteins, 1.5-5% fat, and 2-7% crude fibre and are also rich in vitamins and minerals. In millets are the excellent source of vitamin B, magnesium, and antioxidants. Millet is also called as good source of other dietary minerals like manganese, phosphorus and iron. Millet proteins are very good source of essential amino acids except lysine and threonine but are relatively high in sulphur containing amino acids methionine and cysteine. Milltes contain essential fatty acids likes linoleic acid, oleic acid and palmitic acids found in free form and monogalactosyl, diacylglycerols, digalactosyl diacylglycerols, phosphatidylethanolamine, phosphatidyl serine and phosphatidyl choline in the bound form present in millets. Other fatty acids *i.e*. arachidic acid, behenic acid, erucic acid are found in trace amounts. Millets oil could be a good source of linoleic acid and tocopherols. Millets is an alkaline forming grain that is gluten-free. Vitamin B such as Niacin, folacin, riboflavin, and thiamine and phosphorus are present in millets that play a key role in energy synthesis in the body. Millets are known to be a rich in phenolic acids, tannins, and phytate that act as "antinutrients" However; these antinutrients reduce the risk for colon and breast cancer in animals. It was demonstrated that millet phenolics may be effective in the prevention of cancer initiation and progression *in vitro.* Millets contain many major and minor nutrients like carbohydrate, good protein, fat, dietary fiber, vitamins and minerals as well as antioxidant and phytochemicals. The biochemical analysis of small millets is required, So that we get complete information about protein contents, carbohydrate contents, phenolic contents and also fiber contents in different geographical region in India. Three general types of methods have been developed for isolating and analyzing dietary content of fibers, carbohydrates, phenol and protein in small millets *i.e*: enzymatic-gravimetric, DNS acid, Folin-Ciocalteu's assay method and Lowrys method respectively.

Methods and Materials

Lowry Method

In Lowry method protein assay is a biochemical assay for determine the total level of protein in a solution. The total protein concentration is exhibited by a color change of the sample solution in proportion to protein concentration, which can then be measured using colorimetric techniques. It is named for the biochemist Oliver H. Lowry. Who developed the reagent in the 1940s? His 1951 paper describing the technique is the most-highly cited paper ever in the scientific literature, cited over 300,000 times. The method was first proposed by Lowry in 1951. The Bicinchoninic acid assay and the Hartree–Lowry assay are subsequent modifications of the original Lowry procedure.

The method combines the reactions of copper ions with the peptide bonds under alkaline conditions (the Biuret test) with the oxidation of aromatic protein residues. The Lowry method is based on the reaction of Cu^{+}, produced by the oxidation of peptide bonds, with Folin–Ciocalteu reagent (a mixture of phosphotungstic acid and phosphomolybdic acid in the Folin–Ciocalteu reaction). The reaction mechanism is not well understood, but involves reduction of the Folin–Ciocalteu reagent and oxidation of aromatic residues (mainly tryptophan, also tyrosine). Experiments have shown that cysteine is also reactive to the reagent. Therefore, cysteine residues in protein probably also contribute to the absorbance seen in the Lowry assay. The result of this reaction is an intense blue molecule known as hetero-poly-molybdenum Blue. The concentration of the reduced Folin reagent (hetero-poly-molybdenum Blue) is measured by absorbance at 660 nm. As a result, the total concentration of protein in the sample can be deduced from the concentration of tryptophan and tyrosine residues that reduce the Folin–Ciocalteu reagent.

Enzymatic-gravimetric methods

The original AOAC enzymatic-gravimetric method for the determination of total dietary fiber (TDF) was developed on basis of the joint experience .The enzymatic-gravimetric method measures only a fraction of the fiber components because it can solubilize some of the structural polysaccharides and lignin. In crude fiber methods uses successive acid and alkaline digestion to isolate the insoluble fraction. The fiber method are continues to be used in some regions of the world as well as in the animal feed industry. This method provides a measure of total dietary fiber by enzymatic removal of available starch and solubilization and extraction of a portion of the protein; the remaining residue is dried, weight, and corrected for crude protein and ash contents. In origins of the methods can be traced back to the biochemical approach of measuring the indigestible residues in human foods introduced by Williams and Olmsted in the 1930s, and 1935 and work . These methods use enzymes to remove the digestible component, which was considered more physiological than extraction with chemical reagents as used in the detergent method. In this method the main steps in the AOAC procedure and the recent modification proposed by are outlined in Table 1. In this procedure consists of the following step: (a) weighing of -1 g in a beaker, (b) incubation with enzymes to remove starch and protein, (c) precipitation of soluble polymers with 4 volumes of ethanol, (d) quantitative transfer of sample to a pre-weighed crucible, (e) filtration, (f) weighing of the dry crucible and (g) correction for ash and protein in residues. Blanks are carried through the procedure in parallel. In the modification of the phosphate buffer is replaced with a MES/TRIS (2(N-morpholino) ethanesulfonic acid/tris hydroxymethyl amino-methane) buffer, one of the pH adjustment is eliminated and the total volume is reduced.

DNS acid method

3, 5-Di nitro salicylic acid (DNS or DNSA, IUPAC name 2-hydroxy-3, 5-dinitrobenzoic acid) is an aromatic compound that reacts with reducing sugars and other reducing molecules to form 3-amino-5-nitrosalicylic acid, which strongly absorbs light at 540 nm. In this method was first introduced for detect reducing substances in urine by James B. Sumner and has while been widely used, for example, for quantifying carbohydrate levels in blood. It is mainly used in assay of alpha-amylase. Though, enzymatic methods are usually preferred due to DNS lack of specificity. 3, 5- Di nitro salicylic acid can prepare by the nitration of salicylic acid. This method tests for the presence of free carbonyl group (C=O), the so-called reducing sugars. This involves the oxidation of aldehyde functional group present in, for

example, glucose and the ketone functional group in fructose. At once, 3, 5-dinitrosalicylic acid (DNS) is reduced to 3-amino, 5-nitrosalicylic acid under alkaline conditions:

Oxidation

Aldehyde group ---------->Carboxyl group

Reduction

3, 5-dinitrosalicylic acid ---------->3-amino, 5-nitrosalicylic acid

Since dissolved oxygen can interfere with glucose oxidation, sulfite, which itself is not necessary for the color reaction, is added in the reagent to absorb the dissolved oxygen. In above reaction scheme shows that one mole of sugar will react with one mole of 3, 5-dinitrosalicylic acid. On the other hand, it is suspected that there are many side reactions, and the actual reaction stoichiometry is more complicated than that previously described. The type of side reaction depends on the exact nature of the reducing sugars. Different reducing sugars commonly yield different color intensities; thus, it is necessary to calibrate for each sugar. In addition to the oxidation of the carbonyl groups in the sugar, other side reactions such as the decomposition of sugar also compete for the availability of 3, 5-dinitrosalicylic acid. As a consequence, carboxy methyl cellulose can affect the calibration curve by enhancing the intensity of the developed color.

Although this is a convenient and relatively inexpensive method, due to the relatively low specificity, one must run blanks diligently if the colorimetric results are to be interpreted correctly and accurately. One can resolve the background absorption on the original cellulose substrate solution by adding cellulase, immediately stopping the reaction, and measuring the absorbance, *i.e.* exactly the same procedures for the actual samples. When the effects of extraneous compounds are not known, one can effectively include a so-called internal standard by first fully developing the color for the unknown sample then, a known amount of sugar is add to this sample. This increase in the absorbance upon the second color development is equivalent to the incremental amount of sugar added**.**

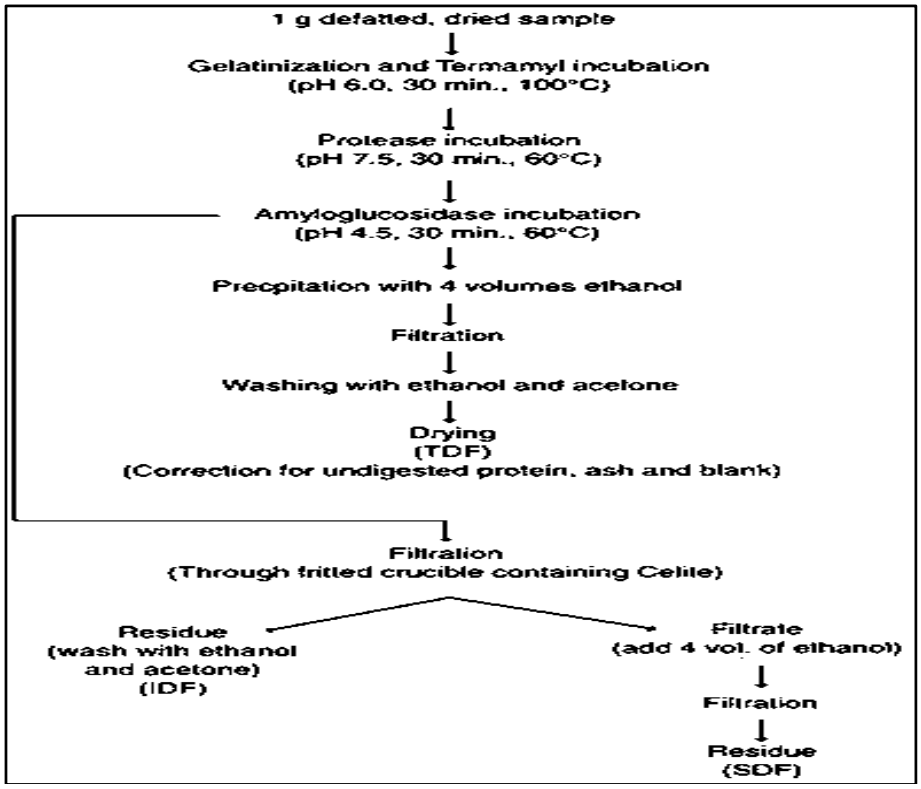

Fig 1: Analysis of Total Dietary Fibre (TDF), Insoluble Dietary Fibre (TDF) and SolubleDietary Fibre (SDF) by AOAC method 32.1.17 (45)

Folin-Ciocalteu's assay

The Folin–Ciocalteu reagent (FCR) or Folin's phenol reagent or Folin–Denis reagent, also called the gallic acid equivalence method (GAE), is a mixture of phosphomolybdate and phosphotung state used for the colorimetric *in-vitro* assay of phenolic and polyphenolic antioxidants. It is named after Otto Folin, Vintilă Ciocâlteu, and Willey Glover Denis.

The reagent does not measure only phenols, but will react with any reducing substance. It therefore measures the total reducing capacity of a sample, not just phenolic compounds. This reagent is part of the Lowry protein assay, and will also react with some nitrogen-containing compounds such as hydroxylamine and guanidine. The reagent has also been shown to be reactive towards thiols, many vitamins, the nucleotide base guanine, the trioses glyceraldehydes and dihydroxy acetone, and some inorganic ions. Copper complexation increases the reactivity of phenols towards this reagent. This reagent is distinct from Folin's reagent, which is used to detect amines and sulfur-containing compounds. A 1951 paper entitled "Protein measurement with the Folin phenol reagent" was the most cited paper in the 1945–1988.

Statistical analysis

Quantitative presented data were conducted and the results were reported as mean values with their respective standard deviations with RBD.

Results and Discussion

Varietals variations with respect to biochemical composition and some of the technological characteristics of the small millets have been recorded. The biochemical analysis done for small millet by lowrys method, gravimetric method, Folin-Ciocalteu's assay and DNS methods. The parameters analyzed are total carbohydrate content, protein content, fiber content and phenolic content in 20 different isolates of small millets. Protein was analyzed following a colorimetric assay after digestion with sulphuric acid. The analyses of the protein content of the different isolates showed significant differences. Available carbohydrates were analyzed after hydrolyzed by sulphuric acid and optical density at 540 nm was determined by the DNS method. The concentration of total phenolic compounds in water extracts was measured according to Supernatant (1 ml) was mixed with 1 ml of 95% ethanol, 5 ml of distilled water, and 0.5 ml of 1N Folin-Ciocalteau reagent and immediately vortexed. After 5 min, 1 ml of sodium carbonate was added and allowed to stand for 60 min in the dark. The samples were mixed again and their absorbances were measured at 725 nm against a 95% ethanol blank. The absorbances were converted to micrograms of catechin per gram of fresh weight of the sample. A calibration curve was generated using (+)-catechin as standard. Results were expressed as mg catechol equivalents per 100g of flour. The soluble, insoluble and total dietary fiber was estimated by using total dietary fiber assay kit (TDF 100-A and TDF 10-C, Sigma, St Louis) based on enzymatic gravimetric analysis by AOAC Methods.

Protein content

The protein content of collected sava millets varied in its composition. Total proteins observed in the year of 2015-16, 2016-17 and 2017-18. The pooled content of total protein across all the varieties was observed in the range of 4.71-7.85%. The protein content of MM-32, MM-15 and MM-17 varieties was observed higher in all sava varieties. While, lowest protein content was found in MM-20.

Similarly protein content was observed in the year of 2015-16, 2016-17 and 2017-18 with kutki variety of millet. Kutki variety showed higher content of protein in comparison to sava variety. The pooled content of protein was observed in the range of 10.31-12.78%. MM-18 was showed higher content of protein while MM-23 showed lowest. The variation detected may be due to their places of origin and therefore referred as genetic.

Phenolic content

The Folin-Ciocalteu's assay used to determine the TPC (total phenolic content) is based on the reducing ability of hydroxyl groups attached to phenolic compounds of the seeds. In this study, the TPC was observedin the year of 2015-16, 2016-17 and 2017-18 with savaand kutki millets variety.The highest TPC pooled content was observed in the variety of sava MM-32 with 7.42 % and kutki MM-23 with the value of 4.26%. MM-1 (6.35%) of sava variety showed lowest TPC whereas in kutki variety MM-31 and MM-29 with the value of 3.26 %.

Fiber content

The estimation of fiber content by enzymatic gravimetric method. This method provides a measure of total dietary fiber by enzymatic removal of available starch and solubilization and extraction of a portion of the protein; the remaining residue is dried, weighed and corrected for crude protein and ash contents. The statistical analysis of small millets produced maximum and minimum value of fiber content in the year of 2015-16, 2016-17 and 2017-18. The analysis of the pooled fiber content of the different isolates showed significant differences. The higher fiber content was observed in MM-3 (13.87) and lowest fiber content in MM-24(11.92) and MM-25 (11.51) in the sava variety of millet. Similarily in the kutki variety of millet showed highest fiber content in the MM-31 variety with the value of 6.28 % and lowest was observed in the MM-28 (4.92%).

Carbohydrate content

The estimation of carbohydrate is done by 3, 5-DNS method. 3, 5-Dinitrosalicylic acid is an aromatic compound that reacts with reducing sugars and other reducing molecules to form 3-amino-5-nitrosalicylic acid, which strongly absorbs light at 540 nm. The analysis of the carbohydrate content of the different isolates of millet in the year of 2015-16, 2016-17 and 2017-18 showed significant differences. The highest carbohydrate content of pooled data showed in sava variety MM-4 (65.85%) and MM-30 (65.62%) while lowest carbohydrate content was observed in MM25 with the value of 57.74. Similarily in the kutki variety of millet showed highest carbohydrate content in the MM-31 variety with the value of 76.82 % and lowest was observed in the MM-28 (72.62%).

Table 1: Protein per cent in genotypes/varieties of Sava

Entries name	Protein per cent			
	2015-16	2016-17	2017-18	Pooled
MM-1	5.87	5.75	5.99	5.87
MM-2	6.31	6.18	6.36	6.28
MM-3	4.93	4.82	4.99	4.91
MM-4	4.87	4.77	4.97	4.87
MM-6	4.95	4.84	5.05	4.95
MM-15	6.28	6.15	6.46	6.29
MM-17	6.28	6.14	6.43	6.28
MM-20	4.71	4.60	4.82	4.71
MM-24	6.28	6.14	6.27	6.23
MM-25	6.28	6.14	6.43	6.28
MM-26	6.19	6.08	6.34	6.20
MM-30	6.19	6.05	6.18	6.14
MM-32	8.73	7.56	7.25	7.85
SE m±	0.37	0.31	0.32	0.24
CD at 5%	1.07	0.92	0.94	0.68

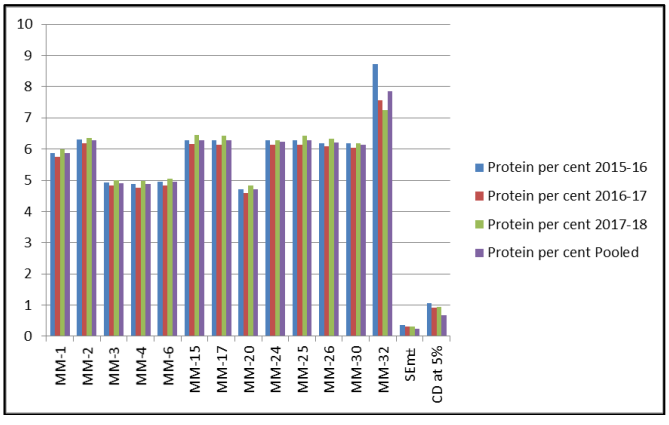

Fig 1: Protein per cent in genotypes/varieties of Sava

Table 2: Carbohydrate per cent in genotypes/varieties of Sava

Entries name	Carbohydrate per cent			
	2015-16	2016-17	2017-18	Pooled
MM-1	62.50	62.24	63.33	62.69
MM-2	61.58	67.07	64.40	64.35
MM-3	58.58	56.78	58.63	58.00
MM-4	66.08	67.07	64.40	65.85
MM-6	60.21	59.31	61.11	60.21
MM-15	58.93	58.05	59.81	58.93
MM-17	59.43	58.54	61.60	59.86
MM-20	59.07	64.40	64.63	62.70
MM-24	59.54	58.65	57.53	58.57
MM-25	58.66	57.01	57.53	57.74
MM-26	60.81	59.90	61.72	60.81
MM-30	65.29	65.31	66.27	65.62
MM-32	59.57	58.68	60.46	59.57
SE m±	1.37	1.91	1.75	1.20
CD at 5%	4.02	5.58	5.13	3.43

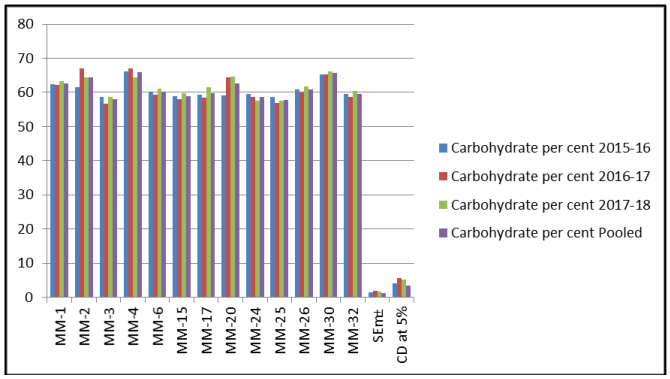

Fig 2: Carbohydrate per cent in genotypes/varieties of Sava

Table 3: Polyphenolic per cent in genotypes/varieties of Sava

Entries name	Polyphenolic per cent			
	2015-16	2016-17	2017-18	Pooled
MM-1	6.40	6.53	6.14	6.35
MM-2	6.52	6.32	7.07	6.64
MM-3	7.07	7.21	6.86	7.05
MM-4	7.07	6.93	6.86	6.95
MM-6	6.71	6.84	6.51	6.69
MM-15	6.48	6.61	6.29	6.46
MM-17	7.50	7.65	7.36	7.51
MM-20	6.52	6.65	6.32	6.49
MM-24	6.89	7.03	6.74	6.89
MM-25	6.64	6.79	6.46	6.63
MM-26	6.79	6.93	6.62	6.78
MM-30	7.03	7.17	6.93	7.04
MM-32	7.35	7.50	7.42	7.42
SE m±	0.21	0.24	0.23	0.16
CD at 5%	0.61	0.70	0.67	0.46

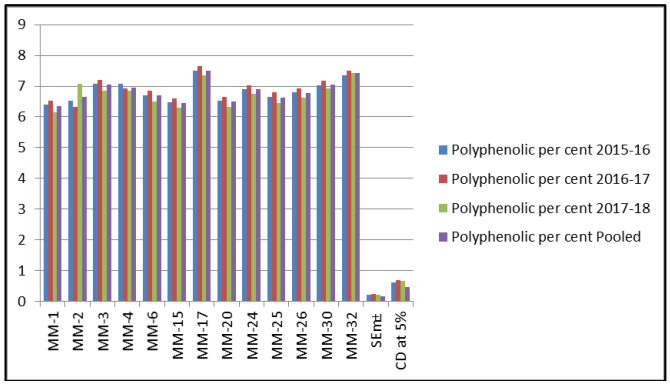

Fig 3: Polyphenolic per cent in genotypes/varieties of Sava

Table 4: Fiber per cent in genotypes/varieties of Sava

Entries name	Fiber per cent			
	2015-16	2016-17	2017-18	Pooled
MM-1	13.60	13.83	14.11	13.85
MM-2	12.42	12.81	13.07	12.77
MM-3	13.59	13.87	14.15	13.87
MM-4	12.41	12.47	12.72	12.54
MM-6	12.34	12.40	12.65	12.47
MM-15	12.62	12.68	12.93	12.74
MM-17	13.51	13.58	13.85	13.65
MM-20	12.95	13.77	14.05	13.59
MM-24	11.80	11.86	12.09	11.92
MM-25	11.39	11.45	11.68	11.51
MM-26	12.74	12.80	13.06	12.87
MM-30	13.29	13.36	13.63	13.43
MM-32	13.39	13.46	13.73	13.53
SE m±	0.36	0.50	0.49	0.32
CD at 5%	1.05	1.45	1.44	0.92

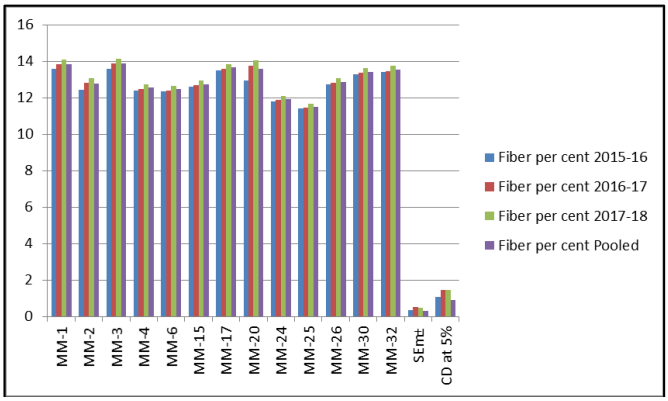

Fig 4: Fiber per cent in genotypes/varieties of Sava

Table 5: Protein per cent in genotypes/varieties of Kutki

Entries name	Protein per cent			
	2015-16	2016-17	2017-18	Pooled
MM-10	12.32	13.16	12.67	12.72
MM-18	12.43	13.19	12.74	12.78
MM-19	10.30	10.92	10.55	10.59
MM-23	10.08	10.36	10.50	10.31
MM-28	10.97	11.64	11.24	11.29
MM-29	10.91	11.58	11.18	11.22
MM-31	10.39	10.56	10.53	10.49
SE m±	0.40	0.56	0.56	0.35
CD at 5%	1.17	1.63	1.63	1.00

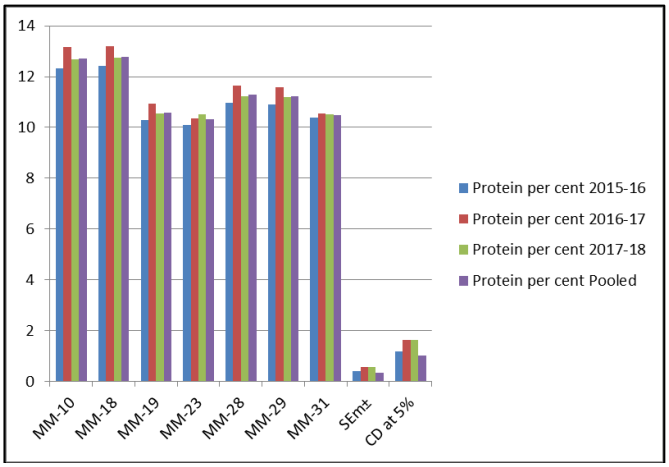

Fig 5: Protein per cent in genotypes/varieties of Kutki

Table 6: Carbohydrate per cent in genotypes/varieties of Kutki

Entries name	Carbohydrate per cent			
	2015-16	2016-17	2017-18	Pooled
MM-10	72.47	73.08	72.74	72.76
MM-18	72.93	73.55	74.27	73.58
MM-19	76.11	76.76	74.47	75.78
MM-23	74.75	75.38	76.11	75.41
MM-28	71.97	72.59	73.29	72.62
MM-29	71.94	72.55	73.44	72.65
MM-31	75.90	76.91	77.66	76.82
SE m±	0.93	1.00	1.00	0.68
CD at 5%	2.73	2.93	2.93	1.93

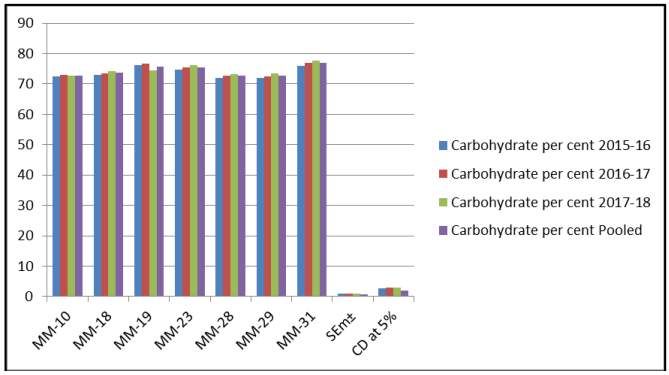

Fig 6: Carbohydrate per cent in genotypes/varieties of Kutki

Table 7: Polyphenolic per cent in genotypes/varieties of Kutki

Entries name	Polyphenolic per cent			
	2015-16	2016-17	2017-18	Pooled
MM-10	3.41	3.58	3.34	3.45
MM-18	4.12	4.33	4.09	4.18
MM-19	3.68	3.87	3.61	3.72
MM-23	4.17	4.43	4.19	4.26
MM-28	2.99	3.14	2.93	3.02
MM-29	3.22	3.39	3.16	3.26
MM-31	3.23	3.39	3.16	3.26
SE m±	0.17	0.19	0.19	0.13
CD at 5%	0.49	0.55	0.55	0.37

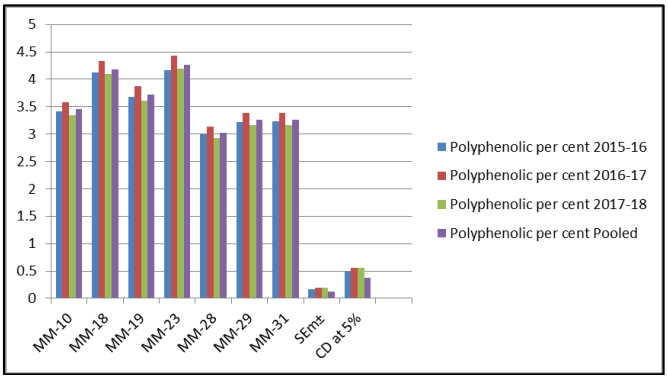

Fig 7: Polyphenolic per cent in genotypes/varieties of Kutki

Table 8: Fiber per cent in genotypes/varieties of Kutki

Entries name	Fiber per cent			
	2015-16	2016-17	2017-18	Pooled
MM-10	5.52	5.63	5.74	5.63
MM-18	5.30	5.40	5.50	5.40
MM-19	4.87	4.97	5.07	4.97
MM-23	5.11	5.21	5.32	5.21
MM-28	4.82	4.92	5.01	4.92
MM-29	4.93	5.03	5.13	5.03
MM-31	6.14	6.28	6.41	6.28
SE m±	0.25	0.24	0.24	0.17
CD at 5%	0.73	0.69	0.69	0.48

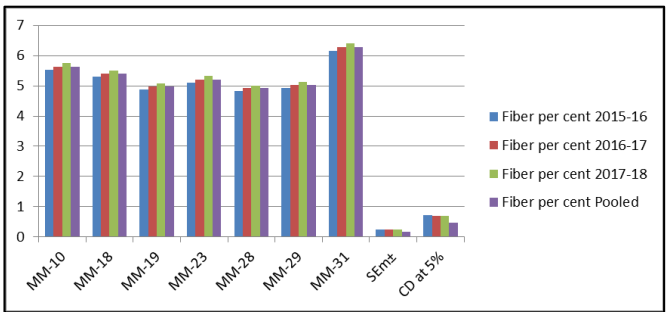

Fig 8: Fiber per cent in genotypes/varieties of Kutki

Conclusions

In this study, biochemical variation in 20 isolates of small millet in nutritive content, mainly protein, carbohydrate phenols and fiber were found. Such information would be of use to identify cultivars suitable for various end uses and assist the plant breeders to improve the grain quality. Millets are staple food source, which are not only providing major nutrients like protein, carbohydrates, fats etc. but also provide ample of vitamins and minerals. In developing countries malnutrition and various health problems like obesity, diabetes, cardiovascular diseases, cancer, celiac disease etc. are most prominent because of inadequate supply of nutrition. This is mainly due to the lack of awareness and knowledge among the people in choosing the kind of food, especially the small millets. Millets are easily available and cheap in cost. Millets contains many major and minor nutrients like carbohydrates, protein, fat, dietary fiber, vitamins and minerals as well as antioxidants and phytochemicals. The importance of this study under takes to concern and to develop specific agenda for these crops which must be recognized as an important food and to introduce the millets as a nutritious food for fulfillment of the nutritional need of the global population and also to find ways to consume the millets effectively and to reduce the problems of malnutrition and other health problems.

The study emphasized on nutraceutical properties of millets and the application of millets as alternative to, cereals, potentially healthy to elaborate therapeutic food products like protein and energy rich diet, diet for diabetes and for the gluten free diet. The study also showed that millets are used as “food medicine” as millets are good source of antioxidants such asphenolic acids and glycated flavonoids. Millet foods are also characterized to be a potential prebiotic and can enhance the viability of probiotics with potential health benefits**.**

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