Molecular Approaches to Understand Nutritional Potential of Coarse Cereals

Amit Kumar Singh[a], Rakesh Singh[a,*], Rajkumar Subramani[a], Rajesh Kumar[a] and Dhammaprakash P. Wankhede[a]

[a]*Division of Genomic Resources, ICAR- National Bureau of Plant Genetic Resources, New Delhi, India*

R. Singh

Abstract: Coarse grains are important group of crops that constitutes staple food for large population residing primarily in the arid and semi-arid regions of the world. Coarse grains are designated as nutri-cereals as they are rich in essential amino acids, minerals and vitamins. In spite of having several nutritional virtues in coarse grain as mentioned above, there is still scope for improvement in quality parameters such as cooking qualities, modulation of nutritional constituents and reduction or elimination of anti-nutritional factors. Besides its use in traditional cooking, coarse grains have been used mainly in the weaning food preparation and other malted food production. Improvement in quality parameters will certainly increase consumer's preference for coarse grains and increase their demand. The overall genetic gain in quality traits of economic importance in the cultivated varieties will enhance their industrial value and simultaneously increase income of farmers growing these varieties. The urgent step for improvement of quality traits in coarse grains requires a detailed understanding of molecular mechanisms responsible for varied level of different nutritional contents in different genotypes of these crops. In this review we have discussed the progresses made in understanding of coarse grain biology with various omics tool coupled with modern breeding approaches and the current status with regard to our effort towards dissecting traits related to improvement of quality and nutritional constituents of grains.

Keywords: Association mapping, Coarse grains, Genomics, Metabolomics, Millets, Proteomics, Transcriptomics, Quality traits.

1. INTRODUCTION

'Coarse grains' is a generic term used to refer cereals other than rice and wheat. It includes barley (*Hordeum vulgare*), oat (*Avena sativa*), maize (*Zea mays*), sorghum (*Sorghum bicolor*), pearl millet (*Pennisetum glaucum*) and minor millets such as finger millet (*Elusine coracana*), foxtail millet (*Setaria italica*), proso millet (*Panicum milaceum*), kodo millet (*Paspalum setaceum*), little millet (*Panicum sumatrense*) and barnyard millet (*Echincloea colona*) [1, 2]. Among coarse cereals, millets are better adopted to infertile soils and important edible crops in semi arid tropical regions of Asia and Africa [3]. On the other hand, barley and oat are widely grown in European and North- American countries where they are used as ingredients in diverse food products as well as feed grain for animals and poultry. Additionally, the larger share of barley produce goes to brewery for its high quality malt. The past few years have witnessed increase in the demand of coarse cereals as food products due to several inherent beneficial health effects. Coarse cereals have nutritional value which is comparable or sometimes superior to rice and wheat [2]. In addition to their high calorific value, they are rich in dietary fibers, minerals, essential amino acids

and also source of bioactive compounds having various health benefits [1, 4]. Despite above mentioned benefits, coarse cereals continue to have small share in world's food basket primarily due to the lower acceptability of their food products. The reasons for poor acceptability of the coarse cereals are the unattractive appearance of their food products, poor digestibility of proteins and starch and presence of several anti-nutritional factors [5-7]. Coarse cereals, except maize, barley and oat, though being the major crops of developing countries have received relatively less research attention than rice and wheat. However, in recent years, there is growing interest in researchers to undertake research in coarse cereals primarily due to their high nutritive value, beneficial health effects and ability to grow in adverse climatic conditions. Furthermore, availability of advanced, high-throughput omics tools has also accelerated fundamental and applied research in crops [8-12]. Application of these tools has promises to unravel the molecular basis of nutritional quality traits in coarse cereals, which will be instrumental for strategic improvement of their nutritional attributes and enhancing their acceptability. Also, the knowledge of molecular mechanisms underlying high content of various nutritional and bioactive compounds may be used for improvement of these traits in other food crops as well. In present article we have reviewed applications of high-throughput omics tools including genomic analysis procedures such as association-mapping and various other func-

*Address correspondence to this author at the Division of Genomic Resources, ICAR- National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi 110 012. India; Tel: 91-11-25842495; Ext: 203; E-mail: rsingh@nbpgr.ernet.in

tional genomics tools that led to better understanding of quality traits in coarse cereals except maize where it has been dealt extensively in other publications [13-15].

2. NUTRITIONAL QUOTIENT OF COARSE CEREALS AND CHALLENGES AHEAD

A balanced human diet contains adequate proportion of various foods that can optimally supply all essential dietary components/nutrients required for overall development. Although, all essential nutrients requirement for proper human growth and development cannot be met from single crop source, and therefore inclusion of adequate quantity of coarse cereals in diet can meet requirements of most of the nutrients as they are rich in minerals, vitamins, crude fibres and essential amino acids. The average nutritional composition of coarse cereals along with rice and wheat is presented in (Table **1**) [16, 17]. It is interesting to note that coarse grains contain higher amount of micronutrients (Fe and Zn), crude fibres (Table **1**) and essential amino acids except lysine and methionine (Table **2**) [18] than primary cereals, rice and wheat. The average fat, crude fibre, ash, iron and calcium content of both sorghum and pearl millet are higher than both rice and wheat. Finger millet is one of the richest sources of calcium (350 mg/100 g edible protein) among known foods and has higher level of crude fibre, minerals and sulphur containing essential amino acids than rice and wheat [17, 19]. The crude fibre content of barley is the highest (15.6) followed by barnyard millet (13.6%) and oats

(11%)%), which is much higher in comparison to rice (1%) and wheat (2%). Barnyard millet contains substantially higher level of iron (18.6%) than other cereals. Protein content of proso millet (12.5), foxtail millet (11.5) sorghum (10.4), pearl millet (11.8) and barnyard millet are comparable to wheat and their essential amino acids composition is generally superior to both rice and wheat [20]. Moreover, coarse grains are rich in several phytochemials as well, which helps in preventing cardiovascular diseases and cancer, lowering blood cholesterol and in managing obesity and type II diabetes [21, 22]. Barley and oat are the rich source of (1-3, 1-4)-*β*-D-glucans commonly referred as β-glucan, a soluble crude fibre implicated in reducing blood cholesterol and managing diabetes. Furthermore, many phytochemicals particularly phenolics found in coarse cereals are good antioxidants [23]. Luteolin, a flavone found in sorghum and oat is known for its antioxidant, cancer preventive and antiinflammatory properties [24]. Avenanthramides, exclusively found in oat, has anti-inflammatory, anti-athrogenic and antioxidant properties [25]. Additionally, some varities of millets and oats are gluten free and therefore considered safe alternative foods for celiac patients [26, 27]. The above mentioned nutritional quality features reflect great potential of coarse cereals to address problem of hunger, malnutrition and nutritional insecurity for the large poor population living predominantly in many Asian and African countries. Although coarse grains have rich repertoire of nutritional constituents, their nutritive value is considerably offset by the presence of one or more anti-nutritional compounds such as polyphenols, tannins, phytate, amylase and protease

Table 1.   Nutrient composition of coarse cereals, rice and wheat (per 100gm edible portion; 12% moisture).

Crops	Carbohydrate (g)	Protein (g)	Fat(g)	Ash(g)	Crude(g) Fiber	Energy (kcal)	Ca (mg)	Fe(mg)	Thiamin (mg)	Riboflavin (mg)	Niacin(mg)
Rice	76	7.9	2.7	1.3	1.0	362	33	1.8	0.41	0.04	4.3
Wheat	71	11.6	2.0	1.6	2.0	348	30	3.5	0.41	0.10	5.1
Barley	77.2	9.9	1.2	-	15.6	352	29	2.5	0.19	0.11	4.6
Oats	66.27	16.89	5.93	2.6	10.6	389	54	4.7	0.76	0.13	0.96
Sorghum	70.7	10.4	3.1	1.6	2.0	329	25	3.9	0.38	0.15	4.3
Pearl millet	67.0	11.8	4.8	2.2	2.3	363	42	11.0	0.38	0.21	2.8
Finger millet	72.6	7.7	1.5	2.6	3.6	336	350	3.9	0.42	0.19	1.1
Foxtail millet	63.2	11.2	4.0	3.3	6.7	351	31	2.8	0.59	0.11	3.2
Common millet	63.8	12.5	3.5	3.1	5.2	364	8	2.9	0.51	0.28	4.5
Little millet	60.9	9.7	5.2	5.4	7.6	60.9	17	9.3	0.41	0.09	3.2
Kodo millet	66.6	9.8	3.6	3.3	5.2	66.6	35	1.7	0.15	0.09	2.0
Barnyard millet	55.0	11.0	3.9	4.5	13.6	55	22	18.6	0.33	0.10	4.2

Source: Hulse *et al.* (1980); USDA nutrient data base.

Table 2.   Essential amino acids content of coarse cereals, rice and wheat (g/100g).

Amino Acids	Sorghum	Pearl Millet	Finger Millet	Proso Millet	Foxtail Millet	Kodo Millet	Barnyard Millet	Barley	Wheat	Rice
Isoleucine	3.9	4.1	4.4	8.1	7.6	3.0	8.8	3.5	3.3	3.8
leucine	13.3	9.6	9.5	12.2	16.7	6.7	16.6	9.8	6.7	8.2
Lysine	2.0	3.4	2.9	3.0	2.2	3.0	2.9	2.6	2.8	3.8
Methionine	1.4	2.5	3.1	2.6	2.8	1.5	1.9	1.6	1.5	2.3
Cystine	1.4	1.8	2.2	1.0	1.6	2.6	2.8	1.6	2.2	1.4
Phenyle alanine	4.9	4.8	5.2	4.9	6.7	6.0	2.2	5.1	4.5	5.2
Tyrosine	2.7	3.3	3.6	4.0	2.2	3.5	2.4	3.6	3.0	3.9
Threonine	3.1	3.1	3.8	3.0	2.7	3.2	2.2	3.5	2.8	4.1
Tryptophan	1.1	2.0	1.6	0.8	1.0	0.8	1.0	1.4	1.5	1.4
Valine	5.0	5.5	6.6	6.5	6.9	3.8	6.4	5.8	4.4	5.5
Histidine	2.1	2.5	2.2	1.9	2.1	1.5	1.9	2.1	2.3	2.4

Source: Gopalan *et al.*, 2007.

polyphenols, tannins, phytate, amylase and protease inhibitors and various other compounds with harmful health effects [28]. These anti-nutritional compounds are one of the main factors implicated for poor digestibility of starch, protein and reduced bioavailability of minerals and other nutritional compounds in various coarse grains [5, 6, 29]. Phytic acid forms insoluble complexes with multivalent cations and proteins rendering them unavailable for absorption by human body [30]. Similarly, high concentration of polyphenols is one of the reasons for low digestibility of proteins in some coarse cereals as they form insoluble complexes with protein and digestive enzymes. [5]. Goitrogen, a type of polyphenol found in pearl millet bran, is reported to cause goitre by inhibiting the conversion of thyroxine to triidothyronine [31]. Avenin protein of oat is reported to induce immune response in some celiac patients [32]. The high β-glucan content of barley is a highly undesirable feature for preparation of good quality malt as it enhances wort viscosity thus reducing beer filterability and also interferes with the enzymes involved in the malting process [33]. Therefore researchers engaged in coarse cereal quality improvement have two fold challenge; i) understanding the complex molecular mechanism involved in synthesis of high amount of various nutritional compounds, and ii) determining the molecular basis of poor digestibility of protein, starch and various other factors responsible for reduced bioavailability of nutritional components.

3. APPLICATION OF HIGH-THROUGHPUT OMICS TOOLS FOR DISSECTING QUALITY TRAITS

The last decade has seen phenomenal development of high-throughput omics tools, which has allowed researchers to perform comprehensive genome scale analysis for obtaining snapshot of diverse molecular processes in plants. The most widely used high-throughput tools include transcriptomics, proteomics, metabolomics and genomics. The genomics approaches includes TILLING (Targeting Induced Local Lesions In Genomes), RNAi (RNA interference) technology, transposon / T-DNA mediated insertional mutagenesis, VIGS (Virus Induced Gene Silencing) and association mapping [8-10, 12, 34-39]. Application of these tools has expedited identification of genes for various complex plant traits and thus has improved our overall molecular understanding of these traits. With increasing focus of society on role of nutritious foods in general well being and healthy life, researchers are pondering on ways and means to improve grain quality traits. Comprehensive knowledge of genetic and molecular basis of grain quality traits is necessary for designing strategies for their further improvement. However, the researcher have great challenges in dissecting the grain quality components as they are complex; controlled by several major and minor genes and also influenced by environmental condition. In the recent years, studies on coarse cereals have demonstrated the effectiveness of various high-throughput omics tools in analysing grain quality traits. Some of the key studies conducted on coarse cereals have been described and discussed in the following sections.

3.1. Barley

3.1.1. Association Mapping

In barley, both candidate gene-based association analysis and genome-wide association study (GWAS) have been applied for understanding molecular mechanisms involved in synthesis and regulation of nutritional components such as (1,3;1,4)-β-glucan, protein and its relation to (expression of) various malting quality associated traits [40-44]. Cai *et al.* [41] reported QTLs for grain protein content on all the chromosomes except 4H in a set of barley collection using GWAS. Further, they found that a major QTL on chromosome 6H, identified as *HvNAM-1a* NAC transcription factor, determined 40% of the total grain protein content. The finding was extremely important as NAC transcription factor have been reported to influence grain protein content by

regulating senescence and protein remobilization. Moreover, there was significant correlation between haplotypes of *HvNAM-1* and *HvNAM-2* and grain protein content, thus providing opportunities to develop high quality barley cultivars [41, 45]. Further association mapping has also identified QTLs/candidates genes for other grain quality traits including β-glucan, amylose and amylopectin content [42, 43, 46]. Houston *et al.* [43] have reported significant marker and trait associations for β-glucan content in a set of 605 elite barley cultivars using GWAS. This study showed genomic regions on chromosome 2 containing clusters of *Csl* (Cellulose synthase like) genes including *CslF3*, *CslF4*, *CslF8*, *CslF10*, *CslF12*, *CslH* and *CslF9* gene on chromosome 2H were associated with β-glucan content, thus reaffirmed that *Csl* genes must be having role in β-glucan synthesis [47-49]. Another GWAS study in barley identified several major and minor genes for β-glucan, amylopectin and amylose content on chromosomes 1H, 5H, 6H, and 7H. *HvCslF6, amo1 and AGPL2, sex6*, and *waxy* were among the major genes identified for β-glucan, amylopectin and amylose content, respectively and β- fructofuranosidase and leucine-rich repeat receptor kinase protein were identified as new candidates for amylopectin synthesis in barley endosperm [42]. Indeed, above studies have demonstrated that *HvCsl* genes must be playing important role in determining β-glucan content of barley. In addition, association mapping studies have identified many QTLs for malting and grain quality traits in barley [40, 44]. Interestingly, a few of the identified QTLs in these studies have been localised in the genomic regions reported to contain QTLs for these traits in earlier studies, which substantiate role of association mapping in underpinning molecular basis of quality traits [50].

3.1.2. Transcriptomics

Transcriptome analysis tools such as suppression subtractive hybridization, microarray, SAGE (Serial Analysis of Gene Expression), cDNA-AFLP (Amplified Fragment Length Polymorphism) and RNA-sequencing have been widely applied to analyze pattern of gene expression during malting and grain development for understanding molecular regulation of barley quality traits [51-56]. A cDNA array based gene expression analysis on 10 barley varieties during malting process identified 13-30 candidate genes for each of the six malting component phenotypes (α-amylase, diastatic power, free amino nitrogen, fine extract, malting protein, and soluble/total protein ratio). Few of the identified candidates were localised in the regions, reported to contain QTLs for malting quality associated traits [57]. Development of Affymetrix 22K Barley1 Gene-chip gave significant boost to transcriptomics studies in barley [58]. Comparative transcriptome analysis of four barley varieties at malting stages and un-germinated seed stage using Barley1 Gene-chip identified genes whose expression was correlated with various malting component traits. Out of 700 differentially expressed genes among the barley varieties, varying number of genes depending on varieties, were correlated with six malting quality traits. Genes encoding carbohydrate metabolism enzymes such as glucosidase, limit dextrinase, sucrose synthase I and fructofuranidase I were predominately expressed [54]. Furthermore, a SSH (Suppression Substractive Hybridization) library of the popular malting variety Morex has identified many differentially expressed genes during

malting process [54]. In another study genetic diversity among the 15 barley good malting quality varieties, developed in a barley breeding program, has been analyzed using Barley1 Gene-chip [56]. Though there were meager gene expression diversity, still diversity existed in elite lines to be used in breeding program to increase malting quality. Nevertheless, new candidates for malting quality were identified by comparing transcriptome of two latest varieties developed in this breeding program [56] Recently, paired-end-RNA-sequencing based transcriptome of developing grains of two hulless Tibetan barley landraces Nimubai and XQ754 has generated wealth of information about the expression pattern of genes involved in biosynthesis of storage compounds such as starch, protein, and β-glucan [55]. The *HvCslF6* gene was constitutively expressed at a relatively higher level in developing grains tissues that indicated its potential role in β-glucan synthesis, which corroborated earlier findings [47, 59]. Conversely, *HvCslH1,* another important gene responsible for β-glucan of barley grain was expressing at relatively low level in seed tissues and might be having role in regulating β-glucan content [55]. Although, this study reported varying expression of genes involved in biosynthetic pathway of various storage compounds; the putative candidates for each of the storage constituents need to be functionally characterized for their definite role.

3.1.3. Proteomics

Initial proteome analyses in barley were mainly performed to either catalogue seed proteins or differentiate varieties based on differences in grain quality proteins using 2-dimensional gel electrophoresis [60, 61]. However, with the availability of advanced high-throughput proteomic analysis tools [62], emphasis shifted towards entire proteome to analyze molecular processes during grain development for dissecting grain and malting quality traits [63, 64]. Proteome analysis of a set of barley varieties including malting and non malting type demonstrated that variations in spots across the varieties was due to single amino acid change which was caused by SNPs in the corresponding genes. A single amino acid substitution (Cys-Arg) was responsible for the variation in the β-amylase enzyme of two malting type varieties Barke and Morex. This study demonstrated that SNPs can link the proteome level changes to the genome and facilitate identification of proteins/enzyme determining various grain quality traits [64]. Moreover, the proteomics data of barley may be used to identify p-QTLs (protein quantitative trait loci) for quality traits as demonstrated in maize [65]. Following this strategy 48 protein spots have been assigned to chromosomes 1H, 2H, 3H, 5H and 7H on the genetic map using a double haploid population derived from the cross of malting (Scarlett) and feed barley cultivar (Meltan) [64]. In another study, two important malting quality QTLs on cromosome 1H and 4H have been validated in set of introgression lines using grain proteomic analysis. Introgression line carrying wild type allele of malting QTL at 4H demonstrated six fold increase in the level of dextrinase inhibitor [66]. Jin *et al.* [67] used comparative proteomics to characterize proteins responsible for malt filterability in two most popular malt purpose variety of barley for developing better cultivars. Earlier, entire seed was analyzed as one proteome irrespective of the purpose of analysis. Currently, proteomics study in crops including barley using various seed tissues (embryo,

endosperm and aleurone) are considered sub-proteomes for obtaining greater insight into underlying molecular basis of grain quality traits [68, 69]. Further, with the availability of draft barley genome sequence, future seed proteomics studies would be able to shed more light on the molecular basis underlying various grain and malting quality traits, as the protein spots can be identified by just sequencing their N-terminal followed by search for corresponding nucleotide sequence in the barley genome assembly.

3.1.4. Metabolomics

Metabolic profiling is of paramount importance in plants especially for unraveling the molecular mechanism of quality traits as the composition of the metabolic profiles of seed not only determines its nutritional value but also the physical features such as texture, hardiness, grain length, etc. In barley, metabolic profiling studies have been mainly performed to analyze the metabolic changes occurring during the malting process [70, 71]. The identified metabolities may be incorporated in to marker aided selection for improvement of varities. However, a recent study has reported metabolic profiling for grain and malting type quality traits by using UTGC-MS (ultra thin gas chromatography coupled- mass spectrometry) [72]. Such study conducted non-targeted metabolic profiling of 72 barley breeding lines (both two row and six row genotypes) grown at two environmental conditions and revealed close co-variation in metabolite concentrations and 20 quality traits. Both genotype and environment influenced concentration and composition of metabolites, which resulted in variation for quality traits. Moreover, metabolites significantly associated with trait of interest have been individually mapped through its biosynthetic protein to the barley genome ,which is likely to improve our understanding of underlying molecular mechanism governing association of metabolites and quality traits [72].

3.1.5. Functional Genomics Approaches

In addition to above mentioned 'Omics' tools, various functional genomics approaches such as RNA interference, insertional mutagenesis, TILLING and transgenic technology have also been employed to understand grain quality in barley. RNAi technique has been extensively applied to decode function of various genes associated with starch synthesis. The starch quality of cereals essentially depends on the proportion of amylose *vs.* amylopectin. The amylose only starch is known to having beneficial health effects and is important desirable quality parameter in cereals [73]. Using RNAi technology, role of starch branching enzymes SBEIIa and SBEIIb in amylopectin synthesis, its chain length distribution and branching frequency in barley has been elucidated [74]. Transgenic barley lines with the both the genes (*SBEIIa* and *SBEIIb*) suppressed accumulated higher amount of amylose, however, suppression of either of gene had little effect on amylose content. This suggested that possible functional interaction of SBEIIb as inhibitory role with other SBE isoforms. [74]. Carciofi *et al.* [75] have developed amylose rich grains by simultaneously suppressing all the starch branching genes (*SBEI, SBEIIa* and *SBEIIb*) using a chimeric hairpin targeting components of each of three genes. The structure of amylose rich grains were normal that indicating that amylopectin was not essential for starch granule crystalline structure and integrity. Additionally, several barley mu-

tant lines have been generated through insertional mutagenesis using the Ac-Ds transposon system and may be potentially exploited for identification of quality related genes [76-78]. Singh *et al.* [78] generated a mutant population in barley for the genomic region containing malting quality QTLs on chromosome 4H through reactivation of Ds element in transposon tagged lines. Molecular characterization of these lines using inverse PCR identified mutants for various genes including β-GAL1, β-amylase-like gene and ABC transporter that may be used as tools for molecular analysis of quality traits in barley. Moreover, many TILLING populations are also available in barley [79-83], which may be exploited for discovering genes associated with quality traits. With the availability of NGS technologies, entire TILLING populations can be parellely sequenced and analyzed to understand mechanisms involved in synthesis and regulation of various quality traits. Further, *HVcslF6* gene, isolated from both mutants (β-glucan less; *bgl*) and wild type barley have been functionally characterized using transgenic approach. Transgenic tobacco plants expressing wild type *HVcslF6* gene demonstrated β-glucan synthesis whereas those expressing mutant form of this gene failed to synthesize even trace of β-glucan; thus implicating its unequivocal role in β-glucan biosynthesis [84].

3.2. Sorghum

3.2.1. Association Mapping

With the availability of sufficient genomic resources, sorghum researchers have started to exploit potential of association mapping for discovering genes and getting detailed molecular level insight of various economically important traits including abiotic stress tolerance, biotic stress tolerance and grain quality [85-89]. A good number of these association mapping studies are on grain quality traits which have mainly focused on two aspects namely, identifying molecular basis of poor protein content and starch digestibility and deciphering mechanism responsible for high concentration of various nutritional constituents including starch, proteins and micronutrients. A candidate gene based association analysis revealed that polymorphisms in genes of starch biosynthesis (*Sh2, Bt, SssI, Ae1,* and *Wx*) and protein content (*O2*) were significantly associated with various grain quality traits of sorghum. While *SssI* and *Ae1* showed associations with peak gelatinization temperature (PGT), which is influenced by amylopectin amount, the *Sh2* was associated with amylose content. Two genes *O2* and *Wx* were associated with hardiness and endosperm texture. However, no association was detected between *O2* and grain protein content. Sequence variation positively associated with starch and protein quality may be exploited for marker assisted breeding to develop improved varieties [90]. Association mapping studies in sorghum gained momentum from development of core / mini core sets of world and regional sorghum collections [91-93], availability of high-throughput genotyping platforms and availability of high density SNP haplotype maps [94-96]. Sukumaran *et al.* [87] applied both candidate genes based and genome wide association analysis (GWAS) in a sorghum collection for mapping of quality traits and has identified significant marker trait association for ten grain quality traits. The two SNPs, one each in starch synthase IIb (*SSIIb*) and *pSB1120* respectively, were significantly associated with

starch content. Further, a SNP in starch synthase IIa (*SSIIa*) was associated with kernel hardness, which suggests its role in enhancing amylose content. The favourable SNPs identified in the above genes may be targeted for improving starch content and associated with physical quality parameters of sorghum grain. Sorghum grain contains tannin that contributes to its nutritional value as it has several beneficial health effects [24]. However, high tannin content of sorghum is considered to be one of the factors responsible for poor digestibility of the sorghum grain proteins [6]. Application of association analysis along with other approaches demonstrated that nucleotide polymorphism in *Tanin-1* (*tan-1*) gene coding for WD-40 protein controlled tannin biosynthesis in sorghum [97]. The above study led to identification of two non-functional alleles of *tan-1*, the *tan-1a* has a 1-bp deletion and the *tan-1b* has a 10-bp insertion in the coding region. This knowledge may be used for developing markers and consequent improvement of protein digestibility and improving nutritional qualities in sorghum. Recently, molecular basis of starch digestibility which is an another limiting factor responsible for poor acceptability of sorghum was deciphered by applying haplotype trait association analysis of a starch metabolism gene pullulanase (*SbPUL*) in a collection of sorghum land races. A low frequency haplotype (*SbPUL-RA*) was demonstrated to be significantly associated with increase starch digestibility regardless of the genetic background [88]. In addition, association mapping has proven equally useful in unraveling mechanism responsible for high content of essential micronutrients such as Zn and Fe. Most recently, Anuradha *et al.* [89] identified genomic regions and candidates for Fe and Zn content using association analysis in a set of sorghum collection that were genotyped by GBS (Genotyping by Sequencing). The significant marker (SNPs) trait associations for Fe content was identified on chromosomes 2, 4 and 8; and for Zn on chromosomes 1, 3 and 5. Physical positioning of SNPs sowing significant associations on the sorghum genome assembly revealed that they were closely placed to the homologues of the various genes governing Fe and Zn concentration reported in other cereals such as rice, barley and maize. The above studies have demonstrated utility of association mapping in improving molecular understanding of sorghum grain quality traits.

3.2.2. Proteomics

Initial proteomic studies in sorghum mainly focused at characterization of grain proteins and developing proteome based cultivar identity. Nevertheless, few studies have used proteomics for understanding molecular basis of grain quality traits [98, 99]. Roy *et al.* [98] investigated seed proteome of a popular inbred and mutant line using 2D-GE coupled with MALDI-TOF-TOF-MS (Matrix Assisted Laser Desorption Ionization-Time of Flight- Time of Flight-Mass Spectrometry). Comparison of protein composition between inbred and the mutant genotypes identified various starch and sugar metabolism enzymes / proteins which may be analyzed for their association for various grain quality traits. Additionally, proteomics studies have also improved our understanding of poor digestibility of sorghum seed proteins. In sorghum major storage proteins are kafirins making 70-80% of total endosperm protein. The α-kafirin constitutes approximately 80% of total endosperm kafirins is a digestible protein but remains unaccessible. The reason for such has

been attributed to its structure and presence of an inter- and intra- sulphide bounded outer layer of indigestible β- and γ-kafirins [5]. Therefore, studies have focused on analyzing proteomes of genotypes with varying level of digestibility due to mutation in kafirin genes [99] or transgenic lines in which these genes have been silenced using RNAi technology [100]. Cremer *et al.* [99] performed comprehensive proteomic analysis in 28 sorghum genotypes containing allelic variations in β, γ and δ kafirin genes identified previously [101]. Significant variations in concentrations of various proteins were detected across these genotypes which indicated possible involvement of at least a few of them in formation of kafirin-starch complex and inter- protein conjugation responsible for the poor protein digestibility. Thioreoxidin, a small protein was absent in the β-kafirin null lines [99]. Various other alcohol soluble proteins were also absent in the β-kafirin null lines. These putative proteins may be targeted for improving digestibility of kafirin proteins in agronomically improved varieties.

3.2.3. Trancriptomics and Metabolomics

Studies using transcriptomics and metabolomics to understand grain quality traits have been limited in sorghum. Nevertheless, few studies have analyzed transcriptome of different sorghum tissues for molecular understanding of other important traits and developing transcritome atlas for facilitating functional genomics analysis [102, 103]. Recently, researchers at RIKEN institute in Japan have developed a sorghum transcriptome database 'Morokoshi, containing sorghum trancriptome data of various tissues generated from their own study as well as 23 other previous studies [104]. The sorghum transcriptome data generated in these studies are good genomic resource for deriving information on synthesis and regulation of the various nutritional compounds including starch, protein and fat.

3.2.4. Functional Genomics Approaches

RNAi technology has proven very useful in underpinning molecular basis of poor digestibility of sorghum proteins [105]. Transgenic plants with suppressed kafirin genes have produced grains with higher *in vitro* digestibility as compared to control plants [100, 106, 107]. These studies endorsed previous findings that suggested kafirins particularly β and γ as the main factor responsible for poor digestibility of sorghum proteins. Although studies on sorghum unanimously agree that suppression of kafirin genes can enhance protein digestibility, but the question remains with regards to number/type of kafirin genes to be silenced/suppressed for obtaining maximum level of protein digestibility. Suppression of the just γ kafirin was unable to enhance *in vitro* protein digestibility of cooked flour samples [100]. Recently, it was demonstrated that co-suppression of only two genes, γ kafirin-1 (25 kDa) and γ -kafirin-2 (50 kDa), significantly enhanced *in vitro* protein digestibility and further improvement in digestibility could be attained with suppression of an additional kafirin gene, α kafirin A-1(25 kDa) [107]. Besides, RNAi technology has been also applied to suppression of lysine keto-glutarate reductase (LKR), the key enzyme involved in lysine degradation. The transgenic lines with *lkr* gene suppressed contained significantly enhanced lysine in whole grain (45.23) and endosperm (77.55%) as compared to their non-transgenic parents [103]. Considering the impor-

tance of RNAi based technology in enhancing nutritional quality of sorghum it has been applied for developing nutritious sorghum under African bio-fortified sorghum program [108, 109]. The other functional genomics strategies in sorghum are still under development as mentioned below. An EMS induced mutant population of BTx623 inbred line of sorghum has been developed and validated for discovery of gene function using TILLING [110]. This population is invaluable genomic resource for TILLING as the genome sequence of BTx623 inbred line is available [111]. Verma *et al.* [112] have developed *Ds* tagged sorghum mutant lines that can be used for identifying and characterizing genes for various traits including grain quality.

3.3. Oat

3.3.1. Association Mapping

Association mapping in oat became feasible with development of high throughput markers, such as DArT and SNP [113, 114]. Newell *et al.* [115] identified three DArT markers significantly associated for β-glucan in a germplasm collection of worldwide origin using GWAS. Interestingly, one marker shared sequence homology to a gene on chromosome 7 of rice close to *cslF* gene family, involved in β-glucan synthesis. Another independent GWAS study using North-American oat elite breeding lines also reported DArT markers significantly associated with β-glucan content and few of them were co-localized in the genomic regions previously reported to contain QTLs for β-glucan content. Physical positioning of significantly associated markers on the rice genome demonstrated that two markers were close to the genes involved in β-glucan synthesis [116]. The above studies clearly suggest that *cslF* gene family must be playing role in determining β-glucan content in oat. The closely linked markers are being exploited for genomic selection and MAS for enhancing the β-glucan content in oat varieties.

3.3.2. Transcriptomics

Application of NGS technologies has greatly facilitated identification of genetic factors responsible for accumulation of high levels of nutritional as well as anti-nutritional compounds in oat. RNA-seq analysis of oat seed developmental stages identified genes involved in biosynthesis of various health promoting substances such as avenanthramides, tocols, and β-glucans [117]. In another study, entire transcripts of root, stem, leaf, crown and various stages of seed development have been sequenced to obtain information on the metabolic pathways operating in these tissues [118]. The expression of β-glucan synthesis genes varied in different tissues as well as during course of seed development. The transcript of *AsCslF4*, *AsCslF6, AsCslF* 8 and *AsCslH1* was highly abundant at later stages of seed development. Moreover, in contrast to barley two additional genes *AsCslF4* and *AsCslF8* were expressed in oat, suggesting involvement of more than one β-glucan synthases in β-glucan biosynthesis which may be either acting concurrently in the same pathway or in different pathway [118].

3.3.3. Proteomics and Metabolomics

Chang [119] analyzed different fractions of oat grain proteins (albumin, globulin and glutelin) using proteomic techniques including RP-HPLC (Reverse Phase High Performance Liquid Chromatography), 2D-GE and LC-ESI-MS/MS (Liquid Chromatography Electrospray Ionization Tandem Mass Spectrometry). The knowledge about various fractions of proteins can be combined with other genomics data for detailed analysis of grain quality traits. Metabolic profiling of oat grain has not been reported yet. However, there are reports that have separately analyzed its nutritional constituents [117, 120], which may be integrated with other omics data for understanding molecular mechanisms of their synthesis and regulation. Separate analysis of tocols in embryo and endosperm tissues of oat during the course of developmemnt revealed different pattern of accumulation of two groups of tocols; tocopherols and tocotrienols [117]. This knowledge, once combined with transcriptomics and proteomics data of seed tissues will be extremely useful in elucidating molecular mechanism responsible for temporal and spatial accumulation of tocols in seed tissues.

3.3.4. Functional Genomics Approaches

Chawade *et al.* [121] have developed a TILLING population in oat and demonstrated its potential in discovering genes for quality traits. Mutation frequency of this population was high as indicated by presence of large number of mutations in two important genes; phenylalanine ammonialyase (*AsPAL1*) and the cellulose synthase-like (*AsCslF6*) involved in lignin and β-glucan biosynthesis respectively. Detailed analysis of these mutant lines pertaining to augment our understanding with regard to underlying mechanism of lignin and β-glucan biosynthesis will be useful for programs related to enhancing nutritional quality [121]. However, other genomic tools in oat could not be exploited due to large and complex genome and lack of efficient transformation protocol.

3.4. Finger Millet

Molecular mechanism responsible for the high calcium content in finger millet grains remains to be one of the challenges before the plant researchers. Recently, transcriptomics studies have given some insight in to the complex mechanism responsible for significantly high level of accumulation of calcium in few finger millet genotypes [122, 123]. RNA-seq analysis of developing spikes of two contrasting finger millet genotypes demonstrated higher level of expression of calcium sensor and transport genes in variety with high calcium content. The differentially expressed genes included calcium sensors and transporter genes such as two CDPKs (Calcium Dependent Protein Kinases), four CIPKs (Calcium Interacting Protein Kinases), one calmodulin gene (CaM), one two-pore channel (TPC) protein, two calcium exchangers (CaX1 and CaX3) and four ATPases [124]. Similar observation were also observed in few of the genes showing high level of expression in calcium rich finger millet genotype, [125]. Studies in plant have mainly focused on calcium exchangers as they are responsible for higher content of calcium in plant. Singh *et al.* [123] identified 82 calcium sensor genes from the transcriptome data of low and high calcium finger millet genotypes using rice genome as a reference. Application of external calcium has found to significantly enhance expression of some of these genes in calcium rich finger millet variety as compared to low calcium variety. Therefore, calcium sensor genes appear to be responsible for

high calcium accumulation in finger millet and constitute important candidates for enhancing calcium content of finger millet varieties. Using RP-HPLC and ESI-MS (Elecrospray Ionization Mass Spectrometry) diverse phenolic compounds which are responsible for the several beneficial health value finger millet grains have been identified [126].

3.5. Foxtail Millet

There has been no reports on application of genomics tools for analyzing grain quality traits in foxtail millet until recent times. However, few studies have used genomics tools for various other traits. Comprehensive genome-wide association mapping for various agronomic traits in foxtail millet has been reported in a set of its 916 diverse varieties [127]. As the agronomic features such as grain weight and panicle numbers have bearing on quality of grains and may be used to obtain their association with nutritional quality. Further core collection representing a small set 153 genotypes of worldwide origin has been developed [128] which may be characterized for quality parameters and associated genomic factors can be identified. Using core collections from India, association mapping were done for key agronomic characters based on SSR markers [129] Transcriptome analysis in foxtail millet have also reported changes in expression of storage compound associated genes during seed development [130, 131]. These transcriptome data may be used to select genes of various pathways involved in biosynthesis and regulation of storage compounds and can be exploited for understanding their role in accumulation of various nutritional compounds. Additionally, transcriptome data from its related species *Setaria viridis* [132] can also serve a valuable genomic resource for analysis of quality traits. Of note, foxtail millet and *S. viridis* are now been realized as experimental models [133, 134].

3.6. Pearl Millet

Availability of genomic resources of pearl millet are still in initial stages and likely to be expedited once its reference genome sequence is available in the public domain [135]. Due to such limitation, no major progress has been made towards understanding of molecular mechanisms of quality traits. Availability of genomic and other omics data would likely to facilitate genome-wide studies in genotypes with significantly high level of the Iron and Zinc content [136]. Transcriptome and proteome analysis of seed and other tissues of these genotypes may also help to generate knowledge on proteins and enzyme responsible for their transport and accumulation in grain. Also the available transcriptome data of pearl millet could be used for identifying target genes for quality traits.

3.7. Other Minor Millets

Nevertheless, as discussed above for other coarse grains, these tools have great potential for deciphering molecular mechanism underlying various quality traits in other millets. Metabolic profiling among three common proso millet (*Panicum milaceum*) varieties, it was observed that variety "Joongjuk" possessed high level of primary metabolites [137]. The metabolic network of proso millet may be reconstructed based on metabolite and transcript abundance data

to get insight of the various pathways and if combined with other genomics may reveal complex regulation involved in synthesis and catabolism of nutritional compounds particularly some of the essential amino acids (leucine, isoleucine, methionine) which are present in higher quantity than wheat [138]. Compared to other glutinous rich cereals, proso millet have no detectable amylase in the seed endosperm possibly due to mutation in the *Waxy* gene [139]. In order to develop glutinous varieties, molecular markers have been developed to identify these *waxy* genotypes, which are of high value [140].

4. STRUCTURAL GENOMICS RESOURCES (GENOME SEQUENCE AND MARKERS)

4.1. Barley

In last decade, low throughput marker techniques including RAPD, AFLP and SSRs were developed and applied for polymorphism study in barley [141-145]. Subsequently, these markers were used to develop consensus molecular maps and mapping of various traits in barley [146]. Application of these markers has facilitated identification of large number QTLs for β-glucan and malting quality traits in barley [147]. However, fine mapping and cloning of these genes could not be achieved due to non availability of high density molecular maps. With application of high throughput detection platform, such as Illumina GoldenGate bead arrays based SNP genotyping platform and DArT markers [148-150] high density maps have been generated. In addition, NGS based genotyping technologies such as restriction site associated DNA (RAD) and genotyping by sequencing (GBS) [151, 152] have been also applied in barley for identifying genome wide sequence variation in barley genome. For the complex genomes like barley, GBS is a method of choice as it allows complexity reduction through targeted sequencing of non repetitive regions. Moreover, an exome capture strategy that selectively sequence the coding regions of the barley genome has been developed for sequence based mapping [153]. International Barley Sequencing Consortium (IBSC) reported construction of a physical map of barley with 4.98 -giga base pairs (Gb) which predicted 36,151 protein coding genes. The barley physical map was also anchored to a genetic map. The integration of barley physical map with genetic map is an important molecular framework for functional genomics studies for identification of grain and malting quality associated genes. In another independent effort, a research group in China has sequenced genome of a Tibetan hullless variety of barley [154]. The draft barley genome assembly of 3.89-Gb contained 36,151 predicted protein-coding genes. With the availability of large number of genomic resources, researchers are better equipped to perform various genomic studies like high resolution association mapping, map based cloning and physical mapping of genes for various quality associated traits.

4.2. Sorghum

Sorghum is one of the extensively characterized crops owing to availability of large number of low and moderate throughput marker resources such as RFLP, RAPD, AFLP and SSR. Some of these markers have been used for map-

ping of grain quality traits [155]. Subsequently, various high throughput markers and genotyping platform such as DArT [156, 157] and SNP assays [158] were developed in sorghum. In addition to above, the advanced sequencing based genotyping approaches such as RAD [159] and GBS [160] have been also applied for discovering genome-wide variations and developing high density maps. However, with the availability of draft sorghum genome sequence [111], re-sequencing of two sweet and one grain sorghum revealed genome wide variations in both the species [94]. In another independent study, a set five genotypes were sequenced, two generated in this study and three of previous study, used to identify large number of genome wide SNPs [94]. Recently, a comprehensive re-sequencing analysis of 44 sorghum genotypes representing accessions from diverse geographical origin and different sorghum species identified 8 millions high-quality SNPs and 1.9 millions indels. Interestingly, the considered set of genotypes included some of the variety with high quality trait, therefore their genomic information would be very useful in dissecting quality traits [96]. Furthermore, an Illumina Infinium iSelect based assay with 2,670 SNPs is available [161]. Application of such high-throughput technologies is expected to improve our understanding of mechanisms of quality traits through high resolution association and bi- parental QTL analysis and accelerate breeding for the desirable quality traits using genomic selection and predictive breeding.

4.3. Oat

One of the initial mapping studies in oat used RFLP markers and identified genomic regions controlling β-glucan content on chromosomes 7 and 11 [162].) Subsequently, an integrated molecular linkage map of oat was constructed using RAPD, RFLP, AFLP and SCAR markers, which assisted mapping of QTLs for various traits including oat β – glucan and protein content [163]. Moreover, some of the less commonly used markers such as CAPS and SCAR were also developed for the regions governing β -glucan and protein content in oat. In later years of the last decade, large numbers of SSR markers were developed in oat [164, 165]. Owing to the large genome size of oat, high-density molecular maps would be required for high resolution mapping of economically important traits. Marker discovery in oat has got boost from high-throughput sequencing technologies [166]. A large number of SNPs have been identified based on sequencing and sequence comparison from a set of cDNA libraries prepared from various tissues of four oat genotypes [159]. Moreover, recently a physically anchored consensus SNP map of oat was also constructed; which constitutes a basic molecular framework for genomic analysis for discovery of genes for quality traits [167].

4.4. Pearl Millet

In pearl millet RFLP and SSR markers are the most widely applied molecular markers for genomic analysis such as assessment of genetic diversity and trait mapping. The first consensus linkage map of pearl millet was constructed using RFLP and SSR markers in four different mapping populations [168]. Recently, consensus molecular map of pearl millet has been constructed using SSR markers in RIL

mapping population [169]. Availability of high-density consensus molecular map is extremely useful as it allow exact positioning of QTLs/ genes in the background of different mapping populations. Other marker techniques such as SSCP-SNPs (Single Strand Conformation Polymorphism – Single Nucleotide Polymorphisms) [170, 171] CISP (Conserved Intron scanning Primers) [172] and DArT [173] have been also applied in pearl millet. However, there are no reports on use of high-throughput genotyping technologies such as SNP based assays and GBS in pearl millet. Genomic studies in pear millet are severely constrained by non-availability of high-density molecular map. Application of high throughput technologies will certainly accelerate discovery of genes/ alleles responsible for various essential amino acids and iron as well as zinc content in pearl millet grains. Pearl millet draft genome sequence is almost ready and likely to be released in the public domain soon [174]. Besides, a HapMap has been also generated using re-sequencing of 963 pearl millet germplasm lines including 580 B and R-lines, 345 pearl millet inbred genotypes association panel and 38 parental lines of mapping populations [174]. These genomic resources will be very useful for analyzing complex traits including quality traits.

4.5. Foxtail Millet

For genomic studies in foxtail millet, largely RFLP and RAPD markers were used [175, 176], until the generation of SSR markers [177-179]. However, these markers still constitute important genomic resource for molecular analysis of economically important traits in foxtail millet. Recently, the application of NGS technologies has greatly accelerated generation of structural genomics resources in foxtail. Foxtail millet genome has been independently sequenced by two groups [180, 181]. The research group at BGI (Beijing Genomics Institute), China sequenced cultivar Zhang Gu with draft genome assembly of 450 Mb representing 89% of the genome. In addition, this group also sequenced foxtail millet variety A-2 and compared it with the genome sequence of Zhang Gu, which allowed identification of several thousand SNPs, InDels and structural variations that may be potentially used for developing markers for genomic analysis [180]. Other research group at the Joint Genome Institute (JGI) of the Department of Energy, USA has independently sequenced foxtail millet genotype 'Yagu1', a green foxtail genotype A-10 and a RIL population derived from cross of Yagu1 and A-10 and have also constructed a high density SNP based molecular map using re-sequencing data of the RIL population [181]. Recently, a foxtail millet SNP haplotype map with 0.8 million SNP was constructed using re-sequencing data of 916 diverse foxtail millet genotypes [182]. This haplotype map may be also employed for discovery of genes for grain quality traits using association mapping based approach. The availability of genome sequence has expedited the development of various sequence based molecular markers such as SSR [183-185], ILP (Intron Length Polymorphism) [186], miRNA-based [187] and transposable elements based markers [180]. The availability of developed marker resources in open access databases such as FmMDb [189], FmMiRNADb [187] and FmTEMDb [186] would expedite genomics-assisted breeding for crop as well as nutrition improvement [188].

4.6. Other Millets

Genomics in proso milllet were earlier limited to genetic diversity analysis using RAPD [190] and AFLP [191]. However, SSR markers in proso-millet have been also developed using SSR enriched genomic DNA libraries [192] and through transferability of SSRs from related species such as rice, wheat, oat, barley and switch grass [193, 194]. Little millet and Kodo millet have only few reports on use of markers and remains uncharacterized at the genome level. Except one study that has used RAPD [190], there is no information on the use of markers for molecular analysis in little millet. Similarly, in kodo millet there are two reports on use of markers for deriving genome level information for assessing genetic diversity and phylogeny [195, 196]. Therefore, immediate focus in both the crops should be on generation of large number of genomic resources, which can be applied for the analysis of quality associated traits. In Barnyard millet the advanced genotyping technique GBS has been used for analysing core set of barnyard millet [197].

5. FUTURE PERSPECTIVES: (UTILIZING COARSE GRAINS GERMPLASM RESOURCES FOR DECIPHERING QUALITY TRAITS)

Over the past decades, many national and international organizations have made large collection of genetic resources in coarse cereals, which are conserved in respective gene banks [198]. These germplasm collections are repertoire of genes/ alleles for economically important traits including biotic and abiotic stresses as well as quality traits. However, in the past these genetic resources have been exploited only to limited extent in crop improvement programs due to their limited agronomic features, unmanageable size, lack of tools for large scale molecular and phenotyping characterization. In recent years, plant breeders have given more emphasis on utilization of germplasm collections in crop improvement through development of core or mini core collection representing the maximum available genetic diversity. Since core are smaller in size and it can be easily characterized for various agriculturally important traits including nutritional parameters. Furthermore, availability of advanced genome analysis procedures such as high-throughput genotyping platforms, NGS based re-sequencing and association mapping has provided a way forward toward exploring these cores and mini-core sets for discovering genes for various traits including quality traits. To give a boost in coarse grain research, ICRISAT have developed core / mini core sets in sorghum [199], pearl millet [200, 201], finger millet [202], foxtail millet [203], proso millet [204] and other minor millets such as barnyard millet, kodo millet, little millet [205]. In barley core sets have been developed from international barley collection [206], Chinese spring barley collection [207], Spanish collection [208] and USDA-ARS National Small Grains Collection. These core and mini core sets can be phenotyped for important grain quality traits and analyzed by above mentioned genomic procedures for identifying genomic regions and understanding molecular mechanism for various quality trait. Already some progress has been made in this direction in few coarse cereals as discussed in above section. Recently, a core collection of sorghum was used to identify candidate genes for high Fe and Zn content using association mapping approach [89]. Further, a finger millet core set (622 accessions) phenotyped for grain quality traits such as Zn, Fe and protein content which can be used for discovering genes for these traits using association mapping approach [209]. To speed up the pace of research in coarse grain a concerted effort is the needed to expand the world food basket to feed ever increasing population and dispel the hidden hunger resulting from micronutrient deficiencies in diet.

CONFLICT OF INTEREST

The author(s) confirm that this article content has no conflict of interest

REFERENCES

[1]   Bouis, H.E. Enrichment of food staples through plant breeding: a new strategy for fighting micronutrient malnutrition. *Nutrition*, **2000**, *16*,701–704.

[2]   Kaur, K.D.; Jha, A.; Sabikhi, L.; Singh, A.K. Significance of coarse cereals in health and nutrition: a review. *J. Food Sci. Technol.,* **2014**, *51*(8), 1429-1441.

[3]   Devi, P.B.; Vijayabharathi, R.; Sathyabama, S.; Malleshi, N.G.; Priyadarisini, V.B. Health benefits of finger millet (*Eleusine coracana* L.) polyphenolsand dietary fiber: a review. *J. Food Sci. Technol.,* **2014**, *6*, 1021-1040.

[4]   Jones, J.M. Grain-based foods and health cereals. *Cereal Foods World*, **2006**, *51*:108–113.

[5]   Duodu, K.G.; Taylor, J.R.N.; Belton, P.S.; Hamaker, B.R. Factors affecting sorghum protein digestibility. *J. Cereal. Sci.,* **2003**, *38*(2), 117-131.

[6]   Sade, F.O. Proximate, antinutritional factors and functional properties of processed pearl millet (*Pennisetum glaucum*). *J. Food Technol.,* **2009** 7(3), 92-97.

[7]   Shashi, B.K.; Sharan, S.; Hittalamani, S.; Shankar, A.G.; Nagarathna, T.K. Micronutrient composition, antinutritional factors and bioaccessibility of iron in different finger millet (*Eleusine coracana*) genotypes. *Karnataka J. Agri. Sci.,* **2010**, *20*(3), 583-585.

[8]   McCallum, C.M.; Comai, L.; Greene, E.A.; Henikoff, S. Targeting induced locallesions in genomes (TILLING) for plant functional genomics. *Plant Physiol.,* **2000**, *123*(2), 439-442.

[9]   Aharoni, A.; Vorst, O. DNA microarrays for functional plant genomics. *Plant Mol. Biol.,* **2002**, *48*(1-2), 99-118.

[10] Bino, R.J.; Hall, R.D.; Fiehn O.; Kopka, J.; Saito, K.; Draper, J.; Nikolau, B.J.; Ute Roessner-Tunali, P.M.; Beale, M.H.; Trethewey, R.N.; Lange, B.M.; Wurtele, E.S.; Sumner, L.W. Potential of metabolomics as a functional genomics tool. *Trends Plant Sci.,* **2004**, *9*(9), 418-425.

[11] Hall, R.D.; de Vos, C.R.; Verhoeven, H.A.; Bino, R.J. In: *Metabolome Analyses: Strategies for Systems Biology;* Seetharaman Vaidyanathan; George G. Harrigan; Royston Goodacre, Ed.; Springer: New York, **2005**; pp. 31-44.

[12] Stylianou, I.M.; Affourtit, J.P.; Shockley, K.R.; Wilpan, R.Y.; Abdi, F.A.; Bhardwaj, S.; Jarod Rollins, Churchill, J.G.; Paigen, B. Applying gene expression, proteomics and single-nucleotide polymorphism analysis for complex trait gene identification. *Genetics*, **2008**, *178*(3), 1795-1805.

[13] Xu, Y.; Skinner,D.J.; Wu, H.; Palacios-Rojas, N.; Araus,J.L.; Yan, J.; Gao, S.; Warburton, M.L.; Crouch, J.H. Advances in Maize Genomics and Their Value for Enhancing Genetic Gains from Breeding. *Int. J. Pl. Genomics,* vol. **2009**, Article ID 957602.

[14] Yan, J.; Warburton, M.; Crouch, J. Association Mapping for Enhancing Maize (Zea mays L.) genetic Improvement. *Crop Sci.,* **2011**, *51,* 433-449.

[15] Riedelsheimer, C.; Lisec, J.; Czedik-Eysenberg, A.; Sulpice, R.;

Flis, A.; Grieder, C.; Altmann, T.; Stitt, M.; Willmitzer, L.; Melch-inger, A. E. Genome-wide association mapping of leaf metabolic profiles for dissecting complex traits in maize. *Proc. Natl. Acad. Sci. U.S.A.,* **2012** *109*(23), 8872–8877.

[16] U.S. Department of Agriculture, Agricultural Research Service. **2014**. USDA National Nutrient Database for Standard Reference, Release 27. Nutrient Data Laboratory Home Page, http://www.ars.usda.gov/ba/bhnrc/ndl.

[17] Hulse, J.H.; Laing, E.M.; Pearson, O.E. *Sorghum and the millets: their composition and nutritive value.* Academic Press: New York, **1980**, pp 81-91

[18] Gopalan, G.; Sastri, B.V.R.; Balasubramanian, S.C. Nutritive value of Indian foods. Revised and updated by Rao, B.S.N.; Deosthale, Y.G. and Pant, K.C. National Institute of Nutrition, Indian Council of Medical Research, Hyderabad, **2007**.

[19] Shobana, S.; Krishnaswamy, K.; Sudha, V.; Malleshi, N.G.; An-jana, R.M.; Palaniappan, L.; Mohan, V. Finger millet (Ragi, *Eleusine coracana* L.): a review of its nutritional properties, processing, and plausible health benefits. *Adv. Food Nutr. Res.*, **2013**, *69*, 1-39.

[20] FAO Amino acid contents of foods and biological data onproteins. FAO Nutritional Series, **1970**, No. *21*, 285–28.

[21] Gupta, N.; Srivastava, A.K.; Pandey, V.N. Biodiversity and nu-traceutical quality of some Indian millets. *Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci.,* **2012**, *82*(2), 265-273.

[22] Suganyadevi, P.; Saravanakumar, K.M.; Mohandas, S. The antipro-liferativeactivity of 3-deoxyanthocyanins extracted from red sor-ghum (*Sorghum bicolor*) bran through P53-dependent and *Bcl-2* gene expression in breast cancer cellline. *Life Sci., F.,* **2013**, *92*, 379e382.

[23] Baik, B.K.; Ullrich, S.E. Barley for Food: Characteristics, Im-provement, and Renewed Interest. *J. Cereal. Sci.,* **2008**, *48*, 233-242.

[24] Dykes, L,.; Rooney, L.W. Phenolic compounds in cereal grains and their health benefits. *Cereal Foods World*, **2007**; 52,105–111.

[25] Behall K. M; Hallfrisch, J. Oats as a functional food for health. In F.H. Webster, P.J. Wood (Eds.), Oats, Chemistry and Technology (second ed.), AACC International Press, **2011**, pp. 255-273.

[26] Arendt, E. K.; Dal Bello, F. Gluten-Free Cereal Products and Bev-erages in (USA): Academic Press, New York, **200**, pp. 321–46.

[27] Comino, I.; de Lourdes Moreno, M.; Real, A.; Rodríguez-Herrera, A.; Barro, F.; Sousa, C. The gluten-free diet: Testing alternative ce-reals tolerated by celiac patients. *Nutrients*, **2013**, *5*(10), 4250-4268.

[28] Saleh, A. S.; Zhang, Q.; Chen, J.; Shen, Q. Millet grains: Nutri-tional quality, processing, and potential health benefits. *Compr. Rev. Food Sci. F.,* 2013, *12*(3), 281-295.

[29] Sharma, A.; Kapoor, A.C. Levels of antinutritional factors in pearl millet as affected by processing treatments and various types of fermentation. *Plant Foods Hum. Nutri.,* **1996**, 49, pp. 241–252.

[30] Kumar, V.; Sinha, A.K.; Makkar, H.P.S.; Becker, K. Dietary roles of phytate and phytase in human nutrition: a review. *Food Chem.*, **2010**,*120*, 945e959.

[31] Klopfenstein CF, Leipold HW, Cecil JE. Semiwet milling of Pearl millet for reduced goitrogenicity. *Cereal Chem.,* **1991**, 68, 177–179.

[32] Lundin, K.E.A.; Nilsen, E.M.; Scott, H.G.;Loberg, E.M.; Gjoen, A.; Bratlie, J.; Skar, V.;Mendez, E.; Lovik, A.; Kett, K.,; Oats in-duced villous atrophy in coeliacdisease. *Gut,* **2003**, *52*, 1649-1652.

[33] Gupta, M.; Abu-Ghannam, N.; Gallaghar, E. Barley for Brewing: Characteristic Changes during Malting, Brewing and Applications of its By-Products. *Compr. Rev. Food Sci. F.,* **2010**, *9*(3), 318-328.

[34] Parinov, S.; Sundaresan, V. Functional genomics in Arabidopsis: large-scale insertional mutagenesis complements the genome se-quencing project. *Curr. Opin. Biotech.*, **2000**, *11*(2), 157-161

[35] Fiehn, O.; Kopka, J.; Dörmann, P.; Altmann, T.; Trethewey, R. N.; Willmitzer, L. Metabolite profiling for plant functional genomics. *Nat.biotechnol.*, **2000**, *18*(11), 1157-1161.

[36] Burch-Smith, T.M.; Anderson, J.C.; Martin, G.B.; Dinesh-Kumar, S.P. Applications and advantages of virus-induced gene silencing for gene function studies in plants. *Plant J.*, **2004**, *39*(5), 734-746.

[37] Travella, S.; Klimm, T.E.; Keller, B. RNA interference-based gene silencing as an efficient tool for functional genomics in hexaploid bread wheat. *Plant Physiol.*, **2006**, *142*(1), 6-20.

[38] McGinnis, K.M. RNAi for functional genomics in plants. *Brief. Funct. Genomics.,* **2010**, elp052.

[39] Plessis, A.; Ravel, C.; Bordes, J.; Balfourier, F.; Martre, P. Asso-ciation study of wheat grain protein composition reveals that gli-adin and glutenin composition are trans-regulated by different chromosome regions. *J. Exp. Bot.*, **2013**, *64*(12), 3627-3644.

[40] Gutiérrez, L.; Cuesta-Marcos, A.; Castro, A.J.; von Zitzewitz, J.; Schmitt, M.; & Hayes, P.M. Association mapping of malting qual-ity quantitative trait loci in winter barley: positive signals from small germplasm arrays. *Plant Genome*, **2011**, *4*(3), 256-272.

[41] Cai, S.; Yu, G.; Chen, X.; Huang, Y.; Jiang, X.; Zhang, G.; Jin, X. Grain protein content variation and its association analysis in bar-ley. *BMC Plant Biol.*, **2013**, *13*(1), 35.

[42] Shu, X.; Rasmussen, S.K. Quantification of amylose, amylopectin, and β-glucan in search for genes controlling the three major quality traits in barley by genome-wide association studies. *Front. Plant Sci.*, **2014**, *15*, (5), 197.

[43] Houston, K.; Russell, J.; Schreiber, M.; Halpin, C.; Oakey, H.; Washington, J. M.; Booth, A.; Shirley, N.; Burton, R.A.; Fincher, G.B.; Waugh, R. A Genome wide association scan for (1, 3; 1, 4)-beta-glucan content in the grain of contemporary 2-row Spring and Winter barleys. *BMC Genomics*, **2014**, *15*(1), 907.

[44] Matthies, I. E.; Malosetti, M.; Röder, M. S.; van Eeuwijk, F. Ge-nome-Wide Association Mapping for Kernel and Malting Quality Traits Using Historical European Barley Records. *PloS one*, **2014**, *9*(11), e110046.

[45] Jamar, C.; Loffet, F.; Frettinger, P.; Ramsay, L.; Fauconnier, M. L.; & Du Jardin, P. NAM-1gene polymorphism and grain protein con-tent in *Hordeum. J. Plant physiol.*, **2010**, *167*(6), 497-501.

[46] Mohammadi, M.; Endelman, J.B.; Nair, S.; Chao, S.; Jones, S.S.; Muehlbauer, G.J.; Ullrich, S.E.; Baik, B.K.; Wise, M.L.; Smith, K.P. Association mapping of grain hardness, polyphenol oxidase, total phenolics, amylose content, and β-glucan in US barley breed-ing germplasm. *Mol. Breeding*, **2014**, *34*, 1229-1243.

[47] Burton, R.A.; Jobling, S.A.; Harvey, A.J.; Shirley, N.J.; Mather, D.E.; Bacic, A.; Fincher, G.B. The genetics and transcriptional pro-files of the cellulose synthase-like HvCslF gene family in barley. *Plant Physiol.*, **2008**, *146*(4), 1821-1833.

[48] Schober, M.S.; Burton, R.A.; Shirley, N.J.; Jacobs, A.K.; Fincher, G.B. Analysis of the (1,3)-β-d-glucan synthase gene family of bar-ley. *Phytochemistry*, **2009**, *70*(6),713-720.

[49] Hu, G.; Burton, C.; Hong, Z.; Jackson, E. A mutation of the cellu-lose-synthase like (*CslF6*) gene in barley (*Hordeum vulgare* L.) partially affects the β -glucan content in grains. *J. Cereal Sci.*, **2014**. *59, 189-195.*

[50] Sreenivasulu, N.; Altschmied, L.; Radchuk, V.; Gubatz, S.; Wobus, U.; Weschke, W. Transcript profiles and deduced changes of meta-bolic pathways in maternal and filial tissues of developing barley grains. *Plant J.*, **2004**, *37*: 539–553.

[51] Druka, A.; Muehlbauer, G.; Druka, I.; Caldo, R.; Baumann, U.; Rostoks, N., Schreiber, A., Wise, R., Close, T., Kleinhofs, A., Gra-ner, A., Schulman, A., Langridge, P., Sato, K., Hayes, P., McNicol, J., Marshall, D., Waugh, R. An atlas of gene expression from seed to seed through barley development. *Funct. Integr. Genomics*, **2006**, *6*, 202–211.

[52] Leymarie, J.; Bruneaux, E.; Gibot-Leclerc, S.; Corbineau, F. Identi-fication of transcripts potentially involved in barley seed germina-tion and dormancy using cDNA-AFLP. *J. Exp. Bot.*, **2007**, *58*, 425–437.

[53] White, J.; Pacey-Miller, T.; Crawford, A.; Cordeiro, G.; Barbary, D.; Bundock, P., Henry, R. Abundant transcripts of malting barley identified by serial analysis of gene expression (SAGE). *Plant Bio-tech. J.*, **2006**, *4*, 289–301.

[54] Lapitan, N.L.; Hess, A.; Cooper, B.; Botha, A.M.; Badillo, D.; Iyer, H.; Menert, J.; Close, T.; Wright, L.; Hanning, G.; Tahir, M.; Law-rence, C. Differentially expressed genes during malting and corre-lation with malting quality phenotypes in barley (Hordeum vulgare L.). *Theor. Appl. Genet.*, 2009, 118(5), 937-952.

[55] Chen, X.; Long, H.; Gao, P.; Deng,G.; Pan, Z.; Liang, J.; Tang, Y.; Tashi, N.; Yu, M.; Transcriptome Assembly and Analysis of Ti-betan Hulless Barley (*Hordeum vulgare L. var. nudum*) Developing Grains, with Emphasis on Quality Properties. *PLoS One*, **2014**, *9*(5), e98144.

[56] Muñoz-Amatriaín, M.; Xiong, Y.; Schmitt, M.R.; Bilgic, H.; Budde, A.D.; Chao, S.; Smith, K.P.; Muehlbauer, G.J. Transcrip-tome analysis of a barley breeding program examines gene expres-sion diversity and reveals target genes for malting quality im-provement. *BMC Genomics*, **2010**, *11*(1), 653.

[57] Potokina, E.; Caspers, M.; Prasad, M.; Kota, R.; Zhang, H.; Sreeni-vasulu, N.; Wang, M.; Graner, A. Functional association between malting quality trait components and cDNA array based expression patterns in barley (*Hordeum vulgare* L.). *Mol. Breeding*, **2004**, *14*(2), 153-170.

[58] Close, T.J.; Wanamaker, S.I.; Caldo, R.A.; Turner, S.M.; Ashlock., D.A.; Dickerson, J.A., Wing, R.A., Muehlbauer, G.J., Kleinhofs, A., Wise, R.P. A new resource for cereal genomics: 22K barley GeneChip comes of age. *Plant Physiol.*, **2004**, *134*, 960–968.

[59] Nemeth, C.; Freeman, J.; Jones, H.D.; Sparks, C.; Pellny, T.K.; Wilkinson, M.D.; Dunwell, J.; Andersson, A.A.; Aman, P.; Guillon F.; Saulnier, L.; Mitchell, R.A.; Shewry, P.R. Down-regulation of the *CSLF6* gene results in decreased (1,3;1,4)-beta-D-glucan in endosperm of wheat. *Plant Physiol.*, **2010**, *152*, 1209-1218.

[60] Østergaard, O.; Melchior, S.; Roepstorff, P.; Svensson, B. Initial proteome analysis of mature barley seeds and malt. *Proteomics*, **2002**, *2*(6), 733-739.

[61] Østergaard, O.; Finnie, C.; Laugesen, S.; Roepstorff, P.; Svennson, B. Proteome analysis of barley seeds: Identification of major proteins from two-dimensional gels (pI 4-7). *Proteomics*, **2004**, *4*, 2437-2447.

[62] Thelen, J.J.; Peck, S.C. Quantitative proteomics in plants: choices in abundance. *Plant Cell*, **2007**, *19*, 3339–334.

[63] Finnie, C.; Bak-Jensen, K.S.; Laugesen, S.; Roepstorff, P.; Svensson, B. Differential appearance of isoforms and cultivar variation in protein temporal profiles revealed in the maturing barley grain proteome. *Plant Sci.*, **2006**, *170*(4), 808-821.

[64] Finnie, C.; Bagge, M.; Steenholdt, T.; Østergaard, O.; Bak-Jensen, K.S.; Backes, G.; Jensen, A.; Giese, H.; Larsen, J.; Roepstorff, P.; Svensson, B. Integration of the barley genetic and seed proteome maps for chromosome 1H, 2H, 3H, 5H and 7H. *Funct. Integr. Genomics,* **2009**, *1*, 135-143.

[65] Damerval, C.; Maurice, A.; Josse, J.M.; Vienne de, D. Quantitative trait loci underlying gene product variation: a novel perspective for analyzing regulation of genome expression. *Genetics*, **1994**, *137*(1), 289-301.

[66] March, T.J.; Richter, D.; Colby, T.; Harzen, A.; Schmidt, J.; Pillen, K. Identification of proteins associated with malting quality in a subset of wild barley introgression lines. *Proteomics*, **2012**, *12*(18), 2843-2851.

[67] Jin, Z.; Li, X.M.; Gao, F.; Sun, J.Y.; Mu, Y.W.; Lu, J. Proteomic analysis of differences in barley (*Hordeum vulgare*) malts with distinct filterability by DIGE. *J. Proteomics*, **2013**, *93*, 93-106.

[68] Bønsager, B.C.; Finnie, C.; Roepstorff, P.; Svensson, B. Germination and radicle elongation in barley tracked using proteome analysis of dissected embryo, aleurone layer and endosperm tissues. *Proteomics*, **2007**, *7*, 4538–4540.

[69] Finnie, C.; Svensson, B. Barley seed proteomics from spots to structures. *J. Proteomics*, **2009**, *72*(3), 315-324.

[70] Frank, T.; Scholz, B.; Peter, S.; Engel, K.H. Metabolite profiling of barley: Influence of the malting process. *Food Chem.*, 2011 *124*(3), 948-957.

[71] Gorzolka, K.; Lissel, M.; Kessler, N.; Loch-Ahring, S.; Niehaus, K. Metabolite fingerprinting of barley whole seeds, endosperms, and embryos during industrial malting. *J. Biotechnol.*, **2012**, *159*(3), 177-187.

[72] Heuberger, A.L.; Broeckling, C.D.; Kirkpatrick, K.R.; Prenni, J.E. Application of nontargeted metabolite profiling to discover novel markers of quality traits in an advanced population of malting barley. *Plant Biotech. J.,* **2014**, *12*(2), 147-160.

[73] Niba, L.L. Resistant starch: a potential functional food ingredient. *Nutri. Food Sci.*, **2002**, *32*(2), 62-67.

[74] Regina, A.; Kosar-Hashemi, B.; Ling, S.; Li, Z.; Rahman, S.; Morell, M. Control of starch branching in barley defined through differential RNAi suppression of starch branching enzyme IIa and IIb. *J. Expt. Bot.*, **2010**, *61*(5), 1469-1482.

[75] Massimiliano, C.; Andreas, B.; Susanne, L. J.; Shahnoor, S. S.; Anette, H.; Alain, B.; Preben, B. H.; Kim, H.H. Concerted suppression of all starch branching enzyme genes in barley produces amylose-only starch granules. *BMC Plant Biol.*, **2012**, *12*, 223.

[76] Singh, J.; Zhang, S.; Chen, C.; Cooper, L.; Bregitzer, P.; Sturbaum, A.; Hayes, P.M.; Lemaux, P.G. High-frequency *Ds* remobilization over multiple generations in barley facilitates gene tagging in large genome cereals. *Plant Mol. Biol.*, **2006**, *62*, 937-950.

[77] Lazarow, K.; Lütticke, S. An Ac/Ds-mediated gene trap system for functional genomics in barley. *BMC Genomics*, **2009**, *10*(1), 55.

[78] Singh, S.; Tan, H. Q.; Singh, J. Mutagenesis of barley malting quality QTLs with Ds transposons. *Funct. Integr. Genomics*, **2012**, *12*(1), 131-141.

[79] Caldwell, D.G.; McCallum, N.; Shaw, P.; Muehlbauer, G.J.; Marshall, D.F.; Waugh, R. A structured mutant population for forward and reverse genetics in Barley (*Hordeum vulgare* L.). *Plant J.,* **2004**, *40*, (1): 143-150.

[80] Talame, V.; Bovina, R.; Sanguineti, M.C.; Tuberosa, R.; Lundqvist, U.; Salvi, S. TILLMore, a resource for the discovery of chemically induced mutants in barley. *Plant Biotechnol J.,* **2008**, *6*(5), 477-485.

[81] Gottwald, S.; Bauer, P.; Komatsuda, T.; Lundqvist, U.; Stein, N. TILLING in the two-rowed barley cultivar'Barke'reveals preferred sites of functional diversity in the gene HvHox1. *BMC Research Notes*, **2009**, *2*(1), 258.

[82] Lababidi, S.; Mejlhede, N.; Rasmussen, S.K.; Backes, G.; Al-Said, W.; Baum, M.; Jahoor A. Identification of barley mutants in the cultivar 'Lux' at the Dhn loci through TILLING. *Pl. Breed.*, **2009**, *128*,332–336.

[83] Kurowska, M.; Labocha-Pawłowska, A.; Gnizda, D.; Maluszynski, M.; Szarejko, I. Molecular analysis of point mutations in a barley genome exposed to MNU and gamma rays. *Mutat. Res.*, **2012**, *52* (70) 738–739.

[84] Taketa, S.; You, T.; Tonooka, T.; Tsumuraya, Y.; Inagaki, Y.; Haruyama, N.; Larroque, O.; Jobling, S.A. Functional characterization of barley betaglucanless mutants demonstrates a unique role for CslF6 in (1,3;1,4)-β-D-glucan biosynthesis. *J. Exp. Bot.* **2012**, *63* (1), 381-392.

[85] Sakhi, S.; Shehzad, T.; Rehman, S.; Okuno, K. Mapping the QTLs underlying drought stress at developmental stage of sorghum (*Sorghum bicolor* (L.) Moench) by association analysis. *Euphytica*, **2013**, *193,* 433–450.

[86] Upadhyaya, H.D.; Wang, Y.H.; Sharma, R.; Sharma, S. Identification of genetic markers linked to anthracnose resistance in sorghum using association analysis. *Theor. Appl.Genet.*, **2013**, *126*(6), 1649-1657.

[87] Sukumaran, S.; Xiang, W.; Bean, S.R.; Pedersen, J.F.; Kresovich, S.; Tuinstra, M.R.; Yu, J. Association mapping for grain quality in a diverse sorghum collection. *Plant Genome*, **2012**, *5*(3), 126-135.

[88] Gilding, E.K.; Frère, C.H.; Cruickshank, A.; Rada, A.K.; Prentis, P.J.; Mudge, A.M.; Mace, E.S.; Jordan, D.R.; Godwin, I.D. Allelic variation at a single gene increases food value in a drought-tolerant staple cereal. *Nat. Commun.*, **2013**, *4*, 1483.

[89] Anuradha, K.; Punna, R.; Shah, T.; Amindala, B. P.; Selvanayagam S , Rathore A, Hash C. T.; Grando S AshokKumar A.; Deshpande, S.; In Genome-wide association analysis for Fe and Zn concentrations in sorghum grains identifies the potential candidate genes for sorghum biofortification In; Proceeding of XXIIIth plant and Animal genome conference ,San Diego CA, January 10-14; **2015**.

[90] de Alencar F.L.; Sine, B.; Chantereau, J.; Fliedel, G.; Rami, J.F.; Glaszmann, J.C.; Deu, M.; Courtois, B. Variability of grain quality in sorghum: Association with polymorphism in Sh2, Bt2, SssI, Ae1, Wx and O2. *Theor Appl Genet.,* *2010 121*(6), 1171–1185.

[91] Deu, M.; Rattunde, F.; Chantereau, J. A global view of genetic diversity in cultivated sorghums using a core collection. *Genome*, **2006**, *49*(2), 168-180.

[92] Casa, A.M.; Pressoir, G.; Brown, P.J.; Mitchell, S.E.; Rooney W.L.; Tuinstra M.R.; Franks, C.D.; Kresovich, S. Community Resources and Strategies for Association Mapping in Sorghum. *Crop Sci.,* **2008**, *48*(1), 30-40.

[93] Upadhyaya, H.D.; Pundir, R.P.S.; Dwivedi, S.L.; Gowda, C.L.L.; Reddy, V.G.; Singh, S. Developing a mini core collection of sorghum for diversified utilization of germplasm. *Crop Sci.,* **2009**, *49*, 1769–1780.

[94] Zheng, L.Y.; Guo, X.S.; He, B.; Sun, L.J.; Peng, Y.; Dong, S.S.; Liu T.F.; Jiang S.; Ramachandran S.; Liu, C.M.; Jing, H.C. Genome-wide patterns of genetic variation in sweet and grain sorghum (*Sorghum bicolor*). *Genome Biol.*, **2011**, *12* (11), R114.

[95] Zou, G.; Zhai, G.; Feng, Q.; Yan, S.; Wang, A.; Zhao, Q. Shao, J.; Zhang, Z.; Zou, J.; Han, B.; Tao, Y. Identification of QTLs for eight agronomically important traits using an ultra-high-density map based on SNPs generated from high-throughput sequencing in sorghum under contrasting photoperiods. *J. Exp. Bot.* **2012**, *63*(15), 5451-5462.

[96] Mace, E.S.; Tai, S.; Gilding, E.K.; Li, Y.; Prentis, P.J.; Bian, L.; Campbell, B.C.; Hu, W.; Innes, D.J.; Han, X.; Cruickshank, A.;

Dai, C.; Frere, C.; Zhang, H.; Hunt, C.H.; Wang, X.; Shatte, T.; Wang, M.; Su, Z.; Li, J.; Lin, X.; Godwin, I.D.; Jordan, D.R.; Wang, J. Whole-genome sequencing reveals untapped genetic potential in Africa's indigenous cereal crop sorghum. *Nat. Commun.,* **2013**, *4*, 2320.

[97]   Wu, Y.; Li, X.; Xiang, W.; Zhu, C.; Lin, Z.; Wu, Y.; Li, J.; Pandravada, S.; Ridder, D.D.; Bai, G.; Wang, M.L.; Trick, H.N.; Bean, S.R.; Tuinstra, M.R.; Tesso, T.T.; Yu, J. Presence of tannins in sorghum grains is conditioned by different natural alleles of Tannin1. *Proc. Natl. Acad. Sci. U.S.A.,* **2012**, *109*(26), 10281-10286.

[98]   Roy, S.K.; Kamal, A.H.M.; Kim, H.R.; Kwon, S.J.; Jang, H.Y.; Ko, J.H.; Kim, J.I.; Ko, T.S.; Xin, Z.; Woo, S.H.; Proteome profiling of seed from inbred and mutant line of Sorghum (*Sorghum bicolor*) [online]. *Aust. J. Crop Sci.,* **2014**, *8*(4), 606-611.

[99]   Cremer, J.E.; Bean, S.R.; Tilley, M.M.; Ioerger, B.P.; Ohm, J.B.; Kaufman, R.C.; Wilson, J.D.; Innes,J.D.; Gilding, E.K.; Godwin, I.D. Grain Sorghum Proteomics: Integrated Approach toward Characterization of Endosperm Storage Proteins in Kafirin Allelic Variants. *J. Agri. Food Chem.,* **2014**, *62*(40), 9819-9831.

[100]   Kumar, T.; Dweikat, I.; Sato, S.; Ge, Z.; Nersesian, N.; Chen, H.; Elthon, T.; Bean, S.; Ioerger, B.P.; Tilley, M. Modulation of kernel storage proteins in grain sorghum *(Sorghum bicolor* (L.) Moench). *Plant Biotechnol. J.,* **2012**, *10*, 533–544.

[101]   Laidlaw, H.K.C.; Mace, E.S.; Williams, S.B.; Sakrewski, K.; Mudge, A.M.; Prentis, P.J.; Jordan, D.R; Godwin, I.D. Allelic variation of the beta, gamma and delta kafirin genes in diverse sorghum genotypes. *Theor. Appl. Genet.,* **2010**, *121*, 1227–1237.

[102]   Buchanan, C.D.; Lim, S.; Salzman, R.A.; Kagiampakis, I.; Morishige, D.T.; Weers, B.D. Mullet, J.E.; Klein, R.R.; Pratt, L.H.; Pratt, M.M.C.; Klein, P.E; Mullet, J.E. Sorghum bicolor's transcriptome response to dehydration, high salinity and ABA. *Plant Mol. Biol.,* **2005**, *58*(5), 699-720.

[103]   Olson, A.; Klein, R.R.; Dugas, D.V.; Lu, Z.; Regulski, M.; Klein, P.E.; Ware, D. Expanding and vetting gene annotations through transcriptome and methylome Sequencing. *Plant Genome,* **2014**, *7*(2).

[104]   Makita, Y.; Shimada, S.; Kawashima, M.; Kondou-Kuriyama, T.; Toyoda, T.; Matsui, M. (2015). MOROKOSHI: Transcriptome Database in Sorghum bicolor. *Plant Cell Physiol.,* **2015**, *56*(1), e6-e6.

[105]   Taylor, J.R.N.; Belton, P.S.; Beta, T.; Duodu, K.G. Increasing the utilisation of sorghum, millets and pseudocereals: Developments in the science of their phenolic phytochemicals, biofortification and protein functionality. *J. Cereal Sci.,* **2014**, *59*, 257–275.

[106]   Da Silva, LS.; Taylor, J.; Taylor, J.R. Transgenic sorghum with altered kafirin synthesis: kafirin solubility, polymerization, and protein digestion. *J. Agric. Food Chem.,* **2011**, *59*, 9265–9270.

[107]   Grootboom, A.W.; Mkhonza, N.L.; Mbambo, Z.; O'Kennedy, M.M.; Da Silva, L.S.; Taylor, J.; Taylor, J.R.N.; Chikwamba, R.; Mehlo, L. Co-suppression of synthesis of major α-kafirin sub-class together with γ-kafirin-1 and γ-kafirin-2 required for substantially improved protein digestibility in transgenic sorghum. *Plant Cell Rep.,* **2014**, *33*(3), 521-537.

[108]   Shewry, P.R. Improving the protein content and composition of cereal grain. *J. Cereal Sci.,* **2007**, *46*, 239–250.

[109]   Da Silva, LS.; Jung, R.; Zhao, Z.; Glassman, K.; Taylor, J.; Taylor, J. Effect of suppressing the synthesis of different kafirin subclasses on grain endosperm texture, protein body structure and protein nutritional quality in improved sorghum lines. *J. Cereal Sci.,* **2011**, *54*, 160–167.

[110]   Xin, Z.; Wang, M.L.; Barkley, N.A.; Burow, G.; Franks, C.; Pederson, G.; Burke, J. Applying genotyping (TILLING) and phenotyping analyses to elucidate gene function in a chemically induced sorghum mutant population. *BMC Plant Biol.,* **2008**, *8*(1), 103.

[111]   Paterson, A.H.; Bowers, J.E.; Bruggmann, R.; Dubchak, I.; Grimwood, J.; Gundlach, H.; Haberer, G.; Hellsten, U.; Mitros, T.; Poliakov, A.; Schmutz, J.; Spannagl, M.; Tang, H.; Wang, X.; Wicker, T.; Bharti, A.K.; Chapman, J.; Feltus, F.A.; Gowik, U.; Grigoriev, I.V.; Lyons, E.; Maher, C.A.; Martis, M.; Narechania, A.; Otillar, R.P.; Penning, B.W.; Salamov, A.A.; Wang, Y.; Zhang, L.; Carpita, N.C.; Freeling, M.; Gingle, A.R.; Hash, C.T.; Keller, B.; Klein, P.; Kresovich S, McCann MC, Ming R, Peterson DG, Mehboob-ur-Rahman, Ware D, Westhoff P, Mayer, K.F.; Messing, J.; Rokhsar, D.S. The Sorghum bicolor genome and the diversification of grasses. *Nature,* **2009**, *457*, 551-556.

[112]   Verma, A.K.; Patil, V.U.; Bhat, R.S. A transiently expressed trans-

posase system to generate Ds-tagged mutants for functional genomics in sorghum. *Plant Cell Tiss. Org.,* **2011**, *107*(1), 181-185.

[113]   Tinker, N.A.; Kilian, A.; Wight, C.P.; Heller-Uszynska, K.; Wenzl, P.; Rines, H.W.; Bjørnstad, A.; Howarth, C.J.; Jannink, J.L.; Anderson, J.M.; Rossnagel, B.G.; Stuthman, D.D.; Sorrells, M.E.; Jackson, E.W.; Tuvesson, S.; Kolb, F.L.; Olsson, O.; Federizzi, L.C.; Carson, M.L.; Ohm, H.W.; Molnar, S.J.; Scoles, G.J.; Eckstein, P.E.; Bonman, J.M.; Ceplitis, A.; Langdon, T. New DArT markers for oat provide enhanced map coverage and global germplasm characterization. *BMC Genomics,* **2009**, *10,* 3.

[114]   Newell, M.A.; Cook, D.; Tinker, N.A.; Jannink, J.L. Population structure and linkage disequilibrium in oat (*Avena sativa* L.): implications for genome-wide association studies. *Theor. Appl. Genet.,* **2011**, *122*(3), 623-632.

[115]   Newell, M.A.; Asoro, F.G.; Scott, M.P.; White, P.J.; Beavis, W.D.; Jannink, J.L. Genome-wide association study for oat (*Avena sativa* L.) beta-glucan concentration using germplasm of worldwide origin. *Theor. Appl. Genet.,* **2012**, *125*, 1687–1696.

[116]   Asoro, F.G.; Newell, M.A.; Scott, M.P.; Beavis, W.D.; Jannink, J.L. Genome-wide association study for beta-glucan concentration in elite North American oat. *Crop Sci.,* **2013**, *53*, 542–553.

[117]   Gutierrez-Gonzalez, JJ.; Tu, Z.J.; Garvin, D.F. Analysis and annotation of the hexaploid oat seed transcriptome. *BMC Genomics,* **2013**, *14*, 471.

[118]   Sikora, P. Mutagenesis and transcriptome mapping in oat and characterization of high β-glucan lines. Doctoral thesis, University of Gothenburg, **2013**.

[119]   Chang Y, W. Isolation and characterization of protein fractions from chickpea (*Cicer arietinum* L.) and oat (*Avena sativa* L.) seeds using proteomic techniques. Ph.D thesis, McGill University, **2010**.

[120]   Brindzová, L.; Čertík, M.; Rapta, P.; Zalibera, M.; Mikulajová, A.; Takácsová, M. Antioxidant activity, β-glucan and lipid contents of oat varieties. *Czech J. Food Sci.,* **2008**, *26*, 163–173.

[121]   Chawade, A.; Sikora, P.; Bräutigam, M.; Larsson, M.; Vivekanand, V.; Nakash, M.A.; Chen, T.; Olsson, O. Development and characterization of an oat TILLING-population and identification of mutations in lignin and β-glucan biosynthesis genes. *BMC Plant Biol.,* **2010**, *10*, 86.

[122]   Kumar, A.; Mirza, N.; Charan, T.; Sharma, N.; Gaur, V.S. Isolation, characterization and immunolocalization of a seed dominant CaM from finger millet (*Eleusine coracana* L. Gartn.) for studying Its functional role in differential accumulation of calcium in developing grains. *Appl. Biochem. Biotech.,* **2014**, *172*(6), 2955-2973.

[123]   Singh, U.M.; Chandra, M.; Shankhdhar, S.C.; Kumar, A. Transcriptome Wide Identification and Validation of Calcium Sensor Gene Family in the Developing Spikes of Finger Millet Genotypes for Elucidating Its Role in Grain Calcium Accumulation. *PloS one,* **2014**, *9*(8), e103963.

[124]   Kumar, A.; Gaur, V.S.; Goel, A.; Gupta, A.K. De Novo Assembly and Characterization of Developing Spikes Transcriptome of Finger Millet (*Eleusine coracana*): a Minor Crop Having Nutraceutical Properties. *Plant Mol. Biol. Rep.,* **2014**, 1-18.

[125]   Mirza, N.; Taj, G.; Arora, S.; Kumar, A. Transcriptional expression analysis of genes involved in regulation of calcium translocation and storage in finger millet (*Eleusine coracana* L. Gartn.). *Gene,* **2014**, *550*(2), 171-179.

[126]   Shobana, S. Investigation on the carbohydrate digestibility of finger millet with special reference to the influence of its seed oat constituents. **2009**. Ph.D. Thesis, University of Mysore, Mysore.

[127]   Jia, G.; Huang, X.; Zhi, H.; Zhao, Y.; Zhao, Q.; Li, W.; Han, B. A haplotype map of genomic variations and genome-wide association studies of agronomic traits in foxtail millet (*Setaria italica*). *Nature genetics,* **2013**, *45*(8), 957-961.

[128]   Vetriventhan, M.; Upadhyaya, H.D.; Anandakumar, C.R.; Senthilvel, S.; Parzies, H.K.; Bharathi, A.; Varshney, R.K.; Gowda, C.L.L. Assessing genetic diversity, allelic richness and genetic relationship among races in ICRISAT foxtail millet core collection. *Plant Genet. Resour.,* **2012**, *10*(3), 214-223.

[129]   Gupta, S.; Kumari, K.; Muthamilarasan, M.; Parida, S.K.; Prasad, M. Population structure and association mapping of yield contributing agronomic traits in foxtail millet. *Plant Cell Rep.,* **2014**, *33*(6), 881-893.

[130]   Jayaraman, A.; Puranik, S.; Rai, N.K.; Vidapu, S.; Sahu, P.P.; Lata, C., Prasad, M. cDNA-AFLP analysis reveals differential gene expression in response to salt stress in foxtail millet (*Setaria italica* L.). *Mol. Biotech.,* **2008**, *40*, 241-251.

[131]   Qi, X.; Xie, S.; Liu, Y.; Yi, F.; Yu, J. Genome-wide annotation of genes and noncoding RNA of foxtail millet in response to simulated drought stress by deep sequencing. *Plant Mol. Biol.*, **2013**, *83*, 459–473.

[132]   Xu, J.; Li, Y.; Ma, X.; Ding, J.; Wang, K.; Wang, S., Tian, Y., Zhang, H., Zhu, X.G.. Whole transcriptome analysis using next-generation sequencing of model species *Setaria viridis* to support C₄ photosynthesis research. *Plant Mol. Biol.*, **2013**, *83*, 77–87.

[133]   Diao, X.; Schnable, J.; Bennetzen, J.L.; Li, J. Initiation of setaria as a model plant. *Front Agri Sci Eng.,* **2014**, *1*, 16-20.

[134]   Lata, C.; Gupta, S.; Prasad, M. Foxtail millet: a model crop for genetic and genomic studies in bioenergy grasses. *Crit Rev Biotechnol.,* **2013** *33*, 328-343

[135]   IPMGSC. International Pearl Millet Genome Sequencing Consortium, ICRISAT. (www.ceg.icrisat.org/ipgsc.html)

[136]   Velu, G.; Rai, K.N.; Sahrawat, K.L. Variability for grain iron and zinc content in a diverse range of pearl millet populations. *Crop Improv.*, **2008**, *35*, 186-191.

[137]   Kim, J.K.; Park, S.Y.; Yeo, Y.; Cho, H.S.; Kim, Y.B.; Bae, H. Park, C.H.; Lee J.H.; Park, S.U. Metabolic profiling of millet (*Paniciim miliaceum*) using gas chromatography-time-of-flight mass spectrometry (GC-TOFMS) for quality assessment*. Plant Omics*, **2013**, *6*, 73-78.

[138]   Kalinova, J.; Moudry, J. Content and quality of protein in proso millet (*Panicum miliaceum* L.) varieties. *Plant Foods Hum. Nutr.*, **2006**, *61*, 45–49.

[139]   Hunt, H.V.; Denyer, K.; Packman, L.C.; Jones, M.K.; Howe, C.J. Molecular basis of the waxy endosperm starch phenotype in broomcornmillet(Panicum miliaceum L.). *Mol.Biol.Evol.*, **2010**, *27*,1478–1494.

[140]   Araki, M.; Numaoka,A.; Kawase, M.; Fukunaga, K. Origin of waxy common millet, Panicum miliaceum L. in Japan. *Genet. Res.Crop Evol.*, **2012**, *59*, 1303–1308.

[141]   Thiel, T.; Michalek, W.; Varshney, R.; Graner, A. Exploiting EST databases for the development and characterization of gene-derived SSR-markers in barley (*Hordeum vulgare* L.). *Theor. Appl. Genet.,* **2003**, *106*(3), 411-422.

[142]   Emebiri, L.C.; Moody, D.B.; Panozzo, J.F.; Read, B.J. Mapping of QTL for malting quality attributes in barley based on a cross of parents with low grain protein concentration. *Field Crop Res.*, **2004**, *87*(2), 195-205.

[143]   Ramsay, L., Macaulay, M., Ivanissevich, S.D., MacLean, K., Cardle, L., Fuller, J., Edwards, K.J., Tuvesson, S., Morgante, M., Massari, A., Maestri, E., Marmiroli, N., Sjakste, T., Ganal, M., Powell, W., Waugh, R. A Simple Sequence Repeat-based linkage map of barley. *Genetics*, **2000**, *156*, 1997-2005.

[144]   Willsmore, K.L.; Eckermann, P.; Varshney, R.K.; Graner, A.; Langridge, P. New eSSR and gSSR markers added to Australian barley maps. *Crop Pasture Sci*, **2006**, *57,* 953–959.

[145]   Li, H.; Vaillancourt, R.; Mendham, N.; Zhou, M. Comparative mapping of quantitative trait loci associated with waterlogging tolerance in barley (*Hordeum vulgare* L.). *BMC Genomics*, **2008**, *9*(1), 401.

[146]   Backes, G.; Graner, A.; Foroughi-Wehr, B.; Fischbeck, G.; Wenzel, G.; Jahoor, A. Localization of quantitative trait loci (QTL) for agronomic important characters by the use of an RFLP map in barley (*Hordeum vulgare* L.). *Theor. Appl. Genet.,* **1995**, *90*(2), 294–302.

[147]   Gao, W.; Clancy, J.A.; Han, F.; Jones, B.L.; Budde, A.; Wesenberg, D.M.; Kleinhofs, A.; Ullrich, S.E. Fine mapping of a malt-ingquality QTL complex near the chromosome 4HS telomere in barley. *Theor. Appl. Genet.*, **2004**, *109*, 750-760.

[148]   Wenzl, P.; Carling, J.; Kudrna, D.; Jaccoud, D.; Huttner, E.; Kleinhofs, A.; Kilian, A. Diversity Arrays Technology (DArT) for whole-genome profiling of barley. *Proc. Natl. Acad. Sci. U. S. A.,* **2004**, *101*(26), 9915-9920.

[149]   Zhang, L. Y.; Marchand, S.; Tinker, N. A.; Belzile, F. Population structure and linkage disequilibrium in barley assessed by DArT markers. *Theor. Appl. Genet.*, **2009**, *119*(1), 43-52.

[150]   Close, T.J., Bhat, P.R., Lonardi, S., Wu, Y., Rostoks, N., Ramsay, L., Druka, A., Stein, N., Svensson, J.T., Wanamaker, S., Bozdag, S., Roose, M.L., Moscou, M.J., Chao, S., Varshney, R.K., Szűcs, P., Sato, K., Hayes, P.M., Matthews, D.E., Kleinhofs, A., Muehlbauer, G.J., DeYoung, J., Marshall, D.F., Madishetty, K., Fenton, R.D., Condamine, P., Graner, A., Waugh, R. Development and implementation of high throughput SNP genotyping in barley. *BMC Genomics*, **2009**, *10*, 582.

[151]   Chutimanitsakun, Y.; Nipper, R.W.; Cuesta-Marcos, A.; Cistué, L.; Corey, A.; Filichkina, T.; Johnson, E.A.; Hayes, P.M. Construction and application for QTL analysis of a Restriction Site Associated DNA (RAD) linkage map in barley. *BMC Genomics*, **2011**, *12*(1), 4.

[152]   Poland, J.A.; Brown, P.J.; Sorrells, M.E.; Jannink, J.L. Development of high-density genetic maps for barley and wheat using a novel two-enzyme genotyping-by-sequencing approach. *PloS One,* **2012**, *7*(2), e32253.

[153]   Mascher, M., Richmond, T.A., Gerhardt, D.J., Himmelbach, A., Clissold, L., Sampath, D., Ayling, S., Steuernagel, B., Pfeifer, M., D'Ascenzo, M., Akhunov, E.D., Hedley, P.E., Gonzales, A.M., Morrell, P.L., Kilian, B., Blattner, F.R., Scholz, U., Mayer, K.F., Flavell, A.J., Muehlbauer, G.J., Waugh, R., Jeddeloh, J.A., Stein N. Barley whole exome capture: a tool for genomic research in the genus Hordeum and beyond. *Plant J.,* **2013**, *76*, 494-505.

[154]   Zeng, X., Long, H., Wang, Z., Zhao, S., Tang, Y., Huang, Z., Wang, Y., Xu, Q., Mao, L., Deng, G., Yao, X., Li, X., Bai, L., Yuan, H., Pan, Z., Liu, R., Chen, X., WangMu, Q., Chen, M., Yu L., Liang, J., DunZhu, D., Zheng, Y., Yu, S., LuoBu, Z., Guang, X., Li, J., Deng, C., Hu, W., Chen, C., TaBa, X., Gao, L., Lv, X., Abu, YB., Fang X., Nevo, E., Yu, M., Wang, J., Tashi, N. The draft genome of Tibetan hulless barley reveals adaptive patterns to the high stressful Tibetan Plateau. *Proc. Natl. Acad. Sci. U. S. A.,* **2015**, *112*(4), 1095-1100.

[155]   Winn, J.A.; Mason, R.E.; Robbins, A.L.; Rooney, W.L.; Hays, D.B. QTL mapping of a high protein digestibility trait in Sorghum bicolor. *Int. J. Plant Genomics*, **2009**, ID471853.

[156]   Mace, E.S.; Xia, L.; Jordan, .D.R.; Halloran, K.; Parh, D.K.; Huttner, E.; Wenzl, P.; Kilian, A. DArT markers: diversity analyses and mapping in Sorghum bicolor. *BMC Genomics*, **2008**, 9, 26.

[157]   Bouchet, S.; Pot, D.; Deu, M.; Rami, J. F.; Billot, C.; Perrier, X.; Rivallan, R, Gardes, L, Xia, L, Wenzl, P.; Kilian, A.; Glaszmann, J.C. Genetic structure, linkage disequilibrium and signature of selection in sorghum: lessons from physically anchored DArT markers. *PLoS One*, **2012**, *7*(3), e33470.

[158]   Sukumaran, S.; Xiang, W.; Bean, S.R.; Pedersen, J.F.; Kresovich, S.; Tuinstra, M.R.; Tesso, T.T.; Hamblin, M.T.; Yu, J. Association mapping for grain quality in a diverse sorghum collection. *Plant Genome*, **2012**, *5*(3), 126-135.

[159]   Nelson, J.C.; Wang, S.; Wu, Y.; Li, X.; Antony, G.; White, F. F.; Yu, J. Single-nucleotide polymorphism discovery by high-throughput sequencing in sorghum. *BMC genomics*, **2011**, *12*(1), 352.

[160]   Morris, G.P., Ramu, P., Deshpande, S.P., Hash, C.T., Shah, T., Upadhyaya, H.D., Riera-Lizarazu, O., Brown, P.J., Acharya, C.B., Mitchell, S.E., Harriman, J., Glaubitz, J.C., Buckler, E.S., Kresovich, S. Population genomic and genome-wide association studies of agroclimatic traits in sorghum. *Proc. Natl. Acad. Sci. U. S. A.*, **2013**, *110*(2), 453-458.

[161]   Bekele, W.A.; Wieckhorst, S.; Friedt, W.; Snowdon, R.J. High-throughput genomics in sorghum: from whole-genome resequencing to a SNP screening array. *Pl. Biotechnol. J.*, **2013**, *11*(9), 1112-1125.

[162]   Kianian, S.F.; Phillips, R.L.; Rines, H.W.; Fulcher, R.G.; Webster, F.H.; Stuthman, D.D. Quantitative trait loci influencing β-glucan content in oat (*Avena sativa*, 2n= 6x= 42). *Theor. Appl. Genet.,* **2000**, *101*(7), 1039-1048.

[163]   De Koeyer, D. L; Tinker, N. A.; Wight, C. P.; Deyl, J.; Burrows, V. D.; O'Donoughue, L. S.; Lybaert, A.; Molnar, S. J.; Armstrong, K. C.; Fedak, G.; Wesenberg, D.M.; Rossnagel, B.G.; McElroy A. R. A molecular linkage map with associated QTLs from a hulless x covered spring oat population. *Theor. Appl. Genet.*, **2004**, *108* (7), 1285-98.

[164]   Becher, R. EST-derived microsatellites as a rich source of molecular markers for oats. *Plant Breeding*, **2007**, *126*(3), 274-278.

[165]   Oliver, R.E.; Lazo, G.R.; Lutz, J.D.; Rubenfield, M.J.; Tinker, N.A.; Anderson, J.M.; Morehead, N.H.W.; Adhikary, D.; Jellen,E.N.; Maughan, P.J.; Guedira, G.L.B.; Chao, S.; Beattie, A.D.; Carson, M.L.; Rines, H.W.; Obert, D.E.; Bonman, J.M.; Jackson, E.W. Model SNP development for complex genomes based on hexaploid oat using high-throughput 454 sequencing technology. *BMC genomics*, **2011**, *12*(1), 77.

[166]   Oliver, R.E., Obert, D.E., Hu, G., Bonman, J.M., O'Leary-Jepsen, E., Jackson, E.W. Development of oat-based markers from barley

and wheat microsatellites. *Genome*, **2010**, *53,* 458-471.

[167] Oliver, R.E.; Tinker, N.A.; Lazo, G.R.; Chao, S.; Jellen, E.N.; Carson, M.L.; Rines, H.W.; Obert, D.E.; Lutz, J.D.; Shackelford, I.; Korol, A.B.; Wight, C.P.; Gardner, K.M.; Hattori, J.; Beattie, A.D.; Bjørnstad, Å.; Bonman, J.M.; Jannink, J.L.; Sorrells, M.E.; Brown-Guedira, G.L.; Mitchell, F.J.W.; Harrison, S.A.; Howarth, C.J.; Ibrahim, A.; Kolb, F.L.; McMullen, M.S.; Murphy, J.P.; Ohm, H.W.; Rossnagel, B.G.; Yan, W.; Miclaus, K.J.; Hiller, J.; Maughan, P.J.; Redman Hulse, R.R.; Anderson, J.M.; Islamovic, E.; Jackson, E.W.SNP discovery and chromosome anchoring provide the first physically-anchored hexaploid oat map and reveal synteny with model species. *PLoS One*, **2013**, *8*(3), e58068.

[168] Qi, X.; Pittaway, T.S.; Lindup, S.; Liu, H.; Waterman, E.; Padi, F.K.; Hash, C.T.; Zhu, J.; Gale, M.D.; Devos, K.M. An integrated genetic map and a new set of simple sequence repeat markers for pearl millet, *Pennisetum glaucum. Theor. Appl. Genet.,* **2004**, *109*(7), 1485-1493.

[169] Rajaram, E., Nepolean, T., Senthilvel, S., Varshney, R.K., Vadez, V., Srivastava, R.K., Shah, T.M., Supriya, A., Kumar, S., Kumari, B.R., Bhanuprakash, A., Narasu, M.L., Riera-Lizarazu, O., Hash, C.H. Pearl millet [*Pennisetum glaucum* (L.) R. Br.] consensus linkage map constructed using four RIL mapping populations and newly developed EST-SSRs. *BMC Genomics,* **2013**, *14*, 159.

[170] Bertin, I.; Zhu, J.H.; Gale, M.D. SSCP-SNP in pearl millet—a new marker system for comparative genetics. *Theor. Appl. Genet.,* **2005**, *110*(8), 1467-1472.

[171] Vengadessan, V.; Rai, K.N.; Kannan Bapu J.R.; Hash, C.T.; Bhattacharjee, R.; Senthilvel, S.; Vinayan, M.T.; Nepolean, T. Construction of Genetic Linkage Map and QTL Analysis of Sink-Size Traits in Pearl Millet (*Pennisetum glaucum*). ISRN *Genetics*, **2013**, 471632.

[172] Feltus, F.A.; Singh, H.P.; Lohithaswa, H.C.; Schulze, S.R.; Silva, T.D.; Paterson, A.H. A comparative genomics strategy for targeted discovery of single-nucleotide polymorphisms and conserved-noncoding sequences in orphan crops. *Plant physiol.*, **2006**, *140*(4), 1183-1191.

[173] Supriya, A.; Senthilvel, S.; Nepolean, T.; Eshwar, K.; Rajaram, V.; Shaw, R.; Hash, C. T.; Yadav, R.C.; Narasu, M.L. Development of a molecular linkage map of pearl millet integrating DArT and SSR markers. *Theor. Appl. Genet.*, **2011**, *123*(2), 239-250.

[174] Varsheny, R.K., Towards developing a reference genome sequence of pearl millet (*Pennisetum glaucum* L.), In; Proceeding of XXIIIth plant and Animal genome conference, San Diego CA, January 10-14; **2015**.

[175] Wang, Z.M.; Devos, K.M.; Liu, C.J.; Wang, R. Q.; Gale, M. D. Construction of RFLP-based maps of foxtail millet, *Setaria italica* (L.) P. Beauv. *Theor. Appl. Genet.,* **1998**, *96*(1), 31-36.

[176] Schontz, D.; Rether, B. Genetic variability in foxtail millet, Setaria italica (L.) P. Beauv.: Identification and classification of lines with RAPD markers. *Plant Breeding*, **1999** *118*, 190–192.

[177] Jia, X.; Zhang, Z.; Liu, Y.; Zhang, C.; Shi, Y.; Song, Y.; Li, Y. Development and genetic mapping of SSR markers in foxtail millet [*Setaria italica* (L.) P. Beauv.]. *Theor. Appl. Genet.,* **2009**, *118*(4), 821-829.

[178] Lin, H.S., Chiang, C.Y., Chang, S.B., Kuoh, C.S. Development of simple sequence repeats (SSR) markers in *Setaria italica* (*Poaceae*) and cross-amplification in related species. *Int. J. Mol. Sci.,* **2011**, *12,* 7835-7845.

[179] Gupta, S.; Kumari, K.; Sahu, P. P.; Vidapu, S.; Prasad, M. Sequence-based novel genomic microsatellite markers for robust genotyping purposes in foxtail millet [*Setaria italica* (L.) P. Beauv.]. *Plant Cell Rep.*, **2012**, *31*(2), 323-337.

[180] Zhang, G.; Liu, X.; Quan, Z.; Cheng, S.; Xu, X.; Pan, S.; Xie, M.; Zeng, P.; Yue, Z.; Wang, W.; Tao, Y.; Bian, C.; Han, C.; Xia, Q.; Peng, X.; Cao, R.; Yang, X.; Zhan, D.; Hu, J.; Zhang, Y.; Li, H.; Li, H.; Li, N.; Wang, J.; Wang, C.; Wang, R.; Guo, T.; Cai, Y.; Liu, C.; Xiang, H.; Shi, Q.; Huang, P.; Chen, Q.; Li, Y.; Wang, J.; Zhao, Z.; Wang, J. Genome sequence of foxtail millet (Setaria italica) provides insights into grass evolution and biofuel potential. *Nat. Biotechnol.*, **2012**, *30*(6), 549-54.

[181] Bennetzen, J. L.; Schmutz, J.; Wang, H.; Percifield, R.; Hawkins, J.; Pontaroli, A. C.; Devos, K. M. Reference genome sequence of the model plant *Setaria. Nat. biotechnol.*, **2012**, *30*(6), 555-561.

[182] Jia, G.; Huang, X.; Zhi, H.; Zhao, Y.; Zhao, Q.; Li, W.; Han, B. A haplotype map of genomic variations and genome-wide association studies of agronomic traits in foxtail millet (*Setaria italica*). *Nat.*

Genet., **2013**, *45*(8), 957-961.

[183] Kumari, K.; Muthamilarasan, M.; Misra, G.; Gupta, S.; Subramanian, A.; Parida, S. K.; Prasad, M. Development of eSSR-markers in *Setaria italica* and their applicability in studying genetic diversity, cross-transferability and comparative mapping in millet and non-millet species. *PLoS One*, **2013**, *8*(6), e67742.

[184] Pandey, G.; Misra, G.; Kumari, K.; Gupta, S.; Parida, S. K.; Chattopadhyay, D.; Prasad, M. Genome-wide development and use of microsatellite markers for large-scale genotyping applications in foxtail millet [*Setaria italica* (L.)]. *DNA Res.*, **2013**, *20*(2), 197-207.

[185] Zhang, S., Tang, C., Zhao, Q., Li, J., Yang, L., Qie, L. Development of highly polymorphic simple sequence repeat markers using genome-wide microsatellite variant analysis in Foxtail millet [*Setaria italica* [L.] P. Beauv.]. *BMC Genomics,* **2014**, *15*(1), 78.

[186] Muthamilarasan, M.; Suresh, B.V.; Pandey, G.; Kumari, K.; Parida, S.K.; Prasad, M. Development of 5123 intron-length polymorphic markers for large-scale genotyping applications in foxtail millet. *DNA Res.*, **2014**, *21*(1), 41-52.

[187] Yadav, C.B.; Muthamilarasan, M.; Pandey, G.; Khan, Y.; Prasad, M. Development of novel microRNA-based genetic markers in foxtail millet for genotyping applications in related grass species. *Mol. Breed*, **2014**, *34*, 2219-2224.

[188] Yadav, C.B.; Bonthala, V.S.; Muthamilarasan, M.; Pandey, G.; Khan, Y.; Prasad, M. Genome-wide development of transposable elements-based markers in foxtail millet and construction of an integrated database. *DNA Res.*, **2014**, *22*(1), 79-90.

[189] Muthamilarasan, M.; Mishra, G.; Prasad, M. FmMDb: A versatile database of foxtail millet markers for millets and bioenergy grasses research. *PLoS One,* **2013**, *8,* e71418.

[190] M'ribu, H.K.; Hilu, K.W. Detection of interspecific and intraspecific variation in Panicum millets through random amplified polymorphic DNA. *Theor. Appl. Genet.,* **1994**, *88*(3-4), 412-416.

[191] Karam, D.; Westra, P.; Nissen, S.J.; Ward, S.M.; Figueiredo, J.E.F. Genetic diversity among proso millet (*Panicum miliaceum)* biotypes assessed by AFLP technique. *Planta Daninha*, **2004**, *22*(2), 167-174.

[192] Cho, Y.I.; Chung, J.W.; Lee, G.A.; Ma, K.H.; Dixit, A.; Gwag, J.G.; Park, Y.J. Development and characterization of twenty-five new polymorphic microsatellite markers in proso millet (*Panicum miliaceum* L.). *Genes Genom.*, **2010**, *32*(3), 267-273.

[193] Hu, X.; Wang, J.; Lu, P.; Zhang, H. Assessment of genetic diversity in broomcorn millet (*Panicum miliaceum* L.) using SSR markers. *J. Genet. Genomics*, **2009**, *36*(8), 491-500.

[194] Rajput, S.G.; Plyler-Harveson, T.; Santra, D.K. Development and Characterization of SSR Markers in Proso Millet Based on Switchgrass Genomics. *Am. J. Plant Sci.,* **2014**, *5*(01), 175.

[195] M'Ribu, H.K.; Hilu, K.W. Application of random amplified polymorphic DNA to study genetic diversity in *Paspalum scrobiculatum* L. (Kodo millet, Poaceae). *Genet. Resour. Crop Evol.,* **1996**, *43*, 203–210.

[196] Kushwaha, H.; Jillo, K. W.; Singh, V. K.; Kumar, A.; Yadav, D. Assessment of genetic diversity among cereals and millets based on PCR amplification using Dof (DNA binding with One Finger) transcription factor gene-specific primers. *Plant Syst. Evol.,* **2014**, *301*(2), 833-840.

[197] Wallace, J.G.; Upadhyaya, H.D.; Vetriventhan, M.; Buckler, E.S.; Hash, C.T.; Ramu, P. The genetic makeup of a global barnyard millet germplasm collection. *Plant Genome*, **2015**, doi:10.3835.

[198] Dwivedi, S., Upadhvava, H., Senthilvel, S., Hash, C., Fukunaga, K., Diao, X., Santra, D., Baltensperger, D., Prasad, M. Millets: genetic and genomic resources. *Plant Breeding Rev.,* **2011**, *35*, 247-375.

[199] Dahlberg, J.A.; Burkee, J.J.; Rosenow, D.T. Development of a sorghum core collection: Refinement and evaluation of a subset from Sudan. *Econ. Bot.,* **2004**, *58*, 556-567.

[200] Bhattacharjee, R.; Bramel, P.J.; Khairwal, I.S.; Reddy, K.N. Establishment of a pearl millet [*Pennisetum glaucum* (L) R. Br.] core collection based on geographical distribution and quantitative traits. *Euphytica*, **2007**, *155*, 35-45.

[201] Upadhyaya, H.D.; Gowda, C.L.L.; Reddy, K.N.; Singh S. Augmenting the pearl millet [*Pennisetum glaucum* (L.) R. Br.)] core collection for enhancing germplasm utilization in crop improvement. *Crop Sci.*, **2009**, *49*, 573-580.

[202] Upadhyaya, H.D.; Gowda, C.L.L.; Pundir, R.P.S.; Reddy, V.G.; Singh S. Development of core subset of finger millet germplasm using geographical origin and data on 14 quantitative traits. *Genet.*

Resour. Crop Ev., **2006,** *53,* 679-685.

[203]   Upadhyaya, H. D.; Pundir, R. P. S.; Gowda, C. L. L.; Reddy, V. G.; Singh, S. *Establishing a core collection of foxtail millet to enhance the utilization of germplasm of an underutilized crop.* Pl. *Genet. Resour.,* **2009.** *7* (2), 177-184.

[204]   Upadhyaya, H. D.; Sharma, S.; Gowda, C. L. L.; Reddy, V. G.; Singh, S. Developing proso millet (Panicum miliaceum L.) core collection using geographic and morpho-agronomic data. *Crop Pasture Sci.,* **2011,** *62,* 383–389.

[205]   Upadhyaya, H.D.; Dwivedi, S.L.; Singh, S.K.; Singh, S.; Vetriventhan, M.; Sharma, S. Forming core collections in Barnyard, Kodo, and Little Millets using morphoagronomic descriptors. *Crop Sci.*, **2014,** *54*(6), 2673-2682.

[206]   Knüpffer, H., van Hintum, Th.J.L. The Barley Core Collection: an international effort. In: Hodgkin T.; A.H.D. Brown, Th. J. L. van

Hintum and E. A. V. Morales (Eds), Core Collections of Plant Genetic Resources, John Wiley and Sons, New York, Chichester, Brisbane, Toronto, Singapore, **1995,** pp. 171–178.

[207]   Hintum, T.J.L., Bothmer von, R.; Visser, D.L. Sampling strategies for composing a core collection of cultivated barley (*Hordeum vulgare*) collected in China. *Hereditas,* **1995,** *122,* 7–17.

[208]   Igartua, E.; Gracia, M.P.; Lasa, J.M.; Medina, B.; Molina-Cano, J.L.; Montoya, J.L.; Romagosa, I. The Spanish barley core collection. *Genet. Resour. Crop Ev.*, **1998,** *45*(5), 475-481.

[209]   Upadhyaya, H.D., Ramesh, S., Shivali, S., Singh, S.K., Varshney, S.K., Sarma, N.D.R.K., Ravishankar, C.R., Narasimhudu, Y., Reddy, V.G., Sahrawat, K.L., Dhanalakshmi, T.N., Mgonja, M.A., Parzies, H.K., Gowda, C.L.L., Singh, S. Genetic diversity for grain nutrients contents in a core collection of finger millet (*Eleusine coracana* (L.) gaertn.) germplasm. *Field Crop Res.,* **2011,** *121,* 42–52.