Biotechnology Advances 31 (2013) 1120–1134

Contents lists available at ScienceDirect

Biotechnology Advances

journal homepage: www.elsevier.com/locate/biotechadv

Research review paper

Achievements and prospects of genomics-assisted breeding in three legume crops of the semi-arid tropics

Rajeev K. Varshney [a,b,c,d,*], S. Murali Mohan [a], Pooran M. Gaur [a], N.V.P.R. Gangarao [e], Manish K. Pandey [a,f], Abhishek Bohra [g], Shrikant L. Sawargaonkar [a], Annapurna Chitikineni [a], Paul K. Kimurto [h], Pasupuleti Janila [a], K.B. Saxena [a], Asnake Fikre [i], Mamta Sharma [a], Abhishek Rathore [a], Aditya Pratap [g], Shailesh Tripathi [j], Subhojit Datta [g], S.K. Chaturvedi [g], Nalini Mallikarjuna [a], G. Anuradha [k], Anita Babbar [l], Arbind K. Choudhary [m], M.B. Mhase [n], Ch. Bharadwaj [j], D.M. Mannur [o], P.N. Harer [n], Baozhu Guo [f], Xuanqiang Liang [d], N. Nadarajan [g], C.L.L. Gowda [a]

[a] International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad 502324, India
[b] CGIAR Generation Challenge Programme (GCP), c/o CIMMYT, DF 06600, Mexico
[c] The University of Western Australia, Crawley 6009, Australia
[d] Crops Research Institute, Guangdong Academy of Agricultural Sciences (GAAS), Guangzhou 510640, China
[e] International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Nairobi 39063, Kenya
[f] Crop Protection and Management Research Unit, US Department of Agriculture - Agricultural Research Service (USDA-ARS), Tifton 31793, USA
[g] Indian Institute of Pulses Research (IIPR), Kanpur 208024, India
[h] Egerton University, Egerton 536-20115, Kenya
[i] Debre Zeit Agricultural Research Centre, Ethiopian Agricultural Research Organization (EARO), Debre Zeit 32, Ethiopia
[j] Division of Genetics, Indian Agricultural Research Institute (IARI), New Delhi 110012, India
[k] Institute of Biotechnology, Acharya NG Ranga Agricultural University (ANGRAU), Hyderabad 500030, India
[l] Jawaharlal Nehru Krishi Vishwa Vidyalaya (JNKVV), Jabalpur 482004, India
[m] Indian Institute of Pulses Research (IIPR), Off-season Research Station, Dharwad 580005, India
[n] Mahatma Phule Krishi Vidyapeeth (MPKV), Rahuri 413722, India
[o] Agricultural Research Station, University of Agricultural Sciences (UAS-Raichur), Gulbarga 585101, India

ARTICLE INFO

Article history:
Received 23 July 2012
Received in revised form 16 December 2012
Accepted 3 January 2013
Available online 11 January 2013

Keywords:
Transcriptome
Molecular markers
Genetic maps
Genomic selection
Molecular breeding

ABSTRACT

Advances in next-generation sequencing and genotyping technologies have enabled generation of large-scale genomic resources such as molecular markers, transcript reads and BAC-end sequences (BESs) in chickpea, pigeonpea and groundnut, three major legume crops of the semi-arid tropics. Comprehensive transcriptome assemblies and genome sequences have either been developed or underway in these crops. Based on these resources, dense genetic maps, QTL maps as well as physical maps for these legume species have also been developed. As a result, these crops have graduated from 'orphan' or 'less-studied' crops to 'genomic resources rich' crops. This article summarizes the above-mentioned advances in genomics and genomics-assisted breeding applications in the form of marker-assisted selection (MAS) for hybrid purity assessment in pigeonpea; marker-assisted backcrossing (MABC) for introgressing QTL region for drought-tolerance related traits, *Fusarium* wilt (FW) resistance and *Ascochyta* blight (AB) resistance in chickpea; late leaf spot (LLS), leaf rust and nematode resistance in groundnut. We critically present the case of use of other modern breeding approaches like marker-assisted recurrent selection (MARS) and genomic selection (GS) to utilize the full potential of genomics-assisted breeding for developing superior cultivars with enhanced tolerance to various environmental stresses. In addition, this article recommends the use of advanced-backcross (AB-backcross) breeding and development of specialized populations such as multi-parents advanced generation intercross (MAGIC) for creating new variations that will help in developing superior lines with broadened genetic base. In summary, we propose the use of integrated genomics and breeding approach in these legume crops to enhance crop productivity in marginal environments ensuring food security in developing countries.

* Corresponding author at: International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad 502324, India. Tel.: +91 40 30713305; fax: +91 40 30713074.
E-mail address: varshney.raj@gmail.com (R.K. Varshney).

0734-9750
http://dx.doi.org/10.1016/j.biotechadv.2013.01.001

Contents

1. Introduction

Legumes form an important constituent of food crops consumed globally and complement cereal crops as a source of dietary protein. In addition to providing important micronutrients to human beings, they also fix atmospheric nitrogen, which consequently increase soil fertility and production of other cereal crops. Legumes are also important source of fodder in many agricultural systems and are grown increasingly on a large-scale in semi-arid tropics (SAT). SAT regions cover many developing countries from Africa, Asia to Latin America, and they are characterized by low and erratic rainfall, prolonged dry seasons, and soils with low fertility. This environment is home to the poor and one-sixth of the world's human population (http://oar.icrisat.org/5283/1/Impact-Flyer-%20Africa.pdf).

Agriculture in the SAT regions is generally undertaken by smallholder farmers and is the mainstay of their livelihood. Among several food crops, chickpea (*Cicer arietinum*), pigeonpea (*Cajanus cajan*) and groundnut or peanut (*Arachis hypogaea*) are the leading legume crops to feed underprivileged living in the SAT, which is also called "habitat of the hungry". As these legume crops are grown in harsh environments and exposed to various biotic and abiotic stresses, their productivity has not increased significantly for the last 50 years (Fig. 1) (FAO, 2012). It is, therefore, important to enhance productivity of these crops to cope up with increased demand by the expanding human population. Although some progress has been made in this direction through conventional breeding methods which may be attributed to insufficient understanding of the underlying genetical or molecular mechanisms conferring resistance/tolerance to biotic/abiotic stresses. Advances in genomics have improved our understanding towards genetic architecture and molecular mechanism for complex traits which led to identification of marker-trait associations for economically important traits in order to enhance selection efficiency in breeding. Tremendous progress made in recent years in genomics research of SAT legume crops namely chickpea, pigeonpea and groundnut has prompted us to review the achievements made so far along with initiatives and future prospects for further genetic enhancement.

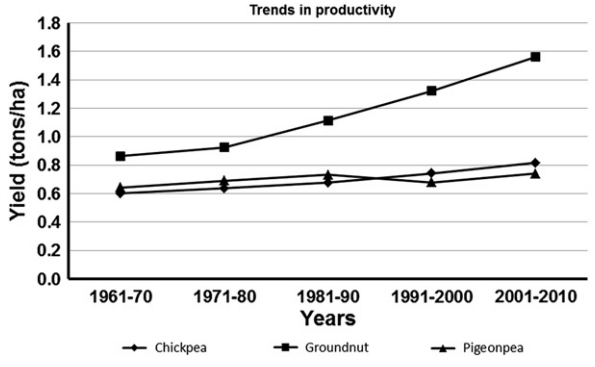

Fig. 1. Trends in crop productivity of three SAT legume crops. Trends in crop productivity during last five decades have been shown in chickpea, pigeonpea and groundnut. Except a small growth in the case of groundnut, in general, the crop productivity has been remained almost stagnant in the SAT legume crops.

2. SAT legume crops and production constraints

2.1. Chickpea (*Cicer arietinum* L.)

Chickpea, also known as garbanzo bean, is a self-pollinated diploid ($2n = 2\times = 16$) crop with genome size of 740 Mb (Arumuganathan and Earle, 1991). The seeds of chickpea are rich in protein (24.6%), carbohydrate (64.6%) and vitamins (Abu-Salem and Abou, 2011). During 2010, chickpea covered a total of 11.9 Mha worldwide with a global production of 10.9 million tons (Mt) and average yield of 913 kg/ha (FAO, 2012). Several abiotic and biotic stresses pose a big threat to high and stable yields of chickpea in the farmers' fields. Among abiotic stresses, terminal drought is a major problem for the crop grown under rainfed conditions as it delays flowering and affects seed yield. In addition to the above, this crop is also sensitive to lower temperature (<10 °C) mainly during reproductive period (Bakht et al., 2006) and to salinity (NaCl) during flowering and podding stages (Flowers et al., 2010). Salinity can affect the root nodules by decreasing their number, size and N_2-fixation capacity. Important biotic stresses affecting chickpea production are, *Fusarium* wilt (FW) caused by *Fusarium oxysporum* f.sp. *ciceri*, reduces yield up to 90% (Singh and Reddy, 1991) and *Ascochyta* blight (AB) caused by *Ascochyta rabiei* (Pass.) Labrousse, may cause total crop loss (Singh and Reddy, 1996). Other biotic stresses of chickpea are *Botrytis* gray mold (BGM) caused by *Botrytis cinerea* Pers. ex. Fr., leaf spot by *Alternaria* spp., black root rot by *Fusarium solani*, *Phytophthora* root rot by *Phytophthora megasperma* and *Pythium* damping-off by *Pythium ultimum*, rust by *Uromyces* and beet western yellow virus (BWYV) causing narrow leaf (Nene and Reddy, 1987). Pod borer or *Helicoverpa armigera* is the major insect pest of chickpea and it feeds on leaves and developing seeds (Sharma et al., 2005). Because of its complex nature and non-availability of good resistance sources in cultivated gene pool,

breeding for resistance to *Helicoverpa* in chickpea has remained a serious challenge (Sharma et al., 2008).

2.2. Pigeonpea (*Cajanus cajan* (L.) Millspaugh)

Pigeonpea is an often cross-pollinated diploid (2n = 2× = 22) crop with 833.07 Mb genome size (Varshney et al., 2012a). Globally pigeonpea is cultivated on 4.75 Mha yielding 3.68 Mt with an average yield of 774 kg/ha during 2010 (FAO, 2012). Pigeonpea supplements the vegetarian diet in developing countries by ensuring high supply of vitamin B, carotene, and ascorbic acid (Miller et al., 1956) which are otherwise deficient in cereals. Being a nitrogen fixing crop, the green manure of pigeonpea offers a natural source for improving soil health by providing organic material rich in nitrogen to the soil (Whiteman and Norton, 1980). The deep root system of pigeonpea also adds adaptive value to the crop making it one of the most tolerant crops of drought prone marginal environments. However, despite of its immense importance in sustainable agriculture and continued breeding efforts directed towards genetic improvement, the global production per hectare of pigeonpea remained static over last three decades. The yield gap between the potential yield and on-farm yield is mainly due to prevalence of various abiotic and biotic stresses in pigeonpea growing areas together with its cultivation in marginal lands with low input supply and lack of efficient management practices (see Varshney et al., 2012b). Among the various diseases, FW caused by *Fusarium udum* Butler, is the most important disease in Indian Subcontinent and Eastern Africa (Saxena, 2008). Occurrence of wilting during pod filling stage causes infection to pigeonpea seeds and yield losses up to 50–70% (Marley and Hillocks, 1996). Another disease severely affecting the pigeonpea yield is sterility mosaic disease (SMD) caused by pigeonpea sterility mosaic virus (PPSMV) causing losses up to 95 to 100% with infection occurring early at <45 days old plants (Kannaiyan et al., 1984). Apart from wilt and mosaic, *Phytophthora* blight caused by the fungus *Phytophthora drechsleri* Tucker f. sp. *cajani* is another important disease that has got the status of economic concern. However, the disease is limited in distribution and witness more severity in short duration cultivars as compared to long or medium duration genotypes (Ratnaparkhe and Gupta, 2007). Among the variety of insects feeding on pigeonpea, the pod borer, *Helicoverpa armigera* (Hubner) is the most damaging pest worldwide and its frequent occurrence often results in complete crop failure. Besides *Helicoverpa*, other pests like *Maruca* (*Maruca vitrata* Geyer), pod sucking bugs (*Clavigralla horrida* Germar) and podfly (*Melanagromyza chalcosoma* Spencer) pose a big threat to pigeonpea production (Shanower et al., 1999). Moreover, infestations from storage pests like bruchids (*Callosobruchus chinensis*) intensify the situation and result in profound seed damage during storage. Among the abiotic constraints, salinity and water logging severely affect the pigeonpea production (Choudhary et al., 2011; Saxena, 2008). The limited success achieved so far in addressing the problem of production constraints is mainly due to complex mechanism underlying these stresses together with the lack of precise and efficient screening techniques.

2.3. Groundnut (*Arachis hypogaea* L.)

The cultivated groundnut, self-pollinated crop with tetraploid (2n = 4× = 40) genome, has originated through a single hybridization and polyploidization event. Successive selection resulted in a highly narrow genetic base of the cultivated species (Young et al., 1996). Even being a tetraploid, cultivated groundnut genetically behaves as diploid due to unusual pairing of AA- and BB- genome chromosomes during meiosis (Stalker, 1991). With the annual production of 37.7 Mt covering 24.1 Mha achieving an average yield of 1564 kg/ha during 2010, this crop stands fourth largest oilseed crop in the world which is cultivated in more than 100 countries (FAO, 2012). The largest producers of groundnut include China and India followed by other countries in Sub-Saharan Africa and Americas. Groundnut seeds are highly nutritious possessing fat (40–50%), protein (20–30%), carbohydrate (10–20%) and several other micronutrients and minerals (Vitamin E, niacin, calcium, phosphorus, magnesium, zinc, iron, riboflavin, thiamine and potassium). It is an excellent cash crop and has multipurpose uses of each plant part in direct consumption, confectionary preparations, cooking oil and a rich source of protein feed for animals (see Pandey et al., 2012a).

Harnessing genetic yield potential in groundnut is severely challenged by several biotic/abiotic stress factors. Among several biotic stresses, early leaf spot (ELS) (*Cercospora arachidicola*), late leaf spot (LLS) (*Cercosporidium personatum*), rust (*Puccinia arachidis*) and groundnut rosette disease (GRD) (groundnut rosette virus, groundnut rosette assistor virus and *Sat*RNA complex) cause up to 50% yield loss. In addition, aflatoxin contamination deteriorates product quality leading to financial loss to farmers and safety issues to consumers. During pod development and seed filling stages, moisture stress conditions increase susceptibility to produce aflatoxin contamination (by *Aspergillus flavus*/*A. parasiticus*). Groundnut bud necrosis and bacterial wilt disease along with nematodes have also been found to be prevalent in some specific regions. Stem and pod rot, caused by *Sclerotium rolfsii*, is a potential threat to groundnut production in many warm and humid areas, especially where irrigated groundnut cultivation is expanding. Although several chemical treatments are available to control these diseases, host-plant resistance is considered to be the best approach. Terminal drought has been the most important abiotic stress reducing the crop productivity very significantly along with deterioration of quality of the produce in groundnut as it predisposes *Aflatoxin* infection in the field.

3. Genomic resources

Although limited genomic resources were available in these legume crops until 2005, significant progress has been made in the development of large-scale genomic resources (Table 1). This has been possible due to financial support and coordinated efforts of several organizations such as CGIAR's Generation Challenge Programme, Bill & Melinda Gates Foundation, Indian Council of Agricultural Research (ICAR) and Department of Biotechnology (DBT) of Government of India, US National Science Foundation (NSF), The Peanut Foundation of the American Peanut Council etc. In brief, these efforts have led to the development of large-scale molecular markers, construction of comprehensive genetic maps, establishment of various marker-trait associations and initiation of molecular breeding in these three crops. Not only this, draft genome sequence has become available in pigeonpea (Varshney et al., 2012a) and similar efforts are underway in chickpea, and groundnut. Coordinated efforts and progress on development of genomic resources can be seen on websites of International Initiative on Pigeonpea Genomics (IIPG, http://www.icrisat.org/gt-bt/iipg/Home.html), International Chickpea Genetics and Genomics Consortium (http://www.icrisat.org/gt-bt/ICGGC/home.htm), and International Peanut Genome Initiative (http://www.peanutbioscience.com). An overview on various strategies to develop genomic resources by ICRISAT and its partners has been presented in Fig. 2.

3.1. Molecular markers and genotyping platforms

Although in recent years a range of marker systems including hybridization-based Diversity Array Technology (DArT) and sequence based markers such as single nucleotide polymorphisms (SNPs) have become available, simple sequence repeat (SSR) or microsatellite marker are still preferred marker system especially for genetics and breeding applications. SSRs exhibit polymorphism in terms of variation in the number of repeat units as revealed by amplification of unique sequences flanking these repeat units. They show co-dominant inheritance and

Table 1
Availability of genomic resources in chickpea, pigeonpea and groundnut.

Common name	Chickpea	Pigeonpea	Groundnut
Species	*Cicer arietinum*	*Cajanus cajan*	*Arachis hypogaea* (cultivated); *A. duranensis and A. ipaensis* (diploid progenitor species)
Ploidy	2n = 2× = 16	2n = 2× = 22	2n = 4× = 40 (cultivated), 2n = 2× = 20 (diploid)
Estimated genome size	740 Mbp	833.07 Mbp	2890 Mbp (cultivated), 1260 Mbp (diploid genome)
BAC libraries	10× Thudi et al. (2011)	11×[†] Bohra et al. (2011)	ca. 5.3×–Diploid (BB); ca. 7.4×–diploid (AA) Guimaraes et al. (2008)
BAC-end sequences	46,270 Thudi et al. (2011)	88,860 Bohra et al. (2011)	182,784 Yüksel and Paterson (2005) and 36,435 Wang et al. (2012)
EST	[¥]44,707 (See Choudhary et al. (2012))	[¥]24,176 Kudapa et al. (2012)	[¥]253,274 (See Pandey et al. (2012a))
SSRs	~2000 (Gujaria et al. (2011), Nayak et al. (2010), Thudi et al. (2011))	4000 (Bohra et al. (2011), Dutta et al. (2011), Raju et al. (2010))	>6000 (See Pandey et al. (2012a))
TILLING population	5000 mutant M_2 lines (Unpublished data)	ca.5000 mutant lines (Varshney et al. (2010c))	3400 mutant M_2 lines Knoll et al. (2011)
DArT clones	5397 Thudi et al. (2011)	15,360 Varshney et al. (2010b, Yang et al. (2006, 2011)	ca. 15,000 Kilian (2008), Varshney et al. (2010a)
454/FLX reads	435,018 Hiremath et al. (2011), 1,931,224 Garg et al. (2011b), 969,132 Jhanwar et al. (2012)	494,353 Dubey et al. (2011)	1000,000 Guimarães et al. (2011)
Transcriptome assembly	103,215 contigs Hiremath et al. (2011), 34,760 contigs Garg et al. (2011b), 37,265 Jhanwar et al. (2012)	48,476 contigs Dubey et al. (2011), 21,434 contigs Kudapa et al. (2012)	–
SNPs	[†]9000	[$]10,000	>2000 SNPs, 768-SNP (see Pandey et al. (2012a))
Mapping populations	30 Upadhyaya et al. (2011), Varshney et al. (2007)	25 Bohra et al. (2011, 2012), Gnanesh et al. (2011), Raju et al. (2010)	Diploid (AA) – 5, Diploid (BB) – 1, Tetraploid –39 (see Pandey et al., 2012a)
Genetic maps	24 (15 inter-specific & 9 intra-specific) Millan et al. (2010), Upadhyaya et al. (2011), Varshney et al. (2007)	Reference genetic map, six intra-specific maps, one consensus map and DArT based maternal and paternal maps Argout et al. (2011, 2011, 2012), Yang et al. (2011)	Diploid (AA)-3, Diploid (BB)-2, Tetraploid-13 maps and one reference consensus map (see Pandey et al. (2012a)
Physical maps	[€]BAC/BIBAC-based, [¤]BAC-based	Not available	Yüksel and Paterson (2005)
Complete genome sequence	In progress	[*]Available Varshney et al. (2012a)	In progress

[†] International Crops Research Institute for the Semi-Arid Tropics, India & University of California (UC)-Davis, USA; EMBRAPA: Brazilian Agricultural Research Organization, Brazil;University of Georgia, USA; University of California, USA; [¤]ICRISAT, India;Osmania University (OU), India; Banaras Hindu University (BHU), India and ICRISAT, India; ICRISAT, India; University of Birmingham, UK; Diversity Arrays Technology (DArT) Pty Limited, Australia; ICRISAT, India; CIRAD, France; Catholic University, Brazil; Diversity Arrays Technology (DArT) Pty Limited, Australia & ICRISAT, [$]India; India & NCGR, USA; EMBRAPA & University of Brasilia, Brazil; Kazusa DNA Research Institute, Japan; UniversidadeEstadualPaulista (UNESP), Brazil; USDA-ARS, USA;EMBRAPA and University of Brasilia, Brazil; [s]in public domain; Frankfurt University; [€]Texas A&M University, USA, Jilin Agricultural University, China, The Hebrew University of Jerusalem, Israel, The Volcani Center, Israel, USDA-ARS and Department of Crop and Soil Sciences, Washington State University, USA, Chinese Academy of Sciences, China; [*]International Crops Research Institute for Semi-Arid Tropics (ICRISAT), India; CGIAR Generation Challenge Programme (GCP), c/o CIMMYT, Mexico DF, Mexico; Beijing Genomics Institute (BGI)-Shenzhen, Shenzhen, China; University of Georgia, Athens, Georgia, USA; National Center for Genome Resources (NCGR), Santa Fe, New Mexico, USA; University of North Carolina, Charlotte, North Carolina, USA; National University of Ireland Galway (NUIG), Botany and Plant Science, Galway, Ireland; University of California, Davis, California, USA; Monsanto Company, Creve Coeur, Missouri, USA; Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, USA; Department of Biology, University of Copenhagen, Denmark; BGI-Americas, Cambridge, Massachusetts, USA; [¥]Public domain (www.ncbi.nlm.nih.gov, 26th June, 2012). [¤]http://www.icrisat.org/gt-bt/ICGGC/GenomeSequencing.htm.

therefore are suitable for genotyping segregating populations (including F_2). Multi-allelic nature of the markers enables them to detect a large number of allelic variants in the germplasm collection (Gupta and Varshney, 2000).

Until recently, development of SSR markers was largely based on screening of SSR-enriched or size-selected DNA libraries, however mining of ESTs (expressed sequence tags) or BAC-end sequences (BESs) have become popular approaches for development of SSR markers. SSR markers developed from ESTs or cDNA sequences are referred to as 'genic SSR' or 'genic markers' (Varshney et al., 2010a). By using a range of different approaches mentioned above, 3000–6000 SSR markers have become available in the target SAT legume crops. For instance, in the case of chickpea, ca. 2000 SSR markers have been developed from genomic DNA libraries (for references see Varshney et al., 2007; Nayak et al., 2010; Gaur et al., 2011), ESTs (Varshney et al., 2009b), 454/FLX transcript reads (Hiremath et al., 2011; Garg et al., 2011a, 2011b,) and BESs (Thudi et al., 2011). Similarly, another set of 487 novel functional markers including 125 EST-SSRs, 151 intron targeted primers (ITPs), 109 expressed sequence tag polymorphisms (ESTPs), and 102 SNP markers has been developed at National Institute of Plant Genome Research (NIPGR) (Choudhary et al., 2012). In the case of pigeonpea, a large number of SSR markers have been developed from BESs and 454/FLX sequences. After mining 88,860 BESs, a set of 3072 SSR markers was developed (Bohra et al., 2011). In addition, 3583 SSRs were identified from ESTs (Raju et al., 2010) and 454/FLX sequences (Dubey et al., 2011; Dutta et al., 2011). Furthermore, by scanning the draft genome sequence of pigeonpea (see later), 309,052 SSRs have been identified (Varshney et al., 2012a) and they can be used to enrich genetic maps with more number of molecular markers and also to tag QTL/genes for important traits. In the case of groundnut, >6000 SSRs have become available by the international groundnut community (see Feng et al., 2012; Pandey et al., 2012a; Wang et al., 2012). After screening ca. 4500 SSR markers on parental lines of several mapping populations, 199 highly informative SSR markers with polymorphism information content (PIC) value of >0.50 were identified (Pandey et al., 2012b). Similarly, more recently a set of 66 highly informative SSRs (>0.5 PIC) with long TC repeats has been reported (Macedo et al., 2012).

DArT marker system is another marker resource mainly used for diversity studies, for saturating linkage maps and also for identifying introgressions from other species. ICRISAT in collaboration with DArT Pty Ltd, Australia has developed DArT arrays with 15,360 features for chickpea, groundnut and pigeonpea crops (see Varshney et al., 2010a). Screening of elite germplasm of the SAT legume crops with these DArT arrays, however, showed very little polymorphism (Thudi et al., 2011). Interestingly, DArT markers have been found very useful for monitoring the genome introgression in the cultivated species of pigeonpea from the wild species (Mallikarjuna et al., 2011).

Because of higher abundance and amenability to high-throughput, SNP markers are becoming popular marker system in several crop

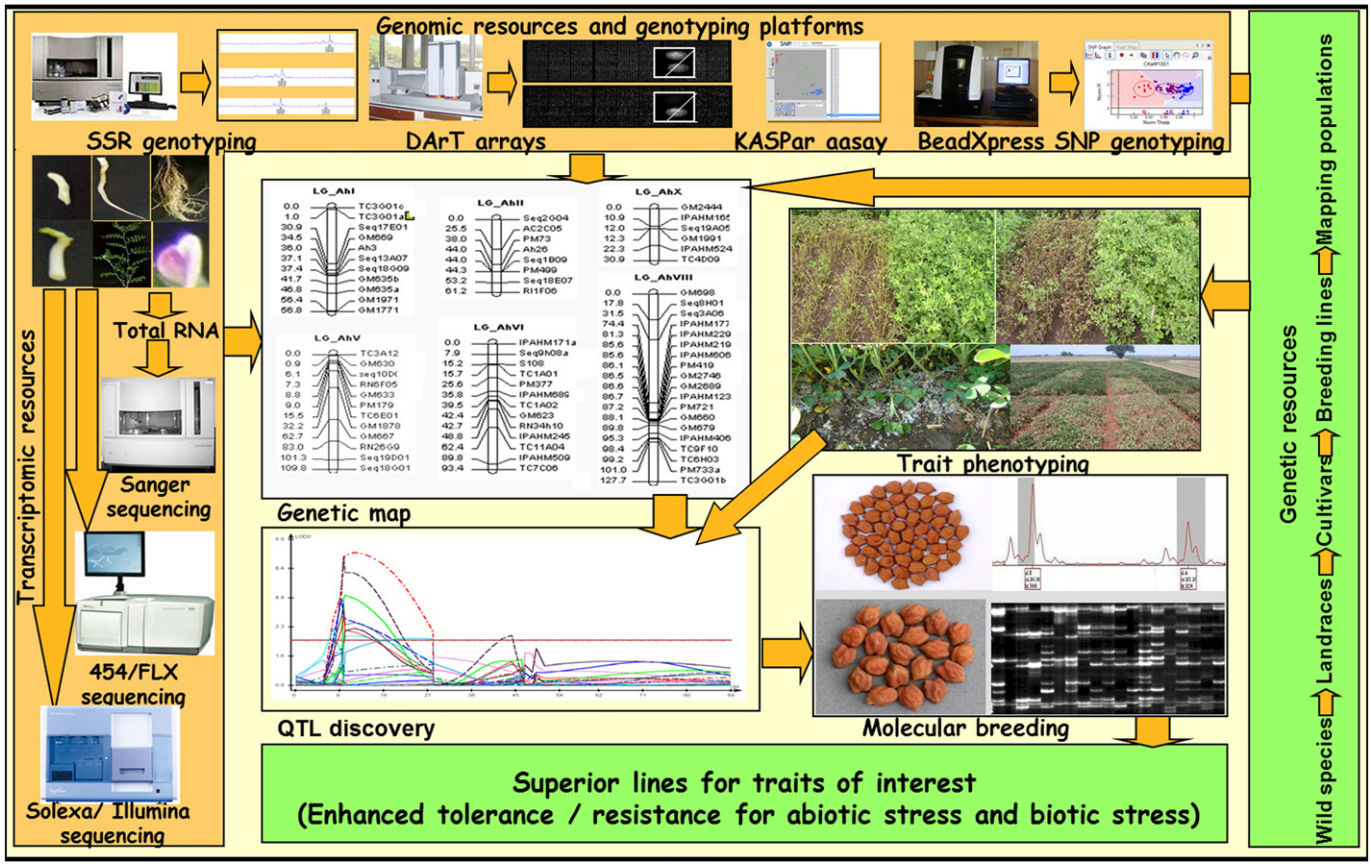

Fig. 2. An overview on coordinated and collaborative efforts on development and application of genomic resources for crop improvement of SAT legumes. This figure shows coordinated and collaborative efforts of ICRISAT and its partners, using high-throughput sequencing and genotyping and modern breeding approaches, to develop and apply genomic resources in crop improvement programs of three legume crops namely chickpea, pigeonpea and groundnut. For instance, genomic and transcriptomic resources such as molecular markers, ESTs, genes, transcriptome assemblies and genome sequences have been developed using Sanger and next generation sequencing technology platforms (*orange colored panel on left*), cost-effective, low-, medium- and high-throughput genotyping platforms have been developed using capillary electrophoresis (SSR genotyping), iScan or BeadXpress systems and KASPar assays (SNP genotyping) and DArT genotyping platforms (*orange colored panel on top*). A range of genetic resources (*green colored panel on right*) are used with genomic resources using different genotyping platforms to develop genetic map (*center top left*) as well as for multi-location phenotyping for traits of interest to breeders (*center top right*). Genotyping data and phenotyping data are used to identify the QTLs/markers associated with target traits using QTL mapping or association mapping approaches (*center down left*). Genetic and QTL information subsequently are used in breeding program using modern breeding programs such as marker-assisted back crossing (MABC) (*center down right*). By using these modern breeding approaches, superior lines for target traits with enhanced crop productivity are generated (*green colored panel on bottom*).

species. In the case of SAT legume crops also, SNPs were identified using a variety of approaches. For instances, Sanger ESTs generated from a range of genotypes were used to identify SNPs in chickpea (Varshney et al., 2009b), pigeonpea (Raju et al., 2010) and groundnut (P. Ozias-Akins, University of Georgia, personal communication). Allele-specific sequencing of candidate genes has also been used for identifying the SNPs, though at a lower frequency, in chickpea (Gujaria et al., 2011). Tentative orthologous genes (TOGs) developed based on sequence similarity from ESTs/ genes of soybean, *Medicago truncatula* (*Medicago*) and *Lotus japonicus* (*Lotus*) species were used for allele-specific sequencing of parental genotypes of mapping populations of chickpea (Hiremath et al., 2012) and pigeonpea (Kassa et al., 2012; Saxena et al., 2012). Next generation sequencing (NGS) technologies including 454/FLX and Illumina/Solexa have been used recently to identify large scale SNPs. For instance based on alignment of ~37 million Illumina/Solexa tags generated from ICC 4958 and ICC 1882 parental genotypes of intra-specific mapping population with a transcriptome assembly of chickpea (see later), 26,082 potential SNPs have been identified (Hiremath et al., 2011). Similarly in the case of pigeonpea, alignment of ~160 million reads generated from 10 parental genotypes against a transcriptome assembly (CcTA version 1.0) has identified a total of 12,141 SNPs in pigeonpea (Dubey et al., 2011). Furthermore, comparison of transcript reads from 12 different pigeonpea genotypes against the genome assembly has resulted in identification of 28,104 novel SNPs (Varshney et al., 2012a). In the case of groundnut, comparison of >350 Mb 454/FLX-sequencing based transcriptome data from 17 tetraploid genotypes against a reference transcriptome of 'Tifrunner' resulted in identification of a total of 8486 SNPs with moderately stringent filtering (http://nespal.org/oziasakinslab/projects/plant-biotechnology-peanut-grasses/peanut-snp-discovery/).

Once SNPs are identified, development of an appropriate SNP genotyping platform is very critical to make the SNP genotyping cost-effective. In the SAT legume crops, a range of SNP genotyping platforms have become available. For instance, University of California-Davis, USA in collaboration with some partner institutes has developed Illumina GoldenGate assays for genotyping 768 SNPs in chickpea, pigeonpea and diploid *Arachis* species. Similarly, The University of Georgia, USA has also developed an Illumina GoldenGate SNP array comprising of 1536-SNPs with high confidence for *Arachis* species. These assays are most suitable when relatively large number of SNPs (>500) need to be genotyped with a large number of samples. However, in the case of certain molecular breeding applications which generally require less number of markers (<400), GoldenGate based SNP arrays are not very cost-effective (see Hiremath et al., 2012). Therefore, VeraCode assays have been developed for 96-plex and 48-plex SNP sets for chickpea and pigeonpea, respectively, which can be used on Illumina's BeadXpress system (RK Varshney et al., unpublished results).

KASPar assay is another important SNP genotyping technology from KBiosciences (www.kbioscience.co.uk) that is very flexible and can be used to genotype any number of samples with any number of SNPs. Very recently, KASPar assays have been developed for 2005 SNPs in chickpea (Hiremath et al., 2012), 1616 SNPs in pigeonpea (Saxena et al., 2012) and 96 SNPs in groundnut.

3.2. Transcriptome and genome assemblies

Transcriptome sequencing is the first step to access the gene contents of a species. In absence of low-cost NGS technologies for sequencing the genome, transcriptome sequencing using Sanger sequencing technology has been a popular approach to access the gene contents in a range of crop species. In the SAT legumes also, Sanger sequencing was used initially to access the transcriptome. For instance, 20,162 ESTs were generated from drought- and salinity-challenged cDNA libraries in the case of chickpea (Varshney et al., 2009b). Similarly, 9888 ESTs were generated from FW and SMD challenged cDNA libraries in the case of pigeonpea (Raju et al., 2010). By analyzing these ESTs together with the then available ESTs in public domain, 9569 and 5085 unigenes were defined for chickpea and pigeonpea, respectively.

To advance development of transcriptomic resources, 454/FLX sequencing was undertaken on normalized and pooled RNA samples collected from >20 different developmental stage tissues of the plant. As a result, 435,018 transcript reads were generated (Hiremath et al., 2011). Analysis of these transcript reads with Sanger ESTs generated by Varshney et al. (2009b) as well as those available in public domain provided comprehensive transcript assembly of chickpea (Ca TA) with 103,215 tentative unique sequences (TUSs) (Hiremath et al., 2011). In another study, Garg et al. (2011b) developed hybrid assembly with 34,760 tentative consensus (TCs). More recently, transcriptome of a wild chickpea, *C. reticulatum* (genotype PI489777) has also been sequenced using GS-FLX Roche 454 NGS technology (Jhanwar et al., 2012). In this study, both *de novo* assembly and reference-based assembly approaches were explored to develop an optimized assembly and 37,265 *C. reticulatum* tentative consensus transcripts were (CrTC) was reported (http://www.nipgr.res.in/ctdb.html).

In the case of pigeonpea, 494,353 transcript reads were generated from normalized and pooled RNA samples collected from >20 different tissues of 'Pusa Ageti' variety (Dubey et al., 2011). Cluster analysis of these transcript reads with the then available ESTs in the public domain resulted in development of the first generation of transcriptome assembly (Cc TA v1) with 127,754 TUSs (Dubey et al., 2011). In another study, 1.696 million 454/FLX transcript reads were generated from 'Asha' variety (Dutta et al., 2011). With an objective to refine transcriptome assembly of pigeonpea, 494,353 454/FLX transcript reads generated by Dubey et al. (2011), 1.696 million 454/FLX transcript reads generated by Dutta et al. (2011) and 128.9 million Illumina reads generated from 12 genotypes were analyzed together with 18,353 Sanger ESTs with better algorithms. As a result, an improved second version of the transcriptome assembly (referred as Cc TA v2) comprising of 21,434 contigs has been developed (Kudapa et al., 2012).

With respect to groundnut, at present, 253,274 Sanger ESTs are available in public domain (http://www.ncbi.nlm.nih.gov/sites/gquery, 26th June, 2012). Most recently The University of Georgia (UGA), USA has developed about 1 million reads representing >350 Mb of transcript sequences, from 17 genotypes using 454/FLX sequencing technology. Analysis of these reads along with publicly available ESTs resulted in a consensus transcriptome assembly comprising 211,244 contigs (P. Ozias-Akins, University of Georgia, personal communication).

The ultimate approach of cataloging all possible genes in a given species is to sequence full genome as it offers three fold advantages: a) enables understanding the genome structure and dynamics in better way, b) enables identification of genes and functional elements responsible for expression of phenotype, and c) provides the genomic tools and platforms for gene mapping, gene isolation and molecular breeding. Further, information gained from sequenced genomes can be very useful for molecular breeding programs in order to develop improved varieties/hybrids. Novel breeding approaches such as marker-assisted recurrent selection (MARS) and genomic selection (GS) (see later) will likely to be feasible in breeding and may be facilitated by genotyping-by-sequencing (GBS) that can be done with a reference genome. The genome sequence also facilitates genome wide association studies (GWAS) leading to the identification of candidate genes or genomic regions for crop improvement.

While genome sequencing was prohibitive earlier especially in less-studied crops, recently available low-cost NGS technologies have allowed initiating genome sequencing in the SAT legume crops. Among three legume crops mentioned in this article, pigeonpea was the first one to have the genome sequence available. International Initiative on Pigeonpea Genomics (IIPG) delivered the draft genome sequence for the pigeonpea by using the Illumina sequence technology (Varshney et al., 2012a). It is the first "orphan crop", the first "non-industrial crop" and the second food legume crop (after soybean) where genome sequence information has been made available. Next generation sequencing (Illumina) was used to generate 237.2 Gbp of sequence. Along with Sanger-based BESs and genetic map sequences were assembled into scaffolds representing ~73% (605.78 Mb) of the 833 Mbp pigeonpea genome. Detailed analysis has resulted in the identification of 48,680 pigeonpea genes. A few hundreds of these genes were found unique to the crop in terms of drought tolerance, an important trait that can be transferred to other similar legume crops like soybean, chickpea or common bean. Comparative analysis revealed that the number of predicted genes in the pigeonpea genome is higher to other sequenced plant genomes, such as those from cucumber (26,682; Huang et al., 2009), cacao (28,798; Argout et al., 2011), grapevine (29,585; Jaillon et al., 2007) and lotus (38,483), but it is comparable to poplar (45,555; Tuskan et al., 2006), soybean (46,430; Schmutz et al., 2010) and medicago (47,529; Young et al., 2011). However, the average number of exons per gene in the pigeonpea (3.59) is less than for soybean (5.80), while average exon (267.39 bp) and intron (536.89 bp) lengths are longer than those for soybean (216.13 bp exons and 419.43 bp introns).

In the case of chickpea, there are two main market classes namely *kabuli* and *desi*. *Desi* chickpeas have colored seeds, a rough coat and are cultivated mostly in the Indian subcontinent, Ethiopia, Mexico and Iran. On the other side, *kabuli* chickpeas possess light colored, smoother seed coat and larger seeds, and is mainly grown in Southern Europe, Northern Africa, Afghanistan, Pakistan, Chile and Indian subcontinent. In general, *kabuli* chickpeas fetch higher price than the *desi* chickpeas. Efforts are underway to sequence the genomes of both types of chickpea. For instance, International Chickpea Genome Sequence Consortium has initiated genome sequencing of CDC Frontier, a *kabuli* variety (http://www.icrisat.org/gt-bt/ICGGC/GenomeSequencing.htm). On the other hand ICC 4958, a *desi* landrace has been targeted for sequencing at NIPGR, New Delhi (http://www.nipgr.res.in/home/home.php).

In the case of groundnut, the Peanut Genome Consortium (PGC), an extension of the International Peanut Genome Initiative (IPGI), has initiated decoding the groundnut genome sequence. Owing to the large genome size (2800 Mb) and polyploid nature, the genetic analysis of groundnut has been challenging. The important goal of PGC is enabling breeders, geneticists, molecular biologists and other researchers to enhance productivity of the cultivated peanut. The peanut genome project (PGP) is different from other genome projects as it is engaged in: (a) development of high density consensus genetic maps that enable markers to anchor chromosomes of wild and cultivated groundnuts, (b) establishing genome wide associations and identifying trait specific genomic segments, (c) characterization of genome wide diversity through identification of genotypic

Table 2
List of markers associated with major QTL/genes for different traits in chickpea.

Traits studied	QTL/genes	Markers linked	PVE (%)	References
Agronomic & yield				
Plant growth habit	*Prostrate*	TA34–TA142	95.2	Aryamanesh et al. (2010)
	Hg/hg	$OPB17_{789}$–$OPAI09_{1651}$	–	Cobos et al. (2009)
Days to flowering	Q3-1	TA6-NCPGR12	22.0	Rehman et al. (2011)
	QTL	TA142–TA64	45.0	Aryamanesh et al (2010)
	QTL	TS29–TA76S	45.2	Aryamanesh et al. (2010)
	DF3	TA142–$OPB17_{789}$	26.0	Cobos et al. (2009)
Days to maturity	Q3-1	TA6–NCPGR12	33.0	Rehman et al. (2011)
Seed coat thickness	QTL_{Tt}	*B/b*–TA61	20.0	Cobos et al. (2009)
Seed size	QTL_{SW1}	GAA47–STMS11	32.0	Cobos et al. (2009)
Seed/pod	*Spp*	UBC465–TA2x	–	Radhika et al. (2007)
Double podding	*Sfl*	NCPGR33–UBC249z	–	Radhika et al. (2007)
Harvest index	Q1-1	H5A08–TA8	13.0	Rehman et al. (2011)
	Q3-1	TA6–NCPGR12	25.0	Rehman et al. (2011)
Abiotic stress				
Root traits	QTL	ICCM0249, TAA170, GA24, STMS11	30.0	Varshney et al. (Unpublished)
Drought tolerance score	Q3-1	TA6–NCPGR12	27.0	Rehman et al. (2011)
Canopy temperature differential	Q1-1	H5A08–TA8	15.0	Rehman et al. (2011)
Biotic stress				
Resistance to *Ascochyta* blight	QTL	$OPAI09_{1276}$–$OPAC04_{1200}$	23.7	Millan et al. (2003), Cobos et al. (2005)
	Ar19	UBC733B–UBC181A	42.5	Rakshit et al. (2003)
	QTL*ar2b*	TA130–TR20	–	Udupa and Baum (2003)
	QTL*AR3*	TR58–TS82	22.6	Iruela et al. (2007)
	QTL*ar1*	GAA47	34.0	Iruela et al. (2006)
	QTL*ar2*	TA146–TA72	21.0	Iruela et al. (2006)
	QTL	TA2–TA146	29.0	Anbessa et al. (2009)
	QTL	STMS11–TAA170	26.0	Aryamanesh et al. (2010)
Resistance to *Fusarium* wilt	*Foc0*	$OPJ20_{600}$–TR59	73.0	Cobos et al. (2005)
	Foc1	TA110–H3A12	–	Gowda et al. (2009)
	Foc2	H3A12–TA96	–	Gowda et al. (2009)
	Foc3	TA96–TA194; TA194–H1B06y	–	Sharma et al. (2004), Gowda et al. (2009)
	Foc4	TA96–CS27; TA96–TR19	–	Sharma et al. (2004, 2005)
	Foc5	TA59–TA96	46.5	Cobos et al. (2009)
Resistance to *Botrytis* gray mold	QTL	TA118–TA159	48.0	Anuradha et al. (2011)
Resistance to rust	*Uca1/uca1*	TA18–TA180	73.7	Madrid et al. (2008)

haplotypes among accessions of the United States Department of Agriculture (USDA), Chinese and ICRISAT germplasm collections and (e) development of high resolution assembly of BACs from a reference population of recombinant inbred lines (RILs). Further PGC aims at: (a) development of a high quality chromosome scale draft of a cultivated species as the reference genome sequence, plus high density maps of both progenitor and synthetic amphidiploid genomes, (b) high-throughput transcriptome characterization of the reference tetraploid cultivar, (c) characterization of gene space in amphidiploid and diploid (progenitor species) germplasm, phenotypic association with mapped genetic markers, and interactive bioinformatics resources for data curation and application in a breeder's toolbox to enable molecular breeding approaches for enhancing peanut yielding ability, optimizing resistance to diseases and insects, tolerance to environmental stresses, and improved quality traits.

3.3. Genetic maps and trait mapping

Availability of large-scale genomic resources, as mentioned above, has led to development of either the first generation or comprehensive genetic maps in the SAT legume crops. Analysis of these genetic maps together with phenotyping of the respective segregating populations for the traits of interest to the breeders has facilitated identification of molecular markers associated with several agronomically important traits.

An inter-specific mapping population derived from a cross ICC 4958 × PI 489777 has been used as a reference mapping population in chickpea and majority of molecular markers have been used to integrate in the genetic map of this population (see Upadhyaya et al., 2011). By using the SSR markers from SSR-enriched libraries and BESs, together with DArT markers and genic molecular markers (GMMs) (Gujaria et al., 2011), an integrated genetic map with 1291 marker loci has been developed (Thudi et al., 2011). In parallel, an advanced gene-rich map of chickpea comprising of 406 loci (including 177 gene-based markers) spanning 1497.7 cM genetic distance has been developed by Choudhary et al. (2011) using the same reference population. By developing large-scale KASPar assays for SNP genotyping, Hiremath et al. (2012) has developed a second-generation genetic map comprising 1328 marker loci including 625 novel CKAMs (Chickpea KASpar Assay Markers), 314 TOG-SNPs and 389 published marker loci with an average inter-marker distance of 0.59 cM. Several intra-specific mapping populations have also been used to identify the markers associated with traits like resistance to FW (Sharma et al., 2004, 2005), AB (Anbessa et al., 2009; Iruela et al., 2007), rust (Madrid et al., 2008), BGM (Anuradha et al., 2011), tolerance to salinity (Vadez et al., 2012) along with seed traits (Cobos et al., 2009) and grain yield (Rehman et al., 2011). Several of these studies have been summarized in earlier reviews (Varshney et al., 2007) and were updated recently by Upadhyaya et al. (2011) (Table 2).

In the case of pigeonpea, only a few SSR markers were available in public domain till 2010. Lack of sufficient DNA markers coupled with less genetic variability were the major obstacle in development of genetic maps in pigeonpea. However, recently substantial advancements have been made in term of generation of large scale SSR and SNP markers

Table 3
List of markers associated with major QTL/genes for different traits in pigeonpea.

Traits studied	QTL/genes	Markers linked	PVE (%)	References
Agronomic				
Fertility restoration	QTL-RF-1	CcM1522–CcM1821	14.85	Saxena et al. (2011), Bohra et al. (2012)
	QTL-RF-2	CcM0047–CcM2332	16.27	Saxena et al. (2011), Bohra et al. (2012)
	QTL-RF-3	CcM2542–CcM1277	20.89	Saxena et al. (2011), Bohra et al. (2012)
	QTL-RF-4	CcM0374–CcM1506	24.17	Saxena et al. (2011), Bohra et al. (2012)
Biotic stress				
Resistance to sterility mosaic disease	qSMD3	CcM2149–CcM0468	12.32	Gnanesh et al. (2011)
	qSMD4	CcM1825–CcM1895	24.72	Gnanesh et al. (2011)
	qSMD5	CcM0970–CcM2485	15.93	Gnanesh et al. (2011)
	qSMD6	CcM0416–CcM2337	10.58	Gnanesh et al. (2011)

using second and third generation sequencing platforms. Availability of more than 3000 SSR markers facilitated development of inter- as well as intra-specific genetic maps using several F_2 mapping populations. The first reference genetic map was developed using an inter-specific population *i.e.* ICP 28 (*C. cajan*) × ICPW 94 (*C. scarabaeoides*) comprising 79 F_2 individuals. This F_2 genetic map consists of a total of 239 SSR markers spanning a map distance of 930.9 cM over 11 linkage groups (LGs) (Bohra et al., 2011). Furthermore, DArT genotyping of the same mapping population also resulted in development of DArT based paternal- (122 unique markers mapped at a distance of 270 cM) and maternal-specific (172 unique loci mapped at 451.6 cM) genetic maps (Yang et al., 2011). Furthermore, after developing KASPar assays for pigeonpea, a dense genetic map comprising 875 SNP loci with an average inter-marker distance of 1.11 cM has been developed on an extended F_2 population (Saxena et al., 2012,). Apart from these inter-specific genetic maps, some more SSR-based genetic maps with low to moderate marker density were made available for cultivated pigeonpea. These intra-specific genetic maps have been developed based on six F_2 populations *viz.* TTB 7 × ICP 7035 (84 loci, 466.97 cM), ICP 8863 × ICPL 20097 (120 loci, 534.89 cM), ICPB 2049 × ICPL 99050 (59 loci, 586.02 cM), ICPR 2043 × ICPR 3467 (140 loci, 881.57 cM), ICPA 2039 × ICPR 2447 (78 loci, 570.53 cM), ICPA 2043 × ICPR 2671 (111 loci, 677.97 cM). Based on these six populations, a consensus map comprising of 339 SSR loci with 1059 cM genetic distance has been developed (Bohra et al., 2012). This represents the first instance of merging multiple genetic maps in pigeonpea.

For trait mapping in pigeonpea, some preliminary mapping efforts have been initiated with F_2 mapping populations. For instance, bulked segregant analysis (BSA) approach was used for mapping of FW resistance with RAPD markers (Kotresh et al., 2006), mapping of SMD resistance with AFLP marker system (Ganapathy et al., 2009), and ideal plant type with RAPD markers (Dhanasekar et al., 2010). Availability of SSR based genetic maps for F_2 populations coupled with extensive phenotyping data facilitated identification of QTLs/marker(s) for various traits of economic importance. For SMD resistance, a total of four major QTLs (qSMD3-qSMD6) were recovered from population TTB 7 × ICP 7035 and one of the underlying QTL 'qSMD4' explained a phenotypic variance (PV) up to 24%. Similarly some minor QTLs, qSMD 1 and qSMD2 (governing PV < 10%) were also discovered from another F_2 population *viz.* ICP 8863 × ICPL 20097 for SMD resistance (Gnanesh et al., 2011) (Table 3). SSR markers associated with SMD resistance offer rapid recovery of SMD resistant genotypes from large segregating populations and would open tremendous opportunities for practicing markers-assisted introgression of resistant allele/QTL(s) into susceptible pigeonpea cultivars. Similarly, to map fertility restoration (*Rf*) genes, genotyping and phenotyping data from three F_2 populations namely ICPA 2039 × ICPR 2447, ICPA 2043 × ICPR 2671 and, ICPA 2043 × ICPR 3467 were also subjected to QTL analyses (Bohra et al., 2012). All four QTLs (QTL-RF-1 to QTL-RF-4) identified from these three populations can be called 'major QTLs' that contributed 14.85% to 24.17% PV. Identification of such linked SSR markers with fertility restoration will supplement hybrid breeding through quick and precise discrimination between B- and R- lines which is otherwise time consuming and labor intensive. In addition, marker-assisted introgression of gene(s)/QTL(s) imparting fertility into the elite genetic background would allow easy conversion of a pure line to a potential restorer in a time saving manner. For facilitating hybrid breeding and adoption, diagnostic SSR markers have also been developed for purity assessment of two hybrids (Bohra et al., 2011; Saxena et al., 2010).

In the case of groundnut, the first SSR-based genetic linkage map for cultivated groundnut was developed on TAG 24 × ICGV 86031 RIL population (RIL-1) (Varshney et al., 2009c). It is now considered as a reference map for cultivated groundnut and has been saturated up to 191 SSR loci (Ravi et al., 2011). Four more genetic maps based on RIL populations segregating for drought tolerance related traits (RIL-2: ICGS 76 × CSMG 84-1 with 119 SSR loci and RIL-3: ICGS 44 × ICGS 76 with 82 SSR loci) and foliar diseases (RIL-4: TAG 24 × GPBD 4 with 188 SSR loci and RIL-5: TG 26 × GPBD 4 with 181 SSR loci) were developed (Gautami et al., 2012a; Sujay et al., 2012). In order to place maximum markers on a single map, one consensus map each for drought tolerance related traits (RILs 1–3; 2840.8 cM) with 293 SSR loci (Gautami et al., 2012a) and foliar disease resistance (RILs 4–5; 1152.9 cM) with 225 SSR loci (Sujay et al., 2012) was developed. Among other genetic maps developed by the groundnut community, an integrated genetic map with 175 marker loci based on three RIL populations (Hong et al., 2010) and another map with 325 marker loci based on two RIL populations (Qin et al., 2012) are noteworthy. More recently, a genetic linkage map consisting of 318 loci onto 21 LGs and covering a total of 1674.4 cM has been developed using an F_2 population (Tifrunner × GT-C20) (Wang et al., 2012). Furthermore, collaborative efforts of several international partners have resulted in the construction of consensus map with 897 SSR marker loci using genotyping data of 11 mapping populations. This map possesses 20 LGs (a01–a10 and b01–b10) spanning a map distance of 3863.6 cM with an average map density of 4.4 cM. This map was divided into 20 cM long 203 BINs and these BINs carry 1 (a10_02, a10_08 and a10_09) to 20 (a10_04) loci with an average of 4 marker loci per BIN (Gautami et al., 2012b). In addition to this, a dense genetic map using F_2 mapping population (Nakateyutaka × YI-0311) with 1,114 loci distributed on 21 LGs covering a total 2,166.4 cM map distance has been developed (Shirasawa et al., 2012). These dense genetic/consensus maps are very useful resource while selecting highly informative and uniformly distributed markers for background selection, construction of new genetic maps and diversity analysis.

In terms of trait mapping in groundnut also, significant efforts have been made. Comprehensive QTL analysis of genotyping data and phenotyping data for drought tolerance traits (e.g. transpiration, transpiration efficiency, biomass, specific leaf area, pod weight, total dry matter, SPAD chlorophyll meter reading, total dry weight, shoot dry weight and harvest index) on three mapping populations detected 153 main effect and 25 epistatic QTLs for drought tolerance related traits (Gautami et al., 2012a; Ravi et al., 2011; Varshney et al.,

2009c) (Table 4). Similarly, QTL analysis based on extensive phenotyping data generated on two RIL populations (TAG 24 × GPBD 4 and TG 26 × GPBD 4) for rust and LLS resistance respectively for 7–8 seasons (2004–2010) at University of Agricultural Sciences-Dharwad (India) and genotyping data (207 marker loci each) resulted in identification of a total of 28 QTLs for late leaf spot (LLS; 10.1 to 67.8% PV) and 13 QTLs for rust (2.5 to 82.9% PV) (Khedikar et al., 2010; Sujay et al., 2012). More significantly, a major QTL each for LLS (62.34% PV) and rust (82.96% PV) resistance were identified. The associated markers for rust and LLS were validated in alternate mapping populations and germplasm set. In addition, QTL analysis using phenotyping data on important nutritional and oil quality generated on TG 26 × GPBD 4 resulted in detection of a total of seven QTLs for protein content (2.5–9.8%), eight QTLs for oil content (1.5–10.2%) and six common QTLs for oleic and linoleic acid (3.3–9.7%) (Sarvamangala et al., 2011). Similarly, two QTLs were mapped for tomato spotted wilt virus (TSWV) using two RIL populations (Qin et al., 2012).

4. Genomics-assisted breeding (GAB)

Genomics-assisted breeding refers to integration and use of genomic tools in breeding practices for developing superior lines with enhanced biotic or abiotic stress tolerance and improved yield. The objective of GAB is to establish and utilize relationship between genotype and phenotype for crop improvement. GAB includes a range of approaches including genomics, transcriptomics and proteomics to identify the molecular markers associated with traits of interest to the breeders that help prediction of phenotype from the genotype to assist breeding (Fig.3). With the advent of next-generation sequencing (NGS) technologies (Varshney et al., 2009c) and high-throughput genotyping technologies (Varshney, 2011), it has been possible to use the genome-wide marker profile/allele data for prediction of phenotype of progenies for selection to the new cycle in breeding programs. To breed for the traits controlled by major QTL/ genes (e.g. disease resistance), marker-assisted backcrossing (MABC) approach has been considered a good approach (Ribaut and Hoisington, 1998). However, majority of traits targeted by breeders e.g. drought tolerance or durable resistance to multiple races of pathogens are controlled by several QTLs or genes. For instance, in the case of groundnut, 153 main effect and 25 epistatic interaction QTLs with small phenotypic variation were identified that confer drought tolerance (Gautami et al., 2012a; Ravi et al., 2011). In such cases, retaining desirable gene combinations or pyramiding of several QTLs through MABC approach is a challenging task (Peleman and Voort, 2003). Hence, marker-assisted recurrent selection (MARS) has been proposed as better approach (Ribaut and Ragot, 2007). Furthermore, genome-wide selection or genomic selection (GS) approach, due to possibility of generating genome-wide marker data through use of high-throughput genotyping or NGS approaches, is emerging as a powerful approach for identifying desirable progenies for making the crosses (Bernardo and Yu, 2007; Jannink et al., 2010). In some cases, superior alleles for a given trait e.g. disease resistance are identified and transferred from the wild species to a leading variety/cultivar. In such cases, advanced back-cross QTL (AB-QTL) approach has been proposed by Tanksley and Nelson (1996) for simultaneous discovery and transfer of superior alleles from wild species to develop improved lines. These approaches are being used in the SAT legume crops for improving a range of traits. Some of these examples have been listed below.

4.1. Marker-assisted backcrossing (MABC)

MABC involves introgression of specific trait(s) from a donor parent into the genetic background of a recurrent parent (generally leading variety) using molecular markers (Hospital, 2005). The product of MABC is a line/cultivar containing only the major gene/QTL from the donor parent, while retaining the whole genome of the recurrent parent (see Gupta et al., 2010). MABC approach generally involves transfer of a limited number of trait loci including transgenes from one genetic background (donor genotype) to the other genetic background (elite variety). This approach can also be used to generate near-isogenic lines (NILs) or chromosome segment substitution lines (CSSLs) for genomics research, which are populations that are often used for genetic analysis of genes/QTLs and alien gene introgressions (Lorieux, 2005; Varshney et al., 2010b). Gene pyramiding is an important application of MABC in which a few different genes for the same trait (e.g. resistance to different races of a pathogen) or for different traits are brought together in one genetic background using molecular markers.

As mentioned earlier, availability of molecular markers associated with traits of interest has provided an opportunity to initiate MABC for some traits in the SAT legume crops. Groundnut is probably the first among three legumes crops discussed in this article in which, MABC has been used to develop and release an improved variety. For instance, markers linked with root-knot nematode (*Meloidogyne arenaria*) resistance were used for introgression through the amphidiploid pathway into cultivated groundnut (Simpson et al., 2001). It was found relatively easy to identify linked markers due to sequence divergence between diploid and tetraploid genomes (Chu et al., 2007; Nagy et al., 2010) in groundnut. DNA fragment carrying nematode resistance was introgressed simultaneously selecting a recessive *AhFAD2B* allele (controls high ratio of oleic: linoleic acid (O/L)) using these linked markers as foreground selection markers (Chu et al., 2011). These efforts led to release of improved Tiftguard variety "Tifguard High O/L" (Chu et al., 2011). As SSR markers linked with resistance to leaf rust have also been identified in groundnut recently, MABC approach has been initiated to introgress a major QTL contributing 82.96% PV for leaf rust into the genetic background of three elite cultivars namely ICGV 91114, JL 24 and TAG 24. By using 2–3 rounds of backcrossing and selfing, BC_2F_3 and BC_3F_2 homozygous lines have been developed at ICRISAT.

In the case of chickpea, two major MABC projects are underway. Under Accelerated Crop Improvement Programme (ACIP) project sponsored by Department of Biotechnology, Government of India, MABC approach is being used for introgressing resistance to two races (*foc2* and *foc4*) independently and pyramiding of resistance to two races (*foc1* and *foc3*) for FW and two QTLs conferring resistance to AB. Jawaharlal Nehru Krishi Vishwavidyalaya (JNKVV), Mahatma Phule Krishi Vidyapeeth (MPKV) and Agricultural Research Station (ARS)-Gulbarga (all in India) are transferring resistance to *foc4* from WR 315 genotype in leading varieties namely JG 74, Phule G12 and Annigeri-1, respectively. Indian Institute of Pulses Research (IIPR), India is engaged in introgressing resistance to *foc2* from the resistant genotype Vijay in to an elite variety, Pusa 256. ICRISAT (India) on the other hand is pyramiding resistances for *foc1* and *foc3* from WR 315 and 2 QTLs for AB resistance from ILC 3279 line into C 214. At present, homozygous $BC_3F_{3:4}$ lines resistant for both FW and AB diseases in the preliminary evaluations are available. Different partner institutes have generated a range of backcross progenies followed by both foreground selection and background selection. In another initiative called as Tropical Legume-I (TL-I) of CGIAR Generation Challenge Programme in collaboration with Bill & Melinda Gates Foundation, significant efforts have been made to develop drought tolerant progenies ($BC_3F_{3:4}$) in the genetic background of JG11, a leading variety in India by transferring a genomic region containing several QTLs for drought tolerance traits from ICC 4958 genotype. Phenotypic evaluation of these lines is underway in India, Kenya and Ethiopia. Inspired by MABC work in JG11 genetic background, IIPR, Indian Agricultural Research Institute (IARI), Egerton University and Ethiopian Institute of Agricultural Research (EIAR) have also initiated MABC program for introgressing the drought tolerance genomic region from ICC 4958 in the leading varieties from their respective regions. While the work at IIPR and IARI is funded through DBT, Government of

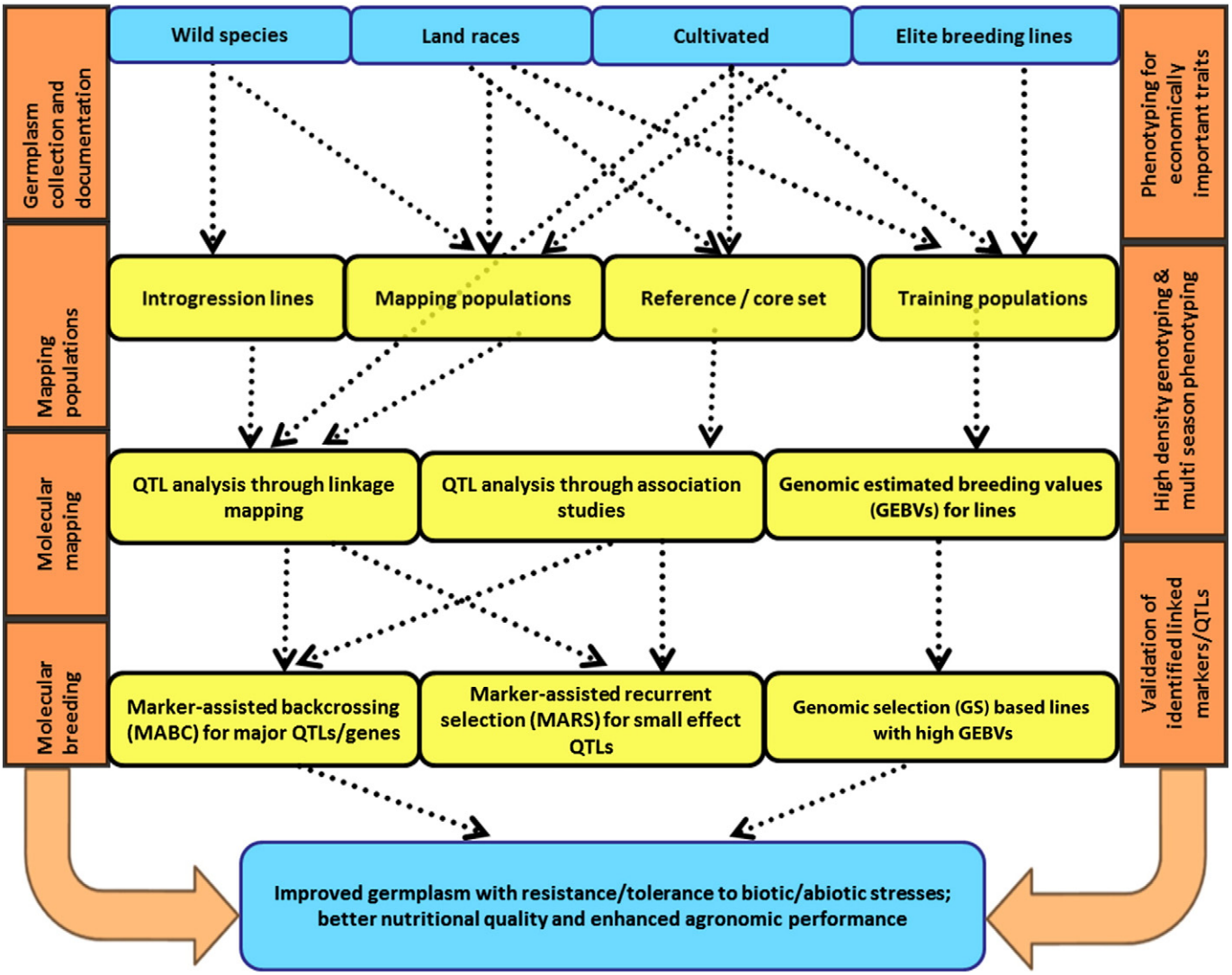

Fig. 3. Integrated genomics and breeding approaches for crop improvement in SAT legumes. This figure presents the use of integrated genomics and breeding approaches in systematic manner for genetics and breeding applications in three legume crops (chickpea, pigeonpea and groundnut). In the first instance, there is a need to utilize existing genetic resources based on phenotyping for target traits. Wild species accessions can be used to develop introgression libraries as well as developing inter-specific mapping populations while landraces and cultivars can be used to in developing mapping populations, reference/core sets and training populations, and elite breeding lines can be included in training populations. Introgression libraries, mapping populations and reference/core sets after genotyping and phenotyping for target traits can be used to identify QTLs or markers associated with traits using linkage or association mapping approaches and QTLs identified so, after validation, can be subsequently used in MABC and MARS approaches. On the other hand, training populations after genotyping and available phenotyping can be used to develop some models to estimate GEBVs. Based on these models, GEBVs may be calculated for progenies coming out from the crosses of lines either from training population or untested populations. Further crossing and selection of candidate lines based on GEBVs can be done in GS approach. By using a combination of MABC, MARS and GS approaches, improved germplasm with enhanced resistance/tolerance to biotic and abiotic stresses as well as with better nutritional quality and enhanced agronomic performance can be developed.

India, TLI-Phase 2 of CGIAR GCP is funding molecular breeding work at Egerton University and EIAR.

4.2. Marker-assisted recurrent selection (MARS)

MARS involves estimation of marker effects from genotyping F_2 or F_3 population and phenotyping F_2 derived F_4 or F_5 progenies, followed by two or three recombination cycles based on presence of marker alleles for small effect QTLs (Eathington et al., 2007). In the first step of MARS, *de novo* QTL identification is carried out initially, i.e. QTLs are identified in the breeding population itself, generally derived from good × good crosses. Subsequently, the lines carrying superior alleles for maximum QTLs are crossed to pyramid superior alleles in one genetic background. Recombined lines are then subjected to a final phenotypic screening to select the best lines for multi-location field testing to release them as varieties. MARS is particularly useful for capturing the several genomic regions especially to target more number of minor as well as major QTLs. Therefore, genetic gain achieved is higher by MARS as compared to the MABC program (Bernardo and Charcosset, 2006).

The recurrent-selection method is routinely used mainly in cross-pollinated crops like maize and, this process can be improved with the help of molecular markers; therefore, the process is called marker-assisted recurrent selection (MARS). While several multi-national companies have been using MARS in crops like maize and soybean, only a few public sector institutes have started to use MARS in crops likes wheat (Charmet et al., 2001), sorghum (Abdallah et al., 2009) and rice (Grenier et al., 2012). Some efforts have been initiated to use MARS in the case of chickpea also, for assembling favorable alleles for drought tolerance using ICCV 04112 × ICCV 93954 and ICCV 05107 × ICCV 94954 crosses. IARI and IIPR also have initiated MARS in

Table 4
List of markers associated with major QTL/genes for different traits in groundnut.

Traits studied	QTL/genes	Markers linked	PVE (%)	References
Agronomic & yield				
Flowering date	qFD02.1	AHGS2736-AHGS1251	19.5	Shirasawa et al. (2012)
Angle of branch	qAB05.1	AHGS2534-AHGS2622	11.9	Shirasawa et al. (2012)
Length of main stem	qLMS04.2	AHGS2155-AHGS3725	19.2	Shirasawa et al. (2012)
	qLMS05.2	AHGS2020-AHGS2450	15.7	Shirasawa et al. (2012)
Length of the longest branch	qLLB06.2	AhTE0697-Ah1TC3H7	21.1	Shirasawa et al. (2012)
	qLLB01.2	AHGS1813b-AhTE1016	14.2	Shirasawa et al. (2012)
Number of branches	qNB06.2	AhTE0967-AhTE0074	15.6	Shirasawa et al. (2012)
Weight of plant	qWP06.2	AhTE0697-Ah1TC3H7	11.8	Shirasawa et al. (2012)
Mature pod wt/plant	qWMP09.2	AHGS0422-AHGS2635	28.1	Shirasawa et al. (2012)
Length of pod	qPL05.1	AhTE0601-AHGS1413	28.2	Shirasawa et al. (2012)
	qPL06.2	AhTE0745-AhTE0826	20.5	Shirasawa et al. (2012)
Pod thickness	qPT07.1	AHGS1803a-AhTE0025	21.7	Shirasawa et al. (2012)
Pod width	qPW07.1	AhTE0025-pPGPSeq2E6b	15.2	Shirasawa et al. (2012)
	qPW08.2	AHGS1286-AHGS2249	25.5	Shirasawa et al. (2012)
Pod constriction	qCP09.2	AHGS0362-AhTE0726	18.1	Shirasawa et al. (2012)
Seed weight	qWS08.2	AhTE0846-AhTE0974	19.1	Shirasawa et al. (2012)
Stem diameter	SD02	pPGPseq2G3–TC7A02	24.1	Liang et al. (2009)
Total dry weight (TDW)	Total DWWW09_AhIX	TC7E04–GM1949	22.39	Gautami et al. 2012a
Harvest index (HI)	HI Control 08_AhIX	GM1922–GM2050	40.1	Gautami et al. 2012a
Shoot dry weight (SDW)	ShDWWS08_AhVII	GM1979–GM1919	22.09	Gautami et al. 2012a
Haulm weight	HaulmWtWW08_IV	TC1D02–TC3E05	33.36	Ravi et al. (2011)
Biomass	ShootBiomass04_XI	GM1971b–Ah193	20.32	Ravi et al. (2011)
Canopy conductance	ISC04_IVa	19H03–PM418	22.24	Ravi et al. (2011)
Biotic stress				
Leaf rust	$QTL_{rust}01$	IPAHM103	55.2	Khedikar et al. (2010)
	$QTL_{R4\text{-}rust}01/QTL_{R5\text{-}rust}01$	GM2009–GM1536	82.27	Sujay et al. (2012)
	$QTL_{R4\text{-}rust}02$	GM1536–M2301/GM207	62.35	Sujay et al. (2012)
	$QTL_{R4\text{-}rust}03/QTL_{R5\text{-}rust}02$	IPAHM103–GM1954	82.96	Sujay et al., 2012
	$QTL_{R5\text{-}rust}03$	RN16F05–GM1988	29.02	Sujay et al. (2012)
Late leaf spot (LLS)	$QTL_{R4\text{-}LLS}01$	GM1573–pPGPSeq2D09	62.34	Sujay et al. (2012)
	$QTL_{R4\text{-}LLS}02/QTL_{R5\text{-}LLS}01$			Sujay et al. (2012)
	$QTL_{R4\text{-}LLS}02/QTL_{R5\text{-}LLS}01$	GM2009–GM1536	67.98	Sujay et al. (2012)
	$QTL_{R4\text{-}LLS}04/QTL_{R5\text{-}LLS}03$	IPAHM103–GM1954	–	Sujay et al. (2012)
	$QTL_{R4\text{-}LLS}04/QTL_{R5\text{-}LLS}03$	IPAHM103–GM1954	42.66	Sujay et al. (2012)
	$QTL_{R5\text{-}LLS}02$	GM2504–GM2746	22.46	Sujay et al. (2012)
Aspergillus flavus invasion	*Af01*	TC11H06–TC4H07	22.7	Liang et al. (2009)
Tomato spotted wilt virus (TSWV)	*qTSWV1*	IPAHM287	12.9	Qin et al. (2012)
	qTSWV2	Seq12F07	35.8	Qin et al. (2012)
Aphid vector of groundnut	QTL	M1-TTG/M-GAA1	76.16	Herselman et al. (2004)
Nematode resistance	*Rma*	S197, GM565	–	Chu et al. (2007), Nagy et al., (2010)
Oil and protein				
Protein content	QTL 1	TC2E05-TC3E02	10.2	Sarvamangala et al. (2011)
	QTL 1	TC6H03-TC11A04	10.7	Sarvamangala et al. (2011)
Oil content		IPAHM103-PM36	10.2	Sarvamangala et al. (2011)
High oleate trait	*FAD2A*, *FAD2B*	aF19/1056R, bF19/R1FAD	89.7	Chu et al. (2007, 2009), Shirasawa et al. (2012)

chickpea by using Pusa 372×JG130 and DCP92-3×ICCV 10 crosses. These efforts are expected to result in superior lines with enhanced drought tolerance.

4.3. Genomic selection (GS)

Genomic selection (GS) or genome wide selection (GWS) unlike MABC or MARS approaches targets identification of superior lines with higher breeding value in a breeding program based on genome-wide marker profile data. As breeding values are estimated using the genome-wide marker data, these are generally referred as genomic-estimated breeding values (GEBVs). In brief, GS employs two populations: (i) 'training population', that is generally comprised of breeding lines that were/ are in use in a breeding program and phenotyping data, not for some traits, but for overall performance (e.g. yield and yield components) are available across the environments, and (ii) 'candidate population', which is generally being used currently by breeders. This population may be derived from the parental lines that are present in the training population. In the first step of GS, all the individuals of the training population are genotyped with large number of markers by considering linkage disequilibrium (LD) in the breeding germplasm collection. Based on historical phenotyping data and genotyping data, statistical models are developed for estimating GEBVs of the lines. Subsequently, marker genotyping data generated on the candidate population are used with the models and GEBVs are calculated for the progenies of the candidate population. Based on these GEBVs, the superior lines are selected for making the next crosses. Though not required, if candidate population is phenotyped, the phenotyping data obtained on this population together with the genotyping data can be used to train/strengthen the model developed based on the training populations. Anyway, the progenies from the selected lines (with higher GEBVs) of the candidate population are genotyped with the same set of markers. GEBVs calculated based on these marker data using the model provides another list of suitable progenies that can either be used for next cycle of crossing or depending on the skills of the crop breeders, can be selfed for field

evaluation in targeted environments and can be advanced for multi-location field trials (Fig. 3).

In summary, GS minimizes time-duration and cost by reducing the frequency of extensive phenotyping and bypasses the need for QTL mapping. GS can also reduce the selection cycle length of a breeding program that could take several seasons to develop reliable phenotypes. However, use of appropriate statistical model is very critical for estimating the GEBVs with higher precision. Among different models of GS, GEBVs predicted using either best linear unbiased prediction (BLUP) or Bayesian methods are more effective according to simulation studies (Bernardo and Yu, 2007). In addition to a Bayesian method, Bayes B, another method called wBSR (weighed Bayesian Shrinkage Regression) which reduces computational burden on MCMC-based Bayesian methods is considered to be a method of choice for genomic selection (Takeshi and Hiroyoshi, 2010). Several studies considered prediction using GS models, for instance Crossa et al. (2010) used two data sets including a historical wheat phenotypic data from trials evaluated in ten environments and another data set pertaining to maize for two diseases (*Exserohilum turcicum* and *Cercospora zeae-maydis*) from five environments. In both the cases models used marker data and a gain in the predictive ability was observed. Other groups including University of Oulu (Karkkainen and Silanpaa, 2012), Cornell University (Jannink et al., 2010), University of Minnesotta (Bernardo and Yu (2007), Hohenheim University (Piepho, 2009) also developed statistical models and/or pursued applications of GS in breeding of some major crops like maize, wheat, etc. Though GS has not been used in any legume species at present, due to availability of: (i) historical phenotyping data on several breeding lines (that can be used for training population), (ii) big linkage disequilibrium (LD) blocks in breeding populations, and (iii) genome wide marker genotyping system like DArT and SNP markers, GS seems to be a deployable approach in coming future at least in chickpea and groundnut.

4.4. Advanced backcross QTL analysis based breeding (AB-breeding)

All above-mentioned molecular breeding approaches (MABC, MARS and GS) are useful only when the superior alleles for the trait of interest are available in the breeding germplasm collection i.e., in primary gene pool. However, presence of genetic variability for a particular trait may not be available in primary gene pool such as resistance to pod borer in the case of chickpea and pigeonpea. In such cases, breeders need to utilize the potential of wild relatives that are considered reservoirs of superior alleles for traits that might have been lost during domestication and breeding. However, this is not a straight forward approach as most breeders are reluctant to use the wild relatives for transferring traits of interest from wild relatives to the cultivars because of not having efficient tracking for desired and non-desired alleles in breeding lines. To solve above problem, advanced-backcross QTL based breeding (AB-breeding) approach is the most suitable for introducing novel alleles from wild relatives to the cultivated species in a controlled manner. In AB-breeding approach, a selected wild species is backcrossed to a cultivar or a variety and then, selection is imposed in segregating BC_2F_2 or in BC_2F_3 population to identify and preserve individuals with desirable traits in the population. Both genotyping and phenotypic data are generated with this segregating BC_2F_2 or BC_2F_3 and, these data sets will be subjected to QTL analysis to identify QTL, QTL associated markers and, also to check whether any of these QTL are involved in trait improvement in the progenies that are preserved. Therefore, AB-QTL strategy involves the parallel discovery and transfer of desired QTL from an unadapted germplasm into selected breeding lines (Tanksley and Nelson, 1996). In addition, AB-QTL strategy postpones the QTL mapping up to BC_2 or BC_3 generations to avoid problems associated with incompatibility and pollen fertility in the initial backcross populations as well as to ensure maximum genome recovery from the recurrent parent. AB-breeding has been initially practiced in a vegetable crop like tomato, where crosses between wild tomato species and elite tomato lines were generated and QTLs for various fruit characters were identified and introgressed successfully (Fulton et al., 2000). The precision of QTL identification increases with a backcross population like BC_2 or BC_3 and, it offers adequate statistical power and ensures sufficient similarity to the recurrent parent. In addition, it also provides an opportunity to select for QTL-near isogenic lines (NILs) in a short time span. Using QTL-NILs, the QTL effects can be established and NILs may serve either as improved varieties or as parents for use in hybridization programs and for studies related to heterosis. In the case of chickpea, attempts were made for making wide crosses using cultivated and wild chickpea (*C. arietinum*×*C. reticulatum*) which resulted in selection of progenies with increased seed yield (Jaiswal et al., 1986). Similarly, Singh and Ocampio (1997) identified transgressive segregation for agronomic traits in an F_2 population of a cross, *C. arietinum*×*C. reticulatum*. These reports offer some opportunities in exploiting genetic variation present in wild species of chickpea and hence, fresh initiatives has been initiated at ICRISAT to introgress stress resistance through AB-breeding approach (N Mallikarjuna, ICRISAT, personal communication).

Realizing the scope for AB-breeding in improvement of pigeonpea, initiatives have been taken at ICRISAT to develop two backcross populations (ICPL 87119×ICPW 29 and ICPL 87119×ICPW 12) for AB-QTL analysis and their subsequent use in AB-breeding. ICPW 29 is an accession of *C. cajanifolius* species and ICPW 12 is an accession of *C. acutifolius* species. At present, for both the crosses, BC_2F_3 seeds have been generated for multilocation phenotyping and selection.

In groundnut, tetraploidization event restricted the sharing of genomic regions between wild and cultivated groups due to difference in ploidy levels, which has created a serious genetic bottleneck i.e., narrow genetic base. Though conventional approaches were used for attempting wide crosses through different ways such as use of autotetraploids and allotetraploids, these efforts seriously posed problems of fertility barrier, linkage drag and, a great difficulty in tracking introgressed alien genomic regions (see Bertioli et al., 2011). Of the above three important barriers, at least the later two (linkage drag and tracking of alien genomic regions) can be efficiently handled by integrating genomics into routine breeding programs to diversify the narrow primary gene pool of groundnut. AB-breeding can help in tracking alien genomic regions and hence, the linkage drag can easily be taken care of. Two major studies by Simpson et al. (1993) and Fa'vero et al. (2006) reported development of three amphiploids using a rage of wild AA and BB genome species like *A. cardenasii*, *A. diogoi* and *A. batizocoi*, *A. ipaensis*, *A. duranensis*, *A.gregoryi* and *A. linearifolium*. More recently, in order to diversify the primary gene pool and conduct AB-QTL analysis, ICRISAT has developed a set of 17 amphiploid and autotetraploid groundnuts (Mallikarjuna et al., 2011). Furthermore, two AB-QTL mapping populations namely ICGV 91114 (cultivated)×ISATGR 1212 (*A. duranensis* ICG 8123×*A. ipaensis* ICG 8206, synthetic amphidiploid) and ICGV 87846 (cultivated)×ISATGR 265-5A (*A. kempff-mercadoi* ICG 8164×*A. hoehnei* ICG 8190, synthetic amphidiploid) have been developed (Mallikarjuna et al., 2011). These populations are segregating for several biotic, abiotic and agronomic traits. A subset of 183 and 184 BC_2F_1 individuals, respectively have also been genotyped with DArT markers to construct genetic maps. These two populations are planned to be phenotyped for several economical traits in multiple locations and seasons. A successful effort for genome-wide segment introgressions from a synthetic amphidiploid (*A. duranensis*×*A. ipaënsis*) to a cultivated variety (Fluer 11) using molecular markers has already been reported (Foncéka et al., 2009). The backcross BC_1F_1 and BC_2F_1 lines carrying the wild genomic segments with maximum recurrent parent genomic regions provided optimal distribution of the synthetic genome introgressions (Foncéka et al., 2009).

Keeping in view, the low genetic diversity in all the three legume crops, this approach is required urgently for diversifying the primary

gene pool with favorable alleles to enhance the chances of further crop improvement. Availability of large number of markers in recent years has ensured limiting linkage drag through stringent background selection and tracking the presence of non-desirable genomic region from the wild relatives.

5. Summary and outlook

Until 2005, there was a shortage of genomic resources in these three SAT legume crops and therefore these crops were often referred to as 'orphan legume crops'. Nevertheless, the collaborative and coordinated efforts of the legume community made during the last 5 years, supplemented with generous financial support from several agencies, however, contributed to development of large-scale genomic resources in these crops. As a result, these crops are no longer 'orphan legumes' and have become 'genomic resource rich' crops (Varshney et al., 2009a, 2010b). These resources have also been used to understand the genetics of traits of several traits and as a result, approaches like MABC, MARS and AB-breeding are being used in these crops. GS seems to be a potential approach to be used very soon in chickpea and groundnut. While genome sequence has become available in pigeonpea, molecular breeding approaches have not yet been initiated as it may take more efforts to identify major QTLs for traits like FW, SMD and fertility restoration.

As genome sequence is expected to become available soon for chickpea and groundnut also, all the three legume crops mentioned in this article will have many more opportunities for practicing GAB. Genome sequence will facilitate re-sequencing of breeding populations, germplasm collections in faster and probably cheaper manner. In addition, availability of genome sequence will enable easy detection of variation at nucleotide level even among closely related parental lines and enhance exploitation of this variation for trait improvement in the breeding program. Analysis of genome-wide allelic data with the phenotyping data on germplasm collections will provide the alleles and haplotypes associated with the traits. These advances will also encourage the legume breeders to develop specialized populations like nested association mapping (NAM) populations or multi-parents advanced generation intercross (MAGIC) populations. These populations may not be just useful for fine mapping of traits but also in the development of lines with enhanced genetic diversity (in the case of MAGIC populations) which is very much required in these low-diversity species. In this case, AB-population will also be very helpful to enhance genetic base of the legumes.

Once genome-wide allelic and haplotype data available on at least leading germplasm including breeding lines, varieties or segregating populations and haplotype-trait association are established, it may be possible for legume breeders to undertake breeding-by-design. Rich databases of genome sequences/haplotypes, phenotypes, marker-trait associations may facilitated legume breeders to select the parental lines and consider different crossing schemes, so that superior lines with enhanced resistance to diseases and tolerance to abiotic stresses with other market quality traits can be generated. It can be anticipated that coming years will be more exciting for integrating GAB tools and approaches in conventional breeding programs. While generating sequence or genotyping data is expected to be trivial, the legume scientists need to work on precise and cost-effective phenotyping and developing the decision support tools and breeders-friendly databases to ensure undertaking integrated breeding approaches for crop improvement.

Acknowledgements

The work presented in this article is a contribution from several research projects sponsored by CGIAR Generation Challenge Programme (Theme Leader Discretionary Grant), The Bill & Melinda Gates Foundation (Tropical Legumes I & II), the Department of Biotechnology (Centre of Excellence and Accelerated Crop Improvement projects), the Ministry of Science & Technology (Indo-German Science & Technology Cooperation and Australia–India Science & Research Foundation projects) and the Ministry of Agriculture (National Fund, Indian Council of Agricultural Research) of Government of India. Thanks are also due to several colleagues at ICRISAT and partners in collaborating centers engaged in genomics and molecular breeding research in SAT legumes.

References

Abdallah AA, Ali AM, Geiger HH, Parzies HK. Marker-assisted recurrent selection for increased out crossing in *Caudatum*–race Sorghum. International Conference on Applied Biotechnology (ICAB), 28–30th September; 2009, Sudan; 2009. [Abstract no. 04].

Abu-Salem FM, Abou EA. Physico-chemical properties of tempeh produced from chickpea seeds. Arab J American Sci 2011;7:107–18.

Anbessa Y, Taran B, Warkentin TD, Tullu A, Vandenberg A. Genetic analyses and conservation of QTL for *Ascochyta* blight resistance in chickpea. Theor Appl Genet 2009;119: 757–65.

Anuradha C, Gaur PM, Pande S, Kishore K, Ganesh GM, Kumar J, et al. Mapping QTL for resistance to *botrytis* grey mould in chickpea. Euphytica 2011;182:1–9.

Argout X, Salse J, Aury J-M, Guiltinan MJ, Droc G, Gouzy J, et al. The genome of *Theobroma cacao*. Nat Genet 2011;43:101–8.

Arumuganathan K, Earle ED. Nuclear DNA content of some important plant species. Plant Mol Biol 1991;9:208–18.

Aryamanesh N, Nelson MN, Yan G, Clarke HJ, Siddique KHM. Mapping a major gene for growth habit and QTLs for *Ascochyta* blight resistance and flowering time in a population between chickpea and *Cicer reticulatum*. Euphytica 2010;173:307–19.

Bakht J, Bano A, Dominy P. The role of abscisic acid and low temperature in chickpea (*Cicer arietinum*) cold tolerance. II. Effects on plasma membrane structure and function. J Exp Bot 2006;57:3707–15.

Bernardo R, Charcosset A. Usefulness of gene information in marker-assisted recurrent selection: a simulation appraisal. Crop Sci 2006;46:614–21.

Bernardo R, Yu J. Prospects for genome wide selection for quantitative traits in maize. Crop Sci 2007;47:1082–90.

Bertioli DJ, Seijo G, Freitas FO, Valls JFM, Leal-Bertioli SCM, Moretzsohn MCM. An overview of peanut and its wild relatives. Plant Genetic Resour 2011;9:134–49.

Bohra A, Dubey A, Saxena RK, Penmetsa RV, Poornima KN, Kumar N, et al. Analysis of BAC-end sequences (BESs) and development of BES-SSR markers for genetic mapping and hybrid purity assessment in pigeonpea (*Cajanus* spp.). BMC Plant Biol 2011;11:56.

Bohra A, Saxena RK, Gnanesh BN, Saxena KB, Byregowda M, Rathore A, et al. An intra-specific consensus genetic map of pigeonpea (*Cajanus cajan* (L.) Millspaugh) derived from six mapping populations. Theor Appl Genet 2012;125:1325–38.

Charmet G, Robert N, Perretant MR, Gay G, Sourdille P, Groos C, et al. Marker assisted recurrent selection for cumulating QTLs for bread-making related traits. Euphytica 2001;119:89–93.

Choudhary P, Khanna SM, Jain PK, Bharadwaj C, Kumar J, Lakhera PC, et al. Genetic structure and diversity analysis of the primary gene pool of chickpea using SSR markers. Genet Mol Res 2011;11:891–905.

Choudhary S, Gaur R, Gupta S, Bhatia S. EST-derived genic molecular markers: development and utilization for generating an advanced transcript map of chickpea. Theor Appl Genet 2012;124:1449–62.

Chu Y, Holbrook CC, Timper P, Ozias-Akins P. Development of a PCR-based molecular marker to select for nematode resistance in peanut. Crop Sci 2007;47:841–5.

Chu Y, Holbrook CC, Ozias-Akins P. Two alleles of *ahFAD2B* control the high oleic acid trait in cultivated peanut. Crop Sci 2009;49:2029–36.

Chu Y, Wu CL, Holbrook CC, Tillman BL, Person G, Ozias-Akins P. Marker-assisted selection to pyramid nematode resistance and the high oleic trait in peanut. Plant Genome 2011;4:110–7.

Cobos M, Fernandez M, Rubio J, Kharrat M, Moreno M, Gil J, et al. A linkage map of chickpea (*Cicer arietinum* L.) based on populations from Kabuli × Desi crosses: location of genes for resistance to *Fusarium* wilt race 0. Theor Appl Genet 2005;110:1347–53.

Cobos MJ, Winter P, Kharrat M, Cubero JI, Gil J, Millan T, et al. Genetic analysis of agronomic traits in a wide cross of chickpea. Field Crop Res 2009;111:130–6.

Crossa J, Perez P, Campos GD, Mahuku G, Dreisigacker S, Magorokosho C. Genomic selection and prediction in plant breeding. J Crop Improv 2010;25:1-23.

Dhanasekar P, Dhumal KH, Reddy KS. Identification of RAPD marker linked to plant type gene in pigeonpea. Indian J Biotech 2010;9:58–63.

Dubey A, Farmer A, Schlueter J, Cannon SB, Abernathy B, Tuteja R, et al. Defining the transcriptome assembly and its use for genome dynamics and transcriptome profiling studies in pigeonpea (*Cajanus cajan* L. Millsp.). DNA Res 2011;18:153–64.

Dutta S, Kumawat G, Singh BP, Gupta DK, Singh S, Dogra V, et al. Development of genic-SSR markers by deep transcriptome sequencing in pigeonpea (*Cajanus cajan* (L.) Millspaugh). BMC Plant Biol 2011;11:17.

Eathington SR, Crosbie TM, Edwards MD, Reiter RS, Bull JK. Molecular markers in a commercial breeding program. Crop Sci 2007;47:S154–63.

Fa'vero AP, Simpson CE, Valls JFM, Vello NA. Study of the evolution of cultivated peanut through crossability studies among *Arachis ipaensis*, *A. duranensis*, and *A. hypogaea*. Crop Sci 2006;46:1546–52.

FAO. Food and Agricultural Organization of the United Nation, FAO Statistical Database. http://faostat.fao.org2012. [accessed on 28th June 2012].
Feng S, Wang X, Zhang X, Dang PM, Holbrook CC, Culbreath AK, et al. Peanut (*Arachis hypogaea*) expressed sequence tag project: progress and application. Comp Funct Genom 2012. http://dx.doi.org/10.1155/2012/373768.
Flowers TJ, Gaur PM, Gowda CLL, Krishnamurthy L, Srinivasan S, Siddique KHM, et al. Salt sensitivity in chickpea. Plant Cell Environ 2010;33:490–509.
Foncéka D, Hodo-Abalo T, Rivallan R, Faye I, Sall MN, Ndoye O, et al. Genetic mapping of wild introgressions into cultivated peanut: a way toward enlarging the genetic basis of a recent allotetraploid. BMC Plant Biol 2009;9:103.
Fulton TM, Grandillo S, Beck-Bunn T, Fridman E, Frampton A, Lopez J, et al. Advanced backcross QTL analysis of a *Lycopersicon esculentum*×*Lycopersicon parviflorum* cross. Theor Appl Genet 2000;100:1025–42.
Ganapathy KN, Byre Gowda M, Venkatesh SC, Ramachandra R, Gnanesh BN, Girish G. Identification of AFLP markers linked to sterility mosaic disease in pigeonpea *Cajanus cajan* (L.) Millsp. Int J Integr Biol 2009;7:145–9.
Garg R, Patel RK, Tyagi AK, Jain M. *De novo* assembly of chickpea transcriptome using short reads for gene discovery and marker identification. DNA Res 2011a;18:53–63.
Garg R, Patel RK, Jhanwar S, Priya P, Bhattacharjee A, Yadav G, et al. Gene discovery and tissue-specific transcriptome analysis in Chickpea with massively parallel pyrosequencing and web resource development. Plant Physiol 2011b;156: 1661–78.
Gaur R, Sethy NK, Choudhary S, Shokeen B, Gupta V, Bhatia S. Advancing the STMS genomic resources for defining new locations on the intraspecific genetic linkage map of chickpea (*Cicer arietinum* L.). BMC Genomics 2011;12:117.
Gautami B, Fonceka D, Pandey MK, Morezsohn MC, Sujay V, Qin H, et al. An international reference consensus genetic map with 897 marker loci based on 11 mapping populations for tetraploid groundnut (*Arachis hypogaea* L.). PLoS One 2012a;7: e41213.
Gautami B, Pandey MK, Vadez V, Nigam SN, Ratnakumar P, Krishnamurthy L, et al. Quantitative trait locus analysis and construction of consensus genetic map for drought tolerance traits based on three recombinant inbred line populations in cultivated groundnut (*Arachis hypogaea* L.). Mol Breed 2012b;30:757–72.
Gnanesh BN, Bohra A, Sharma M, Byregowda M, Pande S, Wesley V, et al. Genetic mapping and quantitative trait locus analysis of resistance to sterility mosaic disease in pigeonpea [*Cajanus cajan* (L.) Millsp.]. Field Crop Res 2011;123:53–61.
Gowda SJM, Radhika PN, Kadoo Y, Mhase LB, Gupta VS. Molecular mapping of wilt resistance genes in chickpea. Mol Breed 2009;24:177–83.
Grenier C, Chatel MH, Ospina Y, Cao T, Guimaraes EP, Martinez CP, et al. Population improvement through recurrent selection in rice. Prospects for maker assisted recurrent selection and genome-wide selection. Plant and Animal Genome XX, January 14–18, 2012, San Diego, USA. Abstract no. W011; 2012.
Guimaraes PM, Garsmeur O, Proite K, Leal-Bertioli SC, Seijo G, Chaine C, et al. BAC libraries construction from the ancestral diploid genomes of the allotetraploid cultivated peanut. BMC Plant Biol 2008;8:14.
Guimarães PM, Brasileiro ACM, Leal-Bertioli SCM, Pappas G, Togawa R, Bonfim O, et al. Comparative 454 pyrosequencing of transcripts from two wild *Arachis* genotypes under biotic and abiotic stress. Plant and Animal Genome XIX Conference, 15–19th Jan 2011, San Diego, USA, Poster no. W362; 2011.
Gujaria N, Kumar A, Dauthal P, Dubey A, Hiremath P, BhanuPrakash A, et al. Development and use of genic molecular markers (GMMs) for construction of a transcript map of chickpea (*Cicer arietinum* L.). Theor Appl Genet 2011;122:1577–89.
Gupta PK, Varshney RK. The development and use of microsatellite markers for genetic analysis and plant breeding with emphasis on bread wheat. Euphytica 2000;13: 163–85.
Gupta PK, Kumar J, Mir RR, Kumar A. Marker-assisted selection as a component of conventional plant breeding. Plant Breed Rev 2010;33:145–217.
Herselman LR, Thwaites FM, Kimmins B, Courtois PJA, Merwe VD, Seal SE. Identification and mapping of AFLP markers linked to peanut (*Arachis hypogaea* L.) resistance to the aphid vector of groundnut rosette disease. Theor Appl Genet 2004;109:1426–33.
Hiremath PJ, Farmer A, Cannon SB, Woodward J, Kudapa H, Tuteja R, et al. Large-scale transcriptome analysis in chickpea (*Cicer arietinum* L.), an orphan legume crop of the semi-arid tropics of Asia and Africa. Plant Biotech J 2011;9:922–31.
Hiremath PJ, Kumar A, Penmetsa RV, Farmer A, Schlueter JA, Chamarthi SK, et al. Large-scale development of cost-effective SNP marker assays for diversity assessment and genetic mapping in chickpea and comparative mapping in legumes. Plant Biotech J 2012;10:716–32.
Hong Y, Chen X, Liang X, Liu H, Zhou G, Li S, et al. A SSR-based composite genetic linkage map for the cultivated peanut (*Arachis hypogaea* L.) genome. BMC Plant Biol 2010;10:17.
Hospital F. Selection in backcross programme. Phil Trans R Soc. 2005;360:1503–11.
Huang S, Li R, Zhang Z, Li L, Gu X, Fan W, et al. The genome of the cucumber, *Cucumis sativus* L. Nat Genet 2009;41:1275–81.
Iruela M, Rubio J, Barro FM, Cubero JI, Milla T, Gil J. Detection of two quantitative trait loci for resistance to *Ascochyta* blight in an intra-specific cross of chickpea (*Cicer arietinum* L.): development of SCAR markers associated with resistance. Theor Appl Genet 2006;112:278–87.
Iruela M, Castro P, Rubio J, Cubero JI, Jacinto C, Milla T, et al. Validation of a QTL for resistance to *Ascochyta* blight linked to resistance to fusarium wilt race 5 in chickpea (*Cicer arietinum* L.). Eur J Plant Pathol 2007;119:29–37.
Jaillon O, Aury JM, Noel B, Policriti A, Clepet C, Casagrande A, et al. The grapevine genome sequence suggests ancestral hexaploidization in major angiosperm phyla. Nature 2007;49:463–8.
Jaiswal HK, Singh BD, Singh AK, Singh RM. Introgression of genes for yield and yield traits from *C. reticulatum* into *C. arietinum*. International Chickpea Newsletter 1986;14:5–8.
Jannink J-L, Lorenz AJ, Iwata H. Genomic selection in plant breeding: from theory to practice. Brief Funct Genomics 2010;9:166–77.
Jhanwar S, Priya P, Garg R, Parida SK, Tyagi AK, Jain M. Transcriptome sequencing of wild chickpea as a rich resource for marker development. Plant Biotechnol J 2012;10:690–702.
Kannaiyan J, Nene YL, Reddy MV, Ryan JG, Raju TN. Prevalence of pigeonpea disease and associated crop losses in Asia Africa and America. Trop Pest Man 1984;30: 62–71.
Karkkainen HP, Sillanpaa MJ. Back to basics for Bayesian model building in genomic selection. Genetics 2012; 191:969–987.
Kassa MT, Penmetsa RV, Carrasquilla-Garcia N, Sarma BK, Datta S, Upadhyaya HD, et al. Genetic patterns of domestication in pigeonpea (*Cajanus cajan* (L.) Millsp.) and wild *Cajanus* relatives. PLoS One 2012;7:e39563.
Khedikar YP, Gowda MV, Sarvamangala C, Patgar KV, Upadhyaya HD, Varshney RK. A QTL study on late leaf spot and rust revealed one major QTL for molecular breeding for rust resistance in groundnut (*Arachis hypogaea* L.). Theor Appl Genet 2010;121: 971–84.
Kilian A. DArT-based whole genome profiling and novel information technologies in support system of modern breeding of groundnut. Proc: 3rd international conference for *Arachis* Through Genomics and Biotechnology (AAGB), 4–8th November; 2008. Hyderabad, India; 2008.
Knoll JE, Ramos ML, Zeng Y, Holbrook CC, Chow M, Chen S, et al. TILLING for allergen reduction and improvement of quality traits in peanut (*Arachis hypogaea* L.). BMC Plant Biol 2011;11:81.
Kotresh H, Fakrudin B, Punnuri S, Rajkumar B, Thudi M, Paramesh H. Identification of two RAPD markers genetically linked to a recessive allele of a *Fusarium* wilt resistance gene in pigeonpea (*Cajanus cajan* (L.) Millsp.). Euphytica 2006;149:113–20.
Kudapa H, Bharti AK, Cannon SB, Farmer AD, Mulaosmanovic B, Kramer R, et al. A comprehensive transcriptome assembly of pigeonpea (*Cajanus cajan* L. Millsp.) using Sanger and second-generation sequencing platforms. Mol Plant 2012;5:1020–8.
Liang X, Zhou G, Hong Y, Chen X, Liu H, Li S. Overview of research progress on peanut (*Arachis hypogaea* L.) host resistance to aflatoxin contamination and genomics at the Guangdong Academy of Agricultural Sciences. Peanut Sci 2009;36:29–34.
Lorieux M. CSSL Finder: a free program for managing introgression lines. http://mapdisto.free.fr/2005.
Macedo SE, Moretzsohn MC, Leal-Bertioli SCM, Alves DMT, Gouvea EG, Azevedo VCR, et al. Development and characterization of highly polymorphic long TC repeat microsatellite markers for genetic analysis of peanut. BMC Res Notes 2012;5:86.
Madrid E, Rubiales D, Moral A, Moreno MT, Millan T, Gil J, et al. Mechanism and molecular markers associated with rust resistance in a chickpea interspecific cross (*Cicer arietinum*×*Cicer reticulatum*). Eur J Plant Pathol 2008;121:43–53.
Mallikarjuna N, Senapathy S, Jadhav DR, Saxena KB, Sharma HC, Upadhyaya HD, et al. Progress in the utilization of *Cajanus platycarpus* (Benth.) Maesen in pigeonpea improvement. Plant Breed 2011;130:507–14.
Marley PS, Hillocks RJ. Effect of root-knot nematodes (*Meloidogyne* spp.) on *Fusarium* wilt in pigeonpea (*Cajanus cajan* (L.) Millspaugh). Field Crop Res 1996;46:15–20.
Millan T, Rubio J, Iruela M, Daly K, Cubero JI, Gil J. Markers associated with *Ascochyta* blight resistance in chickpea and their potential in marker-assisted selection. Field Crop Res 2003;84:373–84.
Millan T, Winter P, Ju¨ngling R, Gil J, Rubio J, Cho S, et al. A consensus genetic map of chickpea (*Cicer arietinum* L.) based on 10 mapping populations. Euphytica 2010;175:175–89.
Miller CD, Branthoover B, Sekiguchi N, Deming H, Bauer A. Vitamin value of foods used in Hawaii. Hawaii Agri Exp Station Tech Bull 1956;30:303–13.
Nagy E, Chu Y, Guo Y, Khanal S, Tang S, Li Y, et al. Recombination is suppressed in an alien introgression in peanut harboring *Rma*, a dominant root-knot nematode resistance gene. Mol Breed 2010;26:357–70.
Nayak SN, Zhu H, Varghese N, Datta S, Choi HK, Horres R, et al. Integration of novel SSR and gene-based SNP marker loci in the chickpea genetic map and establishment of new anchor points with *Medicago truncatula* genome. Theor Appl Genet 2010;120: 1415–41.
Nene YL, Reddy MV. Chickpea diseases and their control. In: Saxena MC, Singh B, editors. Oxon, UK: The Chickpea CAB International; 1987. p. 233–70.
Pandey MK, Monyo E, Ozias-Akins P, Liang X, Guimarães P, Nigam SN, et al. Advances in *Arachis* genomics for peanut improvement. Biotechnol Adv 2012a;30:639–51.
Pandey MK, Gautami B, Jayakumar T, Sriswathi M, Upadhyaya HD, Gowda MVC, et al. Highly informative genic and genomic SSR markers to facilitate molecular breeding in cultivated groundnut (*Arachis hypogaea* L.). Plant Breed 2012b;131: 139–47.
Peleman JD, Voort JRVD. Breeding by design. Trends Plant Sci 2003;8:330–4.
Piepho HP. Ridge regression and extensions for genome-wide selection in maize. Crop Sci 2009;49:1165–76.
Qin H, Feng S, Chen C, Knapp S, Culbreadth A, He G, et al. An integrated genetic linkage map of cultivated peanut (*Arachis hypogaea* L.) constructed from two RIL populations. Theor Appl Genet 2012;124:653–64.
Radhika P, Gowda SJ, Kadoo NY, Mhase LB, Jamadagni BM, Sainani MN, et al. Development of an integrated intraspecific map of chickpea (*Cicer arietinum* L.) using two recombinant inbred line populations. Theor Appl Genet 2007;115:209–16.
Raju NL, Gnanesh BN, Lekha P, Jayashree B, Pande S, Hiremath PJ, et al. The first set of EST resource for gene discovery and marker development in pigeonpea (*Cajanus cajan* L. Millsp.). BMC Plant Biol 2010;10:45.
Rakshit S, Winter P, Tekeoglu M, Munoz J, Pfaff T, Benko-Iseppon AM, et al. DAF marker tightly linked to a major locus for *Ascochyta* blight resistance in chickpea (*Cicer arietinum* L.). Euphytica 2003;132:23–30.
Ratnaparkhe MB, Gupta VS, Pigeonpea. In: Kole C, editor. Genome mapping and molecular breeding in plants: pulses, sugar and tuber crops. Springer; 2007. p. 133–42.

Ravi K, Vadez V, Isobe S, Mir RR, Guo Y, Nigam SN, et al. Identification of several small main-effect QTLs and a large number of epistatic QTLs for drought tolerance related traits in groundnut (*Arachis hypogaea* L.). Theor Appl Genet 2011;122: 1119–32.

Rehman AU, Malhotra RS, Bett K, Tar'an B, Bueckert R, Warkentin TD. Mapping QTL associated with traits affecting grain yield in chickpea (*Cicer arietinum* L.) under terminal drought stress. Crop Sci 2011;51:450–63.

Ribaut JM, Hoisington D. Marker-assisted selection: new tools and strategies. Trends Plant Sci 1998;3:236–9.

Ribaut JM, Ragot M. Marker-assisted selection to improve drought adaptation in maize: the backcross approach, perspectives, limitations, and alternatives. J Exp Bot 2007;58:351–60.

Sarvamangala C, Gowda MVC, Varshney RK. Identification of quantitative trait loci for protein content, oil content and oil quality for groundnut (*Arachis hypogaea* L.). Field Crop Res 2011;122:49–59.

Saxena KB. Genetic improvement of pigeonpea—a review. Trop Plant Biol 2008;1:159–78.

Saxena RK, Saxena KB, Varshney RK. Application of SSR markers for molecular characterization of hybrid parents and purity assessment of ICPH 2438 hybrid of pigeonpea (*Cajanus cajan* (L.) Millspaugh). Mol Breeding 2010;26:371–80.

Saxena RK, Varshney RK, Kavi Kishor PB. Inheritance and marker detection for fertility restorers in pigeonpea. KG Heinrich-Böcking-Str. 6-8, 66121, Saarbrücken, Germany: LAP LAMBERT Academic Publishing GmbH & Co, ISBN: 978-3-8473-0692-4; 2011.

Saxena RK, Penmetsa RV, Upadhyaya HD, Kumar A, Carrasquilla-Garcia N, Schlueter JA, et al. Large-scale development of cost-effective single-nucleotide polymorphism marker assays for genetic mapping in pigeonpea and comparative mapping in legumes. DNA Res 2012;19:449–61.

Schmutz J, Cannon SB, Schlueter J, Ma J, Mitros T, Nelson W, et al. Genome sequence of the palaeopolyploid soybean. Nature 2010;463:178–83.

Shanower TG, Romeis J, Minja EM. Insect pests of pigeonpea and their management. Annu Rev Entomol 1999;44:77–96.

Sharma KD, Winter P, Kahl G, Muehlbauer FJ. Molecular mapping of *Fusarium oxysporum* f. sp. *ciceris* race 3 resistance gene in chickpea. Theor Appl Genet 2004;108:243-1248.

Sharma KD, Chen W, Muehlbauer FJ. Genetics of chickpea resistance to five races of Fusarium wilt and a concise set of race differentials for *Fusarium oxysporum* f. sp. ciceris. Plant Dis 2005;89:385–90.

Sharma HC, Varshney RK, Gaur PM, Gowda CLL. Potential for using morphological, biochemical, and molecular markers for resistance to insect pests in grain legumes. J Food Legumes 2008;21:211–7.

Shirasawa K, Koilkonda P, Aoki K, Hirakawa H, Tabata S, Watanabe M, et al. *In silico* polymorphism analysis for the development of simple sequence repeat and transposon markers and construction of linkage map in cultivated peanut. BMC Plant Biol 2012;12:80.

Simpson CE, Nelson SC, Starr J, Woodward KE, Smith OD. Registration of TxAG-6 and TxAG-7 peanut germplasm lines. Crop Sci 1993;33:1418.

Simpson CE, Krapovickas A, Valls JFM. History of *Arachis* including evidence of *A. hypogaea* L. progenitors. Peanut Sci 2001;28:78–9.

Singh KB, Ocampio B. Exploitation of wild *Cicer* species for yield improvement in chickpea. Theor Appl Genet 1997;95:418–23.

Singh KB, Reddy MV. Advances in disease-resistance breeding in chickpea. Adv Agron 1991;45:191–222.

Singh KB, Reddy MV. Improving chickpea yield by incorporating resistance to *Ascochyta* blight. Theor Appl Genet 1996;92:509–15.

Stalker HT. A new species in section *Arachis* of peanuts with a D genome. Am J Bot 1991;78: 630–7.

Sujay V, Gowda MVC, Pandey MK, Bhat RS, Khedikar YP, Nadaf HL, et al. Quantitative trait locus analysis and construction of consensus genetic map for foliar disease resistance based on two recombinant inbred line populations in cultivated groundnut (*Arachis hypogaea* L.). Mol Breed 2012;30:773–88.

Takeshi H, Hiroyoshi I. EM algorithm for Bayesian estimation of genomic breeding values. BMC Genet 2010;11:3.

Tanksley SD, Nelson JC. Advanced backcross QTL analysis: a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines. Theor Appl Genet 1996;92:191–203.

Thudi M, Bohra A, Nayak SN, Varghese N, Shah TM, Penmetsa RV, et al. Novel SSR markers from BAC-end sequences, DArT arrays and a comprehensive genetic map with 1,291 marker loci for chickpea (*Cicer arietinum* L.). PLoS One 2011;6:e27275.

Tuskan GA, Difazio S, Jansson S, Bohlmann J, Grigoriev I, Hellsten U, et al. The genome of black cottonwood, *Populus trichocarpa* (Torr. & Gray). Science 2006;313:1596–604.

Udupa SM, Baum M. Genetic dissection of pathotype-specific resistance to *Ascochyta* blight disease in chickpea (*Cicer arietinum* L.) using microsatellite markers. Theor Appl Genet 2003;106:1196–202.

Upadhyaya HD, Thudi M, Dronavalli N, Gujaria N, Singh S, Sharma S, et al. Genomic tools and germplasm diversity for chickpea improvement. Plant Genetic Resour 2011;9:45–58.

Vadez V, Krishnamurthy L, Thudi M, Anuradha C, Colmer TD, Turner NC, et al. Assessment of ICCV 23×JG 62 chickpea progenies shows sensitivity of reproduction to salt stress and reveals QTL for seed yield and yield components. Mol Breed 2012;30:9-21.

Varshney RK. Application of next generation sequencing and genotyping technologies to develop large scale genomic resources in SAT legume crops. In: Muralidharan K, Siddiq EA, editors. Genomics and crop improvement: relevance and reservations. Hyderabad, India: Acharya NG Ranga Agricultural University; 2011. p. 1-10.

Varshney RK, Hoisington DA, Upadhyaya HD, Gaur PM, Nigam SN, Saxena KB, et al. Molecular genetics and breeding of grain legume crops for the semi-arid tropics. In: Varshney RK, Tuberosa R, editors. Genomics-Assisted Crop Improvement, 2. Genomics Applications in Crops; 2007. p. 207–41.

Varshney RK, Close TJ, Singh NK, Hoisington DA, Cook DR. Orphan legume crops enter the genomics era. Curr Opin Plant Biol 2009a;12:1–9.

Varshney RK, Hiremath PJ, Lekha PT, Kashiwagi J, Balaji J, Deokar AA, et al. A comprehensive resource of drought-and salinity-responsive ESTs for gene discovery and marker development in chickpea (*Cicer arietinum* L). BMC Genomics 2009b;10:523.

Varshney RK, Nayak SN, May GD, Jackson SA. Next-generation sequencing technologies and their implications for crop genetics and breeding. Trends Biotechnol 2009c;27:522–30.

Varshney RK, Thudi M, May GD, Jackson SA. Legume genomics and breeding. Plant Breed Rev 2010a;33:257–304.

Varshney RK, Glaszmann JC, Leung H, Ribaut JM. More genomic resources for less-studied crops. Trends Biotechnol 2010b;28:452–60.

Varshney RK, Penmetsa RV, Dutta S, Kulwal PL, Saxena RK, Datta S, et al. Pigeonpea genomics initiative (PGI): an international effort to improve crop productivity of pigeonpea (*Cajanus cajan* L.). Mol Breed 2010c;26:393–408.

Varshney RK, Chen W, Li Y, Bharti AK, Saxena RK, Schlueter JA, et al. Draft genome sequence of pigeonpea (*Cajanus cajan*), an orphan legume crop of resource-poor farmers. Nat Biotechnol 2012a;30:83–9.

Varshney RK, Kudapa H, Roorkiwal M, Thudi M, Pandey MK, Saxena RK, et al. Advances in genomics research and molecular breeding applications in SAT legume crops by using next generation sequencing and high-throughput genotyping technologies. J Biosci 2012b;37:811–20.

Wang H, Penmetsa RP, Yuan M. Development and characterization of BAC-end sequence derived SSRs, and their incorporation into a new higher density genetic map for cultivated peanut (*Arachis hypogaea* L.). BMC Plant Biol 2012;12:10.

Whiteman PC, Norton BW. Alternative uses of pigeonpea. Proceed Int Workshop Pigeonpeas, 15–19 Dec 1980, 1. Patancheru, 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics; 1980. p. 365–78.

Yang S, Pang W, Ash G, Harper J, Carling J, Wenzl P, et al. Low level of genetic diversity in cultivated pigeonpea compared to its wild relatives is revealed by diversity arrays technology. Theor Appl Genet 2006;113:585–95.

Yang SY, Saxena RK, Kulwal PA, Ash GJ, Dubey A, Harper DI, et al. The first genetic map of pigeonpea based on diversity arrays technology (DArT) markers. J Genet 2011;90:103–9.

Young ND, Weeden NF, Kochert G. Genome mapping in legumes (Family *Fabaceae*). In: Paterson AH, editor. Genome mapping in plants. Texas: Landes Biomedical Press; 1996. p. 211–77.

Young ND, Debellé F, Oldroyd GE, Geurts R, Cannon SB, Udvardi MK, et al. The *Medicago* genome provides insight into the evolution of rhizobial symbioses. Nature 2011;480: 520–4.

Yüksel B, Paterson AH. Construction and characterization of a peanut HindIII BAC library. Theor Appl Genet 2005;111:630–9.